Hubert-Jean Ceccaldi · Ivan Dekeyser
Mathias Girault · Georges Stora *Editors*

Global Change: Mankind-Marine Environment Interactions

Proceedings of the 13th French-Japanese Oceanography Symposium

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First plenary conference, opening session of the 13th French-Japanese Oceanography Symposium (Colloque franco-japonais d'Océanographie)
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Prof. TAKAI Rikuo, Tokyo University of Marine Science and Technology and Prof. Yvon BERLAND signing of the agreement between the "Université de la Mediterranée" and "Tokyo University of Marine Science and Technology".
From the left, Dr. IZUMI Mitsuru, Vive-President, Director of the Offie of Liaison and Cooperative Research; Dr. TAKAI Rikuo, President, Pr. Roger GIUDICELLI, Vice-President; Prof Yvon BERLAND, President; M. BAMBA Masahiro, Consul général of Japan in Marseille.

Global Change: Mankind-Marine Environmental Interactions

- **Aquaculture**: resource preservation and harmonious management
- **Artificial Reefs**: enhancement and protection of natural environments
- **Biodiversity**: impact of human activities, invasive species, and protection of natural environments
- **Coastal Managements**: interactions between ports and natural environments, ecological and economical consequences
- **Observation of Marine Environment**: emergence of new technologies
- **Physical, Chemical, Biological and Biogeochemical Functioning:** natural and anthropic environments

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Foreword

Son Excellence Monsieur Iimura Yutaka
Ambassadeur du Japon en France
Le 13ème Colloque franco-japonais d'Océanographie s'est déroulé en 2008, l'année célébrant le 150ème anniversaire des relations diplomatiques entre nos deux pays.

Dès la signature du traité franco-japonais de paix, d'amitié et de commerce en 1858, le Japon s'est empreint de l'influence de la France. Notre pays a pu disposer des techniques et des savoirs français dans de très nombreux domaines visant à l'édification d'un Etat moderne tels que la pensée politique, le code civil, l'organisation de l'armée et de la police et l'industrie. La France a également été influencée par le Japon, bien que de façon plus modeste et essentiellement dans le domaine des arts et de la culture. La peinture de style Ukiyo-é et les poteries japonaises sont à l'origine du "Japonisme" qui a joué un rôle décisif dans la genèse de l'impressionnisme et de l'Art Nouveau.

Aujourd'hui il existe entre la France et le Japon des liens forts dans les relations commerciales, les investissements, les échanges entre collectivités locales, l'enseignement supérieur et la recherche, les sciences et les technologies. La coopération dans le domaine de l'océanographie a commencé il y a déjà plus de cinquante ans avec les recherches menées par le bathyscaphe français dans la Fosse du Japon. Les deux Sociétés d'Océanographie franco-japonaises ont largement contribué à la développer.

Je tiens à rendre hommage aux nombreuses personnes qui ont travaillé à l'organisation de ce colloque et apprécie leur passion sans laquelle cette manifestation n'aurait pas eu lieu. Enfin, je tiens à les féliciter du très grand succès qu'elle a rencontré.

M. Yutaka Iimura
Ambassadeur du Japon en France

Translation

The 13th France-Japan Colloquy in Oceanography took place in 2008, the year celebrating the 150th anniversary of the official diplomatic exchanges between our two countries.

Since the signing of the France-Japan Treaty of Peace, Friendship and Trade in 1858, Japan has been marked by French influence. Our country has been able to use French techniques and knowledge in numerous areas in order to edify a modern State, for example, in political thinking, developing a civilian code of justice, organization of the army and police, and in industry.

France has also been influenced by Japan, although more modestly and essentially in the domain of arts and culture. Paintings of Ukiyo-é and Japanese ceramics are linked to the origins of the "Japonisme," which played a decisive role in the genesis of impressionism and Art Nouveau.

Today, this relationship still exists between France and Japan, with strong links in commercial trade, investments, exchanges in the fields of local collectives, upper level education, science, and technology. The cooperation in the field of oceanography began more than 50 years ago with the research involving French bathyscaphs in the Japanese trench. The two Sociétés franco-japonaises d'Océanographie have broadly contributed to that development.

I am paying tribute to the numerous persons who have worked for the organization of the colloquy, and I appreciate their passion, without which that event should not been happened. Finally, I would like to congratulate them for the very large success obtained.

Monsieur le Professeur Fançois Gros
Secrétaire perpétuel honoraire de l'Académie des Sciences

Il m'est particulièrement agréable de rédiger la préface de cet ouvage qui regroupe les textes des communications présentées au cours du 13ème Colloque franco-japonais d'Océanographie. Cette importante réunion scientifique s'est déroulée à Marseille et à Paris entre le 8 et le 12 septembre 2008.

Ce Colloque couvre un champ continu de la recherche en sciences et technologies marines depuis les caractères physiques et chimiques des océans jusqu'aux aménagements du littoral, en passant par la biodiversité, l'aquaculture et les récifs artificiels.

L'Académie des Sciences est attentive à de telles activités qui renforcent les liens existant entre les océanographes français et leurs confrères du Japon. Certaines Fondations de l'Institut se sont d'ailleurs donné pour mission d'aider de telles initiatives comme l'aide apportée par M. le Chancelier Gabriel de Broglie. Le Comité consultatif conjoint franco-japonais que j'ai co-présidé de longues années considère aussi avec un grand intérêt les rencontres de ce niveau qui, grâce à l'établissement de relations personnelles entre chercheurs des deux pays, complètent parfaitement les relations établies entre les administrations et entre les organismes officiels des deux pays.

Ce colloque très ouvert donne surtout des perspectives vers des domaines réellement porteurs d'avenir, comme les biotechnologies marines, la biogéochimie, les technologies d'aquaculture, de nouvelles approches en biodiversité des populations marines, les nouveaux types de récifs artificiels et les tout récents appareillages permettant des observations très fines du milieu marin. Les plus hautes instances de chacun des deux pays – Ambassades, Ministères, Directions nationales et régionales de la recherche scientifique – ne s'y sont pas trompées en accordant leur soutien à cette manifestation. Des perspectives vers la médecine et vers la pharmacie sont déjà ouvertes en nutrition, santé, nouveaux médicaments, cosmétiques, pour ne prendre que quelques exemples. L'établissement, à cette occasion, d'un jumelage entre deux universités, française et japonaise, favorisera grandement le flux de ces échanges, bénéfiques aux deux partenaires. Il pourrait servir d'exemple à d'autres universités.

Aussi suis-je heureux de remercier les deux Sociétés franco-japonaises d'Océanographie, l'Université de la Méditerranée, le Centre d'Océanologie de Marseille et les Comités d'organisation qui, dans chacun des deux pays, ont mis sur pied cette intéressante et si productive manifestation. Elle aura des suites dans le prochain colloque au Japon dans un proche futur et demeurera, j'en suis convaincu, d'une grande fécondité dans un avenir plus lointain. Que les organisateurs, et notamment les Professeurs Hubert Jean Ceccaldi, Imawaki Shiro et Yagi Hiroki soient vivement loués pour cette belle réussite.

Prof. François Gros, Secrétaire perpétuel honoraire de l'Académie des Sciences

Translation

I am particularly glad to draw up this foreword of this book, gathering the texts of the communications presented during the 13th France-Japan Colloquy of Oceanography. This important scientific meeting was held in Marseille, then Paris, between September 8 and 12, 2008.

That meeting covered a continuous field of research in marine sciences and technologies, from physical and chemical characteristics of the oceans, to coastal management, and moving to biodiversity, aquaculture, and artificial reefs.

The French Academy of Sciences pays attention to such activities, as they reinforce the existing links between the French oceanographers and their Japanese colleagues. One of the founding principles of the Institut de France is to help to such initiatives, with the cooperation of Chancellor Gabriel De Broglie.

The joint France-Japan Consultative Committee, which I have co-chaired for many years, is also interested in such high-level meetings because they make the personal relationships easier between researchers of both countries, completing perfectly, by that way, the relationships established between administrations and official bodies of the two countries.

This meeting provided above all perspectives on scientific fields that are very promising for the future, such as marine biotechnologies, biogeochemistry, technologies of aquaculture, new approaches to the domain of biodiversity of marine populations, new models of artificial reefs, and very recent apparatus leading to very fine observations of the marine surroundings. The highest official bodies in each country – Ministries, Embassies, National and regional political and administrative bodies in scientific research – have taken appropriate decisions to help this event with their strong supports. Other perspectives in the fields of medicine and pharmacy have been already opened in nutrition, health, new drugs, and cosmetics, to name a few examples. Signing a document to establish a "twinning" between two universities, French and Japanese, at this point will facilitate greatly the flux of exchanges, for the benefit of both partners. It will constitute a good example for other universities.

Accordingly, I am glad to thank the two Sociétés franco-japonaises d'Océanographie, the University of Méditerranée, the Centre d'Océanologie de Marseille, and the two organizing Committees; in each country, they have set up this very interesting and productive event. It will certainly produce numerous after effects, such as a new meeting in Japan in the near future, and it will induce fruitful work together in the future.

I am glad to express my sincere praise to the organizers, and in particular Professors Hubert Jean Ceccaldi, Imawaki Shiro, and Yagi Hiroki for this nice success.

Monsieur le Professeur Yvon Berland
Président de l'Université de la Méditerranée

L'Université de la Méditerranée et le Centre d'Océanologie de Marseille, dirigé par le Professeur Ivan Dekeyser, ont pleinement participé au 13ème Colloque franco-japonais d'Océanographie qui s'est tenu au mois de septembre 2008. Ce dernier était organisé avec les deux Sociétés franco-japonaises d'Océanographie du Japon présidée par Professeur Imawaki Shiro et la Société franco-japonaise de France, présidée par le Professeur Hubert Jean Ceccaldi.

Ce fructueux colloque permettra de renforcer notablement dans le futur une coopération interuniversitaire qui existait déjà, mais de façon plus modeste, dans le domaine de l'Océanologie.

L'Université se réjouit d'avoir pu apporter son concours à cette belle réussite, notamment en hébergeant les séances d'échanges scientifiques à la Faculté de Pharmacie grâce à son Doyen Patrick Vanelle, et en participant à son soutien administratif.

L'Université se félicite particulièrement d'avoir établi de façon formelle, à la faveur de ce colloque, des accords d'échanges scientifiques et techniques avec les Professeurs Takai Rikuo et Izumi Mitsuru, respectivement Président et vice-Président de l'Université des Sciences et Technologies marines de Tokyo, au cours d'une cérémonie amicale à laquelle ont participé les Professeurs Yvon Berland et Roger Giudicelli, respectivement Président et vice-Président de l'Université de la Méditerranée, plus de quarante congressistes scientifiques japonais, Monsieur Bamba Masahiro, Consul Général du Japon à Marseille et Monsieur Jean-Louis Armand, Conseiller pour la Science et la Technologie de l'Ambassade de France à Tokyo.

De très intéressantes journées scientifiques ont été vécues. Elles conduiront à établir des orientations générales sur les relations entre l'homme et les milieux marins. Elles devraient aussi renforcer les liens entre les équipes de chercheurs des deux pays, en les incitant à travailler ensemble dans le but essentiel de pouvoir partager leurs connaissances.

C'est grâce à ces connaissances nouvelles mises en commun que pourront s'établir et se développer des projets de recherche originaux dans des disciplines qui, compte tenu des problèmes environnementaux très actuels, sont appelés à jouer un rôle primordial à l'échelon mondial.

Prof. Yvon Berland
Président de l'Université de la Méditerranée

Translation

Université de la Méditerranée and Centre d'Océanologie de Marseille, lead by Professor Ivan Dekeyser, have taken a full part to the Colloque franco-japonais d'Océanographie held in September 2008. That meeting was organised by the Société franco-japonaise d'Océanographie of Japan leaded by Professor Imawaki Shiro and by the Société franco-japonaise of France, leaded by Professor Hubert Jean Ceccaldi.

This fruitful meeting will give a new framework to intensify notably the future a cooperation between universities that already exists, and more modestly, in the field of marine sciences.

Our university is very glad to have given its support to this nice success, in particular the sessions of scientific exchanges in the amphitheatres of the Faculty of Pharmacy, thanks to the cooperation of the Dean, Professor Patrick Vanelle, with the help of his efficient administrative staff.

Our University is particularly glad to have formally established, in the framework of that binational meeting, scientific and technical agreements with Professors Takai Rikuo and Izumi Mitsuru, respectively, President and Vice-President of the Tokyo University of Marine Sciences and Technology, during a friendly ceremony, with the participation of the Vice-President Roger Giuducelli and the leading team of the University de la Méditerranée. More than 40 scientists travelled from Japan to attend the meeting. We have been glad also to receive Mr. Bamba Masahiro, General Consul of Japan in Marseille, and Mr. Jean-Louis Armand, Main Advisor for Science and Technology in the French Embassy in Tokyo.

Very interesting scientific meetings took place. They will lead to establishing general directions for the future concerning the relationships between mankind and marine environments. They will also strengthen the links between the teams of the two countries, inciting them to work together with the final aim to share their knowledge.

Thanks to this new knowledge acquired in common, new projects of research will be established and developed in some disciplines having strong links with very contemporary problems at a global scale, at the world dimensions.

Monsieur Tominaga Shigeatsu
Président du Conseil d'administration de la Fondation Franco-Japonaise Sasakawa

"Homme libre toujours tu chériras la mer! La mer est ton miroir; tu contemples ton âme" écrivait Arthur Rimbaud. Ces vers nous rappellent à quel point la mer est précieuse pour l'homme. La mer est aussi, pour l'homme, un moyen de liberté car elle lui permet de se connaître plus profondément. La chérir, l'aimer, la connaître, c'est se comprendre soi-même.

La mer est un miroir, et dans cet échange franco-japonais, connaître signifie, non seulement analyser, étudier les écosystèmes, mais également comprendre le rapport singulier qu'entretiennent, du fait de leur histoire et de leur géographie particulière, la France et le Japon. Le partage des connaissances, l'étude des différences méthodologiques, conceptuelles, permettent aux scientifiques, non seulement de progresser dans leur compréhension de ces objets très complexes que sont les écosystèmes marins et côtiers, mais, en élargissant les champs de recherche aux domaines juridiques, urbanistiques, philosophiques, anthropologiques, la mer, objet d'étude, redevient le miroir où les particularités françaises et japonaises se reflètent et se comprennent.

De nos jours plus personne n'ignore à quel point les écosystèmes marins et côtiers sont fragiles. Les hommes modifient, parfois bouleversent, ces équilibres fragiles. Or, du devenir de ces écosystèmes dépend l'avenir de l'humanité.

Comprendre leur évolution, les causes de ces changements, les raisons des bouleversements est devenu aujourd'hui une nécessité vitale.

Parce que la mer représente tant pour l'humanité, en tant que Président de la Fondation Franco-Japonaise Sasakawa dont la mission est de développer des relations de culture et d'amitié entre la France et le Japon, je suis heureux que nous ayons soutenu ce colloque organisé par les deux Sociétés franco-japonaises d'Océanographie de France et du Japon, et par le Centre d'Océanologie de Marseille. Cette réunion aura permis à plus de 150 chercheurs, principalement français et japonais, de se rencontrer de façon concrète, d'échanger et ainsi de faire progresser le savoir et contribuer à éclairer l'avenir. Les liens entre nos deux pays, dans les domaines de l'océanographie et dans les disciplines connexes s'en trouvent ainsi renforcés. De tous mes vœux, j'espère que les relations nouées pendant ces quatre jours de conférences perdureront.

Pour conclure, permettez-moi de remercier tous les partenaires qui avec nous ont soutenu cet événement ainsi que M. Hubert-Jean Ceccaldi, qui a su trouver les mots et nous convaincre de l'importance des relations franco-japonaises dans ce domaine.

M. Shigeatsu Tominaga,
Président du Conseil d'administration de la Fondation Franco-Japonaise Sasakawa

Translation

"Man of freedom, always you shall cherish the Sea! The sea is your mirror; you contemplate your soul" wrote the French poet Arthur Rimbaud. These verses remind us that the sea is precious for mankind. The sea is also, for any man, a means of freedom because it gives to each one the possibility to know himself more deeply

To cherish it, to love it, to know it is also understand himself.

The sea is a looking glass and, in this France-Japan exchange, to know means, not only to analyze, to study the ecosystems, but also to understand the peculiar relationships to keep alive the exchanges between France and Japan, because there is some resemblance of their peculiar history and geography. The sharing of their knowledge and the study of their methodological and conceptual differences lead the scientists not only to progress in the understanding of these very complex objects of the marine and the coastal ecosystems, but also to widen the fields of research to the domains of law, urbanism, philosophy, anthropology, etc. The sea, as an object of study, becomes again the mirror where the French and Japanese peculiarities are reflected and leads us to understand each other.

Today, nobody ignores the fact that the marine coastal ecosystems are fragile. Mankind modifies and sometimes disrupts these fragile equilibriums. Yet, the evolution of these ecosystems in the future depends on mankind. Understanding their evolution, the origin of these changes, and the reasons for these upheavals are becoming a vital necessity.

Because the sea represents so much for mankind, as I am the President of the Fondation Franco-Japonaise Sasakawa whose mission is to develop relationships in culture and friendship between France and Japan, I am glad to have established the support for this meeting, organized by the two Sociétés franco-japonaises d'Océanographie from France and from Japan, as well as the Centre d'Océanologie of Marseille. This meeting has induced the active presence of more than 150 people, mainly French and Japanese, to meet each other, directly, then to exchange information and to make progress in knowledge and to contribute to improving the future. The links between our two countries in the field of Oceanography and other related disciplines will be strengthened. In all my wishes, I hope that the links established during these 4 days of conferences will continue in the future.

To conclude, let me thank all the partners with whom we have cooperated in support of that event, as well as Mr. Hubert-Jean Ceccaldi, who found the words to convince us of the importance of the France-Japan relationships in that domain.

Monsieur le Professeur Imawaki Shiro
President of the Société franco-japonaise d'Océanographie du Japon

The year 2008 was the 150th anniversary of the cultural exchange between Japan and France, which originated from the trade treaty between the two countries signed in 1858, near the end of Edo era. During the one and a half centuries, we have had very rich exchange activities between the two countries in various fields.

In April 1960, the Japanese-French Oceanographic Society (Societe franco-japonaise d'Oceanographie) was established in Japan in order to stimulate the scientific exchanges between Japan and France in the fields of oceanography and fisheries. The establishment was mostly indebted to the late Professor emeritus Tadayoshi Sasaki of Tokyo University of Fisheries. It was almost 50 years ago. Today, the Society has about 200 regular members. The Society holds its scientific conference in June every year. In 1963, we started to publish a society bulletin "Umi" in Japanese, which is called "La mer" internationally. The bulletin has been issued four times a year. In 1966, we established a Society prize. Its first winner was Professor emeritus Kenzo Takano of the University of Tsukuba, who stayed at Grenoble University in France for several years in the 1960s.

In 1984, the French-Japanese Oceanographic Society was established in France. It was deeply indebted to Professor emeritus Hubert Jean Ceccaldi of "Ecole Pratique des Hautes Etudes." Professor Ceccaldi served as the Director of the Maison Franco-Japonaise in Tokyo for 4 years from 1988 and contributed tremendously to cultural exchanges between the two countries.

The Japanese-French Oceanographic Society in Japan and French-Japanese Oceanographic Society in France have been stimulating oceanographic activities between the two countries. The two societies started to hold joint oceanographic symposia. The first symposium was held in Montpellier in 1983. The present symposium in Marseille and Paris in 2008 is the 13th, and we have almost 40 scientists from Japan.

This is a very brief summary of the history of exchange activities in the oceanography and fisheries fields. Another field of ocean sciences may be ocean engineering. France has been leading the world in deep ocean expeditions. The manned deep-sea vessel, the bathyscaphe F.N.R.S. III, visited Japan in 1958; it was 50 years ago. French and Japanese scientists carried out a deep ocean expedition in Japan Trench in that bathyscaphe. In 1962, another bathyscaphe, "Archimede," visited Japan to carry out a deep ocean expedition in Chishima Trench. Those activities using bathyscaphes were succeeded by IFREMER in France and JAMSTEC in Japan. JAMSTEC built manned deep submersibles called "Shinkai 2000" and "Shinkai 6500" to contribute to deep ocean science.

In the field of physical oceanography, France has been contributing tremendously In the field of satellite altimetry. CNES of France launched a satellite TOPEX/POSEIDON in 1992 in collaboration with NASA of the United States of America. This satellite measured variations of the sea-surface height very precisely. Nowadays, physical oceanographers are addicted to satellite altimetry. Its successor, Jason-1 of CNES and NASA, has already finished its job, and Jason-2, launched in June 2008, is now operating. France is leading in the satellite altimetry observation of oceans.

The next meeting, the 14th Japan-France Oceanography Symposium, will be held in Kobe, Japan, in 2010. Kobe is a sister city of Marseille. The year 2010 is the 50th Anniversary of the Japanese-French Oceanographic Society, and the Symposium is one of the activities of celebrating the anniversary. As the President of the Society, I would like to invite all of you to participate in the 2010 Symposium in Kobe.

Finally, I would like to thank the organizing committee led by Prof. Hubert Jean Ceccaldi and also Prof. Hiroki Yagi of Otaru University of Commerce, Japan, for their perfect preparation of the symposium.

Prof. Shiro Imawaki.
President of the Japanese-French Oceanographic Society

Le maire de la ville de Marseille
Jean-Claude Gaudin

Notre ville a eu le plaisir d'accueillir le 13[eme] Colloque Franco-Japonais d'Océanographie dont le Centre d'Océanologie de Marseille était le principal partenaire. Les échanges et les communications scientifiques de ce colloque et leur publication sont du plus grand intérêt pour Marseille dont la bordure maritime s'étend sur 57 km.

Cette façade littorale représente un espace fragile où se concentrent les enjeux et les défis liés à sa préservation et à sa valorisation. Aussi, dans le cadre de son engagement en faveur de la protection de l'environnement et du développement durable, Marseille a lancé et réalisé d'importants projets.

Géolide, la plus grande station biologique enterrée au monde pour le traitement des eaux usées, est en service. L'implantation de récifs artificiels dans la rade du Prado recrée des conditions favorables à la prolifération de la faune et de la flore. Quant au Parc Maritime des Iles du Frioul, il est intégré dans le réseau Natura 2000.

Le Groupement d'Intérêt Public des Calanques préfigure le Parc National des Calanques, le premier parc national périurbain pour protéger et valoriser les espaces naturels et exceptionnels, terrestres et maritimes, du site. De plus, la Ville a défini le "Plan de Gestion de la Rade de Marseille" destiné à mettre en cohérence les différentes politiques publiques intervenant sur cette zone.

D'autre part, ce colloque a contribué à conforter les liens que notre ville a tissés avec le Japon, en particulier avec la ville-port de Kobé, avec laquelle Marseille a échangé un serment de jumelage il y a près d'un demi siècle. L'accueil de cette manifestation s'inscrit donc dans la vocation d'ouverture et d'échanges de notre ville et la tenue de ce colloque d'intérêt international contribue au rayonnement scientifique et intellectuel de Marseille.

Jean-Claude Gaudin
Maire de Marseille
Vice-Président du Sénat

Translation

Our city has had great pleasure in welcoming the 13th Colloquy of Franco-Japanese Oceanography; the Centre d'Océanologie de Marseille was the main partner.

The exchanges and the scientific communications of that meeting and their publication are of highest interest for Marseille, with its littoral reach 57 km long.

That coastal area represents a fragile space where numerous important stakes and challenges linked to its preservation and its development are concentrated. So, in the framework of the commitment to protect the marine environment and sustainable development, Marseille has launched and carried out important projects.

Géolide, the largest underground biological water treatment facility in the world, is functioning. Immersion of artificial reefs in the Prado Bay create favorable conditions for the proliferation of marine fauna and flora. The maritime natural park of the Frioul Islands is integrated in the Natura 2000 network

The public interest group "Calanques" is a prefiguration of the new natural marine park, the first periurban national park existing, in order to protect and enhance the value of the exceptional terrestrial and maritime natural spaces, of this site. Moreover, the city has defined "Management Planning of the Larger Bay of Marseille" with the aim to add coherence to the different public policies acting within that area.

However, this scientific meeting represent a valuable contribution to strengthen the links with Japan, particularly with the port city of Kobé, with which Marseille exchanged an oath of twinning almost half a century ago. In this, the welcome of this scientific event fits well with the attitude of opening and of exchanges of our city and this international meeting as a concrete contribution to the scientific and intellectual fame of Marseille.

Preface

For a long time, very fruitful dialogues have been established between Japanese and French oceanographers, by means of different kind of contacts. These dialogues and our common scientific meetings are continuing by this Colloque franco-japonais d'Océanographie.

Personally, I have also had the chance to promote these exchanges directly several times with the Société franco-japonaise of Japan, especially when I was Director of Maison franco-japonaise (Nichi-Futsu Kaikan) in Tokyo, for 4 years. It was a nice opportunity to establish new personal contacts, exchanges, seminars, visits to laboratories and research centers, meetings in Japanese municipalities, receiving students for work on their thesis, etc. Such relationships are one of the best ways to understand another approach to scientific problems and to get to know another culture better.

This 2008 scientific meeting in Marseille is the 13th of a rather long series. Fortunately, as a good symbol, it was the year of the 150th anniversary of the trade and friendship treaty established in 1858 between Japan and France. We have received the support of the French Academy of Sciences and the Foundation Louis D., of the Embassy of Japan, of the Maison de la Culture du Japon à Paris, of the Conseil Régional Provence Alpes Côte d'Azur, of the Conseil Général des Bouches du Rhône, of the Municipality of Marseille, of the Faculty of Pharmacy, of the Agence de l'Eau Rhône Méditerranée Corse, of Fondation franco-japonaise Sasakawa, and several other public and private organisations. We are glad to have received numerous members of the Société franco-japonaise d'Océanographie of Japan and a number of Japanese scientists to stimulate personal, friendly, and efficient scientific exchanges between Japanese and French specialists in the oceanography and fisheries fields.

It is also a great pleasure for me to thank President Imawaki Shiro, recently elected President of the Société franco-japonaise d'Océanographie of Japan, and especially, on the Japanese side, a sincere cordial message to my friends of more than 25 years, Professors Yagi Hiroki, Komatsu Teruhisa and Koike Yasuyuki, and Drs. Arakawa Hisayuki and Tanaka Tsuneo. All my sincere thanks also go to colleagues of the Organizing Committee, such as Professor Ivan Dekeyser, Drs. Georges Stora, Yves Henocque, Patrick Baraona, Guy Herrouin, Eric Delort, MMr. Mathias Girault, Maurice Libes, Mrs Catherine Walch, Brigitte Pantat, and Dominique Poirot also for their invaluable efforts in the preparation and the success of this symposium.

We would like to thank especially Professor François Gros, Honorary Perpetual Secretary of the French Academy of Sciences, for his kind and continuous support of our Society within the exchanges between Japan and France in the field of marine sciences and to express our grateful thanks to Mr. Gabriel de Broglie, Chancellor of the Academy, for the grant obtained from the Foundation Louis D.

We are greatly indebted to His Excellency Mr. Iimura Yutaka, for his constant support during the preparation of our program, for his presence and his address during the meeting in Maison de la Culture du Japon à Paris. We are strongly indebted to Ambassador Iimura for having invited the main organizers of this meeting for an exceptional reception at his residence.

Special thanks to Professors Yvon Berland and Roger Giudicelli for the support given to our meeting, for giving us the chance to use some of the material possibilities of the Université de la Méditerranée, and for offering a nice reception to the members of the meeting, and especially to Professor Patrice Vanelle, Dean of the Faculty of Pharmacy of Marseille, for the use of the amphitheatres and the numerous facilities of the Faculty. We are glad to express our gratitude to Mr. Jean-Claude Gaudin, Mayor of Marseille, Vice-President of the French Senate, to Mrs. Dominique Vlasto, European Representative, in charge of the Tourism Office, and Mr. Didier Réault, Municipal Delegate in charge of marine and littoral activities in Marseille City, for the grant and the support given to our meeting in Marseille and for the magnificent reception in the historical City Hall. Mr. Réault kindly gave the first address at the opening of our scientific meeting.

We are very pleased to thank deeply Mr. Pierre Boissery, in charge of studies at Agence de l'Eau, Rhône, Méditerranée, Corse, for the efficient help and the support that benefitted our meeting. Mr. Jacques Saugier, chargé de mission for the Economy, and Mr. Philippe Lassalle, chargé de mission for the Integrated Management of Coastal Zone, in the General Secretary of Regional Affairs of the Prefecture have fortunately supported our meeting by their encouragement and by the allocation of a grant contributing to our budget.

Mr. Tominaga Shigeatsu, President of the Fondation Franco-Japonaise Sasakawa and the Administrative Council of the Foundation, has integrated our multidisciplinary project in the program of their activities and has given important material support for our meeting; we are glad to thank all the members of that Council and also Mr. Eric Mollet for his help. In Marseille, we are glad to thank the two successive General Consuls Mr. Kobayashi Masao and Mr. Bamba Masahiro for their kind help and for their participation tinour to our project. We have obtained the support of the Regional Council of Provence Alpes Côte d'Azur, mainly interested in scientific research and coastal management. We would like to express our thanks to Mrs. Christine Sandel, Conseiller for the coastal zone, then Mrs. Genviève Kalfon and Ms Florence Papini, leading the service Enseignement supérieur et Recherche as well as Mrs. Josette Sportiello, Mr. Benoît Vasselin, and Mrs. Valérie Raimondino, driving the Service "Mer." The Conseil Général des Bouches du Rhône helped by giving us very useful materials.

In Paris, we have been very pleased to cooperate with Mr. Nakagawa Masateru, President of Maison de la Culture du Japon (Nihon Bunka Kaikan), and with Mrs Karine Masneri. We used the magnificent meeting room and the French-Japanese translation facilities for a synthesis of the 3-day meetings in Marseille, then to listen to four conferences of high standard given by Professors Taniguchi Akira from Tokyo University of Agriculture Okhotsk at Hokkaido ("Relationships between mankind and marine environments: example of Japan") and Nakagawa Heisuke from Hiroshima University ("Actual situation and future of aquaculture with examples on fish species in Japanese coasts") as well as Mrs. Catherine Bersani, General Inspector in the French Ministry of Ecology ("New criteria to be used on the evolution of laws in regulation of coastal zones") and Professor Bernard Salvat, from Ecole Pratique des Hautes Etudes ("Actual situation and recent evolution of coral reefs in the world").

We are glad to benefit from the presence of eminent persons such as Mr. Jean-Yves Perrot, President Directeur Général of the Institut Français pour l'Exploitation de la Mer (IFREMER), and Dr Emmanuel Thouard, in charge of international relationships with Asian countries.

We would like to thank also the Ministry of Foreign Affairs for the support of His Excellency Mr. Gildas Le Lidec, Ambassador of France in Japan, and Professor Jean-Louis Armand, Conseiller pour la Science et la Technologie in the French Embassy in Tokyo, who participated personally in our meetings in Marseille and Paris. We are thanking also the Centre National de la Recherche Scientifique (CNRS) and the Institut National des Sciences de l'Univers (INSU) for their interest concerning our 13th Colloquy on Franco-Japanese Oceanography.

We are deeply indebted to Dr. Robert Doe, to Ms Nelly Hemink and to Mrs. Nina Bennink, scientific editors for Springer, for their patience and their help in the publication of this book.

This meeting opens new ways for future cooperative programs and personal exchanges for mutual benefit to the two communities of researchers. The following meeting will be held in Kobe and in Tokyo in October 2010. We are quite glad to pursue with success this fruitful common adventure.

Prof. Hubert Jean Ceccaldi
Président de la Société franco-japonaise d'Océanographie

Contents

Observation of marine environment

Introduction

A Brief History of the Activities Between the Two Sociétés franco-japonaises d'Océanographie

Scientific and technological relationships in the field of marine sciences between France and Japan have been facilitated by dialogues and exchanges established between the Société franco-japonaise d'Océanographie of Japan (founded in 1960 and gathering around 300 members), and the Société franco-japonaise d'Océanographie of France (founded in 1984 and gathering several tens of persons as well as public and private organizations).

These two Societies are mainly associations of researchers and other specialists in marine fields who have expressed their wish to promote efficient and pleasant relationships between Japanese and French individuals as well as organizations, having similar fields of activities.

These Societies enhance friendly and informal relationships and lead very often to the beginning and the basis of cooperative researches.

The Société franco-japonaise d'Océanographie of Japan was founded by the late Dr Sasaki, Professor at the University of Fisheries of Tokyo (Tokyo Suisan Daïgaku), when the French specialists in deep diving with their bathyscaphs came to Japan. Its head office is located in Maison franco-japonaise (Nichi-Futsu Kaïkan) in Tokyo (Ebisu area, Shibuya-ku). Like in other disciplines, there are more than 20 other Sociétés franco-japonaises. Currently, its President is the Professor Imawaki Shiro, former Professor at Kyu-Shu University, who is actually Operating Executive Director in JAMSTEC.

The Société franco-japonaise d'Océanographie of France was founded in 1984 by Dr. Hubert Jean Ceccaldi, Emeritus Professor of the Ecole Pratique des Hautes Etudes, one of the Grands Etablissements of higher education in France and several other specialists in Marine sciences: Y. Henocque, C Mariojouls, F. Simard, J.-M. Thierry, A. Nishikawa, N. Lucas, D. Bailly, The office of the Society is located at the Institut oceanographique in Paris (close to the Luxembourg Gardens).

For several decades, the exchanges between the members of the two Societies have been very fruitful and very friendly. The exchanges reached a maximum during common meetings (colloques, symposiums) alternatively organized in France and in Japan, approximately every 2 or 3 years. It is interesting to summarize them briefly.

First edition in France: Montpellier, 1983

The Colloque franco-japonais devoted to "*Aquaculture*", in the Sciences and Technology University of Languedoc (Université des Sciences et Techniques du Languedoc), plus visits of Montpellier and Etang de Thau (molluscs production). Six specialists from Japan and 21 French participants.

Publication of the communications (152 pages, ISBN 2-906495-00-X)

Second edition in Japan: Sendai, 1984

Colloque franco-japonais devoted to "*Aquaculture*", organized with the help and the support of Kitasato University, the Japanese Society of Scientific Fisheries, Kajima Corporation, and Tokyo Electric Power Co. Visits to hatcheries of fishes and molluscs. Nine French and 34 Japanese participants.

Communications on October 4th

Publication of the communications (102 pages, ISBN 4-9900030-2-0)

Third edition in France: Marseille, 1985

Colloque *pluridisciplinaire* franco-japonais "Les aménagements côtiers et la gestion du littoral" (Coastal management and littoral planning), organized with the support of numerous public and private organizations:

Ministries of Foreign Affairs, of Agriculture, of Environment, of Transportation, of Industrial Conversion and Foreign Trade, then the help of IFREMER, of CNRS, of Conseil régional PACA, of the Municipal Council of Marseille City, of the Office régional de la Mer, of Office de la Recherche Scientifique et Technique Outremer, of Institut National de la Plongée Professionnelle (I.N.P.P.), of Marseille Trade and Industry Chamber, of Naturalia et Biologia Association, of the Fondation océanographique Ricard, of Compagnie maritime d'expertises (COMEX), of Marseille Port Authority, of Ecole Pratique des Hautes Etudes and Centre d'Océanologie de Marseille, etc.

Scientific meetings in Marseille on 16 to 21 September.

There were 24 Japanese scientists were and 320 French specialists attending the meeting. Eight volumes of results were published, amounting to 1,105 pages.

1. Océanographie physique and dynamique sédimentaire (Physical oceanography and sediment dynamics), 193 p. (ISBN 2-906495-02-6)
2. Eaux colorées (Coloured waters, red tides), 109 p. (ISBN 2-906495-03-4)
3. Télédétection, communications, 53 p. (ISBN 2-906495-04-2)
4. Microbiologie des eaux côtières (Microbiology of coastal waters) 99 p. (ISBN 2-906495-05-0)
5. Caractères biologiques des eaux côtières (Biological characters of coastal waters) (ISBN 2-906495-06-9)
6. Récifs artificiels (Artificial reefs), 167p. (ISBN 2-906495-07-7)
7. Exploitation des ressources (resources exploitation) (ISBN 2-906495-08-5)
8. Aquaculture, 187 p. (ISBN 2-906495-09-3)

Fourth edition in Japan: Shimizu, 1988

Colloque franco-japonais "*General oceanography*", held in the Marine Building of Shimizu belonging to the Municipality. Presentation of communications, then visits to aquaculture centers, University Tokai, JAMSTEC, Fisheries Agency, fish market of Yaizu, organized with the help of Japanese Science and Technology Agency, the Ministry of International Trade and Industry (MITI), the French Embassy, the Prefecture of Shizuoka, the société franco-japonaise du Japon, the Maison franco-japonaise de Tokyo (Nichi-futsu Kaikan).

Scientific meeting in Shimizu and Tokyo, on 3 to 7 October.

Twenty French specialists and 50 Japanese attended the meetings.

Publication in 1989 of a special issue of "La Mer" (Journal of Société franco-japonaise d'Océanographie of Japan) tome 27 (3), 165 p.

Notice: This meeting was followed by an important multidisciplinary meeting in the Maison franco-japonaise in Tokyo on October 17, where ten Sociétés franco-japonaises in different disciplines went to explain common areas of research. One hundred French specialists and 300 Japanese attended this meeting in Tokyo, including three conferences in marine subjects.

Fifth edition in Japan: Hiroshima and Higashino, 1989

Series of six French-Japanese colloquys, on the main themes:

"*Founding an algal park in Seto-nai-kai*" (*Création d'un parc algal dans la mer intérieure du Japon*), on the basis of research work of Dr. François Simard (resident in Maison franco-japonaise in Tokyo), between May 16 and March 29, 1990.

These six colloquy-seminars were held in the National establishment for the Merchant Marine in Hiroshima, and in the Cultural Center of Higashino, a city located in Omishima, in Seto-Naï-Kaï, with the help of the municipality, the Hiroshima Prefecture, and the French Group "Aménagements du Littoral."

Ten French specialists and around 100 Japanese attended the meetings.

Publication of a 152-page book by the publishing bureau of Maison franco-japonaise.

Sixth edition in Japan: Tokyo 1990

Colloque franco-japonais: "*Littoral et conflits*" (*Coastline and conflicts*) held in the Maison franco-japonaise.

Communications on 8 October.

Visits to Tokyo by Suisan Daigaku and representatives of several hatcheries with the support of the French Embassy, the Ministry of National Education, IFREMER, and the Société franco-japonaise du Japon.

Six French specialists and 25 Japanese specialists have participated to the meeting.

Texts of the communications and comments have been edited by the publishing bureau of Maison franco-japonaise, 105 p.

Seventh edition in Japan: Tokyo 1990

Colloque franco-japonais: "*Déterminisme du recrutement biologique en mer*" (*Determinism of Biological Recruitment at sea*).

Communications on 13 to 14 November.

Visits of Tokyo Suisan Daigaku and several hatcheries,

With the support of French Embassy, the Ministry of National Education, the Centre National de la Recherche scientifique (CNRS), of IFREMER, and of the Société franco-japonaise d'Océanographie du Japon.

Six French specialists and 25 Japanese specialists have participated to the meeting.

Texts of the communications and comments have been edited in a fascicule of a series published by the bureau of Maison franco-japonaise.

Eighth edition in France: Nantes 1991
Three colloques franco-japonais:

- "*Facteurs déterminants de la croissance en aquaculture*" ("*Détermining factors of the growth in aquaculture*") *Communications November 13th and 14th*
- "*Economie et gestion des Pêches*" (*Economy and management of fisheries*)
- *Co-developpement des pêcheries et des activités de loisir dans les régions côtières* (*Co-development of fisheries and leisure activities in coastal zones*)

Meetings in the halls of IFREMER in Nantes.
Communications from 2nd to 5th
With the support of the Centre National de la Recherche scientifique (CNRS), of IFREMER, of Institut océanographique de Paris, of Union des Océanographes de France, and of the Société franco-japonaise du Japon.
Six Japanese specialists and 30 French specialists have participated to the meeting.
Texts of the communications and comments have been edited as a special issue of the review "*Oceanis*", 1992, vol. 18, fasc. 1: 1–140, and vol. 18, fasc. 4: 371–503.

Ninth edition in Japan: Tokyo 1991
Colloque franco-japonais: "*Flux océaniques*" ("*Oceanic fluxes*").
Communications from 25 to 27 November in Maison franco-japonaise (Nichi-futsu Kaïkan)
Themes of the international JGOFS Program,
With the support of the French Embassy, the Ministry of National Education, the Centre National de la Recherche scientifique (CNRS), of the University of Fisheries of Tokyo (Tokyo Suisan Daigaku), and of the Société franco-japonaise d'Océanographie du Japon.
Eleven French specialists and 21 Japanese specialists have participated to the meeting.
Texts of the communications have been edited as a special issue of the journal "La Mer," published by the Société franco-japonaise d'Océanographie du Japon, tome 29, fasc. 4, 162 pp.

Tenth edition in Japan: Tokyo 1992
Two colloque franco-japonais of Oceanography

- "Biotechnologie et environnement" ("Biotechnology and Environnement")
- "Déterminisme du recrutement biologique en mer" (Determinism of Biological Recruitment at sea).

Communications on August 31st in Maison franco-japonaise de Tokyo (Nichi-futsu Kaïkan)
With the support of French Embassy, the Ministry of National Education, the Centre National de la Recherche scientifique (CNRS), of IFREMER, of Tokyo University of Fisheries (Tokyo Suisan Daigaku) and of the Société franco-japonaise d'Océanographie du Japon.
Six French specialists and 25 Japanese specialists have participated in the meeting.

Texts of the communications and comments have been edited in a multigraph fascicule published by the bureau of Maison franco-japonaise.

Eleventh edition in France: Paris, 1997

Colloque franco-japonais: "*Observations en zone côtière et prévisions à moyen et long terme*" ("*Coastal zone observation and forecast in the medium and long term*")

Meetings in Institut océanographique de Paris

Communications from 6 to 8 October.

Visit the exposition SEAMER in the exhibition hall and fair located in the Porte de Versailles in Paris.

Transfer by TGV to Brittany and reception in Vannes, visit the Morbihan Gulf by boat, the Vilaine River estuary, La Roche Bernard, Auray hatcheries, and back to Paris.

With the support of the French Ministry of Foreign Affairs, Ministry of Education and Scientific Research, Centre National de la Recherche scientifique (CNRS), the Société Daniel Jouvance et Création, and of the Société franco-japonaise du Japon.

Twelve Japanese specialists and 94 French specialists participated to these meetings.

Texts of the communications and comments have been edited as a special multigraph fascicule of summaries and texts of communications.

Twelfth edition in Japan: Tokyo 2005

Colloque franco-japonais: "*Compréhension mutuelle nouvelle pour les recherches en Océanographie et en pêcheries, en France et au Japon*" ("*Mutual new understanding for research in oceanography and fisheries, in France and in Japan*").

Communications and scientific meetings on 5 November at Tokyo University of Marine Science and Technology (or KaiyoDai) and on 6 November at Maison franco-japonaise (Nichi-Futsu Kaikan).

Visits of university laboratories, research vessels, official bureaus, and private enterprises in Onjuku, Abiko, and Shimizu.

With the support of French Embassy, Marine Environment Research Institute, Kaiyodai, the Foundation Sasakawa, and the Société franco-japonaise d'Océanographie of Japan.

Three French specialists and 85 Japanese specialists have participated to these meetings.

Summaries of the communications have been edited in a multigraph fascicule.

Thirteenth edition in France: Marseille 2008

Colloque franco-japonais: "*Global Change: interactions mankind/marine environments*" ("*Le changement global: interactions homme/milieux marins*")

Meetings at the Faculty of Pharmacy of Marseille on 8 to 10 September and at the Paris Nihon Bunka Kaikan (Maison de la Culture du Japon à Paris) on 12 September.

With the support of French Embassy in Tokyo, the Embassy of Japan in Paris, the Municipality of Marseille, The Water Agency Rhône-Méditerranée-Corse, the Regional Council of Region Provence-Alpes-Côte d'azur, Tokyo University of Marine Science and Technology Kaiyodai, the France-Japan Foundation Sasakawa, the Foundation Louis D., and the Société franco-japonaise d'Océanographie of Japan.

Thirteenth Colloque franco-japonais d'Océanographie Marseille, September 8th, 9th and 10th and Paris, September 12th, 2008

The general theme of the present colloquy was:

"*Global Change: interactions mankind – marine environments*" ("*Le changement global: interactions homme – milieux marins*").

In this framework, the session subjects selected for this Colloque franco-japonais were:

- Oceanography, microbiology, biogeochemistry
- Coral reefs
- Biodiversity
- Management of littoral zones and artificial reefs
- Aquaculture
- New techniques of observation in the marine environment

This was suggested during the meeting held in Tokyo in November 2005, organized by the Société franco-japonaise d'Océanographie of Japan; then, it was established after further exchanges by several mails between members of the two Sociétés and the Committee of organization.

Scientific meetings in Marseille took simultaneously in two amphitheatres in the Faculty of Pharmacy of Marseille on 8 to 10 September; then in Paris, in Nihon Bunka Kaikan (House of Japan Culture or Maison de la Culture du Japon à Paris) on 12 September. Forty-two Japanese specialists and 120 French specialists participated in these meetings. Summaries of the communications have been edited in a multigraph fascicule.

During the whole meeting, we were very pleased to welcome Professor Takai Rikuo, President of Tokyo University of Marine Sciences and Technology, Professor Izumi Mitsuru, Vice-President, and the Professor Imawaki Shiro, President of the Société franco-japonaise of Japan.

We were glad also to receive Professor Takagi Ken, a prominent actor in the organisation of the international Salon TECHNO-OCEAN, and the well-known Professor Taniguchi Akira. All the other Japanese participants were eminent in their specialities, and the French members of the Société franco-japonaise d'Océanographie of France were proud to receive them and to meet them during these gatherings.

There were 42 Japanese participants, each one specialized and well known in their own discipline. There were 120 French specialists in marine sciences. These scientists presented 70 oral communications and 50 posters during these 3 days.

These meetings were officially integrated in the manifestations of the 150th Anniversary of the signature of the First Treaty of Friendship between France and Japan.

Mr. Didier Reault, in charge of the relationships between Marseille and the numerous coastal activities, represented the Mayor Mr. Jean-Claude Gaudin, Vice-President of the French National Senate, already engaged in Paris. A magnificent reception was given for all the participants of the Colloquy in the historical reception salons of the City Hall.

In Marseille, the Colloque franco-japonais was officially integrated into the seasonal program: “Septembre en Mer” (“September at Sea”), gathering all events happening at the end of summer in the city during that month.

The President of the University, Professor Yvon Berland, and the Vice-President, Professor Giudicelli, in charge of International Relations of the University, offered a very nice reception in the Presidence building of the University in the Gardens of Pharo.

On that occasion, a twinning and an agreement of scientific exchanges were signed between the two Universities: Tokyo University of Marine Sciences and Technology, and the Université de la Méditerranée, in presence of Mr. Bamba, General Consul of Japan in Marseille,

After these 3 days of meetings in Marseille, the participants moved to Paris on 11 September to attend the second part of the meetings.

On 12 September in the morning, in Paris Nihon Bunka Kaïkan (Maison de la Culture du Japon) participants established a preliminary evaluation of the scientific exchanges in Marseille, then some possible perspectives for future cooperation.

We have been very honored to receive several very important persons who participated in these gatherings, such as His Excellency Mr. Iimura Yutaka, Ambassador of Japan in France, Professor François Gros, Secrétaire perpétuel honoraire de l’Académie des Sciences de Paris, Mr. Jean-Yves Perrot, President Directeur Général of IFREMER (Institut Français pour l’Exploitation de la Mer), Mr. Patrick Baraona, President of Marine cluster (Pôle de compétitivité “Mer”), Mr Tominaga Shigeatsu, President of the France-Japan Sasakawa Foundation and Mr Jean-Louis Armand, Advisor for Science and Technology in the French Embassy in Japan. We do not want to forget the representatives of the different Ministries (Research, Education, Environment, Equipment, Army, etc.) who assisted with these meetings in Paris.

We are very grateful to H.E. Mr. Iimura for having invited the main organizers of this scientific meeting to his residence in Paris for an unforgettable dinner, and to Mr. Nakagawa, Director of The Organisation Committee, wishes to express his gratitude to the organizations and to the persons who gave their support to make this meeting a success. We would like to thank particularly the Municipality of Marseille (Mme Dominique Vlasto and Mr Didier Reault), the Water Agency Rhône-Méditerranée-Corse (M. Pierre Boissery), the Regional Council of Region Provence-Alpes-Côte d’azur, the French Academy of Sciences, the Foundation Louis D., the French Embassy in Tokyo, the Embassy of Japan in Paris and the General Consulate of Japan in Marseille, the France-Japan Foundation Sasakawa, the Dean of the Faculty of Pharmacy of the University of Marseille (Prof. Pierre Vanelle), the Tokyo University of Marine Science and Technology Kaiyodai, and the Société franco-japonaise d’Océanographie of Japan. Particular mention must be made of our colleagues and friends in Japan, the Professors Imawaki Shiro, Yagi Hiroki, Koike Yasuyuki and Komatsu Teruhisa, Arakawa Hisayuki and NNN Tsuneo

Aquaculture

A Few Examples of the Many Approaches to Salmon Resource Creation in Japan

E. Hasegawa, T. Saito, T. Kaga, and T. Suzuki

Abstract The chum salmon *Oncorhynchus keta* (Walbaum) resource of Japan has been dramatically improved by artificial propagation, and this resource increased rapidly in the 1980s. However, an unexpected decrease was observed in 1992, 1999, 2000, and 2008. An accurate resource evaluation of each year's class group is necessary to determine the appropriate method of managing the salmon resource, and the adult return rate is one of the indices that can be used for this purpose. This rate is calculated as a ratio of the number of recurrences (i.e., the total adult returns to rivers and coastal seas) to the number of chum fry stocked, and it has fluctuated around a total mean value of about 3% in Hokkaido and Honshu Islands in recent years. However, the adult return rate also varies by region from 0.1% to about 10%. The return rate of the northern area tends to be higher than that of the southern area, and that of the Pacific Ocean side tends to be a higher measurement of the local resource because the number of salmon caught by set nets, etc., in the coastal sea area catches might not be an accurate measure of the area's resources.

This report introduces an example of a research approach in Japan that attempts to achieve high-ranking stabilization of this accurate adult salmon return rate.

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1 Introduction

In Japan, three species of *Oncorhynchus*, chum salmon *O. keta* (Walbaum), pink salmon *O. gorbuscha* (Walbaum), and masu salmon *O. masou* (Brevoort), are stocked by artificial propagation. Nowadays, the stocking number of chum salmon is about 1.8 billion per 1 year. Their return number is about 60–70 million. Thus, the return rate is about 3% in Japan. However, the adult return rate also varies by region from 0.1% to about 10%. The return rate of the northern area tends to be higher than that of the southern area because the direction of their spawning migrations is from the northern area to the southern area. There are many set nets along the coast of north Japan, and possibly the salmon of southern origin is caught by set nets in the northern area. This might also be one of the important reasons why the survival rate of the juveniles released in the northern area is higher than the population released in the southern area, and also a reason explaining the phenomenon that the return rate of the Pacific Ocean side tends to be higher than that of the Sea of Japan side. The amount of animal plankton consumed by salmon on the Pacific Ocean side is more than that consumed on the Sea of Japan side in spring when they are stocked.

Changes in the status of the resource occur because of deterioration of water quality or over catching. The water quality of the Toyohira River, which flows through the center of Sapporo City, deteriorated and became an unsuitable environment for the fish, but has improved gradually since about 1970 because of expansion of the drainage system. A picture of the Toyohira River, which flows through Sapporo City, is shown in photo 1.

With the improvement of the water quality, the salmon have been able to breed naturally since the latter half of the 1970s. Some of Sapporo's citizens thought,

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Photo 1 Toyohira River that flows in Sapporo City (http://www.welcome.city.sapporo.jp/)

"Let's return the salmon to the Toyohira River," and a movement developed to bring back the salmon at that time. In the spring of 1979, discharge of the fry was recommenced after an interval of about 30 years; the fry became parent salmon in the autumn of 1981 and came back to the Toyohira River. Since then, discharge has continued, and natural egg laying in the Toyohira River has been confirmed every year since 1985. At present, the existence of salmon in Toyohira River is confirmed by both natural egg laying and artificial proliferation. One million salmon fry were stocked every year from 1979–1981. However, the number was reduced upon the confirmation of parent fish's upstream migration and natural egg laying. After 1988, the number was about 200,000. Since 1997, about 1,000 to 2,000 salmon are stocked every year.

The salmon resource in the Rhine River could be recovered using an artificial proliferation method like the one described above. Up to 150,000 salmon were captured in the Rhine before 1890, and these numbers might be reached again.

The following approaches can be used to secure salmon resources. However, they are only part of the approach in Japan.

The bio-resource characteristics of chum and pink salmon were investigated by examining the changes in the rhodopsin-porphyropsin ratio. The visual pigments present in the rod outer segment are of two types: rhodopsin and porphyropsin. Rhodopsin is present in most marine fishes, and porphyropsin is present in most freshwater fishes. The absorption spectrum of the visual pigment in each fish is closely related to the light conditions of their habitat. For example, the maximum wavelength of the absorption spectrum of the visual pigment of a deep-sea fish is shorter than that of a coastal species. Some fish possess both visual pigments in the retina. The proportions of these two visual pigments may be modified by their habitats. Assuming the proportions of these two pigments in anadromous fish are different between fresh and seawater regions, the ratio should indicate the triggering of migration between the two water areas. Furthermore, the difference in the rhodopsin-porphyropsin ratio may be reflected in the difference of their depth of habitat.

In addition, a change in the amount of swimming energy consumed growing up in order to estimate the amount of food necessary so that the released salmon fry might survive in the coastal waters was presumed in this study. This finding is considered useful for deciding on an appropriate number of discharge individuals and time if the food situation on the coast can be understood. In this chapter, it is introduced as the estimation of the swimming energy.

2 Materials and Methods

2.1 Bio-resources Characteristics Considered from the Changes in Rhodopsin Ratio

2.1.1 Materials

The life stages of materials investigated in this report were from eyed egg individuals up to spawning adult chum and pink salmon. Before release, the individuals

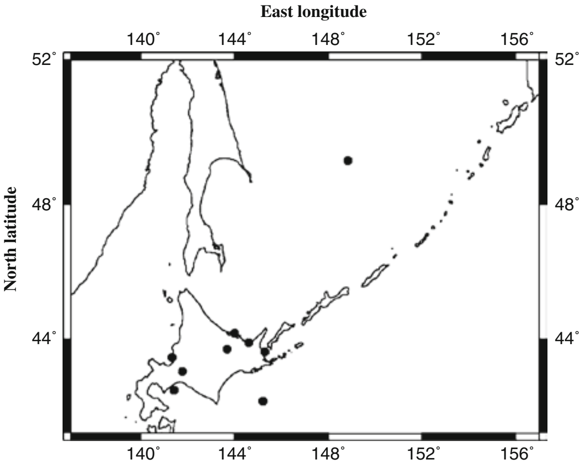

Fig. 1 Research areas for determining the rhodopsin ratio of chum and pink salmon. A, offshore Shibetsu; B, offshore Shiraoi (mouth of the Shikiu River is included); C, Sea of Okhotsk; D, North Pacific Ocean; E, Tokoro River; F, Shari River and set net; G, Atsuta; H, Chitose; I, Kitami

were produced in the Chitose and Kitami branch office of the National Salmon Resources Center hatchery. After release, the juvenile individuals were caught off the coast of Shibetsu and Shiraoi with small round hall, pair trawl, and set nets. Young chum and pink salmon were caught in the Sea of Okhotsk with the trawl net. During feeding or homing migration, salmon were caught in the North Pacific Ocean with a drift gill net in a joint investigation between Japan and Russia. The spawning individuals were caught off the coasts of Shari and Atsuta with the set nets; they were also caught in the Shari River, the Tokoro River, and the Chitose River. The near position of a sampling area is shown in Fig. 1.

2.1.2 Analysis of Composition of Visual Pigment

Visual pigments consist of a chromophore group with a specific protein, opsin, in a lipoprotein complex. Various kinds of pigments with different absorption spectra are distributed over the whole animal kingdom, although opsin differs interspecifically. Spectral sensitivity in scotopic conditions can be analyzed by measuring the absorption spectrum of visual pigment located in the outer segments of the rods. Photosensitive visual pigments are classified into rhodopsin, a vitamin A_1-based pigment, and porphyropsin, a vitamin A_2-based pigment. Most marine fishes have rhodopsin, and most freshwater fishes have porphyropsin. The absorption spectrum of visual pigment that each fish has is closely related to the light conditions in the fish's environment. For example, in the absorption spectrum of a fish living in the deep sea, the λmax (wavelength of peak absorbance) is shifted to the short wavelengths compared to that of a coastal species. Some fishes have both visual pigments in the retina. The proportions of these two visual pigments can change with habitat. For example, the rhodopsin of Ayu *Plecoglosus altivelis* (Temminck & Schlegel) dominates the retina while at sea, whereas porphyropsin is dominant while in fresh water (Hasegawa and Miyaguchi 1997). Λmax of chum and pink salmon is 503 nm in rhodopsin and 527 nm in porphyropsin.

Retinal and 3-dehydroretinal, which are the chromophores of visual pigment, were analyzed by essentially the same method as reported by Suzuki and Makino-Tasaka (1983). An entire eye was homogenized with 200 µl of 1 mol NH_2OH (neutralized with 1N KOH) and 1 ml methanol to form retinaloxime and 3-dehydroretinaloxime. After the solution had been cooled by ice for about 5 min, 1 ml dichloromethane and 0.5 ml of distilled water were added and mixed vigorously. Then, 2 ml of n-hexane was added, and the mixture was centrifuged at 3,000 rpm for 5 min at 4°C. The chloromethane/hexane layer, which is the top part of the solution, was transferred to another tube by a pipette. One milliliter of dichloromethane and 2 ml of n-hexane were added to the residue again. The extraction was repeated two times. The extracts were combined, evaporated under vacuum, and dissolved in 300 µl of elution solvent for HPLC analysis with 7% ether in n-hexane containing 0.075% ethanol. The flow rate of elution solvent was kept constant at 0.6 ml min^{-1} by a Shimadzu LC-10A. HPLC was carried out with a Shimadzu HPLC system SPD-10AV equipped with a Sorbax SIL (4.6 × 150 mm). Peak area was determined by integrating the absorbance at 360 nm with a Shimadzu Chromatopac CR-6A. Examples of the chromatogram of chum salmon just before releasing are shown in Fig. 2.

2.2 Swimming Energy of Salmon Fry

2.2.1 Consumption of Oxygen

A total of 745 chum salmon fry *Oncorhynchus keta* (Walbaum) (53.7 ± 6.7 mm fork length and 1.6 ± 0.5 g body mass; means ± SD) that had been bred in the

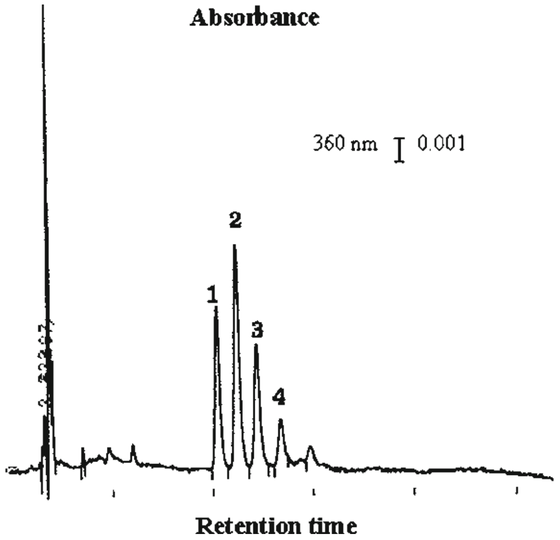

Fig. 2 Example of the chromatogram of chum salmon just before releasing 1,11-cis-retinaloxime; 2,11-cis-3-dehydroretinaloxime; 3, all-trans-retinaloxime; 4, all-trans-3-dehydroretinaloxime

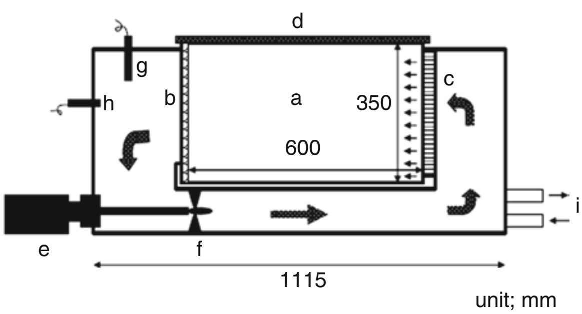

Fig. 3 The experimental apparatus. a, Swimming tank; b, net lattice; c, rectification lattice; d, lid for sealing up; e, motor; f, impeller; g, DO electrode; h, water temperature sensor; i, temperature adjustment circulation valve

Chitose branch of the Salmon Resources Center were used for the experiment. About 150 fry were made to swim for 5 h at water velocities of 4.7, 9.3, 14.1, 18.8, and 28.1 cm s^{-1} in the water tank shown in Fig. 3. The change in the oxygen concentration in the tank was continuously recorded with a DO meter (KRK DC-300, Saitama, Japan). The change of the DO value during the 5 h (300 min) was estimated from the difference between the average of 50 measurements over 5 min immediately after the start of the experiment and the average of 50 measurements over 5 min near the end of the experiment. All fish were replaced at the end of each water velocity trial. All experiments were carried out at a water temperature of 10.2 ± 1.3°C (mean ± SD).

2.2.2 Consumption of Caloric Body Energy

One hundred fifty chum salmon fry were made to swim for 98 h in the water tank at a water velocity of 28.1 cm s^{-1} and a water temperature of 10.0°C. Another 149 individuals that had been bred under similar conditions were used as a control population to measure the total value of body energy before the experiment. The 299 fish used in the experiment were measured (fork length), weighed (body mass), and their condition factor calculated from ([body mass (g)ε(fork length (mm))$^{-3}$] × 10^6). After homogenization, all individuals were freeze-dried, and 1 g sample of each of them was used to measure their body energy using a calorimeter (Ogawa Sampling OSK200, Saitama, Japan).

3 Results and Discussion

3.1 Bio-resources Characteristics Considered from the Changes in Rhodopsin Ratio

Change of visual pigment composition in chum and pink salmon is described in Fig. 4 for every life stage. The first life stage in the hatchery is from the eyed egg until before release. The rhodopsin ratio in each species slightly exceeded 50%. The ratio decreased sharply to 30% at the time of hatching. It increased gradually after that and recovered to nearly 40% at surfacing fry. The rhodopsin ratio continued to increase further until it was released from the start of predation behavior after the fish were moved to the breeding pond, where it reached about 70–80%. This change in composition tendency was the same in both species.

However, the rhodopsin ratio of pink salmon increased at all stages in this life stage compared to that of chum salmon. The second life stage is the period when the fish live in the sea. The between-individual difference of visual pigment composition (standard deviation, SD, of the rhodopsin ratio) of both species was largest

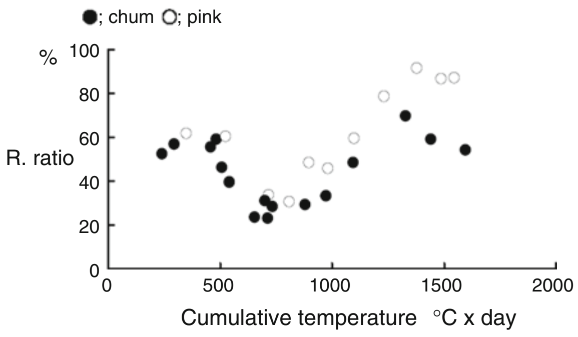

Fig. 4 Composition changes in retinal pigments in the hatchery

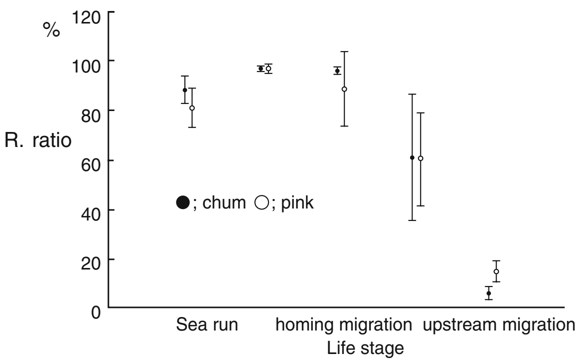

Fig. 5 Composition changes in retinal pigments after release. Sea run comprises two locations: (i) at the mouth of the Shikiu River, offshore Shiraoi and offshore Shibetsu; and (ii) Shari coast and Atsuta coast. Upstream migration comprises the Shari River, Tokoro River and Chitose River, Tokoro River and Chitose River. Five points are the average value, and error bars are standard deviation of all samples in each location

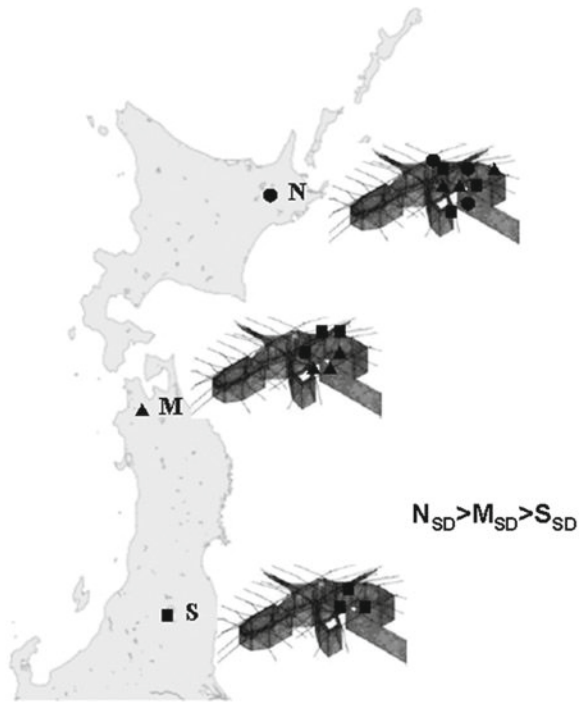

Fig. 6 Standard deviation of rhodopsin ratio reflected in the catch rate of each local origin (N, M, or S)

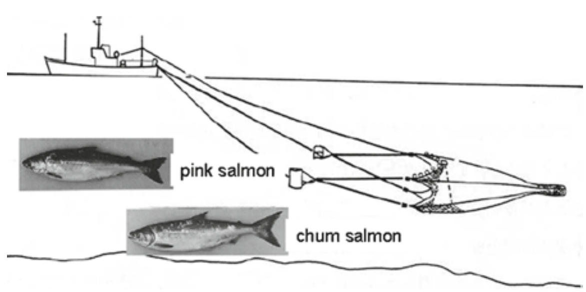

Fig. 7 Difference of swimming depth estimated by the rhodopsin ratio of both species

in the case of the group caught with the set net when they had returned to the coast for spawning at the life stage before upstream migration (see Fig. 5). It is thought that there is a downward tendency in the rhodopsin ratio of the fish just before they ascend to the river for spawning, and its higher value in the fish that go further south is maintained.

In the set net, which catches fish that have carried out the homing migration, individuals that were continuing their migration further to the south could be captured. The number of fish of local origin can possibly be estimated correctly using the difference in the rhodopsin ratio of the individuals caught with each set net laid on the Japanese coast (Fig. 6).

The area of the third life stage is the river, where the fish carry out spawning behavior. The rhodopsin ratio decreased sharply in this period. The rhodopsin ratio of chum salmon was always higher than that of pink salmon in the sea-life stage. Judging from the knowledge about the relation between the spectral sensitivity of fishes and their habitation water depth, the selected water depth of chum salmon in sea water may be deeper than that of pink salmon. The difference between both species in selected water depths may be the factor that should be taken into consideration when estimating the resources of both species in the ocean (Fig. 7).

3.1.1 Swimming Energy of Salmon Fry

Oxygen demand measured at the five swimming speeds is shown in Table 1. From Table 1, at a swimming speed of 28.1 cm s^{-1}, 1.47 mg l^{-1} of oxygen was consumed. The capacity of the tank was 172.18 l, so a total of 253 mg (1.47 mg l^{-1} × 172.18 l) of oxygen was used, which is equivalent to 0.177 l because the weight of 1 l of oxygen is 1.429 g. Furthermore, because the calorific content of 1 l of oxygen is 4,800 cal (Schmidt-Nielsen 1984), the 149 fish consumed a total of 850 cal (4,800 cal l^{-1} × 0.177 l) in 5 h or about 27.4 cal day^{-1} per individual (850 cal (149 individuals)$^{-1}$ × 24 h (5 h)$^{-1}$). The energy usage at the other swimming speeds was similarly calculated. The results, converted to joules, are shown in Fig. 8, where 1 cal = 4.1868 J. The relationship between the amount E (J g^{-1} day^{-1}) of energy consumed during a day and swimming speed U (cm s^{-1}) is given by:

$$E = 20.735U^{0.4519} \quad (1)$$

The coefficient of determination (r^2 = 0.952) of Eq. (1) is greater than that of the exponential equation used by Brett (1964) and Kaufmann (1990), so Eq. (1) was used in the present study for the following analyses.

The experiments described above were designed to estimate the amount of energy consumed by chum salmon fry when swimming using a hydrodynamics model. When a fry swims, it is subject to drag from the water. The drag is calculated in dynes (D) from the expression

$$D = {}_{\rho}S_{w}U^{2}C_{D}/2 \quad (2)$$

where ρ is water density(g cm^{-3}), S_W is wetted surface area (cm^2) of the fry, U is swimming speed (cm s^{-1}), and C_D is the drag coefficient (Webb 1975).

When a salmon fry's shape is assumed to be an ellipsoidal body, the surface area is represented by the elliptic type, considering the wither height, body

Table 1 The characteristics of the experimental fish and their oxygen demand at five swimming speeds

Swimming speed (cm s^{-1})	4.7	9.3	14.1	18.8	28.1
Fork length (cm)	5.2 ± 0.72	5.2 ± 0.73	5.4 ± 0.53	5.4 ± 0.77	5.6 ± 0.50
Body height (cm)	0.91 ± 0.16	0.96 ± 0.18	0.99 ± 0.13	0.98 ± 0.16	1.07 ± 0.11
Body width (cm)	0.57 ± 0.10	0.57 ± 0.11	0.60 ± 0.07	0.58 ± 0.10	0.64 ± 0.09
Body mass (g)	1.44 ± 0.54	1.50 ± 0.55	1.68 ± 0.45	1.67 ± 0.58	1.84 ± 0.43
Number of fish	154	147	148	147	149
Oxygen consumption (mg/1/5 h)	0.54	0.80	0.96	1.26	1.47

Mean ± SD

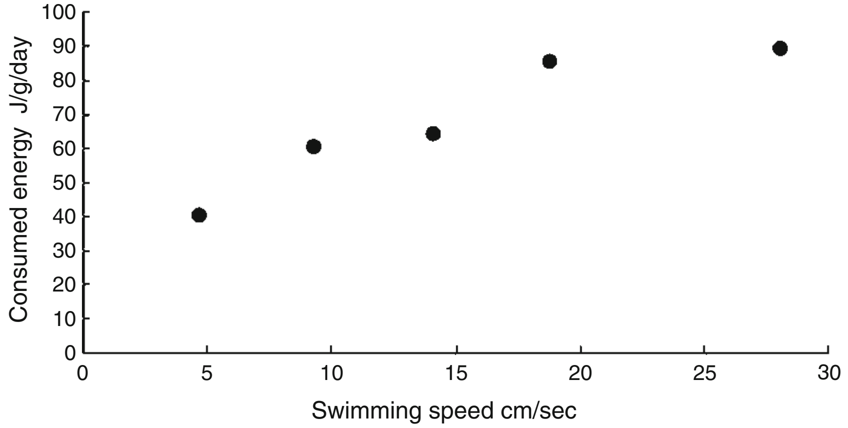

Fig. 8 Relation between E (consumed energy per day per 1 g of body weight) and U (swimming speed) of chum salmon fry

length, and body width. Because body mass is calculated using the wither height, body length, and body width, the area of body surface becomes a function of body mass. Figure 9 shows the relation between S_W and body mass; the equation becomes

$$S_w = 7.9135W^{0.6825} \quad (r^2 = 0.970) \tag{3}$$

The mechanical work (erg) of chum salmon fry done during a day is given by (distance moved [cm] during a day × D[dyn]). Because the distance moved during a day is 86,400 U (cm) (1 day = 86,400 s), and D(dyn) = 1/2 × 1 (g cm^{-3}) × $7.9135W^{0.6825}$ (cm^2) × U^2 (cm s^{-1}) × C_D, the mechanical work done by a fry in a day is $34{,}186.32W^{0.6825}\,U^3\,C_D$ (erg) per unit body mass (g). That is, the mechanical work of $0.03418632W^{-0.3175}\,U^3\,C_D$ (J) is done in a day. If the average value of body height and body mass of fry used in the experiments at each swimming speed is substituted in the theoretical formula (2), the energy consumed during a day is $3.14C_D$ (at U = 4.69 cm s^{-1}), $24.81C_D$ (at U = 9.38 cm s^{-1}), $80.59C_D$ (at U = 14.06 cm s^{-1}), $191.49C_D$ (at U = 18.75 cm s^{-1}), and $627.02C_D$ (at U = 28.13 cm s^{-1}).

When the measured values shown in Fig. 8 are the same as the theoretical value, C_D is the drag coefficient for each swimming speed. That is, it becomes $C_D = 632.05U^{-2.5041}$ ($r^2 = 0.999$) (Fig. 10). When this C_D is substituted in the theoretical formula, the follow equation is obtained:

$$E(U,W)(Jg^{-1}day^{-1}) = 21.607W^{-0.3175}(g)U^{0.4959}(cm\ sec^{-1}) \tag{4}$$

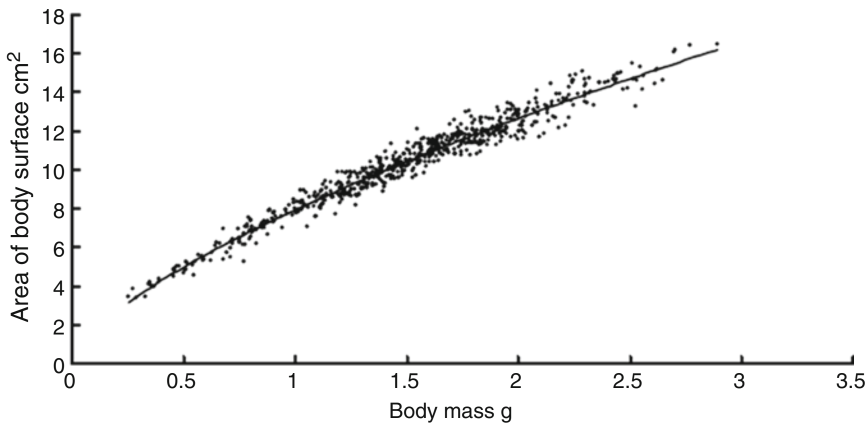

Fig. 9 Relations between body height and body mass (a), and body width and body mass (b)

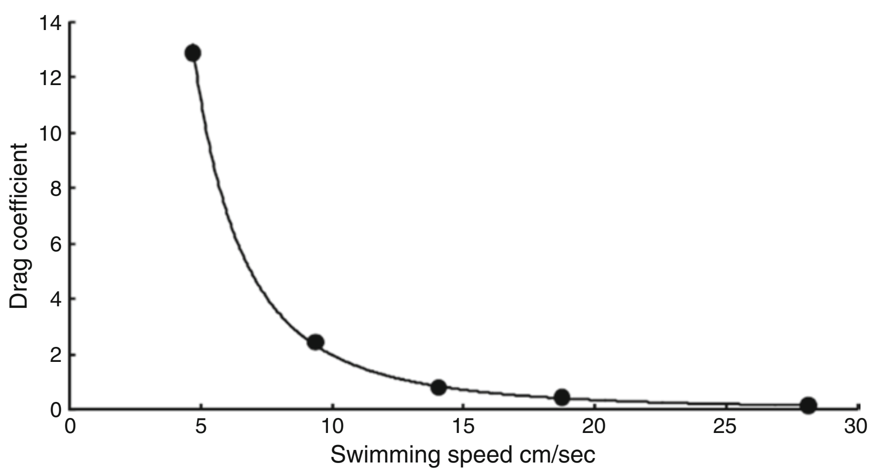

Fig. 10 Relation between drag coefficient and swimming speed of chum salmon fry

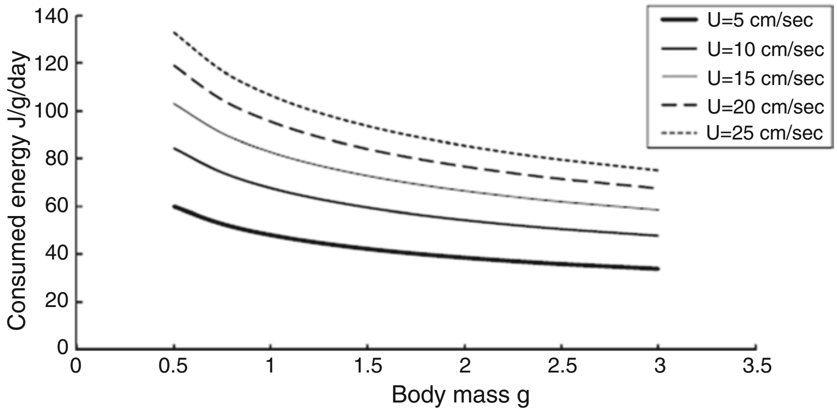

Fig. 11 Relation between energy consumed and body mass at different swimming speeds

This expression gives the energy consumed per unit body mass when the fry swims for 24 h. Figure 11 shows the relationship between energy consumption per day at different swimming speeds and body mass.

Webb (1975) measured a fish's drag coefficient under several conditions, such as varying body size, rigidity, and with the paired fins amputated as possible sources of error in dead-drag measurements at each swimming speed. For the relation between swimming speed and C_D in sockeye salmon *Oncorhynchus nerka* (Walbaum), Brett (1963) indicates that it is comparatively small and the rigidity level is low (length 18.6 cm and 47.9 g in body mass), showing a similar tendency to the result obtained in the experiments described here. It is understood that C_D decreases as swimming speed increases.

Next, the energy consumption value calculated by this theoretical formula is compared with the decreased value of the body energy of the fry measured after they were made to swim in the water tank. The fork length, body mass, and condition factor of the 149 chum salmon fry chosen as the control group (individuals measured before swimming) were 51.3 ± 4.7 mm, 1.5 ± 0.5 g, and 10.7 ± 0.9 (means ± SD), respectively. The equivalent values of the 150 individuals after experimentation were 51.3 ± 4.2 mm, 1.3 ± 0.4 g, and 9.7 ± 0.7 (means ± SD).

An analysis of variance (ANOVA) of the fork length of both groups of individuals was done to determine whether the individuals measured before and after the experiment had been drawn from the same statistical population. Both the ANOVA and a Student's *t*-test of fork length were not significant ($p > 0.05$), and so it was concluded that both groups of individuals had been selected from the same population.

Next, the variance of body mass and the condition factor of both groups were examined by ANOVA, and the null hypothesis that there was no significant difference between these two variables was rejected ($p < 0.05$). Welch's *t*-test of the difference of both mean values was then done, and both mean body mass and average condition factor were found to be significantly different ($p < 0.05$) before and after the experiment, both values being lower after the experiment.

The body energy of both populations measured with the calorimeter was 5,634 cal g^{-1} (dry weight) in the case of the control group (before swimming) and 5,664 cal g^{-1} (dry weight) for individuals after swimming. However, because the water content of the former was 80.3%, and that of the latter 80.8%, the average body dry masses were 0.2955 g (1.5 g × 0.197) before swimming and 0.2496 g (1.3 g × 0.192) after swimming. Therefore, the calorific content was 1,665 cal (5,634 cal × 0.2955) before swimming and 1,414 cal (5,664 cal × 0.2496) after swimming, expressed as calories per individual, so that 251 cal (= 1,050 J) had been used in swimming.

The amount of energy consumed by a fish of W = 1.5 g after swimming for 98 h at U = 28.1 cm s^{-1} calculated using Eq. (4) is 405.6 J in 98 h. On the other hand, if the energy used is calculated by measuring the consumption of calories, a value of 1,050 J per individual is obtained. That is, the estimate of the energy consumed calculated using equation (4) and combining oxygen demand and hydrodynamic theory is not similar to that measured with a calorimeter. However, the value almost corresponded to the energy requirement of the hunger test (unpublished data).

Moreover, the oxygen demand increases exponentially with the water temperature (Brett 1964; Clark and Seymour 2006), so temperature should always be given when energy consumption values are reported. The relationships between a certain amount of the food energy in the sea area and the number of salmon fry will be determined by considering the water temperature, the amount of energy of food, and the energy utilization efficiency, etc., in this research in the future.

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Trials on New Methods for Seed Culture in Japanese Abalones

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Abstract Abalones are one of the most important coastal animal resources in Japan, and the study of their seed culture has a long history of more than 40 years. Most of the produced seeds have been released to the seabeds. The total number released nowadays is almost 3 million (small individuals) per year. However, the annual production of natural abalones has decreased remarkably in the last 10 years.

Still, the culturing of market-size abalones has been started in several regions in Japan, such as *Haliotis discus* in the south and *H. discus hannai* in the north. The production of cultured abalones is slowly increasing. This tendency has the benefits of preventing over catching of natural stocks and reducing the importation of cultured abalone from foreign countries.

Under these conditions, we tried to improve this trend with some experiments on seed culture methods.

1. Using artificial food (a microparticle diet) to replace natural diatoms for feeding post-larval abalones to improve the survival and growth rates
2. Trials for the improvement of artificial production of inter-specific hybrids among three large size species

The result will lead to recommendations for newly developed adult cultures in closed spaces. The possibility of producing regional specialities is suggested.

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1 Introduction

Abalones are one of the most important coastal animal resources in Japan, and the study of their seed culture has a long history of more than 40 years. Most of the produced seeds have been released to the natural seabed for restocking. Currently, the total number of released seed abalones is almost 3 million per year. However, the annual production from natural stocks has decreased remarkably in the last 10 years.

The culturing of market size abalone has been started in several regions, such as *Haliotis discus discus* in the south of Japan, for example, in the Matsuyama region in Ehime Prefecture, and *H. discus hannai* in the north, for example, in Hokkaido and in Kesennuma in Miyagi Prefecture. The production of cultured abalone is increasing progressively. This tendency has the benefits of preventing over catching of natural stocks and reducing the importation of cultured abalone from foreign countries.

Under these conditions, we tried to improve this trend with some experiments on seed culture methods as follows. The first experiment involved replacing natural food with microparticle artificial food to feed post-larval abalones. A new post-larval abalone culture system, Stott's abalone post-larval production system (SAPPS), was tested using commercially available artificial diets. The second experiment involved improving hybrids among the three principal species. These experiments were conducted from 2000 to 2005 at Tateyama Station-Banda, Tokyo University of Marine Science and Technology (former Banda Marine Laboratory, Tokyo University of Fisheries).

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2 First Experiment

We used artificial food (microparticle diet) to replace natural food (diatoms) for feeding post-larval abalones to improve the survival and growth rates.

2.1 Materials and Methods

The experiment was conducted over a period of 4 weeks to test the SAPPS method (Fig. 1) against the Diatom method (Table 1). Two plates from each treatment of SAPPS-Cos (diet supplied by Cosmo, Matsuyama, Japan), SAPPS-Adam (diet supplied by Adam and Amos, Mt Barker, SA, Australia), and Diatom were placed into 10-l flow-through tanks (three treatments replicated three times each). Brood stock at Banda Marine Laboratory was induced to spawn using UV light, and the resulting larvae were used in the trial when they were deemed ready to settle. Once the larvae had metamorphosed into post-larvae (on the experimental plates), the trial was commenced to compare SAPPS (using the two commercial diets) to Diatom in terms of post-larval growth and survival. Food was resprayed onto plates every second day, and measurement for post-larval survival was calculated every second day.

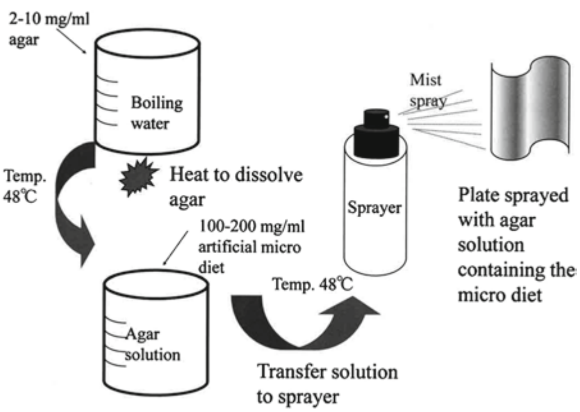

Fig. 1 Stott's abalone post-larval production system (SAPPS)

2.2 Results and Conclusion of the First Experiment

The Cos diet, when compared to the Adam diet, had higher moisture, lipid and ash contents, and lower protein content (Table 2). Final survival of post-larvae was significantly higher in SAPPS-Cos (56.7 ± 11.15%) compared to the other three treatments [9.4 ± 2.7%, 8.5 ± 1.1% and 3.0 ± 2.4% for the Diatom, SAPPS-Adam and PPS-Adam diets, respectively. The final length of post-larvae in SAPPS-Cos (1,065 ± 73 μm) was significantly higher than those in the other treatments (average of 812–883 μm) (Table 3, Fig. 2).

The Cosmo diet was superior to the Adam diet in terms of growth and survival of post-larval abalone. The level of protein in the Adam diet could have been too high, and the level of lipid too low. SAPPS was

Table 1 Main species and density of diatoms that were present on the collector plates of spats

Species	Cell density (%)	Final density (cells/cl)
Navicula spp.	40	8.4×10^4
Nitzschia spp.	3	6.3×10^3
Amphora bigibba	12	2.3×10^4
Entomoneis sp.	3	6.3×10^3
Melosira nummuloides	2	4.2×10^3
Diploneis sp.	1	2.1×10^3
Tabularia sp.	2	4.2×10^3
Amphora spp.	4	8.4×10^3
Cyclophora tenuis	3	6.3×10^3
Others	30	6.3×10^4
Total	100	21 ± 0.3 (1×10^6) (Scott et al. 2004)

Table 2 The size and composition of the different diets

	Diet *		
	Adam *	Cosmo *	Diatom
Particle size – wet (μm)	43 ± 16	38 ± 13	–
– dry (μm)	32 ± 10	28 ± 8	–
Moisture (%)	7.5	6.8	1.6
	On dry matter basis (%)		
Crude protein	54.4	34.3	18.3
Crude lipid	2.5	4.9	8.1
Crude ash	14.1	17.1	52.8 (Scott et al. 2004)

*Diet Adam: supplied by Adam and Amos; Mt. Barker, SA, Australia

Diet Cosmo: supplied by Cosmo; Matsuyama, Japan

Table 3 Results in the three different treatments for *Haliotis discus discus*

Treatment	No. larvae settled	Final Shell length (μm)	Daily growth rate (μm)	Survival rate (%)
SAPPS – Adam	377	841	19	8.5
PPS – Adam	243	812	18	3.0
SAPPS – Cos	334	1,065	27	56.7
Diatom	381	883	21	9.4
				(Scott et al. 2004)

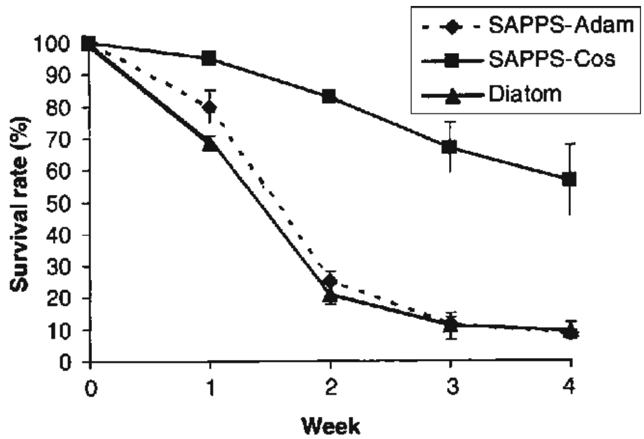

Fig. 2 The average percentage survival (± standard deviation) of postlarval *Haliotis discus* in the three different treatments (trial 1)

superior in terms of growth and survival of post-larvae when compared to the other two methods. This is possibly because SAPPS is a continuous system where artificial food is resprayed onto wet plates that have living post-larvae attached. There is potential for SAPPS to be used instead of, or together with, the current diatom method in the production of post-larval abalone.

3 Second Experiment

Trials were conducted to improve the artificial production of interspecific hybrids among three principal species.

3.1 *Materials and Methods*

Adult *Haliotis discus discus* (HDD), *H. gigantea* (HG) and *H. madaka* (HM) were collected from Tateyama Bay, in the offing of Tateyama Station-Banda. Crossbreeding between these principal species was attempted in several trials from 1985 to 2002. Spawning was induced with ultraviolet-irradiated seawater and heat shock, common methods used in the abalone hatcheries in Japan. The fertilization rates of homologous crosses and the hybrids were estimated. The produced hybrids were reared in separate tanks, avoiding mixing with the other hybrids or pure strains.

For the reproductive viability of the hybrids, HM×HDD and HDD×HM hybrids were induced to spawn. The fertilization rate and number of hatched trochophore larvae were estimated, and development of larvae was monitored to observe progeny viability of F2HMxHDD and HDDxHM hybrids. For the gonad characteristics of the hybrids, gonads of at least 3-year-old abalones were collected from September 2005 to January 2006. The fixed gonads were sectioned at a thickness of 8 μm and stained with Mayer's hematoxylin and eosin. Slides were observed to establish sex, the most abundant gametogenic cell type present and the most advanced gametic cell type present in each abalone. The gametic cell types and maturity stages for male and female abalones were identified following the method of Tomita (1967/68).

3.2 Results and Conclusions

All hybrids responded positively to spawning stimulation [HM×HDD (two males, one female); HDD×HM (four males, three females); HDD×HG (three males, two females) spawned] (Fig. 3). Fertilization rates of homologous gametes of *H. discus discus* and *H. madaka* were on average more than 80%, but they were significantly lower in *H. gigantea*. On the contrary, fertilization rates of heterologous gametes were less than 20% except the reciprocal crosses of *H. discus discus* and *H. madaka* (22.4% in DDxM, 60.8% in MxDD) (Table 4). For the F2 generation, the fertilization rate was averaged at 56% (40.9–68.5%).

In the back-crossing, fertilization rates averaged 45.6% (HDD-HDD×HG), 70.6% (HDD×HG-HDD) and 3% (HG-HDD×HG). The larvae followed the same development stages as described for abalones. At the end of 5 months of rearing, the F2 hybrids were, on average, 9.9 mm (4.94–13.58) in shell length and 0.14 g (0.04–0.3) g in body weight.

Table 4 The fertilization rate of homospecific, heterospecific, hybrid and back-cross (Faruq 2007; Koike et al. 1988)

Parents		No. of	Fertilization rate (%)	
Female	Male	trials	Average	Range
		Homospecific crosses		
DD	DD	6	90	71.4–100
M	M	5	81	19–96.2
G	G	7	34	0.3–86.7
		Heterospecific crosses		
DD	M	3	22	6–65.3
M	DD	3	61	3.4–97.6
G	M	3	2	0.4–8.5
M	G	3	5	1–9.6
G	DD	5	20	0.8–53.6
DD	G	4	1	0.5–4.2
		Hybrid crosses		
DD×M	DD×M	1	59	40.2–87
M×DD	M×DD	1	69	58.9–76.3
DD×G	DD×G	1	41	38.5–45.1
		Back-crosses		
DD	DD×G	1	46	42–53.3
DD×G	DD	1	71	64.7–74.2
G	DD×G	1	3	0.9–5.6

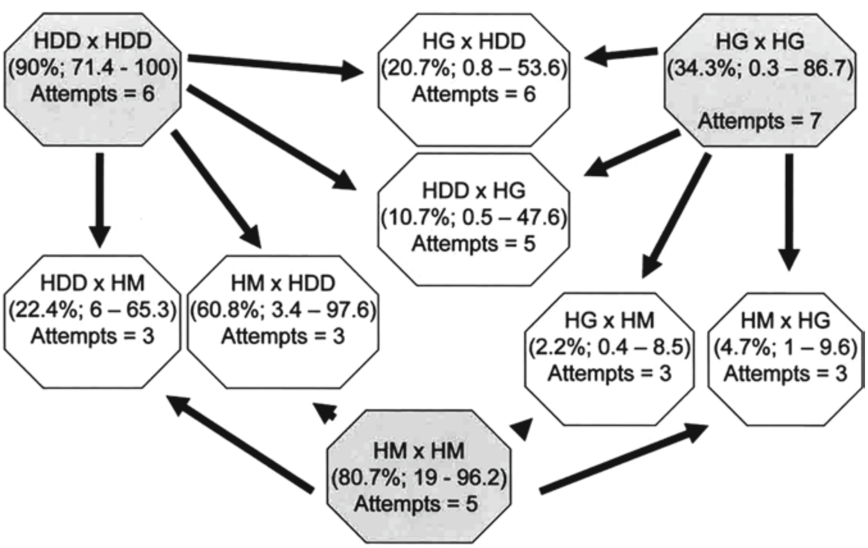

Fig. 3 Gamete combination (Female × Male) and fertilization rate [mean (%); range] of each combination (HDD = *H. discus discus*, HM = *H.madaka*, HG = *H. gigantea*) (Faruq 2007)

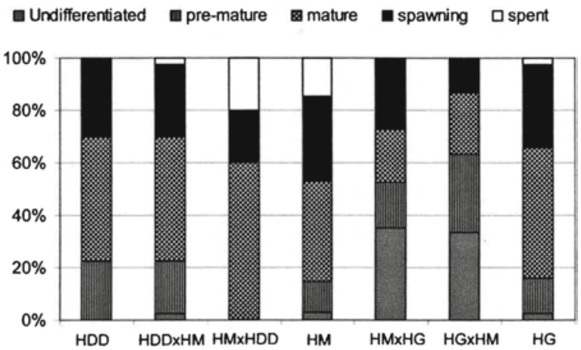

Fig. 4 Maturity stages in the three abalone species and their hybrids (HDD = *H. discus discus*; HM = *H. madaka*; HG = *H. gigantea*) (Faruq 2007)

Most hybrid abalones sampled for gonad analysis could be sexed. Comparison of gonad histology of the parental species with the hybrid suggested that the hybrids had similar gonad development as the parental species (Fig. 4). Hybrids in spawning stage were observed in the spawning season of their parental species from December to January.

It was observed that both male and female hybrids were able to produce and spawn gametes when they were stimulated by the conventional procedures. All four hybrid crosses for which histological observation of gonads was conducted were found to be proceeding through gametogenesis.

4 Conclusion

In conclusion, hybridization among principal abalones *H. discus discus*, *H. gigantea* and *H. madaka* is possible, with crosses between *H. madaka* and *H. discus discus* being relatively easier to accomplish than the other four crosses. The former experiment proved that *H. discus discus* female × *H. madaka* male hybrids had growth performance similar to and in some cases higher than the parental species (Faruq 2007). This can be useful in attempts to produce back-crossed and F2 hybrids to obtain fast-growing strains.

To identify the hybridization and parenthood of the hybrid, analyses of allozymes and mtDNA are effective, and these aspects will be examined in future studies.

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A Multidisciplinary Approach for Anticipating the Presence of Genetically Modified Fish in France

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Abstract Transgenic aquaculture species with improved growth rates are at the premarket stage in the United States and may be produced in countries with less drastic regulations for environmental protection. The French market is widely supplied by imports from all over the world. Regarding the crisis provoked by plant GMO (*genetically modified organisms*) production, rumors of the unauthorized importation of GMF (*genetically modified fish*), even if fortuitous, would undoubtedly have detrimental effects on public confidence, and consequently on fish market and innovation systems.

To anticipate such a situation, the DOG.M.ATIS project (2007–2010), funded by the French Research Agency (A.N.R.), proposes to develop dedicated strategies through multidisciplinary approaches and to deliver:

– A critical analysis of the technological reality and expected evolution of GMF by characterizing the impact of the transgene and transgenesis technique on transgene stability and flow

Development of some methodologies and possible routes of GMF detection, based on detection strategies for plants and establishing specific methods for GM fish

An assessment of the risk of the fortuitous presence of GM fish in our markets by crossing the data from the scientific literature and an expert analysis of filtered statistics of international trade

An assessment of public perception levels, using focus groups for analyzing the opinions of citizens and performing interviews with businesses and NGOs

Development of drafts for dedicated regulations and laws, developed based on an analysis of the current regulations for GM organisms and of the gaps existing within the reality of GM fish and the fish market chain

– A description of examples of GMF and the ethical implications, moving beyond the very classical–theory-driven–scientific descriptions used in the GMO debate up to now

DOG.M.ATIS is a network of scientists in various fields: fish genetics and transgenesis, GMO detection,

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fish market economy, consumer sociology, law, philosophy, and epistemology. We propose to unravel the multiple dimensions of GMF with progressive interdisciplinary approaches that will deliver results to be exploited both by experts in the different disciplines involved and by the overall network.

1 Technological Reality

Since the first publication in 1987, more than 1,400 articles have been published dealing with transgenic fish. The first technique employed was micro-injection into one-cell-stage embryos. It is operant, but lacks efficiency: the first generation is mosaic; neither the copy number nor the genomic location of the transgene is controlled. This leads to (1) the inevitable production of two generations before the transgenic line can be stabilized, (2) the lack of control of transgene expression, and (3) the selection of an integration event. This technique is still the easiest and most employed, and transgenesis has been carried out on the main aquaculture species and model fish. For most of the cases, transgenic aquaculture species are initially produced in research laboratories to study the main functions (growth, reproduction, disease resistance). The number of transgenic model fish lines produced has largely increased in recent years because of gene function and regulation studies. This leads to a strong dynamic for improving transgenesis efficiency: techniques for mass transformation, facilitation of genomic integration, and control of the site of integration are currently being tested. The patents published so far (90 references) serve five different purposes:

- Improved livestock production (40%)
- Bio-protein production (20%)
- Dedicated biosensors, sentinel fish (10%)
- Appearance of ornamental fish (6%)
- New techniques (24%)

The risk of fortuitous presence is a reality, and transgenesis production and purposes will rapidly evolve. Our strategy is:

(i) To quantify the actual risk by merging results obtained so far on transgenic fish with economic data
(ii) To anticipate the evolution of GM fish by characterizing the impact of the transgene and transgenesis techniques on transgene stability and flow

2 Risk of Fortuitous Presence and Effects on the Market

World aquaculture production is growing tremendously (from 1 Mt in the 1950s to 63 Mt in 2005, including plants) and within 5 years will probably comprise 50% of the seafood consumed. The European Union imports 50% of the consumed aquatic products, and half comes from non-European countries.

For finfish, in 2005, the EU (15) imported 5.6 Mt (€16.6 billion) and showed a foreign trade deficit of 1.6 Mt and €6.5 billion. European markets, and notably those in France, are widely supplied by imports from all over the world, among which farmed fish make up an increasing proportion. The import of GM fish, if commercially produced in an exporting country, thus appears to be possible in the near future. Considering the impact of the issue of GM plants, it is very likely that such an event would cause a crisis in consumer confidence.

To estimate the actual risk of fortuitous import of GMF, we use a crossed methodology with several steps:

- Using the available scientific information (gathered in the GMF database and through bibliographic synthesis) and our expertise, we aim to (i) identify the species and countries concerned by trangenesis, and (ii) score the probability of commercial production and export, considering the state of the fish culture sector and foreign trade, and the regulatory framework
- Analyzing the import statistics of fish in France and Europe, we try to point out the trade flows likely affected by GM species at the premarket stage and estimate the possible risk of finding GM fish; this analysis requires a good knowledge of the organization of the European supply chain and of the aquaculture sector in the countries of origin, as the production mode (capture or culture fisheries) is not mentioned, nor the species for some taxa like tilapia, in the foreign trade statistics

Fish is a product known to varying degrees by European consumers. The French are generally do not eat much fish: fish consumption requires socialization, and most consumers look for advice from vendors and need to feel confident about what they buy. When buying fish, they take into account the way the fish was produced and commercialized. Mentioning the existence of GMF within this particularly fragile context could lead to extreme reactions. Therefore, we plan to develop methodologies that will highlight the perceptions of the public and the professionals, and the collective building of risk perception, instead of trying to measure the discrepancy between perceived risks and real risks. We assume that the consumers and actors have practical knowledge that deserves analysis. We aim to try to anticipate the changes in consumer behavior when faced with information from the media, NGOs, government, and businesses.

Regarding the consumers, we analyzed purchase situations by organizing consumer focus groups where we invited the consumers to talk about fish, and then GM fish, following a list of subjects developed in a collaboration among sociologists, economists, and philosophers.

The first results of the focus groups that have already been performed led to three points: (i) a variability in consumer perception of "natural" and "artificial;" (ii) some unusual ideas of consumers about the GMF research sector; (iii) some new forms of criticism of the oppression and marketing characterizing present society, notably concerning health and the environment.

Our study also includes in-depth interviews of economic actors and representatives of NGOs about their perception of GM fish, both as a possible innovative technique in fish culture and as a possible new food product.

3 GMF Regulation

The main concepts of the European directive (2001/18) on the deliberate release of genetically modified organisms into the environment are:

(i) Diversity of the evaluation criteria
(ii) Assessment of possible cumulative effect
(iii) Bio-security monitoring plan
(iv) Traceability
(v) Assessment of the benefits and
(vi) Public consultation

This can hardly be adapted to GMF, not only because none of the new requests can be immediately fulfilled, but also because only confinement is considered and not fortuitous presence. Neither the market chain nor the European network of GMO laboratories is ready to apply regulations 1829/2003 and 1830/2003 for GMO food and feed authorization and traceability. The regulation gap for ornamental fish is even more obvious. The European Council is currently discussing a regulation (SEC2006/421) concerning the use of alien species in aquaculture, which considers only the environmental risk and excludes the case of GMF. Considering the risk of negative perceptions of GMF by consumers, the complexity, opacity, and international dimension of the market, international trade conflicts can be easily foreseen. We propose developing a method to draw up regulation drafts in connection with public and private stakeholders. It includes the following steps:

(i) Identification of the main principles driving the norms concerning GMO.
(ii) Sustaining the production of regulations by the European Commission, and
(iii) Integration of the overall results of DOG.M.ATIS to demonstrate the dynamic interaction between public and private bodies. In addition, we will propose means and ways to ensure GMF traceability

4 GMF Traceability

For plants, analytical methods have to be provided by GMO developers, and traceability has been made mandatory to ensure reliable and cost-effective co-existence frames. Moreover, a modular approach concept has been developed to validate analytical methods in numerous food and feed matrices in a cost-effective way, a concept that is ready to be used in other areas of detection. The specifics associated with GMF detection identified so far are: the variability of the transgene copy number in the host genome, the nature of the transgene, the lack of knowledge of the transgene flanking regions, the large discrepancy of the matrices to be analyzed (from live fish or eggs to freeze-dried compounds and multicompounds food and feed), and the necessity to identify the species and the donor organism for screening methods. We will do this sequentially and on experimental samples (chosen according to the results of the risk assessment study):

1. Test the applicability of the different detection strategies used for plants
2. Analyze their cost and efficiency
3. Test their adequacy to quantify transgene presence
4. Test the possibility of multiplexing with species detection techniques
5. Optimize DNA extraction (efficiency, cost, and purity)

Table 1 Specific objectives/deliveries of the interdisciplinary work of DOG.M.ATIS

Interaction leader	Biology/transgenesis	Chemistry/detection	Law	Economy/sociology	Epistemology	Final deliverable
Biology/transgenesis		– DNA libraries– Features of the transgenes – Transgene stability/flanking regions	– Key elements for producing transgenic lines– Environmental impact/confinement – Case studies	– GMF produced: species and country of origin– Case studies	– Genome integrity– Case studies	
					Technological reality	
Chemistry/detection	– Detection protocol		– Extent and organization of traceability	– Applicability regarding the market chain– Adequacy/risk	– Adequacy/risk (completed)	
						Traceability
Law	– Impact of the GMF at the premarketable stage	– Guidance for setting up traceability		– Regulation agencies surrounding GMF– Key principles to take into account trade and consumer perception	– Key principles to consider GMF representations	
						Regulation
Economy/ sociology	– Structure of the market, trade by species and country, main products	– Species, samples and matrix to be analyzed	– Adequacy regarding market chain and consumers– Regulation agent		– Description of the market chain– Perceptions/ representations of consumer and economical actors	
					Risks of fortuitous presence	
Epistemology	– Impacts of novel transgenesis tech-nique– Definition of biological integrity	– Adequacy of the detection regarding public confidence	– Natural/artificial frontier– Animal welfare/law	– Representations and economical/market risks– Public rationality		
					GMF representations, benefits and risks, common vocabulary	

Our objective is, once protocols have been established, to propose guidance for carrying out GMF traceability taking into account the expected regulation and the features of the fish market chain.

5 GMF and Ethics

The arrival of plant GMO provoked a break. The lack of acceptance of GMO is partly explained by the fact that the technique is perceived as artificial. Biotechnologies in general raise ethical problems, and ethics is indeed a generic science of interdisciplinary frontiers. This means that these problems also have to be considered by biologists themselves. The debates on GMO organized these last years in Europe have been mostly driven by experts having a very classical–theory-driven–representation of science. The evaluation criteria came either from epistemology of the theories or from some classical philosophies of technology. Obviously, assumptions and highlights from epistemologies of models are missing. With fish, we are still upstream from an eventual crisis, but we already can confirm that the consumers have a preconception of what a GMF is. Our objective is to work out a global frame for future debates where both epistemologies of theories and models will be to some extent reunified with bioethics and technology assessment. We will (i) identify the epistemological profiles associated with GMF, (ii) analyze the connections between ethics and epistemology, considering that the epistemological requirement accompanies the ethical questions, and (iii) propose to identify the concepts discriminating the potential impacts of GMF on human representations, using an original methodology of collective work to explore probable forthcoming intellectual strategies for GMF.

6 Conclusions

We propose to unravel the multiple dimensions of GMF with progressive interdisciplinary approaches that will deliver results to be exploited both by experts in the different disciplines involved and by the overall network (Table 1). The integrated chain approach is framed to release original and dedicated methods, novel research objectives in each domain covered by the project, and strategies for stakeholders. We have already identified the ways to interact with the economic actors in the market chain and with the public. The network is already sufficiently productive to stress international connections.

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Shrimp Aquaculture: From Extensive to Intensive Rearing, the Relationship with the Environment and The Key to Sustainability

Jean-Louis Martin

Abstract In this study, we analyze the main characteristics of the different kinds of marine shrimp aquaculture with regards to their potential impact on the surrounding environment and to its sustainability. In order to evaluate the importance of the main features of the impact, we consider parameters related to rearing practices and zootechnical aspects, such as stocking density, as well as the main characteristics of the ecosystem hosting the rearing activities and the receipt of wastes. Two main features, the ecological characteristics of the surrounding environment and the presence or absence of mangroves, are emphasized. At least, based on ecological considerations, we propose a theoretical scheme allowing determination of the best position of the different kinds of aquaculture within an ecosystem in order to minimize the impact and to insure the sustainability of the activity.

1 Introduction

There are two main kinds of aquaculture, extensive and intensive, showing different characteristics concerning their functioning (zootechnical aspects) and their potential relationship with the surrounding environment. For organisms reared in an extensive way, the food comes from the surrounding environment (natural food web). For this kind of aquaculture, the main characteristic required concerning the environment is to be productive in terms of the trophic level. As a rule, extensive aquaculture does not produce wastes. Furthermore, the concentrations of nutrients in the effluents may be lower than in the in-flowing water (Hussenot et al. 2000). For intensive aquaculture, the food comes from the feed pellets added into the ponds. This type of aquaculture produces wastes. The quantity of nitrogen in the wastes can reach up to four times the quantity of nitrogen in the harvested shrimps (Martin et al. 1998; Funge-Smith and Briggs 1998) (see below about FCR). In that case, the main characteristic required by the environment is to be able to eliminate or to assimilate the wastes.

Shrimp aquaculture has an impact on the surrounding environment (Landesman 1994). One of the most important features of the surrounding environment in relation to aquaculture and its potential impact is linked to the notion of confinement (Martin 2004). Confinement is the aquatic ecosystem's capacity to change its water. Then, the level of confinement of an ecosystem is related to different kinds of parameters, such as the hydraulic characteristics (waves, tides, currents) and the geomorphology of the site (from fully closed ecosystems to fully open ones). Confinement plays a leading role in the success of aquaculture. For extensive aquaculture, it will determine the level of nutrient concentrations in the water flowing into the ponds, and thus the capacity of this water to support the natural food web leading to the shrimps. For intensive aquaculture, the level of confinement is in some cases responsible for the accumulation, or not, of the organic wastes released from the rearing ponds into the surrounding environment (Trott and Alongi 2001). As a consequence, for intensive aquaculture, the impact of organic wastes on the environment will depend on the confinement of this environment. Increasing the confinement level leads to an accumulation of organic wastes in the

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surrounding ecosystem and finally to a feedback impact of wastes within the rearing ponds.

2 Results and Discussions

The impact of organic wastes released from shrimp aquaculture on the surrounding environment has been shown (Biao et al. 2004; Das et al. 2004; Sara 2007). It depends not only on the level of its confinement, but also on the quantity of wastes introduced within this environment. This quantity of wastes depends mainly on the intensification of the rearing system. As suggested by Fuchs et al. 1998 there are three possible ways to increase the waste fluxes:

1. The size of the shrimp aquaculture farm: The impact on an ecosystem increases when an important surface of rearing ponds is located near the same ecosystem.
2. The annual number of rearing cycles: for instance, in inter-tropical countries, because of the temperature of the water, three cycles can take place each year).
3. The stocking density (according to the different rearing systems: it can reach up to 100 shrimps m^{-2} pond). The quantity of generated wastes depends on two main factors: the quantity of pellets added into the ponds and the food conversion ratio (FCR).

The FCR indicates the quantity of feed pellets used to produce 1 kg shrimp. Thus, for FCR = 2, for example, 2 kg feed pellets are used to produce 1 kg shrimp. This means that FCR = 2 leads to the formation of 1 kg organic wastes per kg shrimps harvested. Thus, in some sites hosting 1,000 ha ponds with a production of 6–10 t shrimp ha^{-1} $year^{-1}$ and a FCR = 2, the quantity of waste rejected into the surrounding environment ranges from 6,000 to 10,000 t of organic wastes. Some sites can host up to 16,000 ha ponds (Fuchs et al. 1998).

The quality of management of the rearing system may have a huge influence on the formation of wastes (Biao and Kaijin 2007). Thus, the quality of the food (Chim et al. 2001), the rhythm of feed pellet distribution, once or several times per day (Della Patrona and Brun 2007), the water renewal (Lemonnier and Faninoz 2006), and the pond water aeration (Della Patrona and Brun 2007) make a huge difference in the FCR performance (Della Patrona and Brun 2007) and consequently in the formation of wastes. One of the main parameters controlling the FCR is the stocking density. Figure 1a, shows (Martin et al. 1998) that, for ponds of the same zootechnic conditions, the weight of the shrimp at harvest decreased when the stocking density increased. At the same time, FCR increased along with an increase of the stocking density (Fig. 1b). In fact, it has been shown that in earthen ponds, the natural food chain participates to a large extent in the nutrition of the shrimp. Thus, for example, natural prey (copepods, nematods, harpacticoïds, etc.) may represents 42% of the ingested food for *Metapenaeus macieayi* (Maguire and Bell 1981), from 37% to 43% for *Penaeus* japonicus (Reymond and Lagardère 1990), more than 50% for *Penaeus monodon* (Focken et al. 1998), and up to 84% for *Penaeus subtilis* (Nunes et al. 1997).

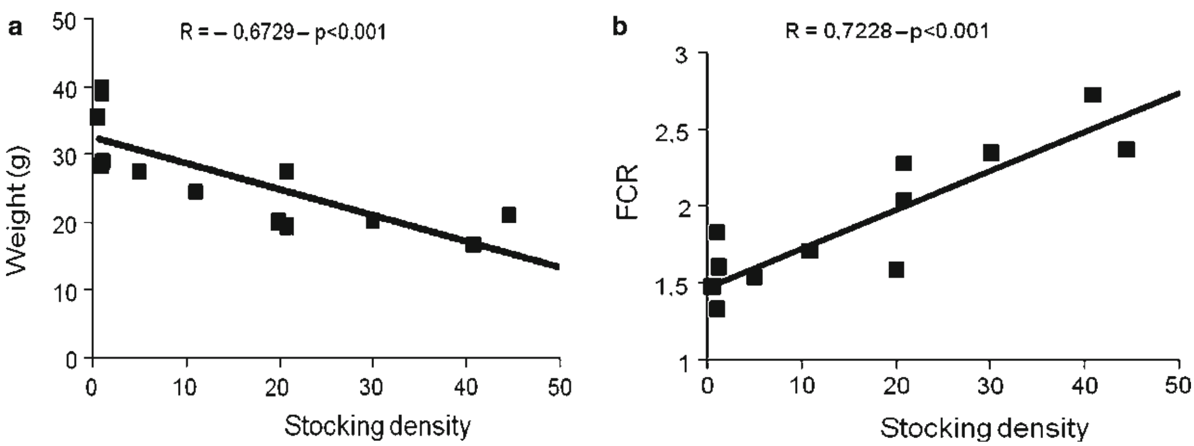

Fig. 1 Relationship between the stocking density and (**a**) the weight of shrimps at harvest, and (**b**) the food conversion ratio (FCR)

For a same species, the variation of the proportion of natural food in the total food intake is inversely proportional to the stocking density (Della Patrona and Brun 2007). This explains the increase of the FCR along with the socking density (Fig. 1b). Before being removed from the pond, with the effluent or through mechanical means, the organic wastes accumulate in the pond at the water-sediment boundary layer level. This accumulation can lead to dystrophy in the pond ecology. Thus, it has been shown (Martin et al. 1998; Lemonnier et al. 2002) that the one of the consequences of the accumulation of organic matter is an increase in the concentration of nutriments in the sediment (Burford and Lorenzen 2004). Figure 2 shows the relationship between the concentrations of N-NH_{3-4} in the 1-cm top layer of the sediment in ponds with increasing shrimp instant biomass. Thus, from very low to very high biomass (up to 800 g m^{-2}), the concentration of N-NH_{3-4} can reach up to 7,000 μM mL^{-1} in pore water. Furthermore, the accumulation of organic matter in the pond leads to a very high consumption of oxygen. Figure 3 shows, as a consequence of this consumption, the redox potential measured in sediment cores sampled in ponds with increasing shrimp instant biomass. A decrease in the redox potential occurs from the lowest instant biomass to the highest one. Thus, for the highest instant biomass, the redox potential ranged from ≈−100 to ≈−300. This very low value explains the high concentrations of nutriments measured in highly intensified rearing ponds (Suplee and Cotner 1996).

Parameters relevant to the quality of the management of the ponds may explain the variations of the quantity of wastes released from the ponds to the surrounding environment. Nevertheless, some parameters that are not dependent on management may have an influence on the rearing performances, and, as a consequence, on the quantities of wastes produced. Thus, for similar management, variations in yield have been noted in relation to the season and are mainly attributed to the difference in salinity and/or temperature and to natural food availability (Scura 1995). A decrease in daylight intensity for several days because of seasonal cloud cover has been shown to lead to a decrease in shrimp production, an increase in the FCR ratio, and an increase in waste formation (Garen and Martin 2002).

The relationship between aquaculture and environment is not only related to the level of confinement of the environment hosting the activity, and to the quantity of wastes introduced within this hosting environment, but also to the presence or not of mangroves. Needing warm water, shrimp aquaculture is mainly located within or near the tropical or equatorial areas. Furthermore, the activity needs huge surfaces to build the ponds. In order to minimize the functioning costs (pumping), the ponds are located very close to sea level. This is why during the huge development of shrimp aquaculture in the 1980s, the easiest and less expensive method for pond implantation was to eradicate the mangroves to build the ponds

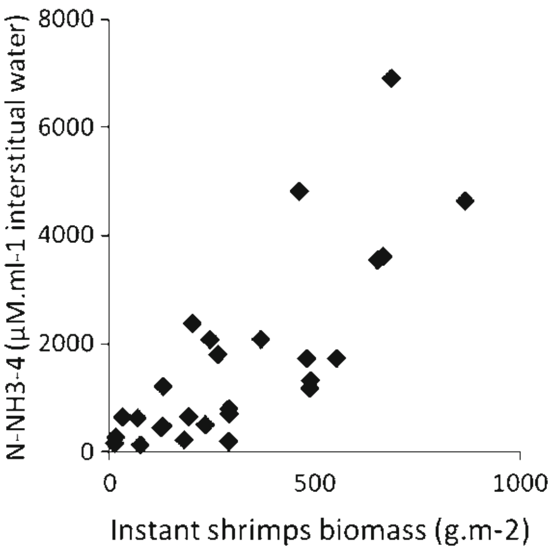

Fig. 2 Relationship between the concentration of N-NH_{3-4} in pore water and the instant biomass in ponds

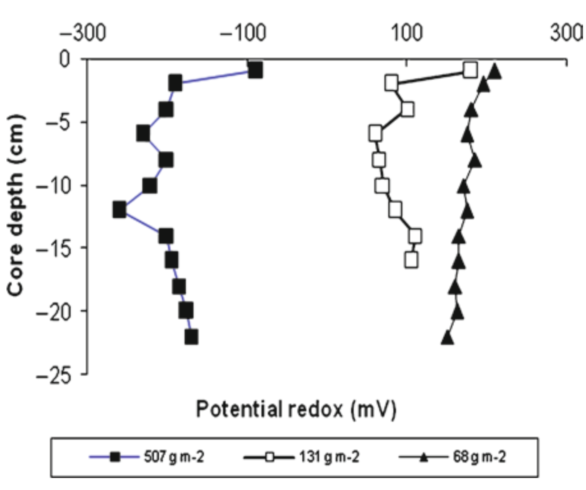

Fig. 3 Relationship between the potential redox values and the instant biomass in ponds

(Primavera 2005; Thu and Populus 2007). It is now well known that mangroves play an important role in the equilibrium of the coastal ecosystem (Blasco 2002).

It is not only a very productive ecosystem (De Graaf and Xuan 1998) showing a very high diversity (Carvajal and Alava 2007), but also a very efficient filter for a large variety of elements, particulate or dissolved, mineral or organic, issued from the catchment basin (Thampanya et al. 2006), or from aquaculture activities (Rivera-Monroy et al. 1999; Gautier et al. 2002). Destruction of mangroves has a huge impact on the surrounding environment (Hong 1996), including soil acidification (Mitra and Bhattacharyya 2003). Mangrove clearance has also been shown to play a leading role in the decreasing performance of coastal marine fisheries (De Graaf and Xuan 1998) and biodiversity (Fondo and Martens 1998). The main environmental impact of shrimp aquaculture is the destruction of coastal mangroves and deterioration of water quality. Many aquaculture activities, located in areas where mangroves had been destroyed, collapsed after a few years of activity. This was mainly due to an increase of the trophic level up to dystrophic conditions in the coastal ecosystem (Populus et al. 2004) under the pressure of the input of organic matter from aquaculture and the input of particles from the catchment basin. The consequences of mangrove destruction on the performances of aquaculture and of the speed of the collapses are directly related to the level of confinement of the surrounding environment (Populus et al. 2004).

The relationship among (1) the two main kinds of aquaculture (extensive and intensive), (2) the capacity of the surrounding ecosystem to change its water (confinement) and, consequently, to accumulate wastes, and (3) as a consequence the trophic level of this surrounding ecosystem is shown in Fig. 4.

We see that there is an inverse relationship between the water change capacity and the potential accumulation of organic matter (wastes). These characteristics determine the trophic level of the hosting ecosystem. In low confined ecosystems with high water capacity

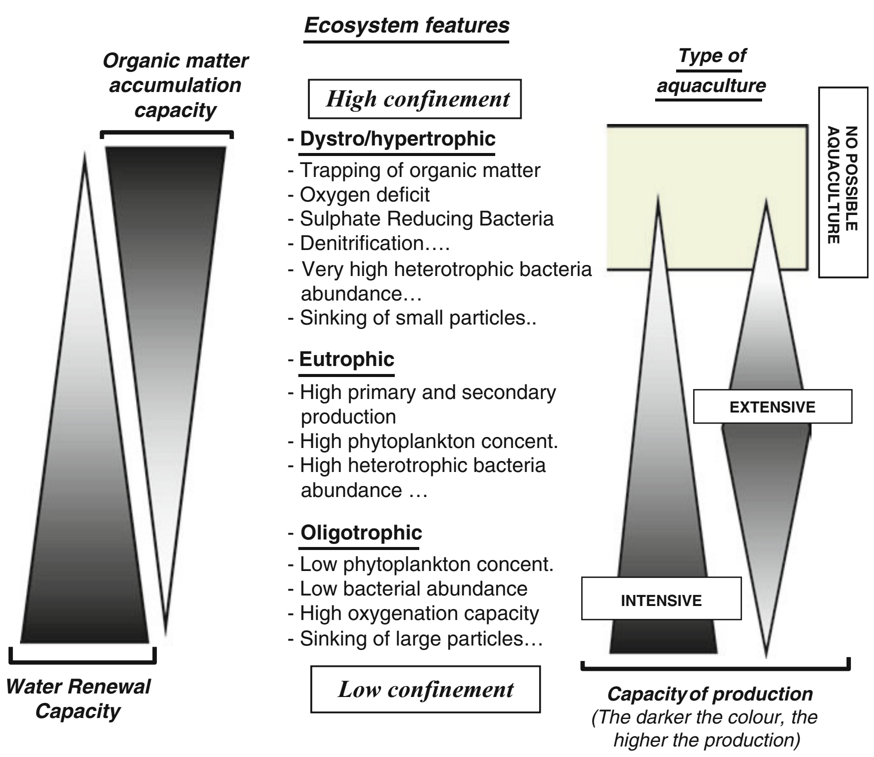

Fig. 4 Positioning the different types of aquaculture and their potential performances, according to the trophic characteristics of the surrounding environment (maximum production is expected in the oligotrophic area for intensive aquaculture, while the maximum occurs in the eutrophic area in extensive production. In the dystro/hypertrophic area, except for some species, aquaculture is not possible)

change, oligotrophic features prevail (low phytoplankton concentration, low heterotrophic bacteria abundance, high oxygenation capacity, etc.).

In this kind of ecosystem, intensive aquaculture that does not need a productive environment may be highly successful and sustainable. In such an environment, when rational management is carried out, no degradation of the environmental characteristics occurs, insuring the sustainability of the activity. On the contrary, ecosystems with a high level of confinement show hyper/dystrophic features, either naturally or under the pressure of the organic wastes released from aquaculture activities. In this kind of ecosystem, aquaculture activity, either intensive or extensive, showed very poor or no performance. In some ecosystems, particularly confined and dystrophic, or those becoming such, no rentable aquaculture activity is possible at all. The degradation of the environmental characteristics because of the input of organic wastes in a confined environment explains the many collapses of shrimp aquaculture activities worldwide in the 1980s (Martin 2004). Concerning extensive aquaculture, the best positioning appears to be in eutrophic ecosystems, where the reared organisms can be fed small organisms, such as meiofauna, grown in the food web developing from the input of nutrients and phytoplankton into the ponds (Reymond and Lagardère 1990). Respecting these considerations must lead (1) to insuring the best choice for site selection, both for intensive and extensive aquaculture, (2) to minimizing the impact of wastes on the surrounding environment for intensive aquaculture, (3) to insuring a satisfactory development of the food web inside the ponds for extensive aquaculture, and (4) finally to promoting the best environmental conditions in order to obtain real sustainability.

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Quality Control of Cultured Fish by Feed Supplements

Heisuke Nakagawa

Abstract The importance of algae, chitin, and laurate, which are widely ingested by numerous varieties of fish living in nature, was confirmed as a feed supplement in cultured fish. These components were added to the composed diet of ayu (*Plecoglossus altivelis*), black sea bream (*Acanthopagrus schlegelii*), and red sea bream (*Pagrus major*) at the same level as found in the stomach content of wild fish.

The effects of these dietary supplements commonly improved the physiological conditions, such as acceleration of lipolysis activity and protein assimilation, depression of adipocytes, resistance to air dipping, and activation of liver function, without depression of growth.

1 Introduction

Advances in fish nutrition have progressed considerably with regard to growth performance. However, much concern has arisen over the quality of fish flesh and health in aquaculture. In response, a variety of feed supplements have been investigated with respect to their effects on the physiological condition and quality of cultured fish.

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2 Materials and Methods

2.1 Fish and Rearing Conditions

In the dietary supplement experiment, food organisms in the stomach content of wild fish were preliminarily analyzed. Fish used for the feeding experiment were 0-year ayu (*Plecoglossus altivelis*), black sea bream (*Acanthopagrus schlegelii*), and red sea bream (*Pagrus major*). The fish in the control group were fed a composed diet. The supplement levels were selected based on the stomach contents of wild fish. For the experimental group diet, certain contents of algae (<5% in dry diet), chitin (10% in wet diet), or lauric acid (0.5% ethyl laurate in dry diet) were supplemented to the control diet.

2.2 Biological Measurements

All the fish in each replicate treatment group were weighed individually. Whole muscle, liver, and adipocytes were obtained from 20 fish in each group. The adipocyte diameter was histologically measured in 50 cells per fish.

2.3 Vitality

For comparing liver function, fish were dipped in water containing anesthetic at a concentration of 0.1% for 50s. Then, the fish were transferred to oxygen-saturated

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fresh seawater, and the recovery time was recorded. For air-dipping resistance, fish were exposed to air dipping for 5 min and returned to oxygen-saturated seawater. Recovery time from the succumbed condition was recorded.

2.4 Biochemical Measurement

The feed, muscle, liver, and adipocytes in the intraperitoneal cavity were frozen immediately after killing the fish and kept in a freezer. Lipid was extracted with methanol-chloroform. The lipid class composition and fatty acid composition were analyzed by an Iatroscan and a gas chromatograph.

3 Results and Discussion

3.1 Micro- and Macro-algae (Nakagawa et al., 2009)

A variety of wild fishes ingest algae, including some primarily carnivorous fish. The primary foods of wild ayu in nature are benthic blue-green algae and diatoms in rivers. *Chlorella* extract as a feed supplement affects physiological characteristics, such as lipid metabolism, disease resistance, carcass quality, and vitality in ayu and yellowtail (*Seriola quinqueradiata*). In the giant (freshwater) prawn (*Macrobrachium rosenbergii*), dietary (cyanobacteria) *Spirulina* elevated growth performance and feed utilization over ranges of supplementation of 5–20% of the diet.

Although the lipid content in the intraperitoneal fat body of cultured ayu was almost 98%, the *Chlorella* extract supplementation reduced the value to 66.7%. In vitro lipolysis activity was superior in the *Chlorella* extract (1%) fed group, suggesting activation of lipid utilization for energy prior to muscle protein consumption. *Chlorella* extract suppressed body weight loss caused by starvation in ayu, because the reserved lipids decreased remarkably and consumption of muscle protein might be suppressed.

The *Chlorella* extract improved the tolerance of hypoxic conditions and liver function distinctly. The preventive action exerted by *Chlorella* extract might involve some internal barrier to infectious disease, such as an inflammatory response and an increase in the number of phagocytes, rather than an immediate effect on the disease as an antibiotic substance.

Spirulina supplementation correlated with marked increases in some key substances for β-oxidation of fatty acids, including hepatic-free carnitine and long-chain acylcarnitine, carnitine palmitoyltransferase activity, etc. Red sea bream fed a diet supplemented with *Spirulina* at a 2% level exhibited elevated protein synthesis, and the stroma (connective tissue) fraction was significantly increased. The muscle protein composition measured by solubility approached that of wild fish. *Spirulina* increased total muscle collagen; the collagen fractions soluble at 20°C and 70°C decreased, but the insoluble collagen increased.

The addition of a very small amount of algal meal has produced a significant increase in the growth and feed utilization of a variety of fish such as red sea bream, Japanese flounder (*Paralichthys olivaceus*), yellowtail (*Seriola quinqueradiata*), ayu, rockfish (*Sebastes schlegeli*), nibbler (*Girella punctata*), and snakehead (*Channa striatus*). The optimum feed efficiency and protein efficiency were attained in black sea bream when the supplementation level of *Ulva* meal was 2.5–5.0% of the diet. Body weight loss of black sea bream during wintering was minimized with supplementation of *Ulva* meal. Growth of Japanese flounder was maximized with *Ulva* at 2% of the diet. Algae also affect other growth indicators when used as dietary supplements.

Supplementation with the following algae, *Ascophyllum, Porphyra* and *Ulva*, at 3–5% in prepared diets elevated the muscle RNA/DNA ratio (protein synthetic activity) and suppressed acid protease activity (protein catabolism) in red sea bream, providing biochemical proof of growth. The effect on growth is due to an acceleration of nutrient absorption by dietary algae.

The addition of a small amount of algal meal to the fish diet can exert considerable effects on carcass quality. While micro-algae generally decrease accumulation of lipid in the muscle, macro-algae often induce an increase in muscle lipid. While muscle lipid of the control group of young yellowtail was 1.6%, supplementation of *Laminaria digitata* at 1% increased muscle lipid to 5.2%. Similar tendencies for increased muscle lipid occurred with supplementation of *Undaria pinnnatifida* and *Ascophyllum nodosum* in the diet of red sea bream and with *Ulva* in black sea bream. Sensory evaluation of fish meat showed that supplementation of macro-algae in the diet generally improved the taste and quality as a whole. Under imbalanced nutritional conditions and inadequate feeding regimes (overfeeding, inadequate feeding frequency, etc.), muscle protein can be consumed in place of the reserved lipids. Fish reared under such conditions cannot endure food shortage.

Supplementation of *Ulva* meal to a red sea bream diet or *Laminaria* meal to a yellowtail diet increased consumption of reserved lipids and suppressed muscle protein consumption. Wild red sea bream and black sea bream ingest algae in nature, and the proportion of algae ingested increases before wintering. The ingestion of algae before wintering could help to activate reserved lipids for energy during the winter period.

Stressors during fish rearing, such as high rearing density, treatment, low dissolved oxygen, water pollution, and nutritional imbalance, can depress disease resistance. Resistance to low oxygen levels was higher in algae-fed red sea bream, black sea bream, and in rockfish. Many algal species are likely to improve vitality in a similar way, because they contain a wide array of macronutrients, micronutrients, and other components.

Supplementation of *Ulva* meal to a prepared diet at 5% elevated phagocytosis in black sea bream. Effects of similar supplementation with *Ulva* were found in the numbers of lymphocytes and granulocytes, agglutinin titer, and hemolytic and bactericidal activities in red sea bream. In addition, bactericidal activity after immunization was enhanced. A nutritional disease involving retardation of growth and high mortality in the young yellowtail is caused by feeding continuously on sardines (*Sardinops melanocticta*). The disease was reduced by supplementation of 0.5% of *Laminaria digitata*. Simultaneous addition of a vitamin mixture enhanced the effect, suggesting a synergistic effect of algae and certain vitamins. Atlantic salmon (*Salmo salar*) fed on a diet supplemented with alginate had high survival and complete hemolytic activity after challenge by *Aeromonas salmonicida*.

3.2 Chitin (Nakagawa et al., 2009)

Crustaceans are important and often the major food organisms of larval fish and contain a considerable amount of chitin in the digesta (2.3–9.3% in dry weight). The growth, feed efficiency, and survival rate were improved by dietary chitin (10% in purified moist diet). The relative intestine length is an indicator of the diversified use of food and nutrients. The intestine of the animals in the chitin group was significantly longer than in the control group. The long intestine presumably prolonged the evacuation time of food in the gut by increasing the surface area. Consequently, growth performance would be improved. The dietary chitin decreased the lipid content and size of lipid droplets in the liver. Low lipid reserves were mainly due to the reduction of the adipocytes accompanied by a reduction in the adipocyte diameter. Reduction of the liver oil globules and the adipocyte size might suggest suppression of lipogenesis and activation of lipolysis. Suppression of body weight loss by the starvation in the chitin group could be due to sparing the consumption of muscle protein.

The dietary chitin significantly reduced the impact of air dipping and improved the recovery time from the succumbed condition. During the treatment, 50% of the fish died in the control group, while the animals belonging to the chitin group recovered completely.

3.3 Lauric Acid (Ji et al., 2005)

A small amount of lauric acid was found in the stomach content of wild black sea bream. Although dietary laurate did not affect growth, survival, and feed efficiency, the muscle ratio was significantly increased. In addition, the hepatosomatic index, adipocyte ratio, and adipocyte diameter were significantly reduced by laurate supplementation. Hepatic lipids were significantly low, and the value was almost half of the control group. Dietary laurate depressed triglycerides markedly, although free fatty acids and phospholipids were hardly influenced.

The dietary laurate significantly reduced the impact of air dipping and quickened the recovery time from the succumbed condition. In the liver function test, the survival from the anesthetic condition was only 40% in the control group, but fish in the laurate group recovered completely.

Feeding laurate to fish led to a proportional decrease of monoenes and a slight increase in n-3 fatty acids in the hepatopancreas. While lauric acid was not found in these organs of the control group, lauric acid and DHA slightly increased in the heart, brain, and eye in the laurate group. Dietary laurate might account for the absorption and/or incorporation of DHA into the fish body.

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Experimental Culture of *Arthrospira (Spirulina) platensis* - Nordsted, 1844

Rija Rakotoarisoa, Alain Riva, and Nardo Vicente

Abstract *Spirulina* was long considered a blue alga (Cyanophycea). As a matter of fact, it is a cyanobacteria, described for the first time by Wittrock and Nordsted in 1844 under the name of *Spirulina jenneri platensis* Nordsted.

Appearing on earth 3.5 billion years ago, it originally produced the oxygen breathed by living organisms. *Spirulina* has been consumed for a long time by many of the world's populations (e.g., in Chad, Mexico, and India).

In different countries in the world, various strains have been described (Paracas, Lonar, etc.). In France, a strain was discovered in Camargue (Planchon 1994) outside of the usual temperature zone where this organism commonly grows. Cultivated out of its natural environment in favorable areas (Indo-Pacific, Subtropical Atlantic, etc.), it can also be grown in greenhouses in temperate countries, such as near the Mediterranean coast.

In recent years, several small producers have appeared in the French Mediterranean zone. Today, it is important to improve the production yield, and many experiments are being carried out in laboratories and greenhouses.

Several strains were tested using water from a brackish pool (Vaccarès in Camargue, in the Rhône River delta), and an experiment with sea-water was also carried out. To obtain good production, it is essential to protect the cultures from contamination by other microorganisms.

Currently, these experiments are being carried out at the Oceanographic Institute Paul Ricard located on Les Embiez Island in close collaboration with university laboratories and other scientific research organizations. Résumé

La spiruline, longtemps considérée comme une algue bleue (Cyanophycée), est en réalité une cyanobactérie décrite pour la première fois par Wittrock et Nordsted en 1844 sous le nom de *Spirulina jenneri platensis* Nordsted.

Apparue sur Terre il y a 3,5 milliards d'années, elle produisit le premier oxygène respirable par les organismes. Elle est consommée depuis les temps les plus reculés par diverses populations du monde (Tchad, Mexique, Inde). Selon la région, on a pu décrire plusieurs souches (Paracas, Lonar, …). En France, une souche a été découverte en Camargue (Planchon 1994) hors des zones de température où ces organismes croissent habituellement. Cultivée à l'air libre en milieu naturel (Indo-Pacifique, Atlantique subtropical), elle ne peut l'être que sous serre en zone tempérée comme près de la Méditerranée.

Au cours de ces dernières années, de nombreux producteurs de spirulines ont ainsi vu le jour en Méditerranée française. Il apparaît aujourd'hui important d'améliorer le rendement de ces cultures. Afin d'atteindre de but, des expériences sont réalisées en laboratoire et sous des serres de type horticole. Plusieurs souches ont été testées en utilisant l'eau d'un étang saumâtre (étang du Vaccarès) et une expérimentation en eau de mer est également en cours.

Afin d'obtenir une production de qualité, il est primordial de tenir compte des contaminations dues à d'autres microorganismes.

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C'est ce type d'expériences qui est conduit actuellement à l'Institut Océanographique Paul Ricard sur l'île des Embiez en collaboration étroite avec des laboratoires universitaires et d'autres organismes de recherche.

1 Introduction

Since 2004, the Oceanographic Institute Paul Ricard has been carrying out research on the cyanobacteria *Arthrospira platensis*, also called *Spirulina*. The main purposes are the scientific study of the nutritional composition of *A. platensis* developed in various conditions, the possibilities of improving the nutritional qualities, and then its potential use to ameliorate malnutrition in developing countries (Fox 1999).

2 Results and Discussion

The cyanobacteria *Arthrospira platensis* appeared on earth 3.5 billion years ago. This edible cyanobacterium has a remarkable nutritional composition. *A. platensis* is composed of proteins (70%), carbohydrates (16%), lipids (6%), minerals (9%), and fibers (7%). It contains vitamins (B1, B2, B3, …, E, including anti-oxidants and β carotene) and several oligo elements, such as Ca, Mg, Cu, Zn, P, and Fe. Eleven photosynthetic pigments are present in these cyanobacteria (mainly chlorophylls and phycocyanine) (Gershwin and Belay 2007).

Because of this interesting composition, *A. platensis* can be considered as an alimentary complement for marasmic children in developing countries.

These cyanobacteria can be found in natural lakes (Chad) and are also cultivated in many countries in the world (India, Africa, France). Cultures of *A. platensis* require specific mediums characterized by a basic pH and a defined salinity. Fresh, brackish, and sea-waters can be used to prepare culture mediums (Jarisoa 2005).

The first stage of the study was to make cultural assays of the cyanobacteria using brackish water from Vaccarès pond.

Results showed that the water had sufficient quality for spirulina cultures considering the physical and chemical characteristics. The most appropriate culture medium for brackish water was Jarisoa's medium, currently used for sea-water. Culture yield was 3.5 g/m²/day (Rakotoarisoa et al. 2009).

The second stage, which took place on Les Embiez Island, was to evaluate the compositional evolution of *Spirulina* through time according to various parameters: strains (Lonar, Toliara, and Paracas), conservation conditions, and culture mediums.

Another subject of the study was culture contamination. Axenic conditions are hard to obtain in order to assure pure strains, particularly for cultures in greenhouses. Contamination can take place in spirulina cultures. Decreasing the risks of contamination is one of the most important aspects of the study.

After determination of the contaminants, finding a way to eliminate them from the culture is the second part of the work.

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Problems Associated with the Recovery on Landings of Black Sea Bream (*Acanthopagrus schlegelii*) Intensively Released in Hiroshima Bay, Japan

Tetsuya Umino, Enricure Blanco Gonzalez, Hidetoshi Saito, and Heisuke Nakagawa

Abstract Stock enhancement programs are conducted worldwide, with Japan leading in marine species. One of the target species for release in the country is black sea bream (*Acanthopagrus schlegelii*). A stock enhancement program for this fish in Hiroshima Bay has been conducted over the last 3 decades. The large number of juveniles released has contributed to fishery recovery.

Nevertheless, the augmentation of landings has led to several problems. The reduction of the market price of the species, an augmentation of the social pressure from oyster farmers demanding the protection of their interests, and losses of the genetic resources of the natural stock of black sea bream are some of the problems encountered.

In this chapter, we discuss the main constraints associated with the increase in the stock biomass. In addition, some lessons learned and recommendations to be considered before and during the development of future stock enhancement programs are given.

1 The Stock Enhancement Programs in Japan

The release of hatchery-reared juveniles into the natural environment as part of stock enhancement programs is very controversial, but still extensively practiced worldwide to recover depleted fishery stocks. The economic feasibility of the releases and the biological impact on the wild stocks remain, especially from a genetic viewpoint, the main drawbacks to applying this management approach. Nevertheless, several studies have demonstrated that stock enhancement programs may increase the stock biomass of fisheries (Fushimi 2001; Kitada and Kishino 2006; Blanco Gonzalez et al. 2008a, b). More than 180 species were released into coastal and marine environments in 64 different countries from 1984–1997 (Born et al. 2004). The magnitude of the releases as well as the number of species varies among countries, with Japan leading in marine stocking.

The history of the stock enhancement programs in Japan dates from the 1870s (Kitada 1999), though it was not until 1963 that the government started these programs, targeting several species, including red sea bream (*Pagrus major*) and kuruma prawn (*Marsupenaeus japonicus*). Stock enhancement programs are now extensively conducted throughout the country to recover depleted stocks of commercially valuable species. The total number of marine fish fingerlings released in 1983 was 35 million, reaching 76 million in 2006 based on the statistics of the Japanese Fisheries Research Agency (JFRA 2008). Regarding the facilities for mass production of the seedlings for release, these are operated either by the central or prefectural/local governments for high migratory and near shore and coastal fisheries species, respectively (Imamura 1999; Fushimi 2001; Sugaya 2005).

The choice of the target species for release is based on the status of the natural stock and the market price. Therefore, small fishery cooperatives and associations are fundamental stakeholders playing a primary role in the management of the fishery resources. Their

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demand for high-priced species for release reinforces the continuation of the stock enhancement programs of the target species.

2 Stock Enhancement Programs for the Main Target Species

Traditionally, in Japan, red sea bream has been the marine finfish species accounting for the largest number of juveniles released in stock enhancement programs (Fig. 1). In 1983, more than 16 million hatchery-reared fish were released throughout the country. Meanwhile, the releases of the other two main fish species for stocking, black sea bream (*Acanthopagrus schlegelii*) and Japanese flounder (*Paralichthys olivaceus*), were ~4 and ~3 million, respectively. In the 1990s, Japanese flounder releases increased drastically (>30 million in 1999), surpassing those of red sea bream. Recently, the number of Japanese flounder juveniles released in stock enhancement programs has stabilized to ~25 million per year, while red sea bream accounts for ~20 million.

On the other hand, releases of black sea bream have decreased to ~2 million after reaching a peak in 1996 (~10 million). In consequence, the relevance of the stock enhancement program for this species dropped to sixth place in 2006, below other species, such as Japanese pufferfish (*Takifugu rubripes*) or Pacific herring (*Clupea pallasii*). In this chapter, we discuss the main constraints associated with the augmentation of landings of black sea bream in Hiroshima Bay, a region where a stock enhancement program has been conducted for the last 3 decades. This bay is the main fishing ground for the species in Japan, and the intensive releases have contributed to the recovery of catches of the species in the bay from <200 t in 1980 to ~400 t in 2005 (Blanco Gonzalez et al. 2008b).

Nevertheless, the different problems associated with the recovery of landings have created several socio-political conflicts that have resulted in the recent suppression of the stock enhancement program.

3 Black Sea Bream Abundance Constraints in Hiroshima Bay

3.1 *Reduction of the Wholesale Price*

The stock enhancement program for black sea bream in Hiroshima Bay started after the drastic reduction in catches of the species registered in the 1960s and 1970s. At that time, black sea bream was a high-value fish in Hiroshima. The wholesale price of the species in 1985 was ~2,000 Japanese yen (JY) kg^{-1} (1 euro = 150 JY), close to the market price of wild red sea bream (~2,500 JY kg^{-1}). These prices were especially high during the first months of the year (~2,800 and ~4,000 JY kg^{-1} for black and red sea bream, respectively), falling to a minimum in summer (~1,200 and ~2,000 JY kg^{-1} for black

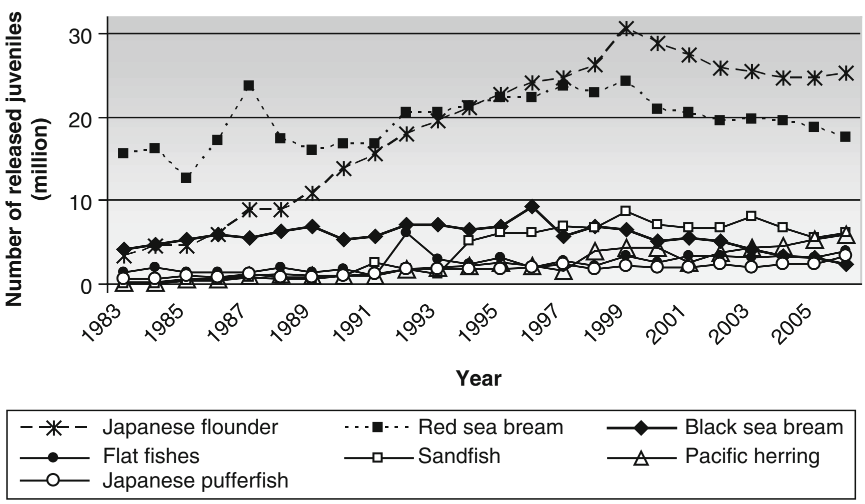

Fig. 1 Main marine finfish species released in Japan (based on the statistics of the JFRA 2008)

and red sea bream, respectively). The market price of the species was inversely correlated to their catch. In winter, landings of black sea bream made up barely ~3 t, in contrast to summer, when > 5 t was registered. Red sea bream presented a similar pattern. Numbers of this species at the Hiroshima Central Wholesale Market fluctuated between ~700 kg and ~4 t in winter and summer, respectively. However, even though the production of cultured red sea bream presented monthly variability as occurs with wild catches, its wholesale price remained constant (~1,800 JY kg^{-1}) throughout the year.

Recently, the numbers of both wild black and red sea bream have increased significantly at the Hiroshima Wholesale Market, from ~26 and ~50 t in 1985 to ~205 and ~78 t in 2006, respectively. However, those increases in landings caused the price of both species to decrease drastically. In 2006, the mean prices of black and red sea bream were ~380 JY kg^{-1} and ~1,000 JY kg^{-1}, respectively. Therefore, the cost efficiency of the releases should be considered. The stock enhancement program for black sea bream in Hiroshima Bay has contributed to increasing the stock biomass (Umino et al. 1999; Jeong et al. 2007; Blanco Gonzalez et al. 2008a). Nevertheless, although the economic benefits of that investment were undoubtedly justified according to the scenario observed in 1985, the actual market price (~380 JY kg^{-1}) may not ensure the continuity of the stock enhancement program. As a result, fishermen demanded that the local authorities replace the releases of the target species for new species with much higher market values, such as Japanese pufferfish, devil stinger (*Inimicus japonicus*), or red spotted grouper (*Epinephelus akaara*).

The increase in the number of large supermarkets is another important factor that may have contributed to the reduction of fish prices. Some decades ago, fresh fish was sold by local fishmongers, maintaining high prices. Recently, most of the small fish stores have disappeared, and the price of the fish sold at the supermarkets has decreased significantly.

3.2 Black Sea Bream as a Predator of Pacific Oyster and Short-Necked Clam

Black sea bream is an omnivorous species, feeding on a wide variety of organisms, including shellfish, shrimps, crabs, small fish, and seaweeds, among others (Saito et al. 2008). Black sea bream's favorite prey is mussels (*Mytilus galloprovincialis*). However, the population of this mussel species in Hiroshima Bay has decreased significantly in recent years, which may be partly explained by the increase in the mean sea-surface temperature (>1°C) registered in the bay over the last 30 years (Takatsuji 2003). Because of the absence of mussels, Pacific oyster (*Crassostrea gigas*) spats were the main prey found in the stomachs of adult black sea bream (Saito et al. 2005, 2008). The change of diet led to an important socio-economic problem because of the high value of the shellfish. Blue mussel meat is not popular in the Japanese cuisine; however, the annual oyster production in Hiroshima Bay is >200,000t, with a value of 38.2 billion JY (Kamiyama et al. 2005). The short-necked clam (*Ruditapes philippinarum*) is another high-value shellfish species in Japan; however, the annual catch of this clam in the country has declined to <40,000 t. In Hiroshima Bay, the short-necked clam is intensively released by several fishery associations, who point to the predation pressure of black sea bream to explain the reduction of their catches in recent years. This situation has led to numerous pressures on government authorities from both oyster farmers and fishery associations demanding the protection of their resources and capital. In consequence, the stock enhancement program for black sea bream in Hiroshima Bay was reduced and then stopped in 2009.

3.3 The Quality of Black Sea Bream Meat

Whenever we talk about Japanese food, it is necessary to mention "sushi" and "sashimi." Fish is mainly eaten raw and represents the primary source of protein for the Japanese people. Therefore, the freshness and quality of the fillets are very important factors considered by the consumers. In addition, marketing is another key component governing people's consumption tendencies. Many species of fish and shellfish are consumed raw daily throughout the country; however, not all varieties have the same level of acceptance. Up to the early 1990s, the meat of black sea bream was highly appreciated, and demand was high for this fish. However, the increase in landings registered in the 1990s produced a surplus of fish, while demand did not increase at the same rate. The main cause for the low demand for raw fish is attributed to the quality of the flesh and the smell of the fish. The low prey selectivity of the species is believed to cause the fish meat to have an unpleasant smell. Moreover, there is another important cultural component. In Japan, the combination of black-and-white is associated with funerals; therefore, the skin pigmentation of the species presents a "bad" image of the fish to the people. Contrarily, the colors white-and-red are associated with festivals and celebration. Therefore, red sea bream, with its characteristic whitish/reddish coloration, is the main species eaten at weddings and can often be seen in worship images, such as the Seven Deities of Good Fortune, because of its "good" image among the people, which keeps it in high demand.

In addition, the low fat content of black sea bream is a reason for the low demand. Most of the landings of the species occur from April–June, when mature females move to shallow waters for spawning (Blanco Gonzalez et al. 2008b). At that time, most of the lipid reserves are consumed for reproduction, and the low fat fish meat is not desired by consumers, decreasing the price and the demand. In Hiroshima Bay, the lipid content of black sea bream marketed during the spawning season was 0.2% (Umino et al. 2009). This value is much lower than that reported for red sea bream, yellowtail (*Seriola quinqueradiata*), or Japanese flounder (Oku and Ogata 2000).

3.4 Other Biological Constraints

In addition to the above-mentioned constraints, our research aimed to provide some scientific evidence of the negative effects of intensive releases of black sea bream in Hiroshima Bay in light of the absence of an exhaustive monitoring and management plan.

Black sea bream juveniles have demonstrated fast and good acclimatization to natural conditions (Umino et al. 1999; Nakagawa et al. 2000; Jeong et al. 2007), and some of the previously released fish were observed to reach maturity (Blanco Gonzalez et al. 2008a).

Nevertheless, the high survival rate of the large number of juveniles released has triggered some genetic problems. The use of a limited number of breeders ($n = 51$) to produce the offspring for release has resulted in a low effective population size ($Ne^* \sim 16$) and an important genetic drift. The high rate of inbreeding detected (~3%) warns us of the necessity to carefully preserve and not compromise the genetic diversity of the species in the bay (Jeong et al. 2007; Blanco Gonzalez et al. 2008a; Blanco Gonzalez and Umino 2009).

Although similar growth rates have been observed in both wild and hatchery black sea bream (Blanco Gonzalez et al. 2008a), a recent study comparing two black sea bream samples from 1983 and 2000 has revealed a significant reduction in size-at-age in recent years (Blanco Gonzalez et al. 2009), suggesting that the intensive releases may have exceeded the carrying capacity of Hiroshima Bay.

This augmentation on the stock biomass may have created intra-specific competition for food, reducing the growth rate and the lipid accumulation on the body of the fish.

4 Lessons and Recommendations

In this chapter, we have reviewed the different problems and constraints of the augmentation of landings of black sea bream after the intensive releases carried out in Hiroshima Bay, Japan, because there is no appropriate existing resource management plan.

The stock enhancement program has confirmed its potential to contribute to restoring the depleted stock of black sea bream. Nevertheless, the implementation of large-scale programs requires a multidisciplinary approach, and all stakeholders should be involved and take active part in the management and decision processes (Lorenzen 2008). Several lessons learned from this experience and a list of recommendations to be considered before and during the development of future stock enhancement programs for this species can contribute to achieving a responsible approach to these programs (Blankenship and Leber 1995).

4.1 Before Implementing the Stock Enhancement Program

Black sea bream has proven to be a good species for stock enhancement. However, important biological and ecological aspects of the species should be investigated before implementing a stock enhancement program on a large scale. Pilot experiments provide relevant information for developing the stocking strategy, although the results achieved may differ greatly with the upscaling of the releases.

Considering that the large number of released black sea bream juveniles come from a limited number of breeders, a primary concern involves the conservation of genetics. The small number of broodstock reared in captivity has resulted in a very low effective population size and high rates of inbreeding in the offspring, compromising the genetic diversity. Therefore, the offspring should be produced by a much larger wild-origin broodstock.

The significant reduction in size-at-age observed in black sea bream is an indicator that the recovery of landings may have exceeded the carrying capacity of Hiroshima Bay for this resource (Blanco Gonzalez et al. 2009). Though difficult, the estimation of the carrying capacity is necessary to manage the local fishery properly.

In addition, global warming and climate change play a key role in explaining long-term variations in marine production. Therefore, the possible enhancement of the black sea bream population associated with variations in the environmental conditions must also be examined. Simulation and prediction models are other very useful tools to be considered for the enhancement programs. Integrating this information with stock assessment studies and other data obtained monitoring the stock biomass would provide a good estimation of the proper magnitude of the releases to be carried out without exceeding the carrying capacity of Hiroshima Bay. To date, more than 20 million black sea bream juveniles have been released since the stock enhancement program began in the 1980s; however, no study has been conducted on stock assessment and the limitations of the bay to support the influx of hatchery fish. The age and size composition of the natural stock should also be evaluated.

Furthermore, little information is available on the exact location of the spawning and nursery areas in the bay as well as the possible factors that may induce the beginning of the spawning season. In addition, the characteristics of the habitats where this species occurs as well as possible intra- and inter-species interactions should be investigated. An understanding of all these factors and the compilation of all the information gathered are essential to establish a proper management plan for the long-term.

4.2 During the Development Process of the Stock Enhancement Program

One of the main problems of the stock enhancement program for black sea bream in Hiroshima Bay has been the lack of a proper monitoring plan. Releases of a given target species require continuous evaluation of the contribution of hatchery-reared juveniles and the possibility of adopting a feed-back strategy to manage the resources. Traditionally, the effectiveness of the stock enhancement program has been based on the recovery of landings. However, this approach is susceptible to annual variations in recruitment in addition to other environmental and climatological factors.

Moreover, results will be obtained in the long-term without understanding the proportion of released juveniles that add value to the stock. A parentage assignment using genetic markers has allowed identifying those fish that were previously released (Jeong et al. 2007; Blanco Gonzalez et al. 2008).

Furthermore, the contribution of the breeders could be evaluated and the genetic diversity monitored. In this regard, it is essential to replace the breeders periodically in order to avoid inbreeding problems and maintain the gene pool of the wild stock.

In case the stock enhancement program succeeds in augmenting the stock biomass, the number of juveniles to be released will probably differ from that estimated at the beginning of the program. The carrying capacity of Hiroshima Bay will continuously fluctuate, as will the number of juveniles released. The stock assessment and carrying capacity analysis allow adjustment of the size of the releases, which should be adopted as part of the feed-back strategy. It should be mentioned that these estimations should not be limited to the intraspecific density-dependent effects, but it should also consider the interactions with other species sharing the same ecosystem.

Finally, new market opportunities to commercialize the target species must be examined. The recovery of the resource should not be accompanied by its devaluation in the market, as occurred at Hiroshima Bay. Therefore, the continuity of the stock enhancement program will be conditioned by its cost-benefit ratio.

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Effect of Wavelength of Intermittent Light on the Growth and Fatty Acid Profile of the Haptophyte *Isochrysis galbana*

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Abstract The haptophyte *Isochrysis galbana* is widely used as a food source for bivalve aquaculture as it is rich in fatty acid. In this study, the effect of different colors of intermittent light on the multiplication of cells of *I. galbana* and their fatty acid composition was investigated. *I. galbana* was cultivated under conditions of white, blue (dominant wavelength; 470 nm), green (525 nm), or red (660 nm) intermittent light (frequency; 10,000 Hz, duty ratio; 50%, light intensity; 52 μmol photons m^{-2} s^{-1}, water temperature; 20°C).

The concentration of cells and fatty acid composition were measured at 6 days after the beginning of cultivation.

Cell concentration of *I. galbana* increased with time. The cell concentration on the 6th day under the blue light condition was significantly higher than in other light conditions. The numbers of cells under white, red, and green light were 65%, 60%, and 40% of the blue light condition, respectively.

I. galbana contains mostly lipids of 16:0, 16:1n-7, and 20:5n-3 (EPA), which account for approximately 50% of the total lipids.

In conclusion, the number of cells on the 6th day under the blue intermittent light was approximately double that under white continuous light, and therefore the amount of fatty acids available is also considered to be approximately double.

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1 Introduction

The haptophyte *Isochrysis galbana* is widely used as a food source for bivalve aquaculture as it is rich in fatty acid. In order to use microalgae as feed, it is necessary to produce a large amount of microalgae within a short time (Phatarpeker et al. 2000). The culture of microalgae is often carried out in a thermostatic chamber to avoid complications resulting from external factors. Therefore, an artificial light source is necessary for culture, and the microalga production efficiency can be controlled by the irradiation conditions with different artificial lights (Tzovenis et al. 2003). On the basis on these points, the optimization of light irradiation conditions is important to produce the maximum volume of microalgae effectively within a short time and to optimize the growth of the phytoplankton species (Toro 1989).

In this study, the effect of different colors of intermittent light on the multiplication of cells of *I. galbana* and their fatty acid composition was investigated.

2 Materials and Methods

The microalgae species used in the experiment was *Isochrysis galbana* of Haptophyceae (Prymnesiophyceae). In this experiment, the strain that was precultured under the conditions of 20°C temperature, F/2 medium (Guillard and Ryther 1962), light intensity of 20 μmol photons m^{-2} s^{-1}, and 12-h light/12-h dark cycle was used. *I. galbana* was cultivated under conditions of white, blue (dominant wavelength; 470 nm), green (525 nm), or red (660 nm) intermittent light (frequency;

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Table 1 Light irradiation conditions

Light color (D.W.)	PPFD	Frequency of intermittent	Duty ratio (%)	L/D
White	104	Continuous	100	12:12
White	52	10^4 Hz	50	24:0
Blue (470 nm)	52	10^4 Hz	50	24:0
Green (525 nm)	52	10^4 Hz	50	24:0
Red (660 nm)	52	10^4 Hz	50	24:0

D.W.: Dominant wavelength, PPFD: photosynthetic photon flax density, L/D: light/dark in x:y hours

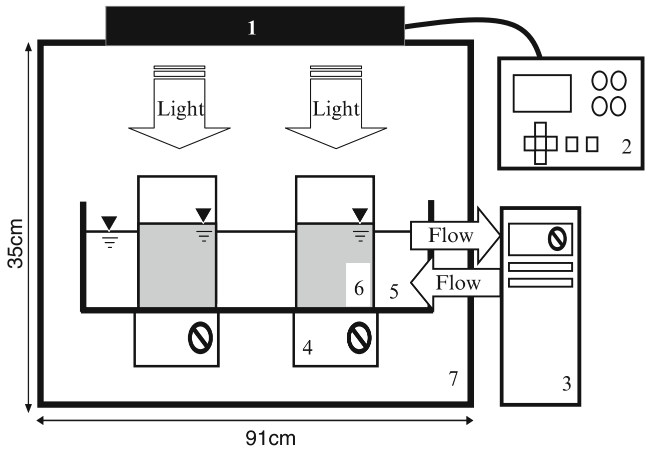

Fig. 1 Sketch of apparatus for experiment. 1, LED panel; 2, LED panel control device; 3, water temperature adjuster; 4, magnetic stirrer; 5, water bath; 6, culture vessel; 7, shadow chamber

10,000 Hz, duty ratio; 50%, light intensity; 52 μmol photons m^{-2} s^{-1}, water temperature; 20°C). The light conditions are shown in Table 1. These light conditions are made by an experimental system, which has a LED panel (CCS Inc.) and a control device (Fig. 1). The concentration of cells, cell dry weight, total lipid, and fatty acid composition were measured at 6 days after the beginning of cultivation.

For the measurement of cell concentration of *I. galbana*, a Coulter Counter Multisizer II (Beckman Coulter Inc.) with a 100-μm aperture tube was used. *I. galbana* cells were recovered from culture medium on the 6th day by centrifugation, freeze-dried, and weighed to have cell dry weight. The extraction of the total lipid was carried out according to the method described by Bligh and Dyer (1959), using a chloroform-methanol (2:1, v/v) solvent system.

Total lipid extracts were fractionated on silica gel cartridge columns (Sep-Pak Classic, Waters) after an activation step with methanol followed by chloroform. Fatty acid composition was determined using gas chromatography as described by AOCS Official Method Ce 1b-89 (1989) with minor modifications.

Results are expressed as mean ± standard deviation. The statistic method ANOVA was used to calculate significant differences (Hochberg 1988).

3 Results

At each lighting condition, the cell concentration of *I. galbana* increased with time. The concentration (Phytoplankton enumerated) under the white intermittent light showed 1.3 times that under the white continuous light on the 6th day. The cell concentration on the 6th day was highest under the blue intermittent light condition (Fig. 2). The concentrations of cells under white, red, and green light were 65%, 60%, and 40% of the blue light condition, respectively. On the basis of these results, the abundance recorded after 6 days under the blue intermittent light was about twice higher than under other types of lights.

The changes of cell dry weight and total lipid weight of *I. galbana* cultured under various color lights are shown in Table 2. The cell dry weight and total lipid weight were significantly higher under the blue light

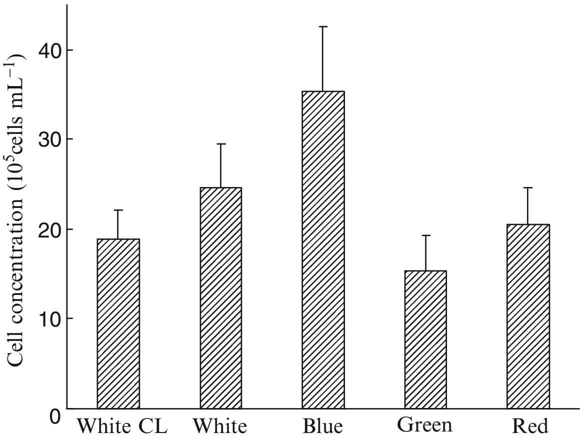

Fig. 2 Cell concentrations of five different light conditions on the 6th day. Values are means. Error bars indicate standard deviations ($n = 16$) (CL: continuous light)

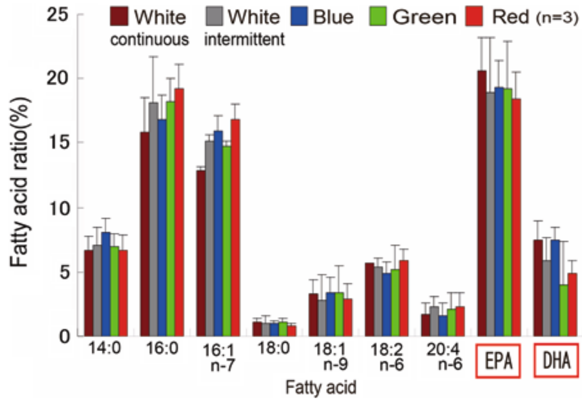

Fig. 3 Selective fatty acid composition of total lipid of cells under different light conditions on the 6th day

Table 2 Cell dry weight and quantity of lipid per cell of *I. galbana* cultured under variously color lights

Light condition	Cell dry weight (g/3 L medium)	Total lipid weight (g/3 L medium)	Cell dry weight/cell (pg $cell^{-1}$)	Lipid weight/cell (pg $cell^{-1}$)
Before cultivation	0.159 ± 0.043a	0.014 ± 0.004a	54.44 ± 10.20a	4.68 ± 1.01a
White continuous	0.279 ± 0.041b	0.021 ± 0.003b	49.14 ± 3.48a	3.76 ± 0.37a
White intermittent	0.317 ± 0.038b	0.027 ± 0.003bc	48.01 ± 6.52a	4.06 ± 0.18a
Red intermittent	0.268 ± 0.044b	0.024 ± 0.004b	47.35 ± 8.14a	3.53 ± 0.46a
Blue intermittent	0.406 ± 0.053c	0.031 ± 0.005c	49.98 ± 4.47a	4.48 ± 0.93a

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cultivation than others. However, the cell dry weight and lipid weight per cell had no significant difference by light condition.

The fatty acid composition of total lipid of *I. galbana* cultured under different color lights is shown in Fig. 3.

I. galbana contains mostly lipids of 16:0, 16:1n-7, and 20:5n-3 (EPA), which account for approximately 50% of the total lipids. DHA (C22 :6n-3) was not significantly different in light condition. The other fatty acids also were not significantly different.

4 Conclusion

The number of cells on the 6th day under the blue intermittent light was approximately double that under white continuous light, and the amount of fatty acids available is also considered to be approximately double. Therefore, the best light condition is blue intermittent light to efficiently cultivate *Isocrysis galbana*.

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Artificial reefs

Profile of Payao (Floating Artificial Reef or Fish Attracting Device) Fisheries of the Philippines

Ricardo P. Babaran and Munechika Ishizaki

Abstract This paper presents the profile of different fisheries that depend on payao (floating artificial reef) in the Philippines. In 2006, payao fisheries production was estimated at about 630,000 Mt, and large tuna species accounted for close to 400,000 Mt of this value.

The three commercial fisheries that depend on payao are purse seine, ringnet, and handlines. Various sustenance fisheries are also based on the floating structure, but many of these are relatively minor.

Purse seines and ringnets are used to harvest large size tunas, small tunas, and small pelagic species; these kinds of gear differ in terms of scale of operations, level of mechanization, and mode of fishing operation, but essentially capture the same species of similar size ranges.

Among handlines, the most dominant handline gear used near payaos has generally been single, large circle hooks that are cast in deep waters (>100 m) to target large yellowfin *Thunnus albacares* and bigeye *tuna Thunnus obesus*. Smaller hooks are also used in relatively shallow waters to capture smaller sized yellowfin tuna.

Meanwhile, multi-hooked handlines with colored lures are particularly designed for small pelagic species, and troll lines mainly harvest juveniles and adults of other (non-tuna) large pelagic species.

Adaptations of fishing techniques to catch various associated species reflect the high level of consolidated fishing efforts near the floating structures.

These techniques, in turn, highlight the growing concerns about overfishing and the sustainability of harvestable resources, as well as the need to regulate the deployment of payaos for effective payao fisheries management.

1 Introduction

A payao (Fig. 1) is an anchored floating structure composed of a bamboo raft with a series of palm fronds suspended underneath it. This structure is anchored generally at depths of about 1 km to serve as a fish-aggregating device (FAD). It was introduced in oceanic waters starting in the 1970s to support the operations of commercial surrounding nets like purse, seine and ringnets to target large tuna species like yellowfin tuna *Thunnus albacares* and skipjack tuna. These structures likewise support the operation of subsistence fishermen operating line gear that also capture small tuna and other small pelagic species. This paper presents the fishing sectors in the Philippines, the contribution of payao fisheries to the national economy, and an assessment of selected payao fisheries.

This presentation also highlights the potential effects of current payao fishing practices on the sustainability of fishing activities based on results of recent studies.

2 Fishery Sub-sectors

Philippine territorial waters cover approximately 220,000,000 ha, while offshore waters are approximately 193,400,000 ha, including the 200-mile exclusive

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Fig. 1 Illustration of a typical payao structure showing its different parts

economic zone (E.E.Z.). Fishing grounds within 15 km are called municipal waters, which are for the exclusive use of small-scale (subsistence) fishermen who use boats less than 3 gross tons (GT). Meanwhile, waters beyond 15 km are generally accessed by commercial fishermen with >3 GT. However, subsistence fishermen also venture to fish in oceanic waters (1 km deep) where payaos are deployed.

Overall, the total fisheries production in 2006 was 4.4 million metric tons, of which the contributions of small-scale and commercial fishermen were 25% and 28%, respectively (BFAR 2006). The landed catch of the almost 1.4 million small-scale fishermen was P59.1 B (36%); this is slightly higher than the contribution of commercial fishermen (P48.6 B, 30%).

3 Payao Fisheries

The design of payaos essentially has not changed since their introduction in the 1970s, but fishing activities involving this floating structure have had an important role in the Philippine economy. Ringnets, "baby" purse seines, and purse seines in overall Philippine fisheries production have remained dominant along with the payao (Babaran 2006). These surrounding nets differ only in terms of the level of mechanization (ringnets are partly mechanized), deck structure of the fishing vessels used (winches for ringnets and baby purse seines; power block for purse seines), and manner of fishing (generally by manual labor for ringnets and mechanized for purse seines). Commercial hook and line fishing, which involves the use of several small-boats that are ferried and deployed from a bigger mother boat, also became a dominant payao fishery.

Subsistence fishermen using various types of hand-line gear also operate near payaos. Some use single hooks and lines lured with chicken feathers, cuttle fish ink, metal plates, or rotating plastic material called a "helicopter." Meanwhile, others use multiple hook hand-lines while remaining attached to the bamboo raft; the same gear may be used as trolling gear by moving the payaos around to capture extranatant species (see, e.g., Dempster and Taquet 2004). Instead of using long-line gear, which usually entangles with the anchor line, subsistence fishermen sometimes release several drifting single hook and line units that are baited with live fish to target large pelagic fish like Spanish mackerel and various species of billfish.

4 Fisheries Production

Production by professional fishermen using purse seine and commercial hooks and lines is documented, but this only covers the three large tuna species. The estimates are not reliable because data-gathering activities were limited (Lewis 2005). Annual tuna production was essentially flat until 1970 and then showed an increasing trend. Their reports reached about 325,000 Mt in 2004 (Williams and Reid 2006); the progressive increase since the 1970s was apparently due to the introduction of payao-aided purse seine fishing in oceanic waters (Fig. 2).

However, this value was still underestimated because catch levels seemed to approach the 500,000 Mt mark in the late 1980s, if catch per unit effort data were used; the catch trend also showed a general decrease from 1990 to 2005 (Babaran 2007).

Purse seines for fishery remained the most dominant gear operating near payaos. The species caught by this gear were mostly skipjack and yellowfin; bigeye tuna formed part of the catch, but the estimated catch is not reliable due to difficulties in differentiating juveniles of bigeye from yellowfin tuna.

Unlike the tuna species, little is known about the other species caught by ringnets and by various

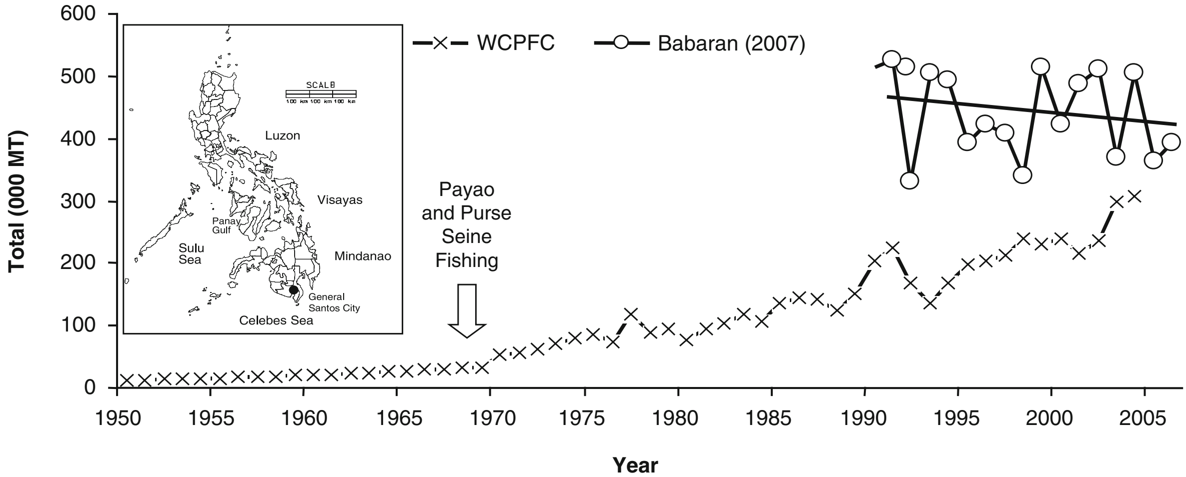

Fig. 2 Estimated total production of large tuna species (skipjack, yellowfin, and bigeye) based on reported landings data (WCPFC), which shows an increasing trend. A recent estimate based on catch per unit effort data (Babaran 2007) shows a declining trend between 1990 and 2005. Insert shows a map of the Philippines, its main island groups, and the main tuna fishing grounds of Celebes Sea and Sulu Sea

handline gear used by individual fishermen for their subsistence.

5 Payao Distribution

The distance between two payaos within Philippine waters is usually very short. For instance, in Panay Gulf in central Philippines, inter-payao distance was generally less than 2 km (Babaran and Biñas, 2001, unpublished data). This condition was due to the unregulated deployment of payao units by private fishing enterprises, which generally decide the distance between payao units based on economic considerations. Consequently, the waters influenced by two adjacent payaos apparently overlapped and made them compete for the same associated fish schools.

6 Catch Composition

Pelagic fish species are usually associated with payaos, but the catch composition according to the different fishing gears used near the floating structure is still not well documented. It was observed in Panay Gulf that about 20 species, including tunas, were captured by ringnets, whereas various handline gears caught 24 different species near payao (Babaran and Biñas, 1999, unpublished data). A similar analysis applied on the landed catch data from purse seines and ringnets in the main tuna fishing port of General Santos City in Mindanao indicated that the two gears captured similar species (Babaran 2006). Based on sampled catch landings of purse seine ($N = 73$), the most dominant species that occurred were skipjack tuna (100%), yellowfin tuna (99%), mackerel scad (89%), and frigate tuna (82%). More than half of the catch was skipjack (53%); other dominant species in terms of biomass were bullet tuna (20%; occurrence = 67%), yellow fin tuna (12.5%), mackerel scad (5.4%), and frigrate tuna (5%). These four species occurred in all sampled landings of ringnet ($N = 23$) and jointly accounted for almost 75% of the landed biomass; bullet tuna, which appeared in majority of the landings (87%), contributed another 21% of the landed biomass for this gear.

These results indicate that although the biomass of non-tuna pelagic species and small tuna (bullet and frigate tuna) was generally lower than those of large tuna species (skipjack and yellowfin), their occurrences were similar to the tuna species. Moreover, this suggests that small pelagics collectively are an important component of the landed catch of both gears.

The catch composition of commercial hook and line gears is distinctly different from those of purse seines and ringnets. This included billfishes and large adults of yellowfin tuna, bigeye tuna, and albacore tuna. Yellowfin tuna was by far the most dominant, both in terms of landed biomass and frequency of occurrence. This result indicates that this gear mainly captures large species.

6.1 *Size Distribution*

Small sized (mostly <30 cm) tuna species of skipjack, yellowfin, and bigeye tuna are usually captured by purse seines near payaos (Babaran 2006). The median length for these three species was between 25 and 26 cm. The median sizes captured by ringnet were slightly smaller (skipjack = 24 cm, yellowfin = 25 cm, and bigeye tuna 21 cm). The small size indicates that individuals of these species caught near payaos are most likely juvenile. However, this probably reflects the nature of Philippine fishing grounds, which are believed to be spawning areas for these species (Lewis 2005). Meanwhile, commercial handline fishermen generally captured bigger sized fish (>60 cm). The median lengths of yellowfin (113 cm) and bigeye (140 cm) tuna indicate that the individuals captured are probably spawning adults of these species.

6.2 *Maturity Stages*

One effect of payaos can be determined from the proportion of juveniles in the catch (Babaran and Biñas, 1999, unpublished data). In the case of ringnet, the maturity stages of selected tuna (skipjack, yellowfin, bullet, eastern little tuna) and non-tuna small pelagic (bigeye scad, roundscad, rainbow runner, and golden trevally) species were also investigated. An examination of over 175 individuals of eight different species indicated that almost all (95–99%) individuals of skipjack, yellowfin, and eastern little tuna were juveniles; only 36% of the bullet tuna were of the same category. Meanwhile, the proportion of juveniles among the captured bigeye scad and roundscad were much lower (5% and 20%, respectively); half of the captured rainbow runner and almost all of the golden trevally were immature.

7 Future Directions

Payao is a traditional device that supports both subsistence and commercial fisheries, enabling them to contribute significantly to the economy. Given this condition, payao fishing will most likely continue in the Philippines. However, adaptations of fishing techniques to catch the various associated species reflect the high level of consolidated fishing efforts near the floating structures. This, in turn, highlights growing concerns about overfishing and the need to regulate the deployment of payaos for effective payao fisheries management. As this study shows, it is also important to consider the impacts of these fishing activities on the sustainability of pelagic resources, especially large tuna species. Clearly, one priority area for research is the effect of the mesh size of purse seine and ringnets on the catch composition and size structure of tuna and small pelagic species. Although increasing mesh size will reduce the catch of juvenile tuna species, this is necessary because doing so may also significantly affect the catch of non-tuna small pelagic species. Results of such a study will provide relevant inputs to a new Philippine regulation that calls for an increase in the mesh size for these two gears.

Regulatory measures are necessary to ensure sustainable harvesting of fishery resources. Therefore, another area for investigation is the minimum distance between payaos. Undoubtedly, deployed payaos in the Philippines are too closely spaced and run counter to the suggested distance of 20 km in open oceanic waters (Holland et al. 1990; Marsac and Cayré 1998). However, since the latter suggestion applies for large tuna species, such as yellowfin tuna, it is probably necessary to propose an alternative distance that considers the economic viability of capturing not just large tuna species, but also nontuna small pelagic species.

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Monitoring of the Artificial Reef Fish Assemblages of the Marine Protected Areas Along the Alpes-Maritimes Coast (France, North-Western Mediterranean)

Pascaline Bodilis, E. Dombrowski, C. Seytre, and Patrice Francour

Abstract Artificial reefs have been deployed within the three marine protected areas located along the Alpes-Maritimes coast (Golfe-Juan, Roquebrune-Cap Martin, and Beaulieu-sur-Mer, France, North-Western Mediterranean). These protected – and no-take areas – were created between 1980 and 1983. They have been fully protected since their establishment, except in 2004 when anthropic activities (except commercial fishing) were exceptionally authorized. Moreover, there have been no park rangers to prevent poaching since 2002.

To carry out long-term monitoring of the marine organism – mainly fish – assemblages established in the artificial reefs, underwater visual censuses (UVC) were carried out in 1987/1989, 1998/2000, and 2008, according to a standardized method. All the fish species present on the artificial reefs were considered to assess density and biomass, but this method is time-consuming (divers formation to UVC techniques).

In 2008, in addition to the traditional UVC, a new sampling procedure of the fish assemblages, the FAST method (*Fish Assemblage Sampling Technique*) was tested. With this method, sampling is based on the presence/absence of a number of given species. This list of species includes only species of interest for commercial and noncommercial fishing.

A comparison of the UVC monitoring shows that density of the fish assemblages increased in the first 10 years, then did not increase during the last decade. These results can be explained by a lack of watching within the protected areas to prevent poaching since 2002 and to the reopening to the anthropic activities in 2004. The real efficiency of the artificial reefs to restore or sustain fish assemblages is then discussed.

Finally, a comparison of the two census methods (UVC and FAST) has enabled us to put forward a simple, low-cost, and relevant protocol to regularly monitor the fish assemblages of the areas.

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1 Introduction

Artificial reefs have been deployed within the three marine protected areas (MPAs) of the Alpes-Maritimes coast (Golfe-Juan, Roquebrune-Cap-Martin, and Beaulieu-sur-Mer, North-Western Mediterranean along the French coasts; see Fig. 1) created between 1981 and 1983 (Charbonnel 1990). These MPAs were created to sustain local professional fishing. These no-take areas have been fully protected since their establishment, except in 2004 when anthropic activities (except commercial fishing) were exceptionally authorized. To carry out long-term monitoring of the artificial reef fish assemblages, underwater visual censuses (UVC) were carried out in 1987/1989, 1998/2000, and 2008.

The aims of this study are to highlight the consequences on fish assemblages of the watching stop and of the brief opening of MPAs to anthropic activities except commercial fishing.

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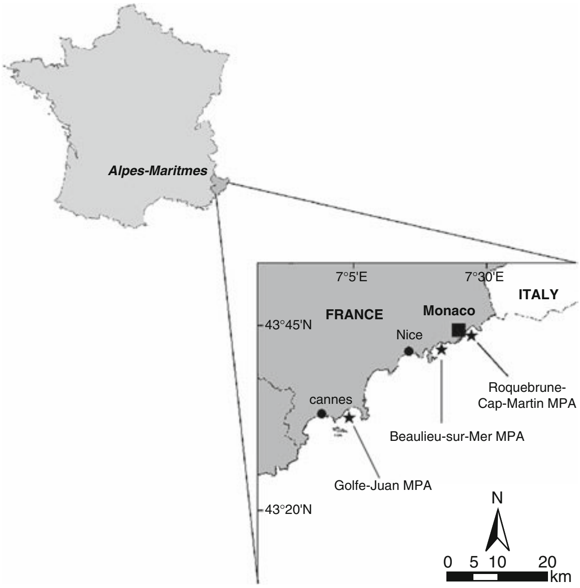

Fig. 1 Map of the situation of the different MPAs studied

2 Materials and Methods

Two underwater visual census methods were used during this monitoring.

The first method is a standardized method (Bortone et al. 2000). This classic method is based on the census of all the species present in and around the artificial reefs studied. Three size classes are retained: small individuals (to 1/3 max Total Length), middle individuals (from 1/3 to 2/3 max TL), and large individuals. It needs to be used by trained divers, and a long dive time is necessary to carry out the census. At each dive, only one reef is censused.

A second method was used for the first time in 2008. This method, called the FAST (*Fish Assemblage Sampling Technique*) method (Seytre and Francour 2008, 2009), has been used in other MPAs with great success (National Park of Port-Cros, Cap Roux, Var, France). This method is based on the presence/absence of several fish species. These species are "target species" that are interesting for professional and leisure fishing. Two size-classes are distinguished: small and middle individuals; large individuals. Several reefs are censused during each dive; these reefs are gathered together in areas. Each area is censused six times. From data obtained after these censuses, six indices are calculated.

Several structures were immerged on the sea-bed of the different MPAs studied. Some reefs, called Bonna reefs (Fig. 2), are giant reefs (158 m^3) made by nine perforated concrete slabs with wide void spaces. They were created to attract pelagic species. But after the first scientific survey in 1987/1989, it was observed that these reefs were not efficient. Thus, it was decided to complexify these reefs. Complexification consists of adding small elements (concrete bricks, tires, etc.) inside the Bonna reef to create several holes of different sizes. Thus, two types of reefs were censused and compared: empty Bonna reefs and complexified Bonna reefs (Fig. 2).

3 Results and Discussions

Consequences on fish assemblages of the watching stop and of the reopening can be seen both in specific richness and density data. The variation of the specific richness during 20 years of monitoring shows two periods of time (Fig. 3).

During the first 10 years, specific richness of fish assemblages living on artificial reefs increased. After the watching stop in 2002 and the reopening to some anthropic activities (2004), this increase of the specific richness has stopped. Opposite results can be observed for natural areas. These areas could constitute a "refuge" for fish.

Two periods of time can also be distinguished for the density (Fig. 4). Mean density per census is described for all species except for planktonophagous species that can mask trends. At Roquebrune-Cap-Martin and Golfe-Juan, the densities increased slightly during the first 10 years and then stopped after the brief reopening of MPAs to anthropic activities.

On the contrary, in natural areas, the densities decreased in the first 10 years and then stopped. The natural areas could then be seen as "refuge areas."

The second objective of this study was to observe the consequences of the complexification of some Bonna reefs, which took place after the first scientific survey in 1987/1989. Specific richness and density of complexified Bonna reefs increased rapidly after complexification (Fig. 5).

Complexification is an important method that allows increasing specific richness, but these species

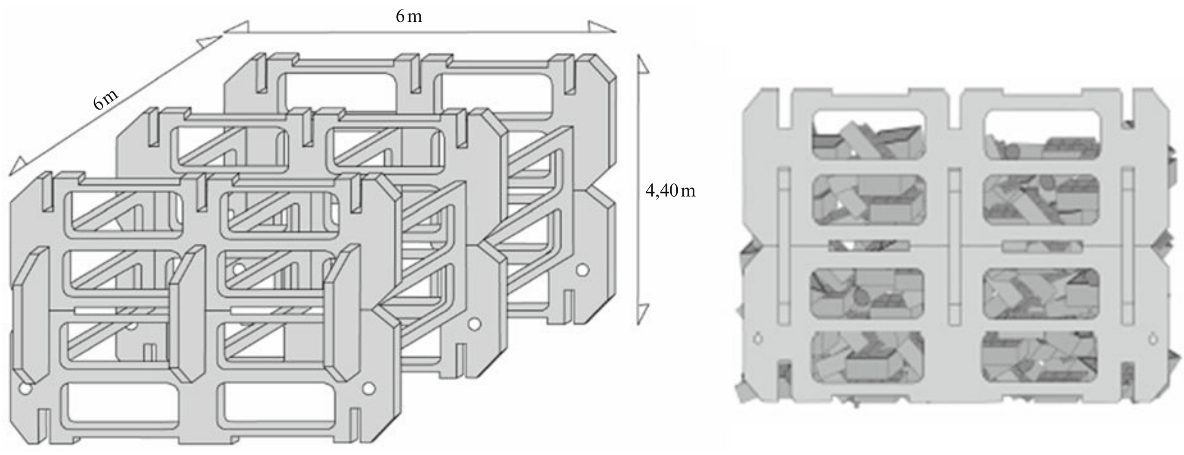

Fig. 2 Empty Bonna and complexified Bonna reefs (Charbonnel and Serre 1999; Charbonnel et al. 2001)

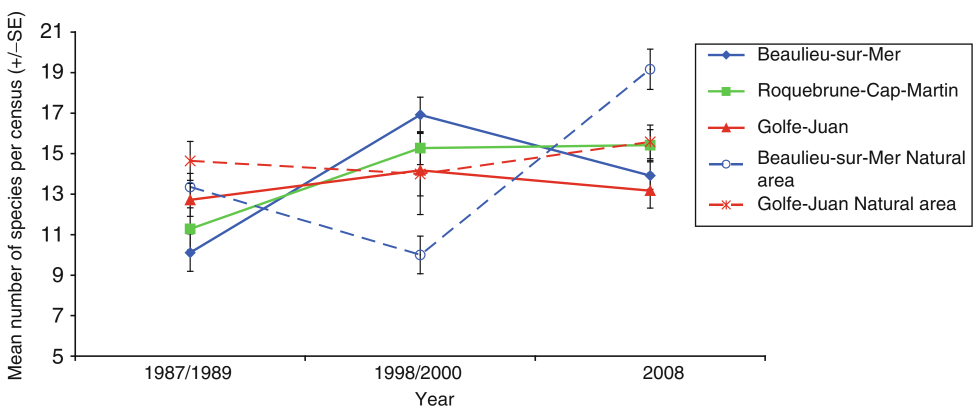

Fig. 3 Evolution of the mean number of fish species during the 20 years of monitoring in the three MPAs studied and in the two natural areas

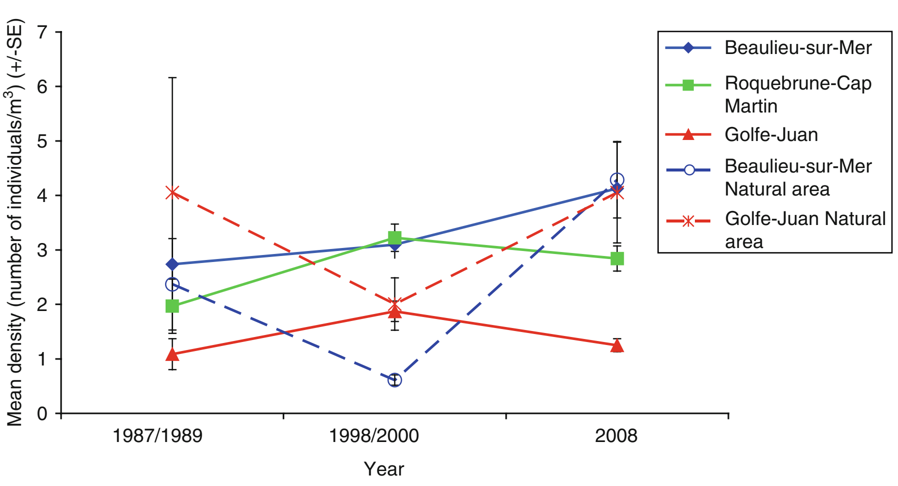

Fig. 4 Evolution of the density of fish during the three sampling periods in the three MPAs studied and in the natural areas

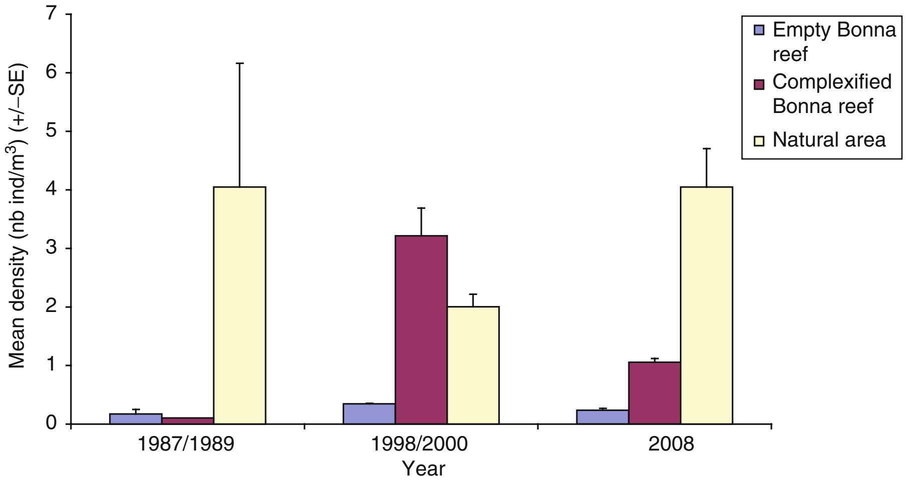

Fig. 5 Comparison of the fish density of different types of Bonna reefs between the three sampling periods

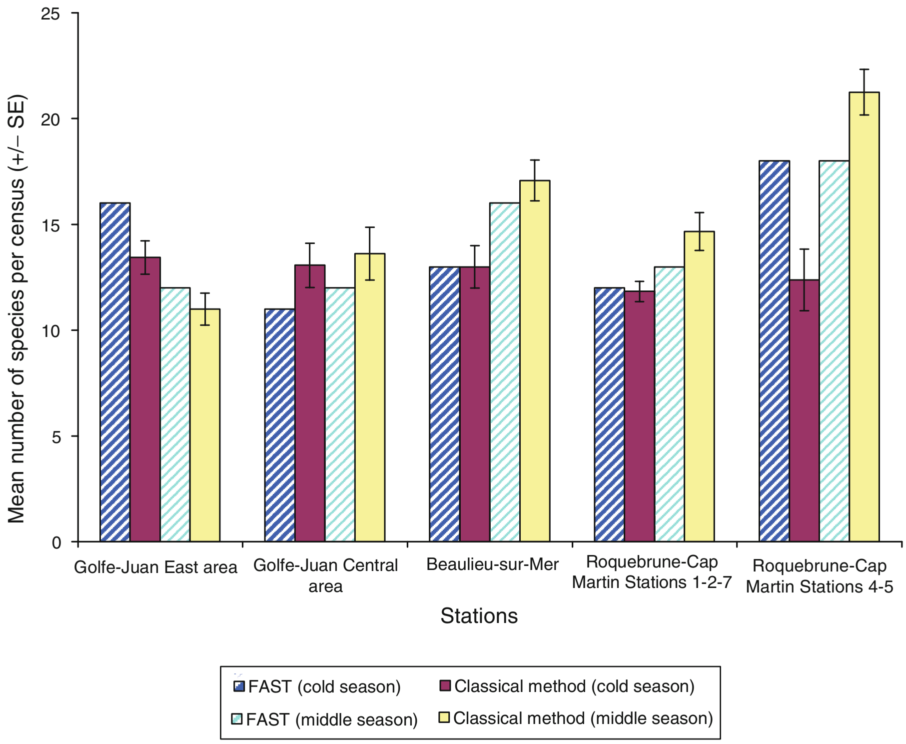

Fig. 6 Comparison of the results obtained for data of specific richness obtained with the classical method and with the FAST method during two seasons of the year 2008 in the different MPAs studied

are for the most part benthic species and are less interesting for professional fishing.

The last objective of this study was to compare results of specific richness and density obtained with the two underwater visual census methods (Classic and FAST) on fish assemblages.

Observed variations of the specific richness of fish assemblages in the different MPAs during the two seasons are comparable between the classic method and FAST method (Fig. 6). The number of species is more important during the middle season (June) in comparison to the cold season (February). Roquebrune-Cap-Martin is the richest area. The proximity of *Posidonia oceanica* seagrass beds near artificial reefs could explain this observation.

Similar results were observed for density and I_m, mean index, an expression of the density and one of the six indices calculated from data obtained during the six dives carried out per area. FAST is an interesting method to realize a fast and easy assessment of fish assemblages living in artificial reefs.

4 Conclusion

Specific richness as density of fish assemblages living on artificial reefs of the three MPAs studied has increased during the first 10 years, then stopped for the last decade. The increase in fish resources observed during the first 10 years of the study was halted by the watching stop since 2002 and the brief reopening of MPAs to anthropic activities in 2004. Despite integral protection since then, the lack of watching does not allow an increase in fish resources. Strong protection of MPAs where artificial reefs are submerged is important to allow the increase in fish resources and specific richness. Complex reefs are a good solution to increase specific richness, but ideal reef structures have been found to attract a great number of individuals of pelagic species. The new FAST method, which was used in this study, gives comparable results to the classical method. This method could be an interesting one for carrying out regular monitoring of fish assemblages of MPAs.

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Artificial Reefs in French Law

Bertrand Cazalet and Bernard Salvat

Abstract Studies on relationships between immersion and the economic exploitation of artificial reefs are relatively rare on the coastlines of France.

The biological effects produced by such installations are beginning to be known and scientifically demonstrated. However, caution is still required when assessing the economic benefits of artificial reefs. They may not be sufficient unto themselves, out of a broader framework of spatial regulation of professional and recreational fisheries and attendance. Each operation of immersion poses specifics problems (technical and practical) and requires obtaining prior administrative authorization, usually very stringent.

The marine environment is protected by the basic principles of public domain; any form of private occupation or use remains systematically temporary, precarious and revocable.

This first aspect is essential in the case of artificial reefs, in which immersion and productive potential are registered over the long term, without any real vocation to be removed from the seabed once their term expired. The administration is still very reserved about the perennity of artificial reefs, a concept against the classic rules in practice for the management of the public domain.

The second aspect lies in the arrangements concerning the exploitation of artificial reefs. Purely private access and use of artificial reefs seem irrelevant, given the potential conflicts they generate and the physical and legal impossibility of recognizing a form of private property of the species they house. The public or "collective" uses are certainly more appropriate to the situation of artificial reefs, they are *de facto* majority, but do not offer legible and uniform mechanisms for implementation.

Ultimately, artificial reefs offer a fragmented vision between different legal systems indirectly relevant to their case. Reflection about a formalization of their status opens up interesting perspectives concerning effectively taking into account the multifunctional specificities of artificial reefs.

1 Introduction

Artificial reefs are solid structures immersed directly and arranged on the seabed, without anchorage[1] in the subsoil. Conventional international law determines a minimum framework for the preservation of the waters and seabed, including the immersion of artificial reefs

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[1]For most artificial reefs located on the French coastline (main land of the hexagon), within the territorial sea and whose legal status is analyzed here. But many models exist around the world, targeting various types of species of pelagic and demersal fishes, shellfishes, crustaceans, and algae. Some "fish aggregating devices" (dispositifs concentrateurs de poissons - DCP in French) have fastening systems, connected in part to a surface float, whose components may "navigate" in the water column. Others may drift over the deep areas, while not having any form of anchor or ground contact.

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and assessment of risks inherent in such operations. At the state level, the design and construction of an artificial reef is now formalized and governed by strict rules, prohibiting use of any form of support to act as "artificial reef" poorly integrated to its natural environment (piles of tires[2], cars, poles, and other industrial waste). However, the laws applicable to the artificial reefs are not uniform (Quimbert 2005a). In accordance with the principles of management and protection of maritime public domain (MPD, or DPM in French: domaine public maritime), multiple conditions exist for the immersion of artificial reefs. The complicated juridical and administrative process involved and the various regulatory requirements keep the artificial reefs in an uncertain and uncomfortable legal situation. To date, we can identify three forms of legal documents authorizing the installation and/or exploitation of artificial reefs. However, it appears that marine farming concessions (see *4*) are no longer used in applications for the establishment of artificial reefs, although they represent the most thorough legal system and the most favorable to the exercise of private interests on the public maritime domain, which is perhaps the reason for its abandonment.

2 Immersion License

Established by Article 4 of the Act of 7 July 1976,[3] the immersion permit or license marks the emergence and implementation of principles established in 1972 by international agreements between Oslo and London. To allow its consolidation (coding), the 1976 Act was repealed by Ordinance of 18 September 2000,[4] itself amended by Ordinance of 18 July 2005.[5] Articles L218-42 *et seq.* of the Environmental Code refer to the London Convention to reaffirm the ban on the dumping of waste, while restricting the scope of exemptions. Indeed, apart from the dumping of vessels[6] and dredging,[7] such permits are no longer possible and seem doomed to disappear, except for those "regularly issued before the publication of Ordinance No. 2005-805 of July 18 2005 ... continue until they expire but shall not exceed ten years" (Article L218-44). Technically, only the reformed vessels are likely to be converted into artificial reefs. The dredgings are composed mainly of sand, gravel, and mud. They are soft sediments, partially biodegradable (organic materials, wood) and gradually dispersed as a result of spilling at sea. They are therefore unsuitable for creating artificial reefs. A number of penalties threaten potential offenders, including up to 2 years imprisonment and a €18,000 fine (article L218-48 and following).

The processing of permit applications is relatively formalized. This type of operation is likely to have a sustainable affect and change the seabed, and therefore first requires an impact study[8] and a public inquiry.[9] The information-gathering process is completed by the consultative Commission Nautique (boating/nautical advisory committee) consisting of ex officio[10] and temporary members.[11] These commissions have been

[2]We can quote a famous American example, presented in the *Thalassa* issue of 19 September 2008. In the 1970s, more than two million used tires were donated by the firm Goodyear and immersed off Ford Lauderdale, Florida. The "mountain" of tires has rapidly spread because of the currents, and the seabed covered has become a true desert of fauna and flora. Removal operations have now been started and should last another 30 years.

[3]Law No. 76-599 of 7 July 1976 on the prevention and suppression of marine pollution by dumping by vessels and aircraft, and the fight against marine pollution. Decree No. 82-842 of 29 September 1982 (JORF, 8 July 1976, p. 4107).

[4]Art. 5. I al. 10 & 11 of the Ordinance No. 2000-914 of 18 September 2000 on the legislative part of the Environmental Code (JORF no. 219 of 21 September 2000, p. 14792, text no. 39).

[5]Ordinance No. 2005-805 of 18 July 2005 on simplification, harmonization and adaptation of policies for water and aquatic environments, fishing and the dumping of waste (JORF no. 166 of 19 July 2005, p. 11760 text no. 42).

[6]Authorization granted by the State representative at sea (*Maritime Prefect*).

[7]Subject to the provisions of Articles L214-1 to L214-4 and L214-10 of the Environmental Code (JORF no. 219 of 21 September 2000, p. 14792, text no. 39).

[8]In accordance with Articles L122-1 *et seq.* and the R122-1 *et seq.* of the Environmental Code (JORF no, 219 of 21 September 2000, p. 14792, text no. 39).

[9]Articles L123-1 *et seq.* and R123-1 *et seq.*, Environmental Code (JORF no. 219 of 21 September 2000, p. 14792, text no. 39).

[10]Permanent and legal members: The Prefect, the Maritime Prefect, the Administrator of Maritime Affairs, the head of the district (Maritime Quarter) concerned and, where applicable, the representative of the Management Committee of the Marine Natural Park.

[11]Five sailors (professional and recreational) and their substitutes and representatives of the maritime service, and the director of the autonomous seaport and the local government service are involved in the project.

created by a decree of 1986[12] in order to examine, *inter alia*, "... any matter requiring consultation browsers sea" (art. 1 al. 3). Final analysis and observations are recorded in the minutes "sent to the maritime service, the seaport authority or the local government service, as appropriate" (art. 7). After obtaining this authorization, the permit holder must observe the safety measures in respect to staff employed to carry out the immersion and others and must also monitor the ships or aircraft involved in the immersion operations. At the request of the administration, the licensee may be obliged to conduct, at his or her own expense, work or study, or to take effective measures for possible consequences and for monitoring facilities. In fact, the administration imposes a regular assessment and consideration of the artificial reef, its effects, and risks to the natural environment, and also to people who use the sea, whether professionally or recreationally.

Inspired by international law, the permit to immerse artificial reefs is not concerned with the biological and economic benefits of installing such "recycled" reefs; only environmental considerations are taken into account. The reef should be properly and sustainably cleaned. Structures of this kind generally have a very large volume in a single mass (vessel) and are sometimes submerged at great depths, far from the shore. The authorization for immersion does not include the right to exploit the biological potential of the reef, which would be for the sole recipient of the title. Indeed, the installation of a reef of such a template is not, generally, the only private initiative and does not imply the granting of a concession. We speak, preferably, of a consensus among the State, project supervisor, company, contractor (permit holder), and the various stakeholders involved through nautical committees (fishermen, environmentalists, boaters, local elected officials and politicians, etc.).

Accordingly, the permit suggests a final and permanent immersion of materials (except in case of serious environmental problems), without any form of prescription for site remediation and removal of the reef. An artificial reef designed in this way would therefore be directly integrated into the maritime public domain as real estate (immovable property) by destination (the French term is *immeuble par destination*).

[12]Decree no. 86-606 of 14 March 1986 on boating (nautical) commissions (JORF of 19 March 1986, p. 4623).

3 Granting Use of Maritime Public Domain Outside of Ports

Described in French as CUDPM (concession d'utilisation du domaine public maritime en dehors des ports), it replaces the "containment concession" that has existed since 1979.[13] Founded in 2004,[14] this authorization allows a very broad definition of the concept of *use* of the MPD (marine public domain), thus opening the door for artificial reefs. The concession is applicable to various forms of work, facilities, and/or uses, subject to assigning them to public use, to a public service, or to an operation of general interest (Becet 2004). After a rather conventional administrative instruction procedure,[15] the CUDPM is granted by order of the Prefect (art. 11) for a maximum of 30 years, with implementation conditions strictly regulated by Articles 8 and 9 of the decree. First, the dealer must be able to guarantee "the effective reversibility of changes in the natural environment." Due to the precarity of authorizations for MPD occupation, when they arrive at their term or if permission is withdrawn prematurely by the administration, the owners[16] are forced to repair, restore, or rehabilitate the occupied sites. For this purpose, the licensing authority requires financial guarantees according to the estimated potential cost of such operations. Second, the decree of 2004 states that the CUDPM is not constitutive of "real rights" (property rights) and does not confer the commercial property to its owner.

Do we consider the use of CUDPM as the most appropriate legal system for the situation of artificial reefs? The original purpose of containment and the associated concessions was precisely to allow for implantation around beach areas[17] in order to "win land from the sea" (Quimbert 2005b). The immersion

[13]Decree no. 79-518 of 29 June 1979 relating to containment concessions and use dependencies of maritime public domain maintained in this area outside ports (JORF of 1 July 1979, p. 1593). Repealed by Article 13 of Decree 2004 (JORF no. 76 of 30 March 2004, p. 6078, text no. 40).

[14]Decree no. 2004-308 of 29 March 2004 (JORF no. 76 of 30 March 2004, p. 6078, text no. 40).

[15]Request to the Prefect, the followed including an impact study, a public inquiry, and the convening of an *Advisory Boating (nautical) Committee*. See Articles 6 and 7.

[16]Individual person or private law corporation.

[17]Article 1 of Decree of 2004 states that CUDPMs are distinct from beach concessions *sensu stricto*, but are discussed in Decree no. 2006-608 of 26 May 2006.

of materials and layout work on the seabed are not mentioned in the Decree of 2004. The duration of the concession for 30 years (non-renewable) does not seem to be compatible with the physical characteristics and long-term goals of artificial reefs.[18] Because of the weight and volume of the modules used, the development of artificial reefs is time-consuming, difficult, and costly. The reefs are a series of stacks and casings of the grantor, which in the end is a mass that is too heavy and dense. Eventual dismantling of a reef after several decades for the rehabilitation of the site would obviously be destructive and counterproductive, except for bringing to light any evidence of the failure of the the biological and economic effectiveness of the submerged structure. This conjecture remains improbable because the very purpose of artificial reefs is to recreate shelter and attachment points as close as possible to the natural habitats[19] rich in biodiversity. In addition, they are usually installed in naturally poor areas or, more frequently, where the seabed has been depleted and degraded by multiple anthropogenic pressures. In this case, the dismantling of an artificial reef in a good state of conservation would be absolute nonsense, negating all benefits sought for and brought to the restoration of the marine environment.

Finally, in the first article of Decree of 2004, we note the oddity that "the provisions of this Decree shall not apply (...) for marine culture (farming) exploitation." However, the legal possibility to immerse artificial reefs by either of the two procedures demonstrates the contrary. This measure determines the necessary scope of CUDPM to avoid any risk of confusion with other models of concessions, but more importantly illustrates the inadequacy of these concessions in the area of artificial reefs, a category that is still unclassifiable and therefore unclear.

4 Authorization of Marine Culture Exploitation

Used for a long time in the case of artificial reefs, the authorization of marine culture exploitation (farming) has not been feasible since the 2000s. Being relatively old,[20] this authorization does not concern, as its name suggests, the immersion of artificial reefs. Its extension to the latter is the result of a flexible interpretation of the definition of the activity of "marine culture" and its contents. *Sensu lato*, marine cultures include shellfish farming (oyster and mussel cultures) and continental/marine fish farming, related to a general legal system of an agricultural[21] nature. Article 1-§1 of Decree of 1983 specifies that these farms are "for the purpose of organic production, including capture, breeding (farming), refining, processing, storage, packing, and shipping of seafood." The idea of biological productivity through capture and breeding (farming) is extrapolated to the case of artificial reefs in terms of economic profitability. Indeed, the installation of new habitats attracts, protects, especially juveniles, and creates areas of concentration and settings for species (fish, crustaceans, algae, shellfish).

Permits are issued by the Prefect (administrative instructions) following similar methods to those desecribed above, but also involving a marine culture committee (art. 3). The title of occupancy, valid for a maximum of 35 years, which is renewable once, is accompanied by specifications laying down conditions for the occupation and use (form of installations and recommended improvements), the nature of the cultures and technology being proposed, the amount of the fee for occupation, the presence of easements, etc. According to Art. 5, the beneficiaries are individuals or private law corporations. The allocation to public law corporations (state, local government service, public institution, etc.) shall be admissible only for the purposes of "collective actions" contributing to the development of aquaculture.

[18]Even though the administration does not share this view and is very cautious about the durability of artificial reefs: See legal update following the publication of decree No. 2004-308 of 29 March 2004, entitled "The legal feasibility of the implantation of artificial reefs on the coastline", *Response of Direction du Transport Maritime, des Ports et du Littoral (Directorate of Maritime Transport, Ports and Coastline) to a Prefect (maritime service) dated 18 October 2000*, April 2004, p. 3.

[19]The research and efforts towards maximum integration, even an imitation, led some authors to prefer the expression "artificial habitats" to that of artificial reefs. See S. Pioch, *Les "habitats artificiels" : élément de stratégie pour une gestion intégrée des zones côtières ? Essai de méthodologie d'aménagement en récifs artificiels adaptés à la pêche artisanale côtière*, Thèse de doctorat de géographie, Université de Montpellier III Paul Valéry, mai 2008, 289 p.

[20]Decree no. 83-228 of 28 March 1983 (JORF of 25 March 1983, p. 918).

[21]Article L311-1 of the Rural Code (JORF of 23 July 1993).

Resolutely oriented (directed) towards the development and recovery of marine activities, marine culture concessions recognize “real rights” (property rights or real property) or “quasi-real”[22] rights of their operators. These rights, expressed in Articles 9 to 14 of the decree, based on the personal nature (Art. 5.2) of the permit and its exploitation, give the right of use and enjoyment (*usufruct*) exclusively to its “owner.” Thus, in the owner’s absence, he or she may temporarily transfer its use to another operator. Many dealers can form a private corporation to undertake the exploitation of the structures. As long as the title is valid, the beneficiary may request the transfer of his or her rights to a third party. The exchange of equivalent concessions between two operators is also possible. Finally, the transfer to a surviving spouse or direct-line heirs helps to overcome the difficulties involved in the sudden death of the concessionaire (holder). All these possibilities of change are of course regulated and subject to prior approval of the administration. As in the case of CUDPM, the authorizations of marine cultures suffer from the same threat related to the prescription and the rehabilitation of the site at the end of the concession. A “classic” and normal exploitation of marine culture (shellfish tables, poles, nets, and aquaculture ponds) does not pose serious problems of decommissioning. This is not the same for an artificial reef installed several tens of meters deep, weighed down by the organic concretions, and that can be removed at the cost of the concession holder. The ordinary life of a reef is not ephemeral, the results are revealed slowly, and such a structure is not intended to deconstruct.

Aquaculture or shellfish activities open the possibility for operators to be the exclusive owners of their live products. Indeed, a fish trapped and bred (for farming) within a fence or shells mounted on ropes, wires, or pickets belong to the holders of concessions, compared to a fish or a crustacean colonized within an artificial reef, which still retains its status as *res communis*. The animals belong to anyone until they are captured by the first fisherman, a holder of the concession or not. An operator or a group of farmers who have exclusive access and use of the reef cannot be considered the owner of species that live there, since no physical barrier could prevent the animals from moving outside of the reef. Therefore, it is easy to imagine the conflicts that can arise among different stakeholders in the sea, whether professional fishermen, boaters, or scientists. How can we effectively define the surface contours of an artificial reef? How can we monitor and articulate pragmatic uses and their users? How can we find a balance among the protection of an artificial reef, especially at the beginning of an installation, and easement of passage and attendance over the concession.

5 Conclusion

Since a letter from the Minister of Agriculture and Fisheries on 17 March 2000, “the Decree of 22 March 1983 cannot be used to authorize the establishment of artificial reefs.” However, the letter states that according to Article 19 of the Decree and in exceptional circumstances, such authorization may be granted for artificial reefs with an “experimental purpose (test) for the protection, conservation, and regeneration of the seabed” and be exempted from payment of the fee. Only interprofessional agencies (Law of 2 May 1991) and scientific organizations can benefit from these special permits. In the end, the new legal system of the CUDPM[23] is now favored by the administration for future artificial reef projects. This reversal has not been consolidated in legal terms, such as by adding an amendment to the Decree of 2004 on CUDPM to incorporate artificial reefs permanently. The term “artificial reef” has still not been added, also not to any other regulation in force. Currently, the administration has begun a process of legal “requalification” (in CUDPM) of artificial reefs installed under the

[22]Expression of Quimbert, M. (2005) “Les récifs artificiels: autorisation d’immersion, régime d’occupation du domaine public et cadre d’exploitation en droit français”, Revue juridique de l’environnement, 2/2005, p. 129.

[23]The ambitious project in Marseille, located off the Prado Bay, is exemplary of the limits of CUDPM (concession d’utilisation du domaine public maritime en dehors des ports) to determine the conditions of exploitation after the immersion. For now, the reef has been permanently installed since July 2008 and classified as a strict nature reserve (no-take, no-entry) for 2 years to become progressively (a) a colonized habitat and productive. This period was used by the city of Marseille (holder of the reef concession) to encourage various stakeholders to reach agreement on conditions for future exploitation of the reef ... if no agreement, the strict nature reserve will be renewed 2 more years by the City. The CUDPM offers no predetermined solution or framework in terms of management and use of artificial reefs.

framework of marine cultures. By way of comparison, and by extension, we mention the example of Japan, unanimously recognized by experts as the world reference for artificial reefs. Traditionally, the priority of access and use of the sea is given to fishermen. The Japanese legal framework allows the creation of property rights for the management and exploitation of artificial reefs. The arrangements for the exercise of these rights are based on groups of fishermen, identified and representative of the same village, cooperative, or region. The Japanese model is conceived as a seaward extension of land use rights, with a vision of rural development and a fruiting seabed (Cazalet 2009).

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Contribution to the Planning of the Research in Artificial Reefs Programs

Hubert Jean Ceccaldi

Abstract The final aim of man made structures is to simulate some parts of the natural environment where the populations of different species have evolved and where they are living. These structures may be set down on the bottom of the sea, set at mid depth in the water mass, and even floating at the surface.

In terms of ecological protection and species consumed by mankind, is it possible to obtain better results than natural environments to obtain increased profits from natural production carried out in the euphotic zone?

Systematic assays have to be done to understand more clearly the effects of the factors affecting the attraction, the presence, and the growth of different species living inside and around these artificial habitats.

The research subjects are numerous and are different from one site to another. Among them, specific studies have to be developed on the effects of reef sizes on the presence of certain species not only inside the reefs, but in also in its wake, in the close sediments, above the reefs, and on the surrouning ecosystems.

Surface studies have to be developed as well, in terms of micro- and macro-rugosity, chemical nature (adhesive biological cements), optimal sizes of microholes, and habitats, for instance. Effects on successive bacterial populations have to be known as well.

Ecological studies are necessary concerning the water quality of the currents flowing through the artificial structures; the number and nutritional quality of particles transported inside the reefs, then consumed by filter-feeder species, various larvae of vertebrates and invertebrates, and the vagile fauna. Microphytobenthos, endogenous fauna, and bioturbation need to be understood with some precision.

The production of eggs and larval forms of different species and their exportation in the surrounding ecosystems have to be evaluated.

Physical aspects of the presence of these artificial structures, modifications of local currents, induced turbulences, and modification of granulometry are also other subjects of researches; local hydrology controls organic production and biological oceanography: these elements have to be known with precision in each site.

Chronobiological and seasonal behavior of different species of fishes, crustaceans, and molluscs (larval, and adult forms) associated after some time into and around these artificial structures need to be described not only concerning their presence, but also their behavior, their stomach content, their physiology.

Nutrition studies may give various subjects of comparative experimental approaches in species living in nature and maintained in large aquariums.

Even if artificial reefs concept is known since a rather long time, numerous subjects of researches have to be developed to make them producing more than nature, and, in the same time, to protect natural environment.

1 Introduction

Coastal management begin to be a general problem in littoral zones of almost every country. Since a long time, very often these areas have been misused and even destroyed. To reconstitute valuable ecosystems,

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the managers, public, and sometimes private are thinking to immerse artificial structures to give some habitats to fish populations. The observation of various wrecks attracting numerous fishes lead to the conclusion that it is sufficient to set down various materials to "enrich" the bottom of the sea, and to attract marine flora and faunas.

In fact, there is a long history of these submarine managements as for instance the immersion of bamboos and other vegetal materials along the coasts of Japan, Korea, and China. A national program of such installations have begun since more than 40 years in Japan, which is more advanced in the world in that field.

They set down these structures at mid depth in the water mass and even floating in the surface, in order to simulate some parts of the natural environment where the populations of different species have evoluted and where they are living.

Sometimes, in different countries, some advisors are recommending to immerse old ships, as battle ships, aircraft carriers, and other ancient vehicles to create a favorable submarine environment. The examples are numerous, as for instance the aircraft carriers USS America in Virginia coasts (2005) and the USS Oriskany in the Gulf of Mexico (2006). Old school buses and metro wagons have been also used in these purposes.

These approaches are too rough to be really adapted at the fragile equilibrium of the coastal ecosystems, delicate and complex. For an ecosystemic point of view, the question is rather: Is it possible to obtain better results, in terms of species living their biological cycle in these artificial habitats, consumed by mankind and constituting an ecological protection, than natural environments to obtain a better profit of the natural production synthesized in the euphotic zone?

Systematic experimental assays and detailed observations have to be made to understand more clearly the effects of the different factors affecting the attraction, the presence, and the growth of different species living inside and around these artificial habitats.

In this respect, the term of "artificial reef" (in French: "récif artificiel") seems to be inappropriate and it would be better to use the term of "marine – or submarine – habitat."

The subject of researches are numerous and are different from one site to another.

2 Themas of Research

2.1 Coastal Oceanography

As the artificial structures are put down on the bottom of the coastal waters, the managers have to know what will be the oceanographical characteristics of the areas where these structures are located. Local sea water hydrology controls organic production and biological oceanography.

That means the currents, the temperature, the nutrients, the turbulences, etc., explaining not only the organic production but also the distribution of the nutritive particles produced in euphotic zone, arriving on the artificial structure immersed, where the filter feeders are collecting them.

2.2 Physics

Sizes and designs of reefs are very important factors. Some of them are constructed near the bottom and, on the contrary, other ones may occupy a part of the water mass, for instance in the case of few Japanese enterprises who constructed structures of 35 m in height.

Such a physical and technical evolution in their sizes lead to new kind of problems, as their construction, their immersion, their stability, and the observation in situ of the numerous organisms inhabiting in these artificial structures.

Studies on reefs surfaces have to be developed as well, in terms of micro- and macro-rugosity, chemical nature (adhesive biological cements of different larvae during their metamorphosis), optimal sizes of microholes, and habitats for instance. The effects of this character on successive attached bacterial populations have to be known as well.

Modifications of the natural sites where the artificial structures have been set down is also an important factor, because their presence modifies, more or less, the natural physical conditions: physical aspects on currents modifications, on induced turbulences in the water masses, etc.

2.3 Sedimentology

Modifications of granulometry under the influence of the artificial structures in each site are also other subjects of researches these elements have to be known with precision. As they are obstacles to the transport of suspended materials, the sediments are modified mainly close to the reefs: a gradient is setting up showing the largest and the heaviest particles the closest ones.

Accordingly, the distribution of the benthic endogenous species varies greatly. Another consequence is the gradient in the chemistry of the sediments, the bioturbation, and the oxygenation of the different parts of the affected surfaces. They have to be studied in detail, using for instance optrodes technology.

2.4 Marine Biology

Even if some are quite well known, many marine species, even the more popular ones, have unknown biological cycles. Frequently, the adult forms are well described and are observed by divers and scientists. Nevertheless, their larval forms are generally much less known, and their biology, their habits, the fields where they are living and growing, where they gather, what they eat at different steps of their growth are, in numerous cases, unknown.

At the opposite point of the biological cycles, we have a few information concerning the condition of reproduction, the environmental factors affecting the growth of the ovary in the females of different species, the process of mating and fecondation of the ovulas, the embryogenesis, the dispersion of the eggs, etc. What is true for the current species is much more true in less represented species, especially the noncommercial and the less spectacular ones.

In straight connection with these new knowledge in marine biology, ecological studies are necessary concerning the effects of water quality and of the currents flowing through the artificial structures; filter-feeders species under the form of various larvae of vertebrates and invertebrates, as well as the vagile faunas are mainly concerned. Microphytobenthos, endogenous faunas, bioturbation processes need to be known with more precision than actually.

2.5 Physiology

Although we may gather a good knowledge concerning the biology of numerous species living in north mediterranean coasts, our frequent ignorance in their physiology is evident. Very often, the optimal conditions of their growth, the evolution of their food at different steps of their biological cycle, etc., explain major factors of their presence and their distribution is not very known.

On the other hand, their chronobiological and seasonal behavior of different species of fishes, crustaceans, and molluscs (in their larval and adult forms) specially when they are associated in the same site after some time into and around these artificial structures need to be described not only concerning their presence, but also their behavior, their stomach content, their respiration, nutrition, excretion among the major ones.

Collection of observations and detailed growth studies in artificial conditions of these larval forms and of the species of small sizes have to be developed if we wish to understand the food chains, the trophic networks and, in other words, the functioning of the local ecosystem established around and inside the immersed artificial structures. In other terms, ecophysiology and ecobiochemistry will be used in this field as well as in aquaculture (Ceccaldi 1982), for the same reason: the detailed explanation of the adaptation of living organism to a changing environment.

2.6 Nutrition

The animals living in straight relationships with the artificial structures are mainly consuming, directly or indirectly, the nutritive particles produced in the euphotic zone, as well as (or) the algae attached on the immersed surfaces and growing using nutrients present in the sea water where they are living.

Number, biomass, and nutritional quality of particles transported inside the reefs have to be known in detail, quantitatively and qualitatively in their biochemical composition: protein, carbohydrate, lipid, mineral composition are the most important.

Enzymatic studies are necessary (Ceccaldi 2006) to begin to understand the ability of the different species to efficiently use the natural production.

Nutrition studies may also give various subjects of comparative experimental approaches in species living in nature and maintained in large experimental aquariums.

2.7 Ecosystems and Role as Seminatural Park

Implantation of artificial structures are modifying the natural ecosystems, at least locally. A new equilibrium is established after some time, several months, even after several years. It is necessary to follow these evolution and the managers of the affected areas have to compare the zone of implantation and the evolution of the ecosystems existing in the closest homologues zones.

It seems perhaps paradoxal to treat the subject of "natural" park in a chapter dealing with "artificial" reefs and we prefer to use the expression "seminatural" park when a field of reefs is located in a protected area. Among them, specific studies have to be developed on the effects of the reefs – sizes, volumes, shapes – in the surrounded ecosystems, particularly on the presence of the preexisting species, not only inside the reefs, but in its wake, in the close sediments existing in the surrounding areas, above the reefs, and even rather far from them.

On the other hand, the implantation of artificial habitats happens close to human activities and these zones are very often under the direct – or indirect – influence of the industry and various managements.

In a few words, the reefs have to be integrated without perturbations in the coastal areas where they have been set down. When they are exploited by fishermen, they may use the new and diversified habitats of the reefs, especially if they are sufficiently complex and, as a consequence, they reconstitute the natural populations' specific of the local hydroclimate.

The production of eggs and larval forms of different species and their exportation in the surrounding ecosystems have to be evaluated.

The quantitative evaluation of the different species filling the space in and around the reefs is a rather difficult problem, as the mobile species are moving, as they follow some chronobiological and seems to disappear at some hours of the day, as there are some seasonal variations in the presence and the absence of some species. The detailed study and the exploitation of these living resources linked with the reefs have to take into account these particular characters. Observations in situ of some species by divers and/or scientists do not necessarily mean that the fishermen will fish the observed species in the same place.

2.8 Economy of These Submarine Animal Habitats

Economy of the implantation of artificial reefs is far to be established in good conditions. There are no precise methods to evaluate the cost of investment and the cost of return, in terms of efficiency. Generally, the cost of the fabrication and the installation on the seabed come from public budgets.

Fishermen and private companies are not rich enough to spend their money to increase the complexity of the bottom of the ocean to gather more marine animals and vegetal species. In Japan, where the fishermen are well associated to the decisions and the exploitation of the artificial structures, different levels of administrative and political levels are concerned; also the fishermen themselves are participating in the investment – at a modest level – but they are directly concerned indeed.

2.9 Management

Installation of artificial reefs on the seabed and the sea shores have to be integrated in the coastal management and have to follow the local rules as well as the general ones.

Survey and police among their fisheries will become a necessity. But a strong basis of specific education is also necessary, when the cost of the survey becomes too high in regard to the resources collected.

Concurrence with new constraints and occupation of the sea coasts by the offshore wind propellers implanted in the seabed. Their basis play unavoidably a role of artificial reef.

2.10 Relationships with the Fishermen

This aspect is certainly more sensible and depends essentially on the culture and the practices of the corporation of fishermen in the different countries. In Japan, where the fishing activities on shores are regulated mainly by cooperatives ruled by a long tradition, the exploitation of artificial reefs fields is regulated by fishermen themselves. They are knowing more or less roughly the presence of such or such species and they keep the resource generally in a good condition.

In other countries, the fishermen are rather competing or even fighting among them to catch the more possible in the resource.

There are necessarily a planification in the vocational training and in the pedagogy of that corporation to make them adjust their behavior and their activities to the existing and limited resource existing inside and around the reef fields.

2.11 Roles in Touristic Diving Activities

Artificial reefs are very attractive elements for tourists, specially the divers using bottles and/or the apneists. They know that, as in and around wrecks, they are sure to observe rich marine fauna and flora. Some of them are diving to hunt fishes and collect animals.

Some countries have taken the decision to sink old boats and warships, metro used wagons to make – they assert – new artificial reefs. They do not represent specific and specialized habitats for marine animals, even if all wrecks attract fishes. In other countries, some artificial wrecks have been immersed to attract tourists.

2.12 Juridic Status

This status is far too established. Some simple – and fundamental – questions are actually without reply: Who invests? If the aim of the reefs is oriented to be exploited by fishermen, do they have the obligation to invest? Themselves? Their cooperative? Other professional associations?

When the structures are set up on the bottom of the sea, who is their owner; the closest municipality? The State? The main investor? The fishermen exploiting them?

When the marine products living into or near the artificial reefs are caught, who is their owner? The fishermen? The designer of the reefs? The main investor? The administration in charge of the marine coastal areas?

If a diver visiting the artificial reefs structures has an accident inside the reef and dies into the sea, who is responsible? The designer of the reefs? The constructor?

Finally, as scientists and lawyers, we have to establish new forms of rules concerning the property, the exploitation, the safety, the ecology of natural environment where these reefs are constructed. They have to be really general, i.e. to be applied in any country, because the rules of the ecology and the protection of the environment are concerning any coastal areas, any part of the natural environment.

3 Conclusions

Artificial reefs concept is known since a long time. Engineers and coastal managers are usually thinking that this subject is well known. Marine biologists and marine ecologists, who are more familiar with the complexity of ecosystems and their natural processes, recently recognized the necessity to make progresses in new fields of research.

New programs have to be developed in coastal oceanography, in physics, in sedimentology, in sea water and sediment chemistry, in marine biology of larval and adult forms, in physiology, in nutrition, and in ecosystems. Some relatively neglected aspects have also to be improved, as their role as small natural park, the economy of these submarine animal habitats, their importance in the coastal management, the relationships with the fishermen, their roles in touristic diving activities, and their juridic status.

These subjects of researches have to be developed to make these structures produce more and better than nature, and, at the same time, to protect the natural environment.

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Artificial Reefs in the Cote Bleue Marine Park: Assessment After 25 Years of Experiments and Scientific Monitoring

Eric Charbonnel and Frédéric Bachet

Abstract The Côte Bleue Marine Park (PMCB) was one of the precursors in France concerning the deployment of artificial reefs (AR). For 25 years, the Park has led several varied operations by using different kinds of modules: between 1983 and 2004, seven types of architecture for production reefs and five architectures for protection reefs were studied, for a total AR volume of 4,884 m^3, and represent an investment of €480,000.

The purpose of the AR deployment policy of the Park was to experiment and test various types of modules, during small-scale operations, but including several phases of immersion and scientific field surveys. These experiments allowed development of the conception of modules and adapting them to the local context, targeting several objectives. Moreover, they are used to support small-scale artisanal coastal fishing by two complementary aspects:

(1) The promotion of biological production in impoverished seabeds and sustaining professional artisanal fishing, with 2,684 m^3 of modules arranged in chaotic heaps in five areas, and increasing the biodiversity and the available fishing resources.

(2) The protection of priority natural habitats (*Posidonia* meadows and coralligenous rocks) and to manage and share both resources and fishing grounds among fishermen, with 326 heavy obstacles designed to protect against illegal trawling within the 5.5 km offshore limit. These 2,200 m^3 of anti-trawling reefs are spread along 17.5 km, creating barriers perpendicular to the coast. Since they have been installed, a significant decrease in illegal trawling has been observed.

In the Côte Bleue Marine Park, the two categories of AR (production and protection reefs) are interrelated in the two protected marine areas (integral reserves of 295 ha where all kinds of fishing activities, scuba diving, and mooring are prohibited).

These management tools have worked in an additional way and contributed to the preservation of the traditional small-scale fisheries in the Côte Bleue territory (for about 60 fishermen) at a time when these fishing activities are decreasing in the nearby zones.

1 Introduction

The Côte Bleue Marine Park ("Parc Marin de la Côte Bleue", or PMCB) was one of the precursors in France concerning the deployment of artificial reefs (AR). For 25 years, AR deployments have concerned both ecological aspects and promotion of fisheries and resources, with two complementary kinds of AR: protection reefs against illegal trawling on the coastal band and production reefs, for a gross volume of 4,884 m^3. The aims of this paper are to provide some assessments of the experiments conducted; to describe some aspects of AR as tools for integrated management of marine resources and sustainable fisheries; and to confirm the crucial influence of reef design, architecture, and layout on the biological effectiveness of AR.

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2 Presentation of the Côte Bleue Marine Park

The PMCB, located east of Marseille (Fig. 1), was created in 1983 with the same objectives as terrestrial natural parks: (1) protection of the natural environment, (2) preservation of traditional fisheries and participation in better management of resources, (3) increasing public awareness and education about the environment, and (4) promotion of experiments and research.

The PMCB is a public institution involving five cities of the coast "Côte Bleue" (Martigues, Sausset-les-Pins, Carry-le-Rouet, Ensuès-la-Redonne, and Le Rove), local governments ("Région Provence Alpes Côte dAzur," "Département des Bouches du Rhône"), and professional organizations of fishermen ("Comités Locaux des Pêches Maritimes" and "Prudhomies" of Marseille and Martigues).

The PMCB operates along the 40-km coastline of "Côte Bleue" up to 4 km offshore, between the gulf of Fos and Marseille. The Park manages two marine protected areas (MPA), no-take zones where all kinds of fishing activities, diving, and anchoring are prohibited: the Carry-le-Rouet reserve (85 ha protected since 1983) and Cap-Couronne reserve (210 ha protected since 1996; Fig. 1). For 25 years, the PMCB has led several programs of AR deployment, at an experimental scale, for a gross volume of 4,884 m^3. The two categories of AR used (production and protection reefs) are interrelated in the two integral reserves. Additionally, they work in an additional way towards the improvement of marine biodiversity and fishing resources.

Since the creation of PMCB 25 years ago, educational training has been created for schooled children, with activities at sea during 4 days per school. More than 22,000 children have attended these sea programs. Since 1994, an underwater pathway in MPA has welcomed the public during visits organized by the Park in the summer.

3 Description and Objectives of Artificial Reef Deployment

For 25 years, the Park has led several and varied operations by using different kinds of modules: between 1983 and 2004, seven types of architecture for production reefs and five architectures for protection reefs were studied (Fig. 2), for a total AR volume of 4,884 m^3, and represent an investment of €480,000 (Table 1). Professional fishermen are often the originators of AR projects, which can involve funding of up to 50% by the EU (EFF, European Fisheries Fund) and local governments. The goal of the AR deployment policy of the Park was to experiment and test various types of modules, during small-scale operations, but including several phases of immersion and scientific field surveys. These experiments allowed development of the conception of modules and adapting them to the local context and targeting multiple objectives. Moreover, they are used to support small-scale artisanal coastal fishing by two complementary aspects:

(1) The promotion of biological production in impoverished seabeds and sustaining artisanal fishing,

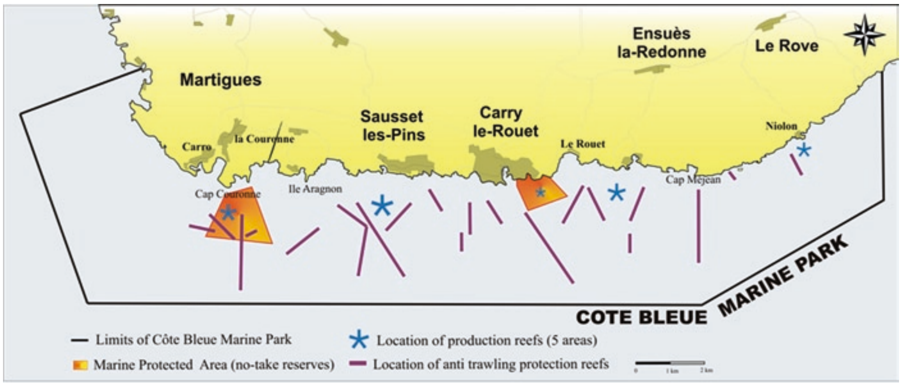

Fig. 1 Presentation map of the Cote Bleue Marine Park territory: a concession of 9,873 ha delivered on December 15, 2003 along 40 km of coastline to 4 km offshore, between Marseille and Fos

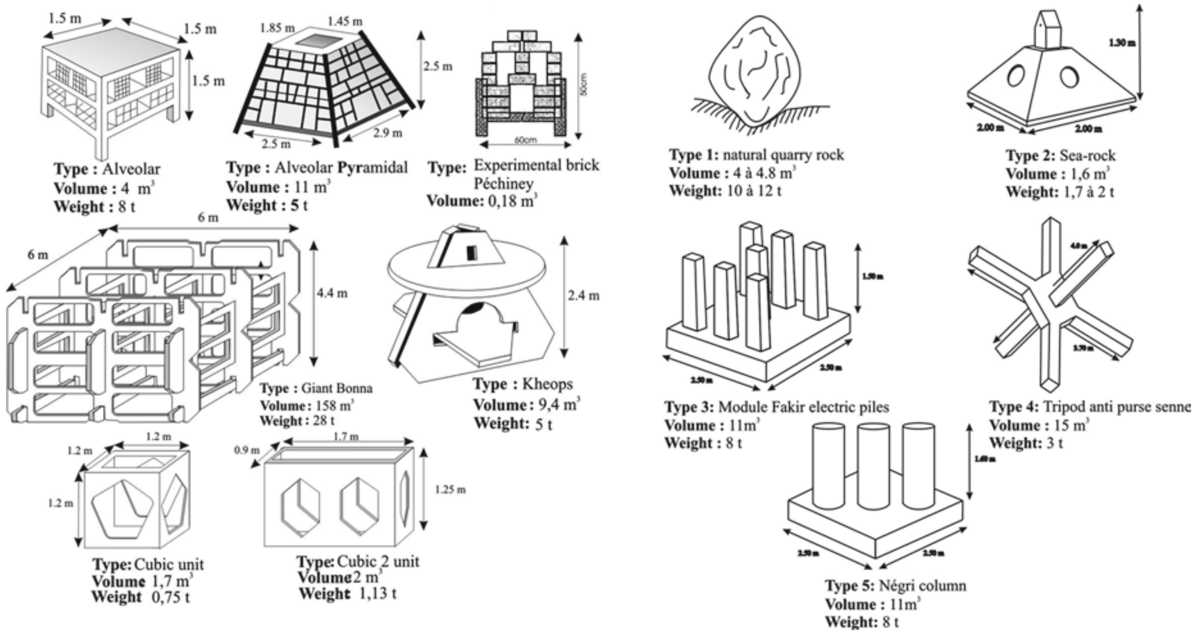

Fig. 2 Presentation of the two main categories of artificial reefs used in the Parc Marin de la Côte Bleue: on the left, seven models of production reefs (the small cubic modules are deployed into chaotic heaps) and on the right, five kinds of antitrawling reefs deployed one by one

Table 1 Description of artificial reef deployments on the five areas of the Côte Bleue Marine Park

Areas/volumes (m³ of A. Reef)	Year of deployment	Description of artificial reefs
Niolon-Le Rove, 319 m³	1985, 1989	– 319 m³ of production reefs, with three types (83 small cubic modules of 1.7 m³ + ten cubic modules of 2 m³ + one large unit of 158 m³)
Ensuès-la-Redonne, 677 m³	1985, 1989	– 546 m³ of production reefs (112 small cubic modules of 1.7 m³ + 20 cubic modules of 2 m³ + two large units of 158 m³)
	1990	– 131 m³ of protection reefs (17 quarry rocks of 11 t (4.4 m³) + 36 pyramidal “sea-rocks” of 1.56 m³, weight 2 t)
Carry-le-Rouet(no take reserve of 85 ha), 1,093 m³	1983	– 225 m³ of production reefs (36 alveolar modules, among 27 table unit of 4 m³ and nine pyramidal unit of 13 m³)
	1986,1990	– 190 m³ of protection reefs (36 quarry rocks of 11 t (4.4 m³) + 20 pyramidal “sea-rocks” 1.56 m³ of 2 t)
	2000	– 678 m³ of protection reefs (40 heavy column modules of 12.5 m³ weight 8 t + 12 tripods antipurse senne 14.8 m³ of 3 t)
Sausset-les-Pins, 1,442 m³	1985, 1989	– 1,241 m³ of production reefs (195 small cubic modules of 1.7 m³ + 60 cubic modules of 2 m³ + five large units of 158 m³)
	1986,1990	– 201 m³ of protection reefs (30 quarry rocks of 11 t (4.4 m³) + 44 pyramidal “sea-rocks” 1.56 m³ of 2 t)
Martigues-Couronne (no-take reserve of 210 ha), 1,205 m³	1996–1997	– 1,001 m³ of protection reefs (91 “Fakir” electric piles of 11 m³)
	1997	– 148 m³ of production reefs (87 small cubic modules of 1.7 m³)
	2004	– 56 m³ of experimental reefs (six modules “Khéops” of 9.4 m³)

with 2,684 m³ of modules arranged in chaotic heaps on five sandy areas, destinated to create new habitats similar to natural rocky reefs with high structural complexity for fish fauna and intended to increase the biodiversity and the available fishing resources.

(2) The protection of priority natural habitats (*Posidonia oceanica* meadows and coralgal banks called "coralligenous"), with 326 heavy obstacles designed to protect against illegal trawling within the 5.5 km offshore limit. These 2,200 m^3 of anti-trawling reefs are spread along 17.5 km of barriers stretching off the coast, arranged in perpendicular lines, and insure the protection of bottom nets and long lines from being damaged by trawlers and consequently reduce conflicts among fishermen and allow them to share the fishing grounds and resources.

There is no particular regulation on the majority of reefs. Most ARs are open to fishermen (both professional and recreational) and divers, except those located inside the two no-takes reserves, where fishing is prohibited. It represents 16% of the volume of production reefs and 26% of the number of protection reefs.

Several kinds of modules were tested (Fig. 2), both for production and protection effects. Most of the production reefs are composed of small concrete cubic modules of 1–2 m^3, gathered into chaotic piles of 60–100 m^3, and also bigger units (158 m^3) inspired by Japanese technology, but having a very low habitat complexity.

Concerning antitrawling reefs, five kinds of modules were used in the PMCB (Fig. 2). First experiments were conducted in 1986 with heavy quarry rocks, the first trial in the Mediterranean Sea. To be efficient, these protection reefs have to weigh a minimum of 8 t. These reefs are deployed one by one, separated from each other by 50–200 m. They are being arranged by lines, creating barriers perpendicular to the coast (17.5 km of barriers within the 3 km area, Fig. 1).

4 Main Results of Scientific Monitoring

After 25 years of AR experimentation in the PMCB, there is a wide variety of kinds of reefs (production, protection), module architecture, depths, environmental conditions, management rules (inside/outside MPA), etc. Several scientific monitorings were organized, using underwater visual census techniques, adapted to the specificity of AR (Charbonnel et al. 1997).

4.1 Protection Reefs

The most characteristic example of a successful protection reef is shown in Fig. 3, with the path of illegal trawling plotted before and after reef deployment, and the creation of the no-take reserve in Cap-Couronne (1997). The result has been a decrease of fishing pressure on the coastal band by removal of illegal

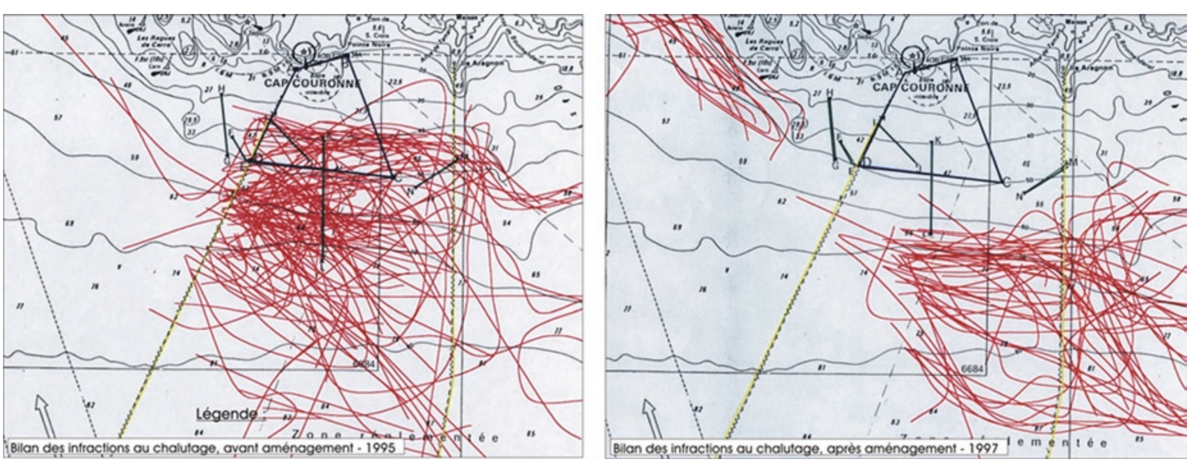

Fig. 3 Results of protection reef efficiency; on the left, before and, on the right, after reef deployment in the Parc Marin de la Côte Bleue, around the vicinity of Cap-Couronne integral reserve (red lines correspond to illegal trawling paths within the 5.5 km area)

Table 2 Evolution of the fish assemblages on two reef types (large unit of 158 m³ and small cubic units of 1.7 m³ deployed into chaotic heaps of 119 m³) for the area of Sausset-les-Pins between 1987 and 2000 (Charbonnel et al. 2000)

Reef type (volume: unit/reef)	Large unit (158/158 m³)			Cubic unit in chaotic heaps (1.7/119 m³)		
Year of survey	**1987**	**1993**	**2000**	**1987**	**1993**	**2000**
Total number of species	24	24	31	28	35	41
Mean number of species	7.3	8.0	11.5	10.8	13.6	16.8
Density without pk (individual/m³)	0.12	0.14	0.32	0.93	1.52	1.04
Total density (individual/m³)	–	11.8	7.2	–	27.7	76.4
Biomass without pk (g/m³)	5	21.5	12.4	116	306	155
Total biomass (g/m³)	–	269	108	–	2,918	2,396

Pk = planktivorous species (*Chromis, Boops, Spicara*) and *Coris julis*; – = data unavailable

trawling activities offshore. The efficiency of anti-trawling reefs allowed a better sharing of space and resources among fishermen by supporting traditional small- scale fisheries (bottom gillnet and trammel net, hook on line) with much more selective techniques (catching only adults and thus facilitating conservation of fishing resources), whereas trawlers caught all-sized species, particularly juveniles, who had not yet reproduced. This is a crucial point in the stock dynamics of necto-benthic species: mortality because of catching juveniles is the principal factor of the falling of catches.

The other essential effect of protection reefs is to preserve the most productive and fragile natural habitats (*Posidonia* meadows and coralligenous banks) from mechanical destruction by trawlers. These damages have important ecological and economic repercussions, because these habitats serve for spawning, nurseries, recruitment, and feeding areas for most part of the exploited resources.

4.2 Production Reefs

Concerning production reefs, monitoring showed that the fish assemblages associated with AR are similar in species composition, density, and biomass to those occurring on natural reefs made of rocky bottoms, and most often with superior performance because of the multimodal aspect and higher heterogeneity (Charbonnel et al. 2000, 2002). Availability of shelters may be more important, because food resources are generally not limited, because most species feed on the surrounding natural habitat. Colonization of AR can proceed quickly (2–3 years), but maturation of reef assemblages can take many years, and some reefs still show evolution after 10–15 years of deployment (Table 2, Fig. 4). Duration of assemblage maturation depends on the size and complexity of the AR.

For example, on a small AR of 150 m³ in Cap-Couronne (Fig. 4), the specific richness of fish assemblages increased regularly between 1995 and 2004, from 7 to 29 species (TNS, total richness) and from 2.6 ± 0.9 to 15.7 ± 2.1 species per census (MNS). Biomass was multiplied by a factor of 46: from 2.2 kg in 1995 (before reef deployment) to 100.7 kg in 2004.

The monitoring on Sausset AR between 1987 and 2000 showed that colonization can proceed over a longer period of time before reaching the maximum carrying capacity. The specific richness still increased on both reef types (total richness × 1.3 to 1.5, mean richness × 1.6) on these ARs aged 11–14 years in 2000 (Table 2). Fish assemblages are increasingly complex, and this

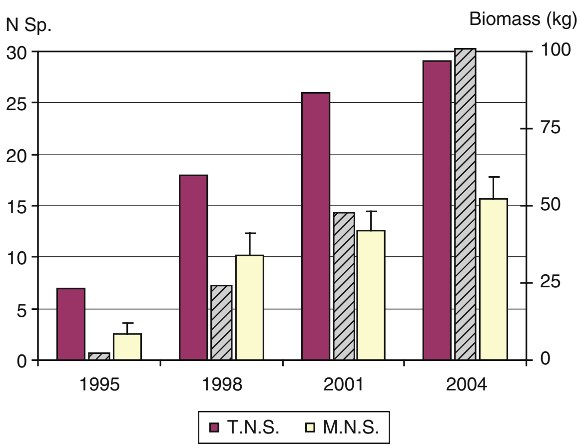

Fig. 4 Evolution between 1995 (before reef deployment) and 2004 of the specific richness of fish assemblages [total number of species (TNS), mean number of species (MNS) per census and standard deviation],and biomass (hatched) on a 150 m³ artificial reef located in the integral reserve of Cap-Couronne (Jouvenel et al. 2006)

process of slow colonization continues, with turnover and the appearance of new species, as well as a higher proportion of permanent species (based on their frequency of occurrence), suggesting a higher temporal stability of the species assemblage; especially chaotic heaps of small cubes offer numerous small interconnected chambers.

After 13 years, by comparing 2000 to 1987, the density and the biomass have also increased over time, according to the available resources. The best results were obtained in the 1993 survey, particularly connected to the abundance of Sparids. By comparison, the monitoring in 2000 was conducted with low temperature, inducing rarefaction of Sparids, and a decline in density and biomass when removing planktivorous species and wrasse *Coris julis*.

The structure of fish assemblages clearly differed according to the reef type: cubic units offer more complex habitats, with greater diversity in size of shelters. This model of AR presents a greater species richness (×1.5) and greatly increased density (×10) and biomass (×22) compared to a large unit reef (see Table 2) with vast undivided empty chambers, which are ineffective, because they have no natural equivalent.

This example and other French field work showed that reef design is a crucial factor in AR effectiveness. AR architecture and layout should be adapted according to the behavior and the habitat requirements of targeted fish species.

Research and experiments are necessary to improve reef efficiency, in relationship with studies of natural habitat effects. A reef aimed at maximizing its associated biodiversity and fish fauna should be heterogeneous and should have a high structural complexity.

The use of a mixture of different kinds of materials of different sizes and void spaces deployed in irregular piles facilitates the creation of a network of complex cavities of small interconnected chambers, which appear to provide benefits for many fish species – both predators and preys – that may find features in the multimodal pattern that suit their specific requirements (Charbonnel et al. 2002).

The reef architecture and module design determine not only the global performance of the reef (species richness, abundance, and biomass), but also the identity of species that are particularly fitted for exploiting this new resource.

It is important to notice that ARs worked as MPAs and have similar "reserve effects" with:

(1) Increasing biodiversity and global species richness, because of higher frequency of rare target species
(2) Increasing abundance of individuals, particularly those of species targeted by fishing
(3) Recovery of balanced demographic structure, with a higher frequency of large individuals, which are potential spawners

All these phenomena are similar to those observed in marine reserves (Francour 1994; Harmelin et al. 1995; Harmelin 1999; Charbonnel et al. 2000, 2001), and the same supposed beneficial effects on fish numbers seen in the peripheral area can be attributed to AR. The increase in the mean individual weight suggests that reef maturation induces an increase in the stock of large adults (Charbonnel et al. 2000).

There is also an evident ecological benefit, because spawning productivity of larger individuals is much higher than that of small ones.

Finally, similar to reserves, ARs also have a refuge function by virtue of their physical presence, without imposing the need for any particular protective management, such as fishing restrictions.

5 Conclusions

In the Côte Bleue Marine Park, the two categories of AR combining production and protection effects are not dissociable from of the two MPAs (no-take reserves). These are complementary tools, used in an additional way. All these management tools have contributed to the preservation of the traditional small-scale fisheries on coastal bands of the PMCB, using selective techniques. They have allowed maintaining the same number of artisanal fishermen for 25 years (about 60) at the same time that these fishing activities are decreasing in nearby zones, in a general context of full exploitation of resources.

AR may constitute a response to numerous problems concerning coastal resources, overexploitation of fisheries and ecosystem degradation. AR and MPA (particularly no-take zones) represent efficient tools for marine resources management and can maintain a sustainable development of fisheries and fishermen. In a general context of high fishing pressure, AR and MPA are good tools for the organization and management of activities on the coastal band. Because of the lack of respect of regulations and lack of repression, the PMCB implemented antitrawling reefs, which have been very efficient and allowed a better sharing of space and resources because of the decreasing fishing pressure on the coastal band with the removal of trawling activities offshore. These protection reefs are good tools for the preservation of priority habitats as defined by Natura 2000.

However, ARs are not a miracle tool. They represent only one facet of the overall management, which must take into account all phases of the life history of the over-fished species and more especially their spawning areas and nurseries. In the actual context of rule changes in Europe, with new management of fisheries (Common Fisheries Policy, CFP) and the implementation of the ecological network Natura 2000, ARs are well-adapted tools for integrated management of coastal marine resources and sustainable fisheries.

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Artificial Reefs in Marseille: From Complex Natural Habitats to Concepts of Efficient Artificial Reef Design

E. Charbonnel, François Carnus, Sandrine Ruitton, Laurence Le Direac'h, J.-G. Harmelin, and J. Beurois

Abstract With the deployment of 27,300 m^3 of artificial reefs (AR) and a €6 million budget, the project "Récifs PRADO" is the largest artificial reef ever realized in the Mediterranean Sea. This large-scale program is dedicated to the restoration of an active artisanal fishery in a zone where *Posidonia* beds, a highly productive habitat, have previously been destroyed.

Marine biologists conceived the design of these ARs, benefiting from the feedback of field experience of the colonization of ARs immersed in the French littoral for more than 20 years. The core knowledge was based on years of underwater visual observations of fish behavior and visual censuses of fish assemblages on both artificial and natural habitats. The selected concept was to copy the most performed natural benthic habitats in order to optimize the biological efficiency of AR. Combining architectural complexity, module design and urbanism (laying out of modules on the bottom) was considered to be the key to AR effectiveness.

The reef deployment relied on the creation of horizontal and vertical discontinuities in heights, sizes, and volumes, thanks to a great variety of reef types and shapes, as well as diverse arrangements and horizontal spacing of reefs.

Three years were needed for conceiving the different reef types, the achievement of the technical and administrative folders, and overcoming the numerous technical and administrative problems.

Six types of modules of different shapes, sizes, volumes, and materials were specially designed for this project. To optimize the reef habitat diversity, these modules were complexified by the addition of several types of small filling materials (bags containing oysters shells, breeze blocks, and octopus pots used for fishery) and floating immersed ropes.

Piles of quarry blocks of variable sizes were also used, to reconstitute natural rocky boulders, which represent the most suitable habitat for many target species like groupers and sea breams.

One of the purposes of this project was to arrange the different modules by grouping them in "hamlets" and "villages" (diffused urbanization concept). These six "villages" of triangular shape were linked together by series of reefs ("functional connections"), functioning as biological corridors and stepping stones for fish and propagules.

The locations of peripheral natural habitats (*Posidonia* meadows and rocks) were taken into account in the arrangement of the villages for optimizing a rapid colonization of the AR. Reef structures were deployed between October 2007 and July 2008 in two areas of 110 ha: one closed to every kind of fishing and the other restricted to artisanal fishing.

In total, numerous compromises and adjustments were applied to the pristine ideal ecological reef project to cope with legal, economic, environmental, and social constraints.

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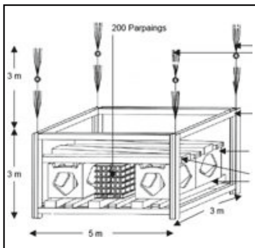
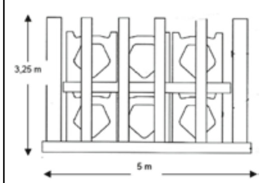
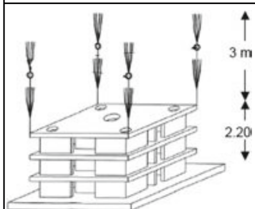
Moreover, this operation is a real success in terms of cooperation and effort among numerous maritime actors, such as marine biologists, planners, state and engineering offices, professional fishermen, concrete manufacturers, public workers, local and regional authorities, and all stakeholders concerned in the coastal management, to realize the best reef structures adapted to the local context and to reach several objectives.



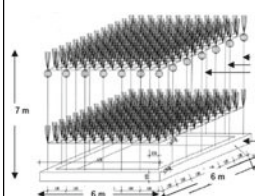
Reef type "metal basket". This huge reef (187 m^3) is constituted by a metal frame (3 baskets measuring 5 x 5 x 3 m) filled by several, differently-sized materials (4 concrete cubes of 1.7 m^3, 10 concrete piles, 200 breeze blocks per basket).

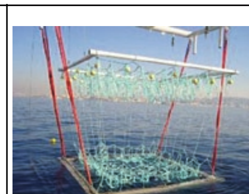


Reef type "fakir basket"
This reef (82 m^3, 5 x 5 x 3 m) is constituted by 26 peripheral concrete piles, filled by several materials of different sizes (16 concrete cubes of 1.7 m^3, 200 breeze blocks).

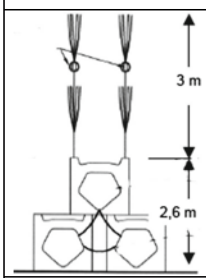


Reef type "chicane"
This reef (19 m3, 4 x 2.4 x 2 m) is constituted by a concrete frame with 2 floors of mazelike galleries, connected with five holes. This reef was specially designed for sea breams ("rague à sars").



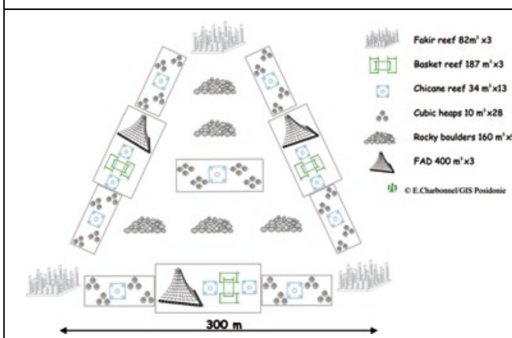


Reef type "floating ropes"
This reef (6 x 6 x 7 m) is constituted by a concrete frame with two floors of interconnected floating ropes. This reef was designed for attracting the small planktivorous fishes and their predators.

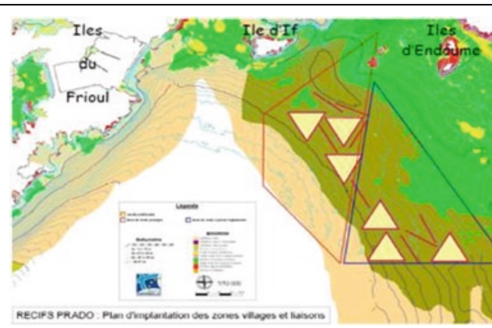


Reef type "cubes pile"
This reef (10 m^3, 2 m high) is an assemblage of six concrete cubes of 1.7 m^3, with small void space between the cubes.


Reef type "quarry rocks"
This reef (300 tons, 160 m^3, 20 x 4 x 2 m) is constituted by piles of quarry blocks of three different sizes. This type of habitat is most suitable for many target species like groupers and sea breams.



Representation of a "village" (diffused urbanisation concept) and space arrangement among the six types of modules.



Location of A.R. in the Prado bay of Marseille, arranged in six "villages" of triangular shape, connected by functional connection, working as biological corridors.

The Dubai Underwater Observory Projects and Turtle Rehabilitation Unit

Etienne Clamagirand

Abstract The new, important, and rough onshore urbanization of the Arabian Gulf close to the Dubai Emirate has the consequences of perturbations and sometimes the disappearance of the natural environment of its coasts.

The Dubai authorities launched original projects to facilitate the observation of underwater life by their own population and by tourists along the littoral. At the same time, they decided to take great care of some of the endangered marine species.

Architeuthis, a private architecture bureau, planned and studied original systems to enhance the observations of the sea bed around Dubai. It also created and constructed two types of new underwater habitats to protect and increase the number of marine species, especially the marine turtles landing and living on the seashore.

Marine turtles used these habitats. The design of these original structures is useful and efficient.

1 Introduction

In almost all countries, to protect their own environment, people need to learn about the composition and characteristics of the marine fauna and flora living under the surface of their coastal waters.

In several countries, the biology of marine turtles has been studied in detail as, for instance, in the Seychelles Islands (Hitchins et al. 2004), Malaysia (Pilcher and Lamri 1999), and Saô Tomé (Graff 1996). Recently, Luschi et al. (2007) have shown that marine turtles use geomagnetic fields to orient their trajectory when homing to their spawning areas and coming back to their beaches.

Despite a nice synthesis by Witzell (1983) concerning the biological data on the hawksbill turtle *Eretmochelys imbricata* (Linnaeus 1766), the biological cycle of marine turtles is not known in detail, particularly when they are living in the open sea, far from the coast.

When they are living near the sea shore, they are often eating different species of sponges, even some toxic ones (Meylan 1998). It has been established recently that marine turtles return to spawn in the same place, on the same sandy beach where they have already spawned.

In the Mideast, the general problem of protecting marine life has attracted the attention of the authorities in many countries. This is the case in Abu-Dhabi Emirate, where an environmental strategy has been established by the agency responsible for the environment and protection of flora and fauna, applied some strict rules of protection, and surveyed the living marine resources. It is also, for example, the case in Lebanon, where similar rules and scientific studies are in progress.

It is also the case in Dubai, where the Office of Protection of wild flora and fauna has been authorized to study marine turtle populations (*soulahfat bahriat* in local language) using tagging and satellite surveys.

These chelonians are endangered; several solutions have been introduced, such as protection of sea shores where they release their eggs, protection of the very young turtles after they hatch when they are moving to the sea, construction of artificial beaches to induce the females to spawn, specific festivals when the hatching of eggs takes place, etc.

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The new and rough onshore urbanization of the Arabian Gulf close to the Emirate of Dubai has caused the disappearance of the natural ecological characteristics of the coasts and nesting areas of marine turtles. A few of them have washed ashore, sick and scared, and have been brought by concerned people, fishermen, and divers to Dr. Kevin Hyland at the Wild Life Protection Office of Dubai to be treated and cured before they are released back to the sea.

Also new habitats are being created to facilitate the arrival and the survival of particular species, in this case, the turtles. This general policy is quite helpful for marine species and also increases the reputation of Dubai among countries protecting the natural environment.

Some marine turtles living close to the sea shore or even living in enclosures sometimes suffer from aerophagia because of the food they eat close to the coasts. This common disease of the chelonian group makes them float continuously. As a consequence, during the day, the hot sun may burn the top of the carapace. The turtles need to dive. These turtles have found an appropriate shelter under an artificial structure providing shade during the warmest hours of the day.

2 Achievements

2.1 First Attempts

(1) Computer-generated image of the turtle shelter
(2) Built turtle shelter
(3) The first turtle in the shelter

After veterinary treatment in a special tank, for the final stage of rehabilitation, the turtles are transferred to a special enclosure located in an important hotel, the Mina Al Salam Hotel (five stars), in the town of Jumeirah (Jumeirah Madinat). The manager of the National Marine Aquarium, Mr. Warren Baverstok, at the Burj al Arab, asked Architeuthis to design a specific artificial reef, which is in fact an underwater habitat for the turtles.

Architeuthis studied, designed, and installed the artificial reef for turtle rehabilitation based on a "Fractal module," and this new habitat was fully accepted by the turtles. The Architeuthis turtle rehabilitation unit is a true environmental success story.

2.2 Positioning

The immersion of artificial reefs is a heavy and expensive operation. The Fractal is an artificial reef concept designed by Architeuthis with the purpose of reducing fabrication and immersion costs.

Because of its specific geometry, like the Hexapora modules, the Fractal is also a landscaped reef that provides the following advantages:

- Fabrication adapted to the volume of the studied reef: easy molding realized industrially or manual molding realized with a cheap and easy-to-use matrix.

The compound is of GRC (glass reinforced concrete), a neutral composition for marine biology.

- Lighter modules, leading to an easier manipulation in different kinds of weather on land or under water.
- Various possibilities of three-dimensional fittings making possible a complete adaptability to the immersion site.
- Simplified laying procedures leading to light use of local maritime resources.
- A huge global volume considering the volume of an element.
- A surface to be colonized, reaching approximately 3.5 times the ground surface.

The Fractal reefs are designed to suit biological, architectural, and landscape purposes.

With naval resources being one of the most important financial parts of the immersion of artificial reefs, Architeuthis has developed specific concepts for its immersion. They guarantee lower costs and a good ratio of immersion timing/number of modules laid.

An analogy with molecular chemistry leads us to better understand this type of reef and lets us imagine the architectural diversity of the outcome.

3 Technical Description

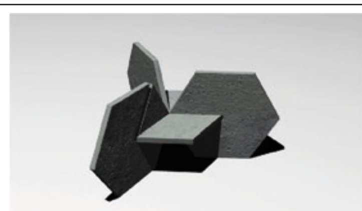
This innovative concept of these artificial reefs consists of jointing hexagonal slabs slit along half of a diagonal. The technical innovation consists of a pressure-sealed system, canceling every mechanical actuator. An anchoring system will guarantee the reef's stability against the swell and possible heavy currents. The jointing possibilities lead to the creation of fractal geometry.

Considering this new concept design, it is obvious that the possibilities in biological, landscape, and architectural areas are fully sufficient to lead to a final geometry that will match both the seabed geometry and the requirements for scuba diving activities.

4 Positioning Procedures

As described, the Fractal module is designed to reduce the immersion costs. To minimize the diving resources during the laying, Architeuthis has developed an immersion process that regroups 100 modules. The design is a crown with a star in the middle that covers a surface of 100 m^2. In one working day, it is possible to lay 600 m^3 of reef.

In the permanent search for eco-conceptions that respect the environmental, Architeuthis has developed this concept with the possibility to bring every group of modules back to the surface, preventing any degradation of the seabed (www.seaturtle.org).

SUBSEA PROJECTS & PRODUCTS	FRACTAL ®	HEXAPORA ®
Underwater observatory Subsea leisure park Touristic submarines	Handy & high-performance Artificial reef	Landscape artificial reef
 *Monaco underwater observatory project*	 *Four piece assembly*	 *Type of Hexapora module assembly*

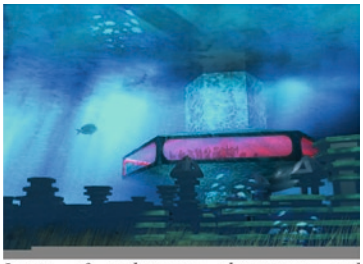


Image of underwater observatory and landscaped reef

Reef immersion in Dubai

Hexapora assembly in Monaco

Landscaped reef project

Star type Fractal design

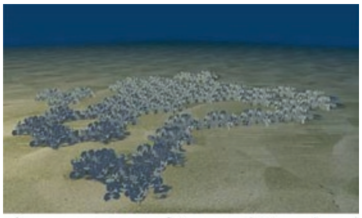
Hexapora in Monaco water (Larvotto protected area)

Underwater marine park

« Pentafrac » in Cannes Mediterranean sea

Computer generated image of Hexapora reef garden

Six places tourist submarine SMAL 5



Computer generated image of Fractal reef field

One year later in Monaco water

Two places tourist submarine SMAL 2

5 Discussion

Concerning the underwater observations, we have designed new types of underwater observatories permitting the inhabitants of Dubai and the tourists to have a direct look at the fauna and the flora of the coasts.

Today, there are several observation towers undersea, like those constructed in Japan in Okinawa, Kushimoto, Genkaï, Ashizumi, and Kalzima. Most of them have been built with common construction planning and materials. Often, they destroy the subsea environment where they have been constructed. The Diatomées project was built in a shipyard and towed to location and ballasted to minimize the installation troubles substantially. Our projects try to avoid these defects.

Concerning the artificial habitats and the turtle rehabilitation unit, we have created new habitats presenting a better shelter than those found by the animals in nature. The door is open to design other new ones that facilitate the presence of benthic food such as sponges, for instance, survival, biological cycles, and new shelters, not only for the turtles but also for other zoological groups.

6 Conclusions

We have designed and constructed new types of observatory systems in artificial habitats specialized for the turtles (*Eretmochelys imbricata*) living on the Dubai coasts.

The models we have designed are well accepted by the turtles: it represents one more step in the coastal management of the sea shore.

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Immersion of Artificial Reef in Ohya Island: Lessons From New Experiences

Eric Delort and Didier Grosdemange

Abstract An immersion of artificial reefs was carried out in the coastal area of Île d'Yeu (in English: Ohya Island).

The immersion chart voluntarily used various modules in order to use several experimental fixtures and different depths.

Large and huge single modules, chaotic arrangements, and organized arrangements of small modules at two different depths were chosen by the company to try several practical solutions.

Different surveys were made using a side-scan sonar, monobeam echosounder, underwater photo and video, and biofouling surveys.

1 Preliminary notice

This chapter relates the results of experiments developed by the authors in the framework of the company "In Vivo" during 4 years. The company had to survey the biofouling arrangement and the structural evolution. Several pieces of equipment were used: side-scan sonar, monobeam echosounder, experimental tiles, underwater photo, and video carried by divers.

2 Introduction

On French Atlantic coast, there has recently been an increase in interest in artificial reefs and marine habitats (Ceccaldi 2000; Ceccaldi and Nakagawa 2003).

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This immersion of artificial reefs was carried out at Île d'Yeu (Ohya Island). The immersion chart used different types of experimental modules showing several fixtures and different depths. The French government, European Community, and the Region of Normandy were the main financial supporters of these immersions. Fishermen held control of the entire project from the start in 2000. The zones were chosen in order not to disturb the fishing operations. The first aim of this project was to protect the coastal fishing areas from trawlers and their devastating consequences.

Then, the fishermen oriented their program toward better use of the sea shore in order to increase the production of marine species, and particularly the commercial species. A preliminary and comprehensive survey became necessary, and a specific study was carried out.

3 Experiments

3.1 General Framework

During the first step of the following study, several measures concerning the characteristics of the immersion site provided precise data concerning the quality of sand, the bathymetry, and several oceanographic characteristics.

Then, surveys by underwater divers were developed for 3 years by observation of experimental tiles; a side-scan sonar recorded and an underwater settled camera followed the fouling settlements, the fish assessment, and the mechanical structures of the artificial reefs.

The side-scan sonar showed the exact position of the reefs after their immersion as well as the way they gradually buried themselves in the sediment.

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The whole plan of immersion was based on an experimental method to choose the best depth and the best arrangement of different types of modules in order to obtain the best zone and the best plan for better organic development on the basis of the food chain existing under the surface.

The significant height of the tide in Île d'Yeu did not allow immerging the modules at a depth of less than 20 m, and the second zone at a depth of 45 m was chosen as a place not too directly influenced by tide and not too deep for the observations by the divers.

3.2 Steps of Experiments

The experimental tiles followed the settlement of larvae of different marine species on the artificial reefs. Several tiles were immersed with the reefs at different positions: on the top and on the lateral faces. Every year, three tiles (top, front of the stream, and on the side of the stream) were removed from the different immersed structures. The settlement of the fouling, the time spent for their fixation, and the coverage observed on the different sites and at the different depths were determined.

The underwater dives surveyed the structure of the modules, the way they were immersed, and the colonization by fishes and lobsters. All these surveys allowed us to appreciate:

- The settlement process: it was fast and complete within 3 years with some differences between the two depths.
- The chaotic arrangements allowed the best colonization for vagile species compared to the organized arrangements. This information was linked with the immersion system, which is cheaper and easier for chaotic arrangement.
- Huge modules were usually fragile and can be buried naturally faster.

These experiments and the different surveys were used for a new immersion of artificial reefs off the coast of Etretat in Normandy: the chart, the modules structures, the modules surfaces, the technical characteristics, the immersion system, and the reef arrangement were optimized.

The immersion was carried out in 2002 at Île d'Yeu in two zones close to the island. One zone is at 45 m depth and the other at 20 m depth.

At each zone, different modules were used: small modules and large modules. The small modules were installed with two different arrangements: chaotic and organized arrangements. A complex of ten small modules were used to form every chaotic or organized arrangement.

In addition, a ring all around the field of the experiment was established with numerous units of small modules to create a curve of the protected system. These elements have been immersed to prevent the harmful effects of fish trawlers. All such activities are forbidden inside the concession of the artificial reef (diving, fishing, etc.). Only scientific activities are allowed: underwater counting, diving, and sampling.

Figures 1 and 2 show the different arrangements: chaotic and organized arrangements. Figure 3 shows the large module used at – 20 m and – 45 m depths.

Only scientific activities are allowed: underwater counting, diving, and sampling.

The Figures. 1 and 2 and 2 show the different arrangements: chaotic and organized arrangements. The Figure 3 shows the large module used at – 20 m and – 45 m depths.

Figure 4 shows all the different modules before their immersion on the same site.

3.3 Following Observations

Once the reefs had been immersed, a complete monitoring operation was established for 5 years. The main characteristics of the survey were:

- Benthic fouling species fixed on the different surfaces
- Light reaching the reefs
- Water quality (suspended matter, chlorophyll, temperature)
- Position of reefs at the beginning of the experiments and at different periods of time
- Side-scan sonar
- Detailed bathymetry
- Fish assessment around and inside the reefs
- Mechanical structure evolution
- Buried structures
- Photo and video taken
- Settled video

One of the main factors to monitor was the fouling (the survey of fish assemblages was given to another company). If the principal biological function of

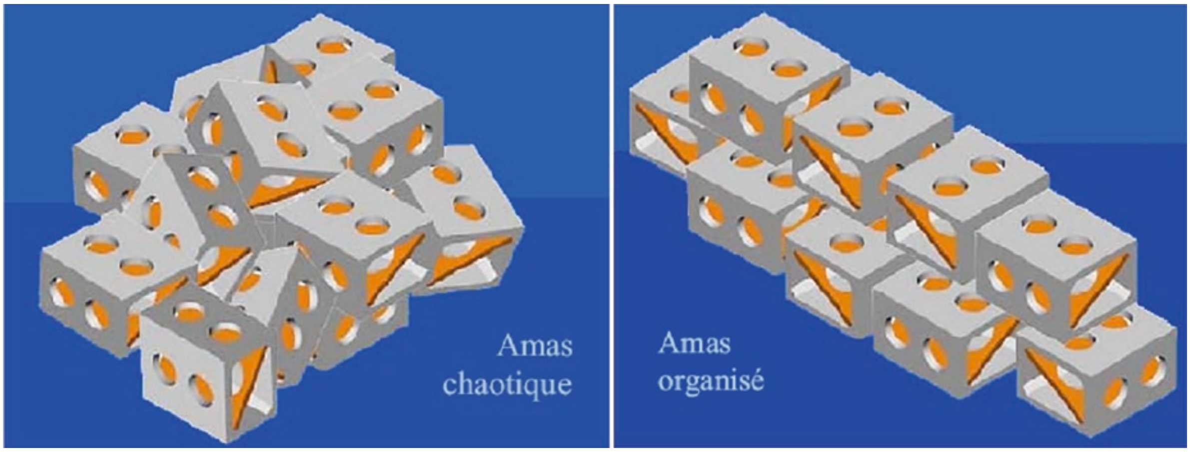

Fig. 1 Organized and chaotic assemblages of artificial reefs in Ohya Island

Fig. 2 Organized assemblages of reefs before their immersion

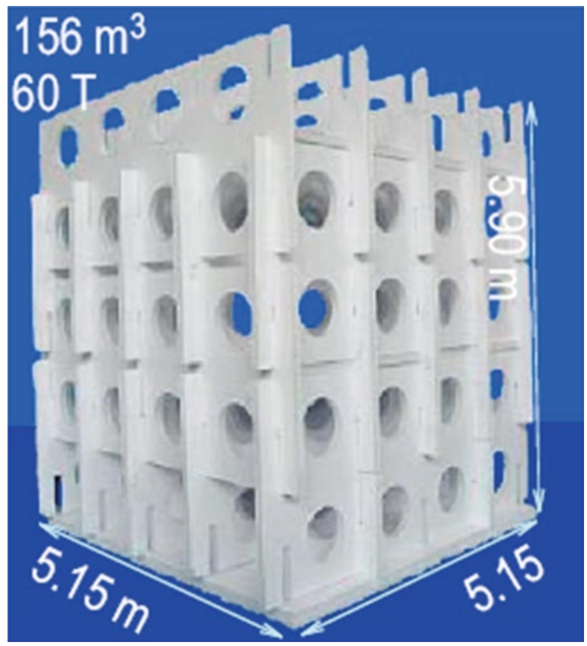

Fig. 3 Large module immersed in the experiments

artificial reefs (AR) is to increase production of interesting commercial species, it needs to have an important production of settled animal and vegetable organic matter as food for high stages of the food chain (Delort et al. 1995, 2000; Delort 1999).

The fouling monitoring was carried out using experimental tiles. These tiles (see Figs. 5 and 6) were installed on the reef at three different positions: on the top, in front of the main current, and in the direction of the main current.

All the tiles were settled on the reef before immersion, so the company only had to collect three tiles a year.

The main results of the fouling settlement are given in Figs. 7 and 8.

These results allowed us to develop a settlement study of species on artificial surfaces. The first step was to examine the recruitment coming from benthic larvae in the water column. Then a predominance step was carried out, leading to the main pioneer species. During the next step, the substratum was modified; this stage is defined as a predestination step. After this phase, a maturation step was developed, leading to the presence of all species on the reef, but with a small size and number. Then, the final step was when the reef became well balanced, and the ecosystem became mature and dynamic.

Another monitoring phase was carried out using side-scan sonar survey. Figures 9 through 12 show

Fig. 4 Different modules before immersion

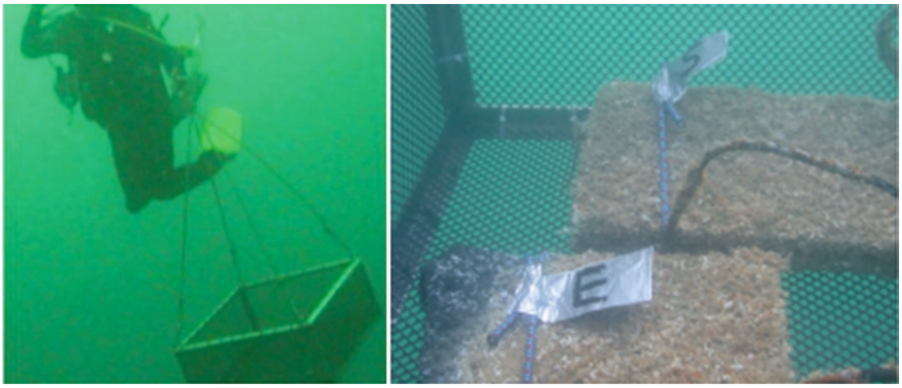

Fig. 5 Diver observing immersed structures, and experimental tiles

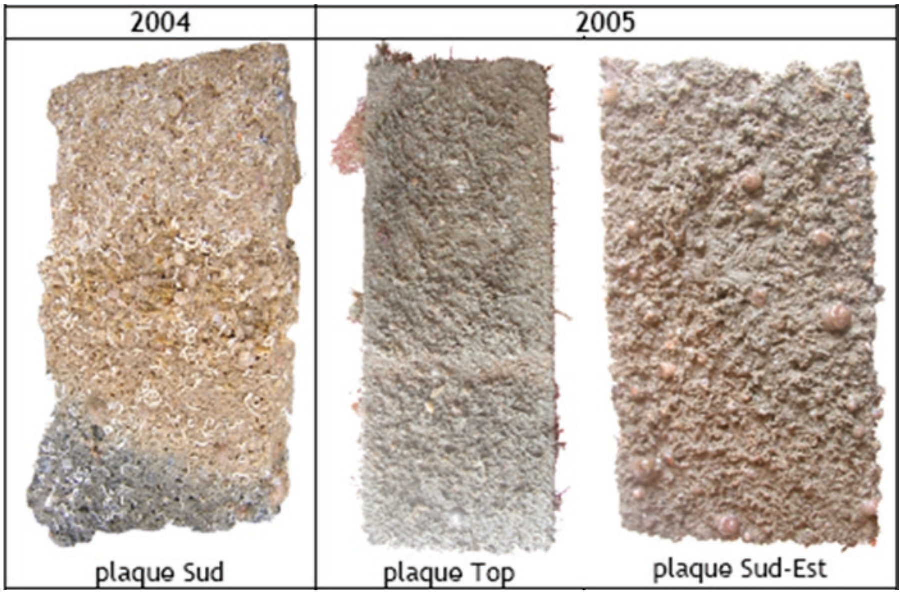

Fig. 6 Experimental tiles in 2004 and 2006

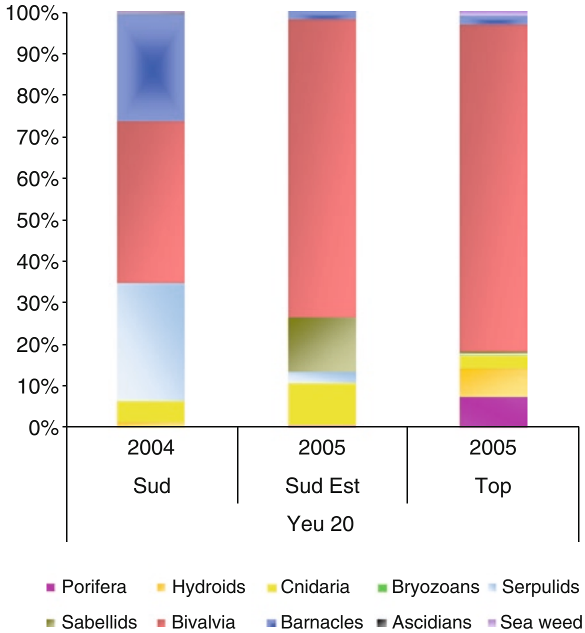

Fig. 7 For three tiles at 20 m depth, relative abundance of settled taxa (number of individuals/m²)

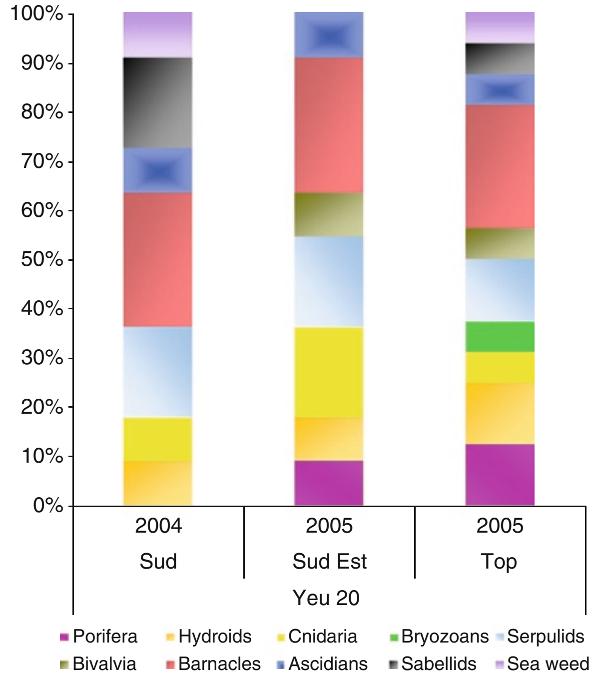

Fig. 9 For three tiles at 20 m depth, relative specific richness of settled taxa (number of species)

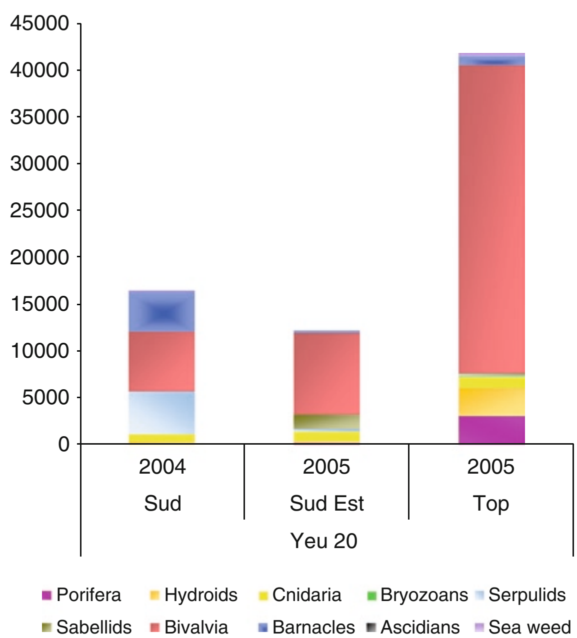

Fig. 8 For three tiles at 20 m depth: total abundance of settled taxon (number of individuals/m²)

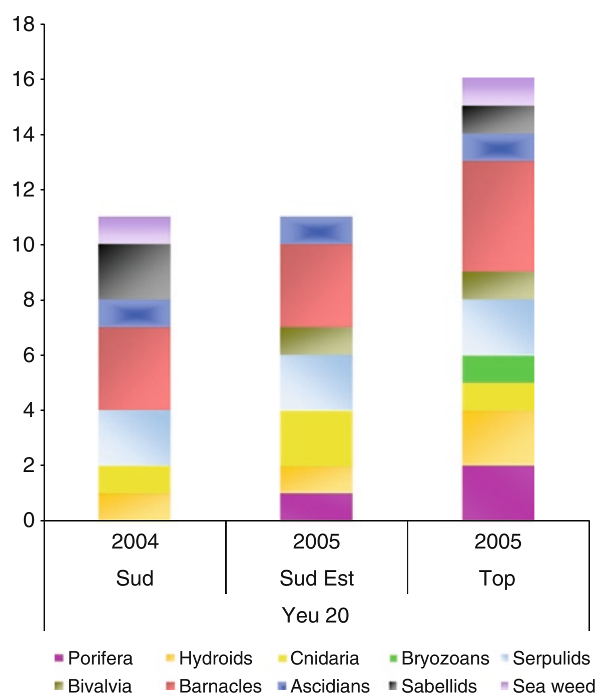

Fig. 10 For three tiles at 20 m depth, specific richness of settled taxa (number of species)

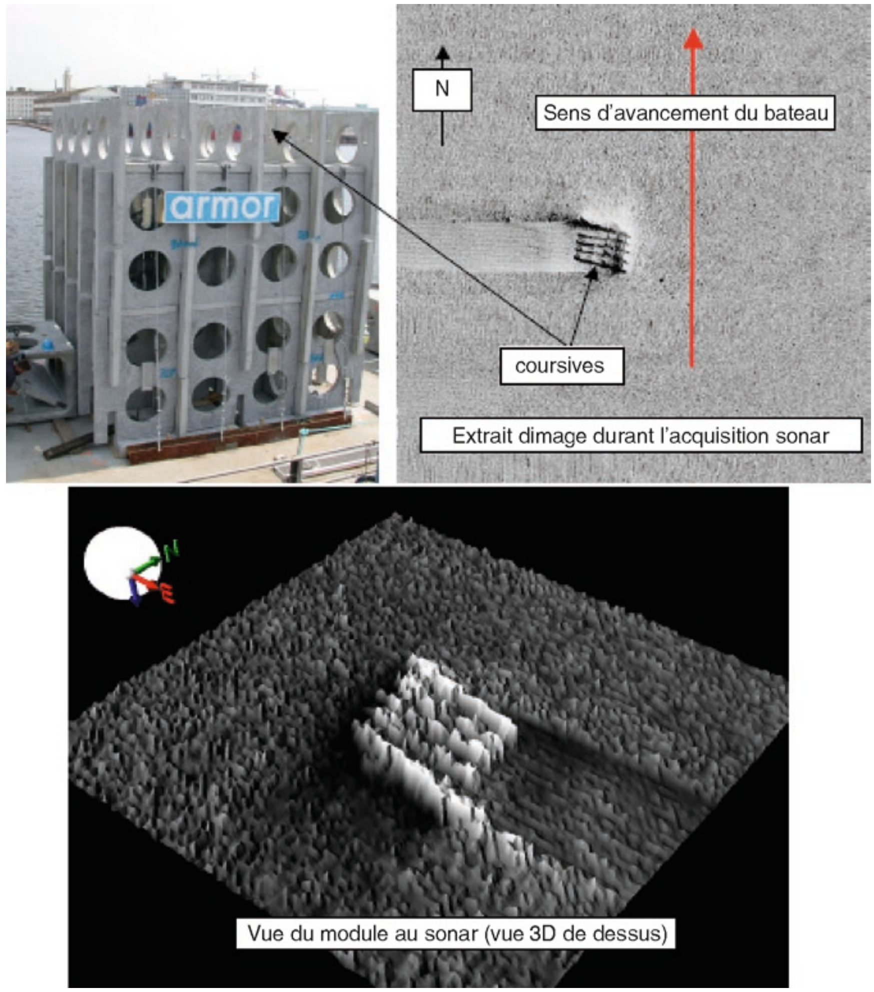

Fig. 11 Side-scan sonar image from the large module. 2D and 3D images. The next pictures were obtained at the same reef, but 1 year after immersion. They show very clearly the burring problems at 20 m depth and not at 45 m depth

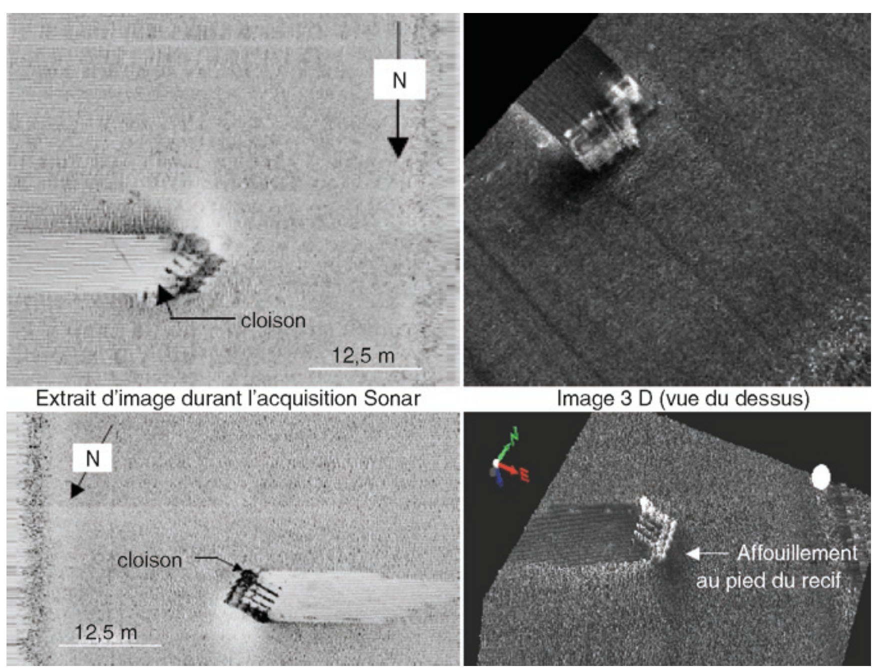

Fig. 12 Images of large reef at 20m depth extracted from side-scan sonar survey

data from 2004 on the large reef at 20 m depth. The 2D and 3D images allowed us to survey the exact position and the complete structure.

4 Conclusions

The settlement process of artificial reefs was fast and complete within 3 years with some differences between the two depths. A hundred percent of the surface of the reefs was covered before less than 2 years. The species settled were similar to the ones described in the literature.

Chaotic arrangements allowed the best colonization for vagile species. All reefs installed with an organized arrangement did not allow a large colonization.

The chaotic arrangement is easier and cheaper to immerse. Moreover, it settles more quickly on the sea floor.

Huge modules are fragile and can be buried naturally faster.

Other research and surveys are necessary to understand the role and the efficiency of the artificial reefs and artificial habitats as a new method to manage the seashore.

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Development of Small and Lightweight Artificial Reef for *Fukutokobushi (Haliotis diversicolor diversicolor)*

Keige Ebata, Akira Higashi, Akihiro Shiomitsu, Seiichi Saisho, and Toshimitsu Ikeda

Abstract Abalone is one of the important fishing resources in Japan. Fishing of abalone has been decreasing annually continuously. We have tried a new type of artificial reef and habitat, made in concrete, for abalones. Experiments and observations by divers have been done in Taneshima, a Japanese island on the coasts of Kyu-Shu.

1 Introduction

Abalone is one of the important fishing resources in Japan. Fishing of abalone has been decreasing annually. Tane Island (Taneshima), located in southern Japan, is famous for *Fukutokobushi* (*Haliotis diversicolor diversicolor*), a species of abalone. *Fukutokobushi* fishing is also decreasing; for example, the amount of that species was approximately 80 t in 1981, but only a few tons at present. One of the reasons explaining the decrease in abalone fishing is probably the decrease of habitat size because sand and stone have been covering the rock surfaces where abalones live.

2 Materials and Methods

This study aimed to restore the habitat of abalone by placing concrete blocks on the seabed. The concrete blocks, as shown in Fig. 1, are designed to be wavy and lightweight in order to enable the formation of rock shelters between the blocks and seabed; such a structure serves as a habitat for abalone and supports algae growing on the surface of the concrete blocks. They constitute bait, and food for the abalone can grow on their surface. These concrete blocks are designed to be small and lightweight in order to increase the efficiency of harvesting abalone. In the fishing ground where the concrete blocks had been placed on the seabed, abalones were efficiently caught just by turning the blocks. Therefore, these concrete blocks have several functions: not only as artificial reefs, but also as fishing gear to enhance the catch of *Fukutokobushi*.

3 Results and Discussion

The field experiments were conducted in the coastal waters of Tane Island. The artificial reefs were developed by placing 300 concrete blocks on the seabed of the research field on 28 March 2005. Ten juveniles per concrete block were released on 20 June 2005.

The number of *Fukutokobushi* living on the bottom side of concrete blocks and algae growing on the top were measured directly by a scuba diving scientist (see Fig. 2). The experiment site is flat, and concrete blocks are immersed between 0.5 and 3 m.

Six months after release, observations showed that the number of *Fukutokobushi* had decreased sharply and algae had grown up on the top side. *Fukutokobushi* were released in the adult stage, and the average shell length reached over 50 mm, which is commercial size, for approximately 1 year after release. The habitat was restored by developing artificial reefs made of concrete

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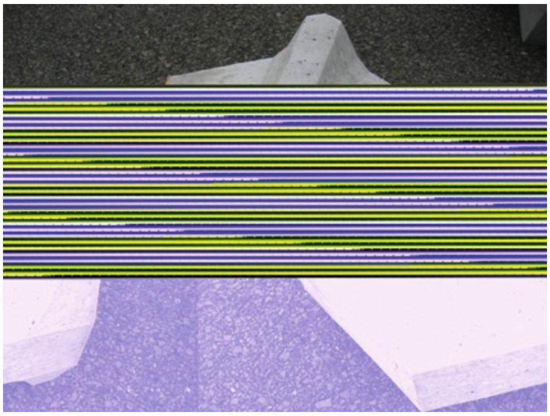

Fig. 1 The concrete block used in this experiment. Length 49 cm, width 42 cm, height 25 cm, submerged weight 30 kg

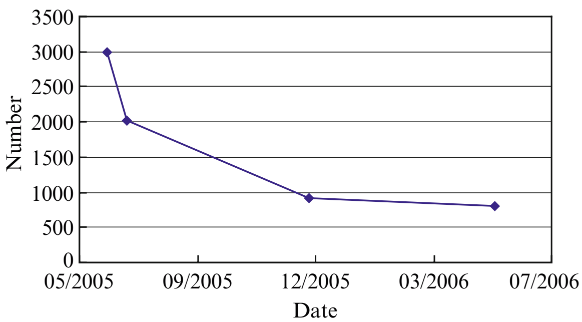

Fig. 2 Evolution of numbers of abalone *Haliotis diversicolor diversicolor* living on the lower side of the special artificial reefs

blocks in the field where *Fukutokobushi* had not existed before we conducted the experiment (see Fig. 3).

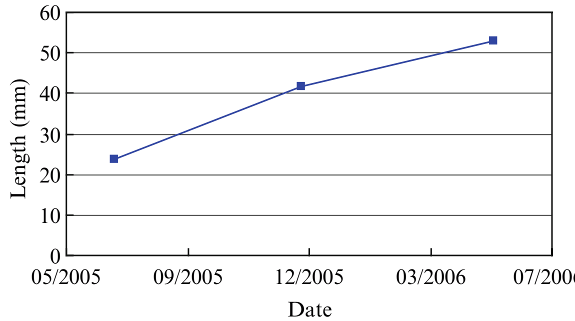

Fig. 3 The average of shell length of *Fukutokobushi* (*Haliotis diversicolor diversicolor*) after 14 months of growth

4 Conclusion

The survival rate of the abalone species is about 27%. As the result of this field experiment, fishermen have begun to make fishing grounds for *Fukutokobushi* by developing the artificial reefs in the coastal waters of Tane Island.

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Assessment of the Effect of Artificial Reef on Fish Distribution: The Combined Use of Acoustic Data and GIS

Akira Hamano

Abstract Artificial reefs have been constructed in numerous coastal areas off Japan to enhance coastal fisheries. The setting up of a method to assess the effectiveness of these artificial reefs is essential for controlling and managing these projects.

The distribution of fish schools around reefs is frequently characterized by significant variability with respect to time and space dependent on the environmental conditions. This study aims to find methods that provide a better understanding of the spatial distribution of fish schools around reefs. The pilot study area was located in the Japan Sea off Nagato, Yamaguchi Prefecture. A survey methodology was developed that combines the acoustic method and Geographic Information System (GIS) techniques.

These GIS techniques were used to demonstrate the distribution of schools on the two- and three-dimensional map. Hydroacoustic and oceanographic observations were conducted from 1996 to 2001, and 1 year of data on fish distributions and oceanographic conditions in the reef area were collected to assess the effects of artificial reefs.

The relationship between the spatial distribution of fish schools and environmental factors, including the bottom topography, was assessed in visual form using GIS techniques.

These results show the effectiveness of the combined use of GIS and the acoustic method to assess the distribution of fish schools around artificial reefs.

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1 Introduction

A large number of artificial reefs have been constructed along the coastal area of Japan for the enhancement of coastal resources (Uchida et al. 1995). The type of fishing development grounds required in order to achieve "sustainable fishery production" and a "fishery harmonized with ecology" is an ongoing issue (Kakimoto 1998).

The focus of the fishery infrastructure improvement project in Japan is shifting from coastal to offshore areas, and a balance between the conservation of the environment and fishing ground development is the most important issue. Research and analysis methods producing accurate data on the spatial distribution of fish schools around reefs are required to aid decision-making.

As fishing ground developments expand, artificial reefs are increasing in size and depth, and sampling by fishing or diving observations or conventional acoustic surveys can no longer assess their effectiveness. To address this problem, a survey methodology was developed that combines the acoustic method and GIS technology (Meaden 1996). This study focuses on this integrated research method, in particular how to correctly collect horizontal and vertical information and build a database of quantitative data.

2 The Assessment of the Fishing Ground Development in the Horizontal Plane

This section provides an example of the assessment of the effects of large-scale artificial reef clusters built in a 4,000-ha area offshore of Nagato, Yamaguchi Prefecture (Fig. 1).

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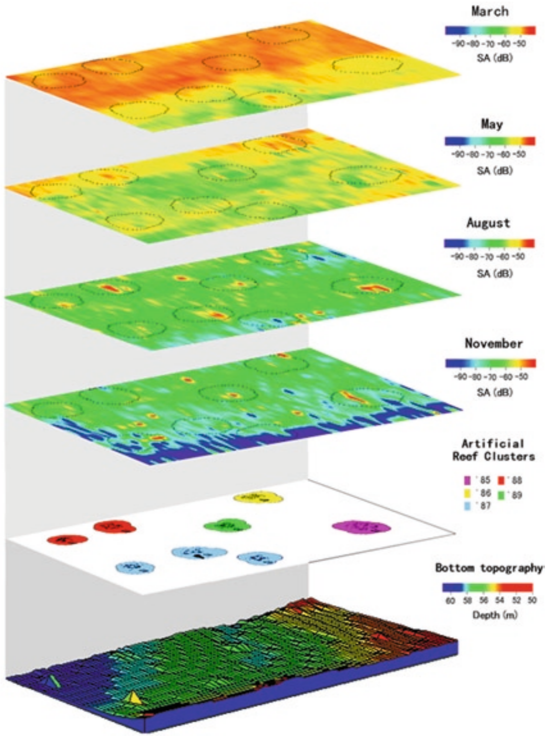

Fig. 3 Multiple layer expression of seasonal (March, May, August, and November) changes in the distribution of the area backscattering strength (Sa) representative of fish school distribution, the 200 m buffering range from reef clusters (○), and a three-dimensional illustration of bottom topography (area C) (after Hamano et al. 2001)

3 The Assessment of the Fishing Ground Development in the Vertical Plane

Collection of three-dimensional information becomes important as the size and depth of artificial reefs increases. Acoustic survey, using an underwater acoustic remote-sensing method, can provide three-dimensional information, such as the distribution and behavior patterns of various fish and bottom topography, unlike two-dimensional data collected from satellite remote sensing. Fish species move through three-dimensional marine space over short time periods. Therefore, statistical interpolation based on data gathered from the conventional vertical echo-sounder does not provide an accurate school distribution. A method of measurements using sonar to collect 3D data on school distribution and size that does not rely on this statistical method is instead suggested. Here, sector-scanning sonar was used to observe the distribution of fish schools during daytime.

This study introduces the combined use of scanning sonar data and GIS for the visualization of semiquantitative three-dimensional images of fish schools (Hamano et al. 2005). The school information acquired using this method was integrated with 3D GIS display techniques to show the spatial distribution of fish

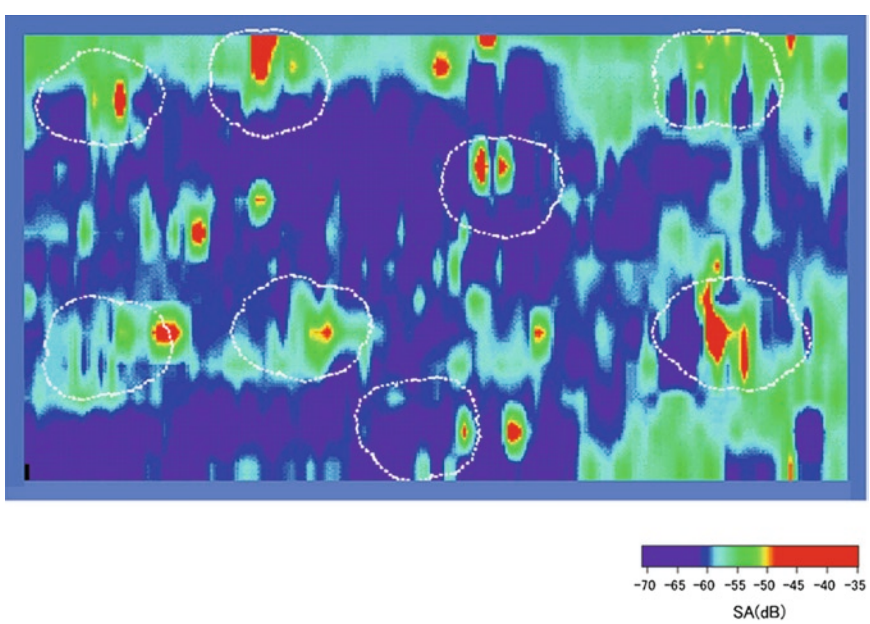

Fig. 4 Map of area C showing the three-season average (May, August, November) of the area backscattering strength (Sa) overlaid with a 200-m range around artificial reefs (○) (after Hamano et al. 2001). Red marks indicate the fish school assembling around reefs during daytime

schools near the artificial reefs or fishing ground areas (Fig. 5). It was found valuable to view the 3D image from different distances, directions, and altitudes to appreciate fish school shapes and their location with respect to each other and their environment. From this visualization, it was found that the distribution of schools was concentrated around the sea hill called *Futashima guri* and the artificial reefs shown in Fig. 5. About 56% of the fish schools were distributed in the upper 20 m above the seabed (Fig. 6).

A three-dimensional visualization of individual schools is shown in Fig. 7. For this example, school extent in depth, athwartship, and alongship dimension are 46 × 109 × 47 m^3. School volume was estimated to be 66.2 m^3. To identify the species that were observed around the large reef shown in Fig. 7, jigging was carried out by a chartered commercial fishing boat. The fish caught were predominantly carangids (Japanese horse mackerels). The average of the total length and body weight of the samples (41 individuals) was determined to be 11.9 cm and 15.5 g, respectively.

4 Discussion

Projects on the development of fishing grounds using artificial reefs are and will continue to be a major issue in coastal fishing for Japan. A balance between the conservation of the environment and fishing ground development is the most important issue of this national project. The intensive use of coastal fishing grounds and the control and management of marine resources are important. As the area and depth of fishing grounds expand in the future, a method for the evaluation of fishing grounds despite the time and location is a key

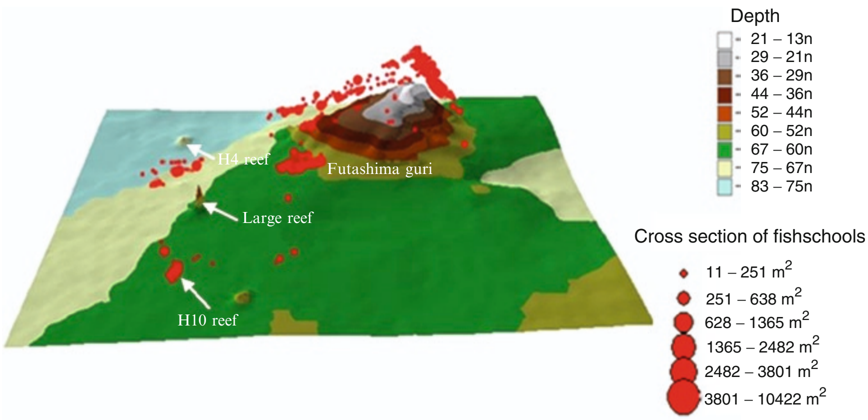

Fig. 5 Three-dimensional visualization of fish schools (*red circles*) overlaying the bottom topography

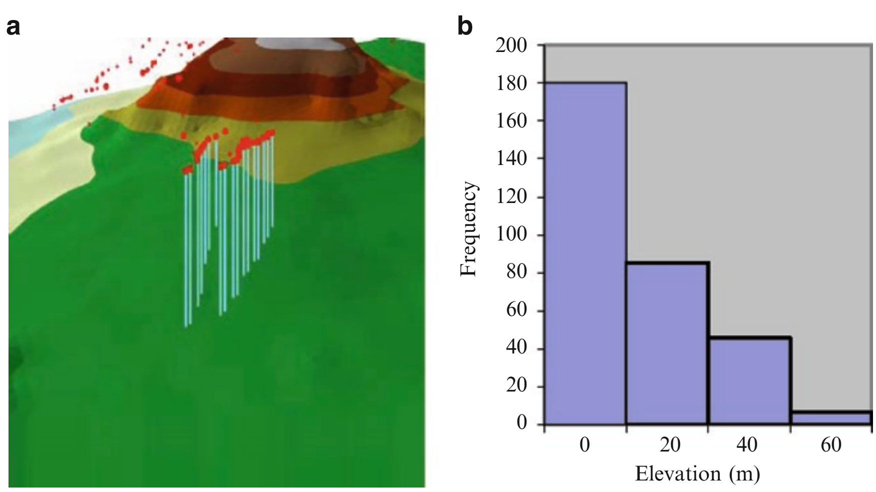

Fig. 6 (**a**) Fish school locations are shown in red; elevation of schools above the seabed is indicated by vertical bars. (**b**) Frequency distribution of fish school elevation above the seabed

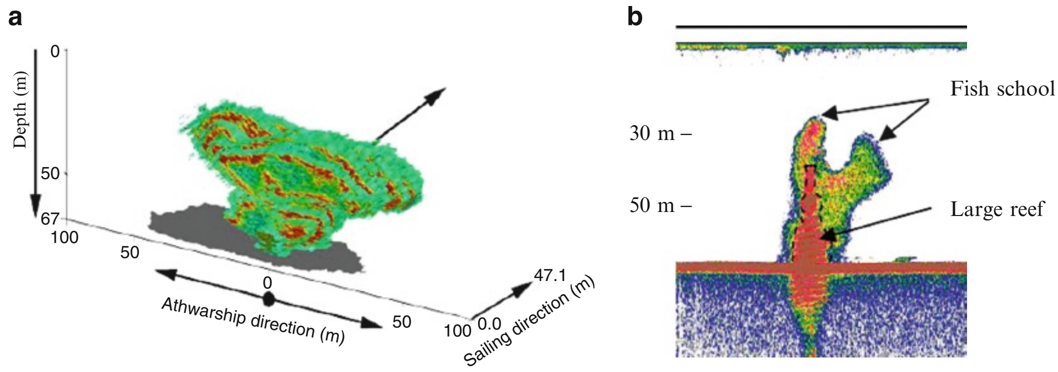

Fig. 7 (**a**) Three-dimensional school image obtained at the large reef, which was constructed from the scanning sonar information for the school. School density increases from *green* to *yellow* to *red*. The *gray* area gives the projection of the school on a horizontal plane, which is located at the maximum depth shown. (**b**) Echogram of fish school assembling around the large reef obtained by the quantitative echo sounder simultaneously

requirement. Until recently, however, it was hard to find methods that provide a better understanding and management of spatial distribution of schools around reefs and establish a relationship between them. To settle this problem, we explored the combined use of acoustic information from sonar and GIS-based 3D visualized software.

Our results illustrate the importance of spatially referenced sonar information for the visualization and understanding of fish schools, the relation between fish schools, and their relation with the bottom. If measured plankton and oceanographic features are added easily, the approach also will be valuable for the planning of assessment surveys and, as we show, for the monitoring and assessment of coastal fishing grounds that involve artificial reefs.

Our work offers a greater understanding of fish school distributions within the marine environment; it provides valuable fundamental information for building effective fishing grounds and reefs.

Acknowledgments The author is thankful to the late professor Dr. H. Kakimoto from the National Fisheries University for his constructive advice. He also thanks the late Mr. K. Uchida and Mr. T. Nakamura and students from the Laboratory of Fisheries Instrumentation, the National Fisheries University, for assistance during the field survey. He is also grateful to Dr. H.Tanoue and Dr. R.Kieser for their helpful discussion.

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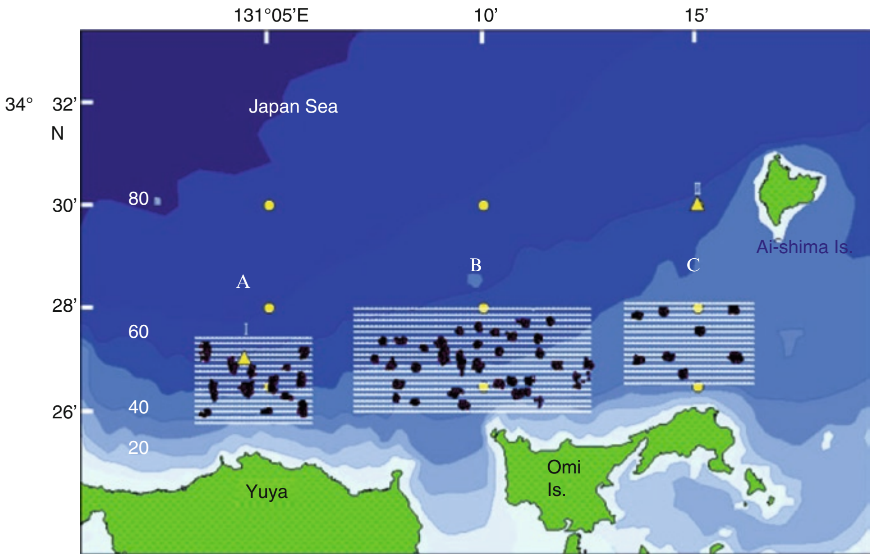

Fig. 1 An outline of the survey area and its vicinity off Nagato, Yamaguchi Prefecture, showing the location of artificial reef clusters (●), echo survey transects (- -), CTDO casts (○) (after Hamano et al. 2001)

The data shown in Fig. 2 are the precise positions of artificial reefs measured by side-scan sonar from 1991 to 1997 and fish school distribution measured by echo surveys from 1996 to 2001. Side-scan sonar and a laser positioning system, of which 2,172 were used in the analysis (Fig. 2), verified a total of 11,539 artificial reef locations.

Figure 3 represents a multilayer view of seasonal (March, May, August, and November) relationships between the exact locations of the artificial reefs obtained by side-scan sonar and the area backscattering strength (Sa) representing the fish school distribution (Maclenan and Simmonds 1991). In this figure, the bathymetry, including fish reefs, is shown on the bottom layer and fish reef distributions above it. The area backscattering strength (Sa) was used as an index to show the school distribution and is shown on the upper layers of Fig. 3. Up to 75% of fish schools were distributed within a 200-m range over −50 dB/m^2 from an artificial reef (Fig. 4), although the area of the 200-m range comprises only 23% of the total researched area. This shows that fish schools tended to aggregate in the vicinity of the artificial reefs. In summary, combining the artificial reef locations and acoustic information in a GIS database can provide quantitative information and speedy analysis of fish aggregation around a reef.

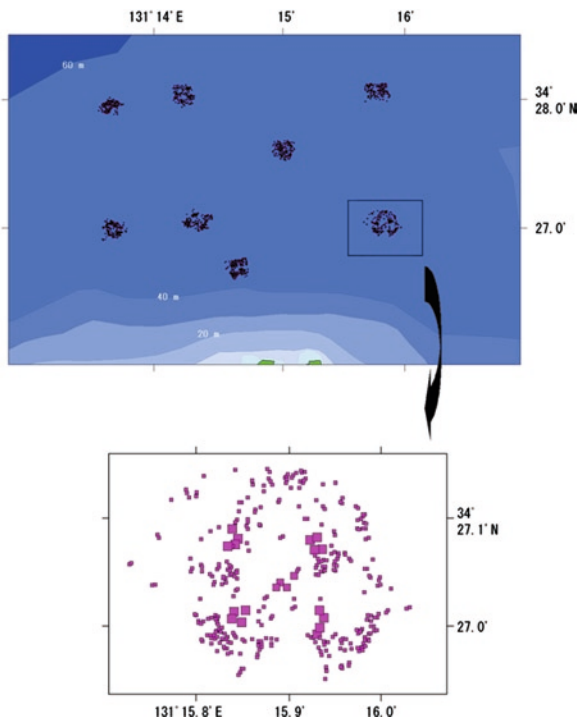

Fig. 2 Detected distribution of component blocks of reef clusters in area C from Fig. 1. Dots indicate the positions of component blocks detected by side-scan sonar (after Hamano et al. 2001)

Evaluation of Artificial Reefs Impact on Artisanal Fisheries: Necessity of Complementary Approaches

Philippe Lenfant, Jérémy Pastor, Nicolas Dalias, and Patrick Astruch

Abstract Currently, fisheries in all oceans are declining as a consequence of overfishing and other phenomena like global warming. Artisanal fisheries in the Mediterranean Sea are following this trend, which is intensified by the high price increase of oil. In this context, some management strategies need to be proposed to maintain both natural stocks and human activities.

Complementary to the quotas and marine protected areas, the artificial reefs may be one part of the solution. Concerning the fisheries context, artificial reefs (AR) can induce new habitats, more or less complex, to allow the development of rocky fish (adult and/or juvenile). Species from sandy bottoms also profit from these artificial structures because they provide protection of adjacent zones from illegal trawling. We study some artificial reefs in the south of Lion's Gulf (near Le Barcarès/Leucate, Mediterranean Sea) constituted by six "villages" of 12 ha (300 × 400 m). Each zone contains 28 modules of three types: pipes, cubes, and construction debris.

We use different methods to evaluate the effectiveness of artificial reefs:

- Visual count by diving
  Observation of commercial landing in each harbor
- External tagging of fish

Visual census showed a significantly higher species richness and density on ARs than on the control site. Abundance, biomass, and LPUE data (Landings Per Unit of Effort) issued from artisanal fisheries landing surveys were not significantly different around the AR system from other fishing grounds of the French Catalan coast. The tagging experiments on *Diplodus sargus* suggested that the connectivity of demersal fish populations must be taken into account to evaluate the influence area of ARs and thus their indirect impacts on artisanal fisheries. This study highlights the potential of combining methods covering different spatial scales to evaluate direct and indirect impacts of ARs on artisanal fisheries. Methods for the evaluation of AR efficiency are discussed.

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1 Introduction

In a general context of fisheries decline due to overfishing (Lauck et al. 1998; Castilla 2000; Agardy 2003; Pauly and Watson 2003) and to other phenomena such as climate change, it appears crucial to implement sustainable management of natural resources by conciliating conservation and exploitation purposes. In the Mediterranean Sea, artisanal fisheries are particularly impacted by fish stock declines (Lleonart and Maynou 2003; Colloca et al. 2004) because of the high fishing pressure and the increasing price of oil over the past decade. Different management tools are already being used as fishing quotas, marine protected areas (Claudet et al. 2006; Claudet and Pelletier 2004; Ashorth and Ormond 2005), and more recently, artificial reefs (Baine and Side 2003).

Although the principle of attracting fish with different immersed objects has been known since the seventeenth

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century, artificial reefs have truly been used as a management tool only since 1980. Today, ARs are not only used to increase local fish production (Bohsack et al. 1994; Relini et al. 1994; Grossman et al. 1997; Pickering and Whitmarsh 1997; Ramos et al. 2006), but also to mitigate the impacts of wastewaters (Antsulevich 1994; Leihonen et al. 1996) or aquaculture (Angel et al. 2005; Gao et al. 2008) and to restore or protect degraded habitats (Clark and Edwards 1994; Pickering et al. 1998; Reed et al. 2006).

Numerous studies have already focused on the evaluation of the efficacy of ARs as management tools for local resources, using different methodological approaches. Most of them use experimental fishing to define the potential impact of ARs on artisanal fisheries catches (Santos and Monteiro 2007), but sometimes direct visual census by scuba divers (Leitao et al. 2009) is used.

The main aim of this work was to evaluate the effect of the AR system of Leucate and Le Barcarès on fish community structure and to see how this pattern is reflected in artisanal fisheries. Scuba visual census was adopted to provide information concerning fish assemblages, and a fisheries landings survey was performed to test the direct influence of the AR system on artisanal fisheries. The landings data will be analyzed for different locations depending on the substratum type and will provide a variability analysis of catches at a local scale. A visual tagging experience of a target species (*Diplodus sargus*, Linnaeus 1758) was used to give insights concerning the connectivity of AR habitats into a natural ecosystem.

2 Materials and Methods

2.1 Study Site and Artificial Reef System

This study relates to an artificial exploitation reef (ER) system located along the French Catalan coast in the NW Mediterranean Sea (Fig. 1a). The specificity of this AR system rests on its location along a spatially very heterogeneous coastline, which represents the natural and artificial fragmentation of marine ecosystems well. The sandy Catalan coast is framed in the North by Cap Leucate and in the South by the Vermeille Coast, and is dotted with isolated rocks and artificial structures such as the studied AR system. This AR system was established in 2004, offshore between Leucate and Le Barcarès, on sandy ground and is composed of six reef groups called "villages."

These villages run parallel to the coastline along the 15- and 30-m isobaths (Fig. 1b), and they each consist of 28 reef sets of concrete. Each reef set being at a distance of 50 m, a village occupies a total area of about 120,000 m² (400 m long × 300 m wide). Three

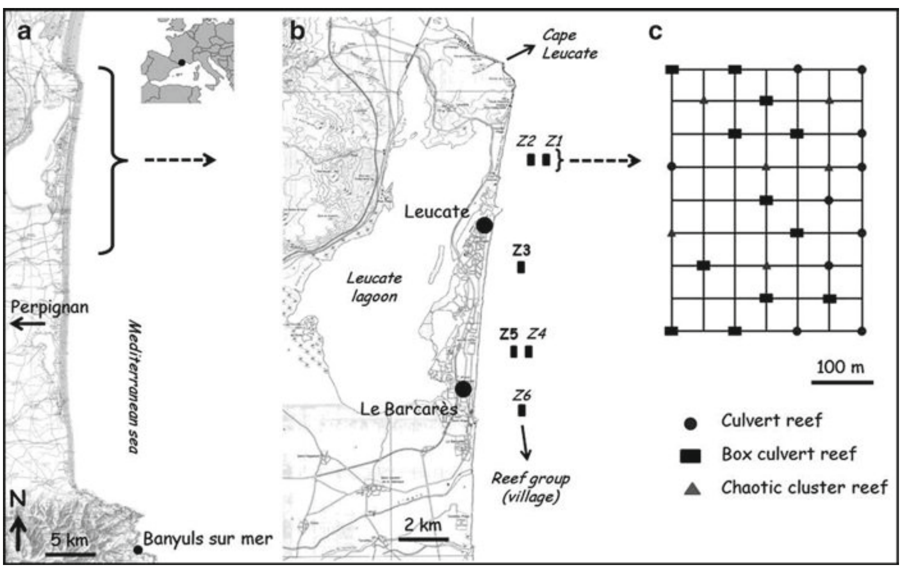

Fig. 1 (**a**) Study site and location; (**b**) position of the six reef groups, which constitute the studied AR system; (**c**) scheme of the positioning of the reef sets of an AR village

different reef sets are used: culvert reefs, box culvert reefs, and chaotic clusters, which are distributed in the "villages" according to the following scheme (Fig. 1c). This study was carried out on four chaotic cluster reef sets of two of these "villages" Z3 (17.5 m deep) and Z5 (18 m deep). A natural rocky reef, located 8 km North of Z3 and 11 km North of Z5, was selected as control site.

2.2 Data Sampling and Analysis

2.2.1 Underwater Visual Census (UVC)

The number of fish and invertebrates per species were recorded on ARs and the control sites by a scuba visual census in spring 2007. To avoid bias due to the high relative abundance of fish species belonging to the *Blennidae* and *Gobidae* genus, they were excluded from analyses in this study. Visual census was performed along a 40-m-long and 5-m-wide (200 m²) transect on four zones with similar depth as the AR systems, selected randomly on the control location. Another visual census method had to be adapted to the specific design of the AR structures. A complete inventory of each reef set (84 m²) was performed by one diver. The fish count was performed in three steps, to catch species of different mobility: (1) the very mobile fish species, (2) then the species near the reef, and finally (3) inside the cavities of the reef were recorded. Density data (individuals per m²) were used for the analyses, to compare UVC counts of the AR and control site. Prior to data analysis, species richness and densities were log-transformed to reduce the weighting of abundant species and increase that of more rare species. Fish assemblage structure of the different locations was compared by a similarity analysis using the PRIMER software package (Plymouth Marine Laboratory, UK; Clarke and Warwick 1994). The Bray-Curtis similarity matrix was used to generate a hierarchical cluster analysis. The major fish species contributing to dissimilarities among locations were identified by a similarity percentage analysis (SIMPER). A one-way ANOVA followed by a post hoc Tukey test was performed on richness and densities to test the differences among locations (significance threshold: $p < 0.05$). To estimate the contribution of commercial fish species to the fish assemblage structure, the analyses have been performed on two datasets, a complete one and one excluding noncommercial species.

2.2.2 Landings Per Unit of Effort (LPUE)

A survey of artisanal fisheries landings along the French Catalan coast (between Leucate and Port-Vendres) was undertaken in spring 2007 (April–June). During this survey, gear type, fishing location, species richness, abundance, and weight were recorded. Biomass, fishing effort, and LPUE were calculated. With the GPS position of deployment location of gears, the LPUE data (g · m² · h) were geo-referenced on a map of the study site and analyzed with a geographical information system (GIS). We used a grid (1 × 1 km) to maintain the confidentiality of exact net position.

2.2.3 Tagging

A visual tagging experiment was carried out in Summer 2006 on 54 individuals of white sea bream (*Diplodus sargus*) associated with the ARs. Fishes were captured at night by divers using landing nets and were externally tagged with T-bar anchor tags (FD-68BC, Floy Tag®) below the dorsal fin. The recapture data were obtained by professional and recreational fishermen tag return, but mostly through regular recapture dives. The capture and recapture locations of tagged fishes were identified on a GIS map of the study site to obtain the displacement distance, and the number of days between recaptures was calculated.

3 Results

3.1 UVC

We identified 29 fish and 6 invertebrate species during the scuba visual census on the three studied locations. The species recorded are listed for the different locations in Tables 1 and 2. The hierarchical cluster analysis of fish assemblage structure showed a distinct separation between counts at the control site and the AR systems (Z3 and Z5) at less than 10% similarity (Fig. 2). On the contrary, no separation was visible between the two AR sites. The analyses of one-way ANOVA on

Table 1 List of fish species at each location. AR corresponds to both AR systems Z3 and Z5 together. The noncommercial species are marked with a star

Location	C	Z3	Z5	AR
Diplodus sargus	+	+	+	+
Pomadasys incisus	−	+	+	+
Dicentrarchus labrax	−	+	+	+
Phycis phycis	−	+	+	+
Trisopterus sp.	−	+	+	+
Conger conger	+	+	+	+
Scorpaena notata	−	+	+	+
Scorpaena porcus	+	+	+	+
Mullus surmuletus	+	+	+	+
Diplodus vulgaris	+	+	+	+
Serranus cabrilla	+	+	+	+
*Serranus hepatus**	+	−	−	−
*Ctenolabrus rupestris**	+	+	+	+
*Coris julis**	+	−	−	−
Symphodus tinca	+	−	−	−
Diplodus annularis	−	+	+	+
Atherina sp.	−	+	−	+
*Chromis chromis**	+	+	−	+
Diplodus puntazzo	+	−	−	−
Trachurus sp.	−	+	+	+
Sciaena umbra	−	−	+	+
Sarpa salpa	−	+	−	+
Spicara maena	−	+	−	+
Spicara smaris	−	−	+	+
*Boops boops**	−	+	+	+
*Oblada melanura **	+	+	+	+
Spondyliosoma cantharus	−	+	+	+
Sparus aurata	−	+	−	+
Pagellus acarne	−	+	+	+

Table 2 List of invertebrate species at each location. AR corresponds to both ARs systems, Z3 and Z5 together. The noncommercial species are marked with a star

Location	C	Z3	Z5	AR
Octopus vulgaris	−	+	+	+
Palinurus elephas	−	+	+	+
Portunus puber	−	+	+	+
*Galathea squamifera**	−	−	+	+
*Palaemon serratus**	−	−	+	+
*Stenopus spinosus**	−	+	−	+

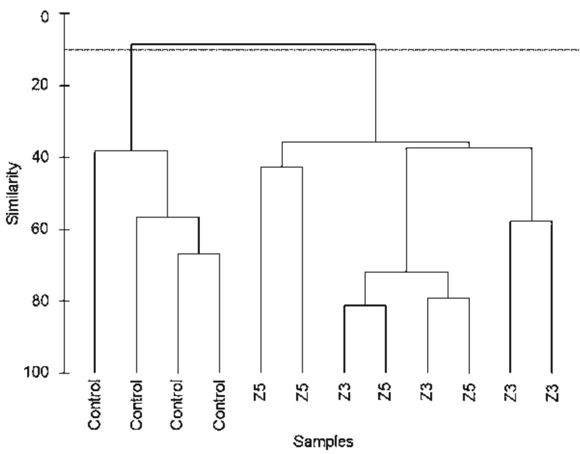

Fig. 2 Hierarchical cluster analysis (group average) of all samples for the three different locations

species richness and density showed a significant difference between locations, with *p*-values of 0.032 and 0.000, respectively (Table 2). Density at the control location was significantly lower than for both AR villages Z3 and Z5 (*p*-value = 0.000), with mean densities between X and X-fold lower than for the control location (Table 2).

Species richness was only significantly higher than in the control location in Z3 (*p*-value = 0.036). Among the AR villages Z3 and Z5, no significant differences were observed (*p*-value = 0.614). The dissimilarity percentage between the control location and the two AR locations reached 90% for Z3 and 92% for Z5. Between Z3 and Z5, the dissimilarity percentage of the species assemblage reached 55%. The bogue *Boops boops* was the species contributing most to dissimilarity between assemblages, with a dissimilarity percentage exceeding 22% for all locations. Common two-banded and white sea bream also contributed highly to the dissimilarity percentage among locations.

3.2 LPUE

During the fisheries landings survey in Spring 2007, 90 species were identified, including nine invertebrate and six elasmobranchii species. From the 35 species counted by UVC, all were present in artisanal fisheries landings, except the black squat lobster *Galathea squamifera* and the two shrimp species *Palaemon serratus* (common prawn) and *Stenopus spinosus* (Mediterranean boxer shrimp). The grid of mean LPUE for the different fishing locations is shown in Fig. 3. The one-way ANOVA and the post hoc Tukey analyses revealed no significant differences among the different locations.

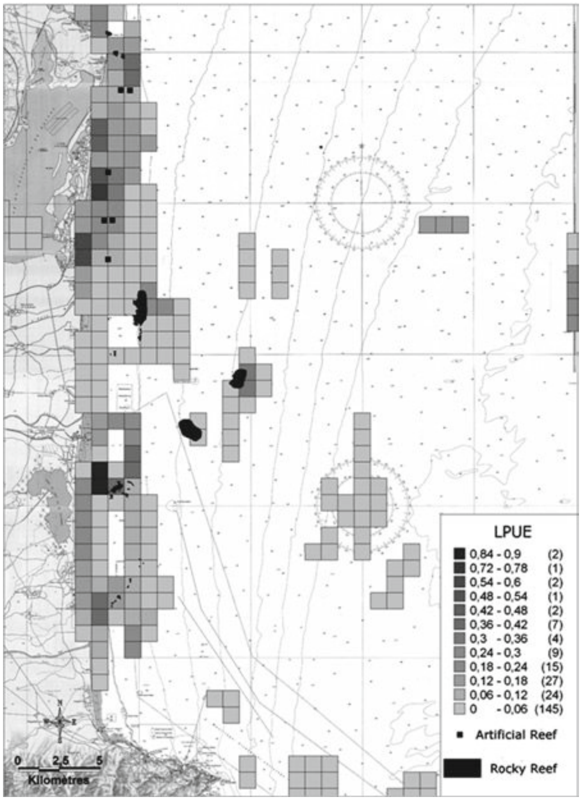

Fig. 3 Grid of mean LPUE of the different fishing locations

Table 3 Recaptured white sea bream tagged on the ARs of Leucate and Le Barcarès

Recapture location	Distance (km)	Time (day)	Size (mm)
AR	0.2	20	385
AR	0.2	5	350
AR	0.2	5	208
Leucate channel	6.7	11	210
Port La Nouvelle	20	15	220

3.3 Tagging

Out of the 54 white sea bream tagged on the ARs, 5 individuals were recaptured (Table 3), representing a recapture rate of 9%. Three were found by divers on another reef set as for the capture, 5 and 20 days after capture (Table 3). The two other fish were recaptured by fishermen. One moved toward the Leucate channel of the Leucate lagoon (Fig. 4), and the other moved 20 km north to Port La Nouvelle (Fig. 4, Table 3).

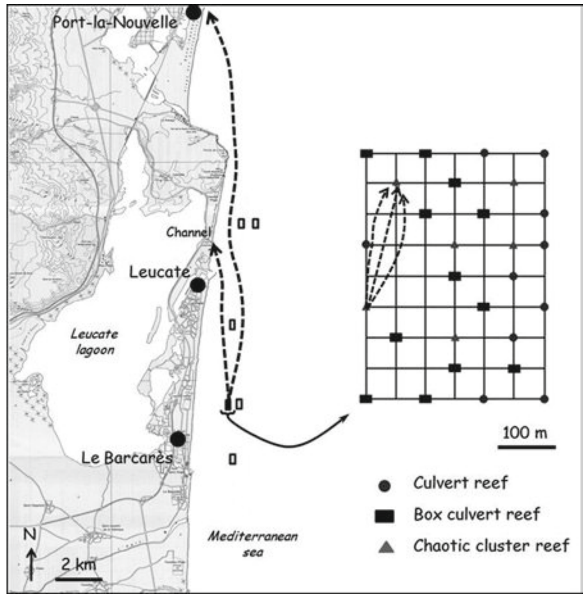

Fig. 4 Displacements of the recaptured tagged white sea bream (*dotted line*) outside of the AR village and inside the village between reef sets

4 Discussion

As mentioned in many AR studies (Alevizon and Gorham 1989; Fabi and Fiorentini 1994; Grossman et al. 1997; Fabi et al. 2004; Arena et al. 2007; Santos and Monteiro 2007; Dupont 2008), the fish assemblages associated with the studied AR structures were significantly different from those associated with the control site, especially regarding fish densities, which were significantly higher on ARs. The comparison between both AR villages, Z3 and Z5, located at a similar depth but at different distances from the control location, provides evidence that the distance to the closest natural rocky reef (control location) is not a factor influencing fish assemblages (richness and density). Under a purely attraction hypothesis of fishes from natural rocky reefs to the artificial reefs, it would have been likely to see an abundance gradient with distance from the control location. The ARs of Leucate and Le Barcarès were thus probably not attracting fishes away from the control location at the Cap Leucate.

Out of the 35 invertebrate and fish species recorded during UVC, only 13 were present on the control site. None of the censused invertebrate species were seen in the control location. However, the dissimilarity analysis of the fish assemblages of the different sites showed that the species contributing most to dissimilarity are those present in all locations but in different proportions. Excluding the bogue *Boops boops*, for which a high contribution to the dissimilarity percentage of the community assemblage is due to its gregarious behavior and not necessarily linked to the substrate type, the species contributing most to dissimilarity between locations is the white sea bream *Diplodus sargus sargus* followed by the common two-banded sea bream *Diplodus vulgaris*. The low abundance of common two-banded and white sea bream on the control location explains the important contribution of these species to dissimilarity of species assemblages between control and AR locations. According to the FAO statistics (FAO 2004), the white and the common two-banded sea bream are two of the most commercially important species in southern Europe's fisheries.

This UVC study provides evidence that the colonization of the studied AR is by fish and invertebrates of more or less commercial value. The high abundances of some demersal fish species on the ARs compared to the Cap Leucate explain the differences in the community structure between the artificial and natural hard bottoms. As in many studies (Charbonnel et al. 2002; Sherman et al. 2002; Brotto et al. 2006; Grober-Dunsmore et al. 2008; Lan et al. 2008), higher fish densities can be related to a higher structural complexity of the habitat, providing better shelter. The high fish abundance and the high contribution of commercially valuable fish species (sparids) on the ARs of Leucate and Le Barcarès, compared to the control site highlight the potential role of these artificial structures to fisheries enhancement.

During the fisheries landings survey of Spring 2007, 90 species were recorded out of the 300 fish, crustacean, and mollusc species identified in the Gulf of Lion (Aldebert 1997). Despite the small sampling area and the selectivity of fishing gear, nearly a third of the species richness of the Gulf of Lion could be sampled through the artisanal fishing survey. The Mediterranean fisheries, except for large pelagics, are characterized by fragmented fleets with a large number of landing points and multispecies catches (Lleonart and Maynou 2003), explaining in part the high species richness observed in our study. Furthermore, the studied location is a very heterogeneous environment, composed of a rocky coast in the south, partly surrounded by posidonia seagrass meadows and a sandy coast in the south, interspersed with artificial and natural hard bottoms. This mosaic of substrates could probably play a role in the high species richness observed in the sampled area (Gratwicke and Speight 2005).

All species sampled by UVC were also identified in the fisheries landings, except four of them. The species, exclusively recorded by UVC, correspond to rare species, such as the bastard grunt *Pomadasys incisus* (Pastor et al. 2008) and small crustaceans mostly living in cavities, such as the Mediterranean boxer shrimp, *Stenopus spinosus,* and the black squat lobster *Galathea squamifera*, all with no commercial value and thus not targeted by fishing gear. Contrary to what could have been expected from the UVC results, the analyses of the fisheries data do not show significant differences in catches surrounding the ARs and the other fishing locations.

The method of Underwater Visual Census (UVC) is mostly used in AR research to describe the community structure associated with artificial vs. natural habitats (Bayle-Sempere et al. 1994; Bombace et al. 1994; Coll et al. 1998; Seaman and Jensen 2000; Chou et al. 2002; Zalmon et al. 2002; Arena et al. 2007; Dupont 2008). In fact, this method provides insight into the fish community directly associated to the artificial or natural hard bottoms, by the sampling of demersal and benthic species, which are not necessarily available to fishing gear. As confirmed by the fish species sampled in this study, this sampling method is however limited to the census of pelagic and very vagrant demersal fish species, which have a high commercial value. In complement to the UVC method, the LPUE data provide information of the direct repercussions of fish community structure on fishing yield, with a greater selectivity for pelagic and demersal species from homogeneous grounds (Godoy et al. 2002; Zalmon et al. 2002). The differences in the results of the LPUE and the UVC survey point out the necessity of having insights into both the fish population dynamic and the fisheries dynamic to better understand and manage resources.

The absence of differences in catches between the Cap Leucate and the AR system could be due to the patchy distribution of reef sets and the high relief of reef structure. The most commonly used fishing gears in the study zone are gillnets and trammelnets, which cannot be used close to rocky bottoms and especially not on high relief bottoms. For this reason, most nets are deployed at least at 200 m from the hard bottom structures. A study by Alevizon and Gorham (1989) shows the increase in local resident reef fishes associated to ARs but no effect on fishes dwelling in nearby nonreef habitats, which would have been more available to net gear. Unpublished experimental fishing with different fishing gear showed differences in the catchability of sea bream by nets and long-lines, which seem to be a much more efficient fishing gear on the ARs of Leucate and Le Barcarès. As indicated in Leitao et al. (2009) for the Algarve, over the years, long-line fishing has been widely abandoned by local artisanal fishermen on the French Catalan coast in favor of net gear, which is easier to operate.

Despite the reward offered for the capture of tagged white sea bream, only two fish were returned by fishermen. A study by Abecasis et al. (2009) with capture-recapture experiments on white and two-banded sea bream showed similar difficulties with the return by fishermen, with a recapture rate between 3% and 4%. Although it may be time consuming, regular recapture dives at strategic sites seem to be a good alternative for the recapture of tagged sea bream and to avoid the loss of information on these individuals. Despite the few recaptures, the tagging experiment demonstrated the connectivity between artificial reef sets within the same "village" as well as between the AR and habitats outside this area. The displacement of one of the recaptured sea bream to the Leucate channel could show the potential role of the adjoining lagoon as feeding habitat (Kjerve 1994; Maci and Basset 2009). Since the white sea bream is one of the most highly contributing species to differences in fish density between the control location and the AR system, this preliminary study emphasizes the necessity of considering the life-history connectivity of fish populations and their habitat usage to better understand the role of these artificial structures in fish habitats and the processes driving the increase of fish abundance close to ARs. According to Sheaves (2009), "life-history connectivity" is the sum of all migrations and dispersals among multiple habitats necessary to the achievement of the fish life cycle, such as spawning migrations, dispersal of eggs and larvae, migrations to join the adult population or feeding migrations, and migrations to habitat refugia. Recent advances in tagging with electronic devices limit the difficulties of studying the spatial dynamics of highly mobile fishes, as shown for the common two-banded and white sea bream in Abecasis et al. (2009). Further research on habitat use and displacement patterns of white sea bream would thus be required to better understand the role of ARs in the habitat of coastal fishes and to find an adequate way to evaluate the indirect impact of ARs on fish production.

5 Conclusion

As an overall conclusion, we can say that the AR system of Leucate and Le Barcarès is efficient for fish aggregation, including commercially valuable species. However, fishing yield is not enhanced in the vicinity of the ARs compared to the other fishing locations. This preliminary study highlights the importance of using complementary approaches to evaluate the impacts of ARs on artisanal fisheries. The results of the tagging experiment of white sea bream shows the importance of considering the "life-history connectivity" of fish populations to evaluate the influence area of these reefs and to study the impact of ARs on artisanal fisheries.

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Operation Prado Reefs: A Model for Management of the Marseille Coast

Emilia Medioni and Jean-Charles Lardic

Abstract In 2000, the City of Marseille launched the "PRADO REEFS" program involving the construction and submersion of underwater ecological habitats designed to repopulate, in a few years, previously unproductive seabeds. It was a national-scale pilot project; in fact, nearly 30,000 m^3 of reefs have been submerged at depths of 25–30 m, over an area of 200 ha, off the coast of Marseille.

This €6 million project was conducted in close dialogue with all stakeholders concerned. Representative of the "Plan de Gestion de la Rade de Marseille" (Marseille harbor management plan), launched in March 2006, helped to establish relations of trust and cooperation with all ocean management bodies and users, particularly with professional fishermen.

The concrete results of this successful cooperation were an outside funding reaching 80% of the total investment. Significant discussions and technical studies, based on two decades of experimentation, were carried out, resulting in the definition of seven types of reefs, adapted to the natural and ecological conditions existing around Marseille and guaranteeing the widest possible diversity of crevices, shelters, and attachment sites for marine organisms.

After obtaining regulatory approvals from the Prefecture and following the public consultation in 2006, works began in March 2007, for completion in the summer of the year 2008. As planned from the project outset, an 18-month moratorium period came into force on the submersion of the last reef, giving the users the opportunity to define the future management of the whole area jointly.

Scientific monitoring will start in 2009 in order to follow the development of various organisms into the reefs and to gain greater knowledge regarding the local functioning of the marine environment and its relationship with these submerged structures. The "PRADO REEFS" project is a true sustainable development project, an example of what a community can do to protect and enhance its environment and support its socioeconomic activities.

1 Introduction

The rehabilitation of the soft sea bottom of Prado Bay is an important operation in territory restoration, one that fits in with a program for sustainable management of the Marseille coast. The aim of this project is to increase natural resources and ensure the permanence of human activities on the coastal areas.

With this program, the city of Marseille has decided to act in a global and coordinated method by initiating a voluntary approach in favor of the Management Plan for the Roadstead of Marseille (Plan de Gestion de la Rade de Marseille, known by its French acronym P.G.R.M.).

This restoration policy and management plan consist mainly of reaching an adequate balance between:

– Natural environments that must be preserved due to their exceptional value in terms of ecology and scenery (e.g., the classified Calanques site)
– Sites that have become definitively "artificial" to satisfy the needs of maritime activities (such as ports)

Between these two extremes are intermediary urban spaces, such as the sites of Prado Bay and the Frioul archipelago (islands of the Gulf of Marseille), and sites

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with ecological and economic potential that have heretofore been neglected, but that now could be developed and enhanced.

The dual advantage of restoring former marine productive zones that have disappeared (such as dead seabed matter that causes the rise of the lower limit of living *Posidonia* beds by several meters) is that it directly benefits users of the sea, particularly fishermen, and relieves pressure on the sensitive and threatened natural zones that have been damaged by various overuses.

2 General Principles

The main goal of the artificial reef immersion project is to increase the diversity and stability of marine resources and the local ecosystems in the soft bottom of Prado Bay.

The basic idea is to provide new hard-substrate undersea habitats, immersed between 25 and 30 m below the surface, which are adapted to the ecological needs of a greater number of marine organisms. With a massive input of rocky-like habitat, the productivity and biological diversity of the current sandy bottom will increase considerably, along with the value for ecology and fishing activities (Bernard et al. 1999; Lefèvre et al. 2000).

Various research projects carried out in the Mediterranean coasts over the past two decades are now considered to be sufficiently conclusive (Bregliano and Denis 1985; Ceccaldi 1994; Jouvenel et al. 2005), providing support for the Marseille project to adopt a very ambitious qualitative and quantitative objective from the start, including interesting exchanges with scientists from Japan.[1]

This has made the project more significant at the national level: the immersion of a volume of nearly 30,000 m^3 of reefs, for a total investment of €6 millions (40% from Europe (European funds for fisheries), 30% from Agence de lEau Rhône Méditerranée et Corse (the Mediterranean Rhone and Corsican Water Agency), 20% from the city of Marseille, and 10% from the Conseil Régional Provence Alpes Côte dAzur (Regional Council in Provence, Alps and Côte dAzur). This objective was met in 2008 with the immersion of the last of the 406 artificial reefs.

From the start, the city of Marseille has built its project on the active collaboration of all partners, brought together into a Scientific and Technical Monitoring Committee that reunites state institutions, scientific and expert groups, and stakeholder representatives (Juppeau and Kahoul 2004). Its essential mission is to discuss and validate each main step in the project, at the scientific, technical, and administrative levels (Bernard et al. 1999; Lefèvre et al. 2000; Noël 2002; Sartoretto et al. 2002; BCEOM 2002; CREOCEAN 2004).

Over the past 9 years (2000–2008), the committee has held nine plenary meetings at Marseille City Hall.

From the very start of project development, the involvement of professional fishermen has resulted in active participation of the Local Committee of Maritime Fishers (Juppeau and Kahoul 2004), transfer of European financing to structural funds destined for fishing, the application of a ban on all forms of fishing during the consultation period until December 31, 2010 in the regulated fishing zone, and their willingness to undertake self-surveillance of reef zones and sustainable and reasoned management of the halieutic resources.

3 Anticipated Organization

In agreement with the main partners involved, it was finally decided to limit the sector of immersion to two zones:

- **A sanctuary zone** of 110 ha, in which all uses are forbidden outside of surface navigation.
- **A regulated zone** of 110 ha, in which fishing will not be allowed during the transition period until December 2010. This delay will be used to develop management of this zone with all stakeholders.

Prado Bay has many positive features for this type of project: easy access and surveillance; protection afforded by the Frioul archipelago against waves from the open sea; the existence of nearly permanent marine currents that change direction depending on the dominant winds (mistrals, eastern winds); proximity of a vast *Posidonia* bed and significant natural rocky zones that guarantee rapid colonization by local submarine

[1]Notice that the Colloque franco-japonais dOcéanographie held in Marseille in September 16–21, 1985, has published a whole volume of NNN pages (tome no. 7) on the subject of artificial reefs.

flora and fauna; the possibility of restoring former plant bed zones that have disappeared in a recent past and which form vast flat bottoms that are favorable to reef installation (CREOCEAN 2004); the existence of a ferry way (Marseille-South developed zone) that enables a sanctuary for marine fauna to be included in this zone without engendering new constraints.

4 Description of Reef Architecture

The objective is to provide marine plant and animal life with the features of hard substrates, a range of ecological habitats that are as large and diversified as possible (dimensions of cavities between a few decimeters and a few millimeters) to provide food sources and shelter to all stages of biological cycles of the different species (Tables 1 and 2).

Biological effectiveness is provided by the system known as "chaotic piles," which has proven to be valuable in the Mediterranean. But the legal obligation to plan for the eventual possibility of reef removal (due to the temporary nature of territory acquisition in the public maritime domain) has led to significant constraints. As well, studies have been done on the design of large-volume units (58–306 m^3) that are solid and stable on the bottom, and that are easy to manufacture and immerse (as well as to remove).

Ecological optimization of these structures, which are organized on land, essentially focuses on the interior three-

Table 1 Number and volume of the different models used

Type of reef	Number	Total volume (m^3)	Percentage (%) Number	Percentage (%) Volume
Piles:	245	10,000	34	37
– Concrete cubes (ACB)	202	3,100	28	11
– Quarry rocks (ABR)	43	6,900	6	25
Units:	142	8,300	20	30
– Metal basket reef (RPA)	21	4,725	3	17
– Fakir basket reef (RPF)	21	1,575	3	6
– Chicane reef (RC)	100	2,000	14	7
Floating ropes:	337	9,000	46	33
– Low (FB)	323 (×4)	5,500	44	20
– High (FH)	14	3,500	2	13
	730	27,300	100	100

Table 2 Data on the weight and volume of the reefs

Unit or pile	Volume (m^3)	Weight (t)	Volume and total weight
Piles of 6 Concrete cubes (ACB) + low floating ropes (FB)	Volume of one cube = 1.7 m^3 Pile of 6 cubes ± FB = 25 m^3	1 cube = 0.750 t 4.5 t + linkage	202 ACB = 5,200 m^3 (= 1,000 t)
Piles of quarry rocks (ABR)	160 m^3	500 t	43 ABR = 6,900 m^3 for 21,500 t
Metal basket reef (RPA) + low floating ropes (FB)	1 element = 45 m^3 RPA = all 3 elements piled up + FB = 266 m^3	16 t 48 t	21 RPA = 5,600 m^3 for 1,000 t
Fakir basket reef (RPF)	75 m^3	48 t	21 RPF = 1,575 m^3 for 1,000 t
Chicane reef (RC) + low floating ropes (FB)	50 m^3	15 t	100 RC = 5,000 m^3 for 1,500 t
High floating ropes (FH)	252 m^3	Ballast = 6 m^3 of concrete = 14 t	14 FH = 3,525 m^3 for 200 t of ballast
TOTAL			27,300 m^3 26,200 t

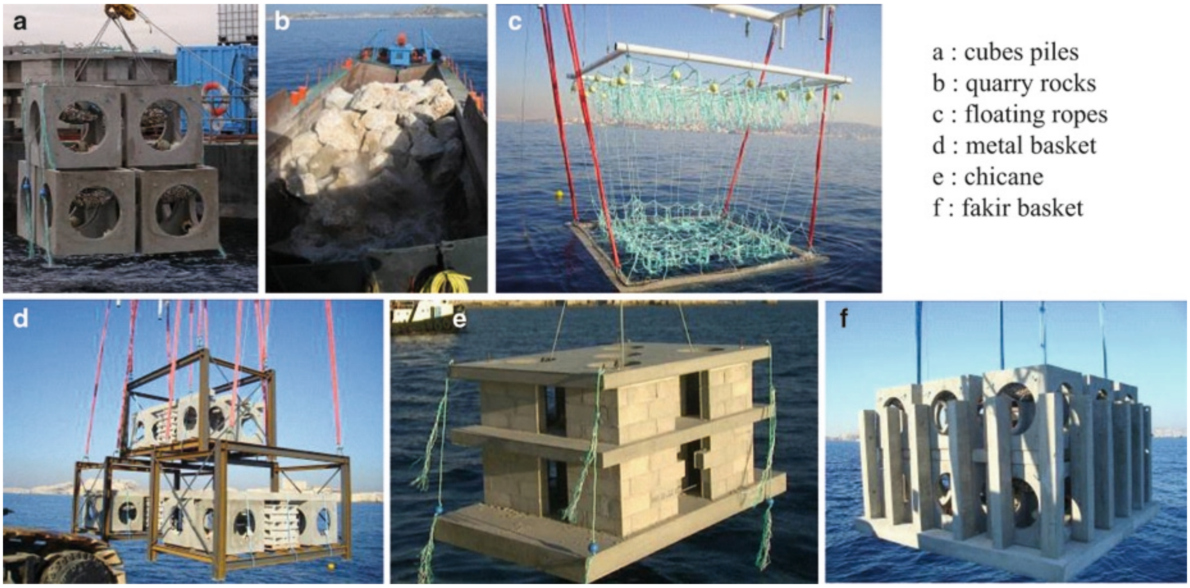


Fig. 1 The different types of artificial reefs used in the PRADO REEFS OPERATION in Marseille

dimensional complexity and uniformity obtained with the materials used in their fabrication (concrete, steel, synthetic cords for guard lines) and the interior packing using units of different sizes and functions (1.7 m^3 concrete cubes, blocks, oyster shell nets, octopus pots).

Project studies have led to three overall types of reefs being retained:

- Large volume "basket" models
- Piles of concrete cubes
- Quarry rocksand two types of guard lines:

- "High" floating ropes, which are independent units of two floors
- "Low" floating ropes, which are always combined in groups of four; there are three specific models: the metal basket reef, the chicane reef, and the piles of concrete cubes (Fig. 1).

5 Plan for Implantation

The general objective is to favor the distribution of a large number of reef units (surface effect) rather than a great concentration in the form of a few big piles and to facilitate accessibility by mobile fauna to all lower parts of the reef units, as well as communication among the reefs using linkages.

Two overall types of unit grouping were thus adopted, enabling optimal occupation of all available surfaces (220 ha): the villages (six triangles of 300 m on each side, made of 59–65 units) and linkages (eight segments of 300 m in length made up of nine units).

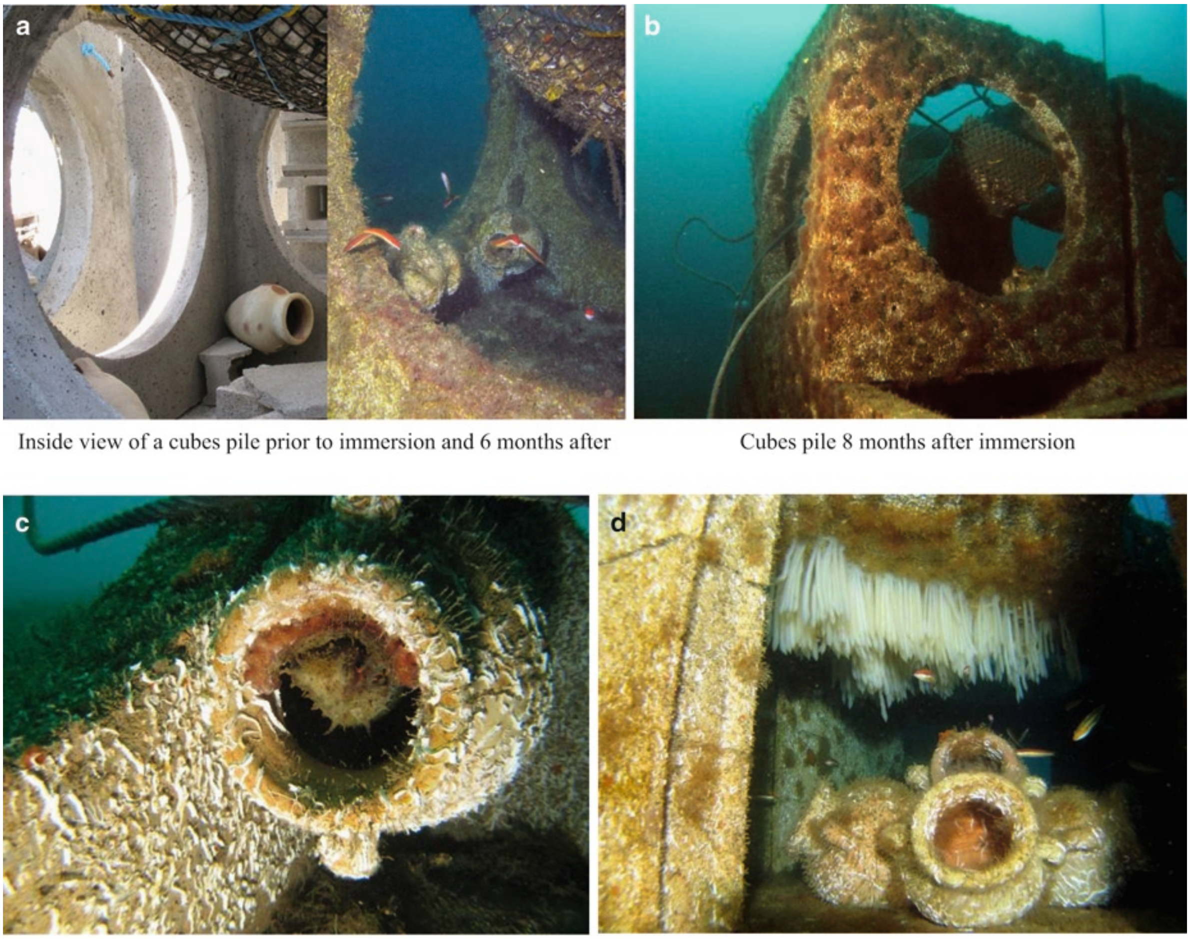
6 Initial Results (Fig. 2)

7 Monitoring and Development Program

The territory acquisition decree anticipates initial scientific reef monitoring over a period of 10 years, based on the measurements and observations carried out over the different "stage zero studies."[2]

Monitoring is based on the use of two complementary methods: experimental fishing (by professional fishers

[2]État zéro de la pêche artisanale aux petits métiers dans la baie du Prado (rade sud de Marseille) Lot 1 – (opérations de pêche) attribué à l' Association de la Pêche Professionnelle pour la Préservation de l' Environnement Marin (APPPEM). Contrat Ville de Marseille –APPPEM no. 04.0860 du 12.07.2004.
Lot 2 – (Responsable scientifique) attribué à la société P2A Développement. Contrat Ville de Marseille – P2A Développement no. 04.0886 du 09.08.2004.



Inside view of a cubes pile prior to immersion and 6 months after

Cubes pile 8 months after immersion

Octopus inside a pot

Squid "bridge" on the ceiling of the first level of a chicane reef

Fig. 2 Colonization of artificial reefs in Prado Bay (Marseille) 8 months after immersion

and supervised by an expert in halieutics) and direct population inventories using a visual reporting technique in undersea diving. Control monitoring of reef stability and wear will also be carried out during diving expeditions.

Provisional planning of this first scientific and technical monitoring phase is suggested the table below, with regular follow-up during the first 5 years, then a report 10 years after reef immersion.

The stakeholders in the project have hoped to go further than the compulsory monitoring program. An operation of this scope is an opportunity to develop a research program aimed at a better understanding of ecosystem function in "artificial reefs" and its relationship to the environment. The scientific community and its partners have therefore been solicited to suggest other types of monitoring of the zone.

The marine science center in Marseille and the Commission Environnement et Biologie Subaquatiques du Comité Départemental 13 (Bouches du Rhône Department) de la Fédération Française dEtudes et de Sports Sous-Marins (Subaquatic Environment and Biology Commission of Department Committee 13 of the French Federation of Undersea Studies and Sports) have therefore suggested additional means of monitoring organic matter, plankton communities, genetic origins of populations, and biological surveillance.

Other means of scientific monitoring are being discussed with new partners.

A socioeconomic study will evaluate the impact of these reefs on activities and uses, in particular the economic spin-offs for professional fishing. The study will use surveys and analysis of specific indicators.

The involvement of professional fishers is essential, as it will enable monitoring of their catches over a set period to compare them with the period before the reefs were set up.

8 Regulations and Management

Fishing, diving, and anchoring are forbidden throughout the zone until December 31, 2010 to allow species and food chains to establish themselves naturally.

During this period, consultation with stakeholders will help define the modes of use over the regulated zone (south zone, outside the channel). The north zone (inside the navigation channel) will be set up as a sanctuary where all activities are forbidden. The existence of two differently managed zones will assist in evaluating a conservation zone and population evolution.

9 Conclusion

From the moment of conception to the immersion of the last reef, the immersion of nearly 30,000 m^3 of artificial reefs in Marseille has taken 10 years. The city of Marseille and its partners have mobilized and involved themselves in carrying the operation PRADO REEFS successfully to term, the first project of this scope in France. The artificial reefs are an excellent tool for sustainable management of the coast and coastal activities. Spearheaded by the Plan de Gestion du Littoral de Marseille (Management Plan for the Marseille Coast), a program of Gestion Intégrée des Zones Côtières (Integrated Coastal Zone Management), this unifying project should open the way for other restoration zones to support small coastal fishing endeavors, restore irretrievably degraded zones, or make certain sites more accessible for scuba diving.

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Swimming Behavior of Juvenile Yellowfin Tuna *(Thunnus albacares)* Around Fish Aggragate Devices (F.A.D.S) in the Philippines

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Abstract Three juvenile yellowfin tuna (22–26 cm FL) were monitored around a payao (FAD: Fish Aggregating Device) in Panay Gulf, the Philippines. A receiver was attached to the anchor line of the payao, and ultrasonic transmitters were implanted into the abdominal cavities of the fish. Juvenile yellowfin tuna swam within a limited shallow range during nighttime and dived to deeper waters, maximum 105 m, during daytime, then moved away from the payao at midnight. One juvenile yellowfin tuna was recaptured 2 days after release by handline in the same payao, and two juvenile yellowfin tuna were recaptured 12 days after release by ringnet in another payao over 3 km away. These recaptures indicate that the fish were active enough to feed and swim, and also show the feasibility of undertaking telemetry studies on juvenile yellowfin tuna.

1 Introduction

Most telemetry studies on tuna behavior around FADs involve adult fish using ultrasonic transmitters and data loggers. These studies have already revealed relatively consistent patterns in the swimming behavior of adult tuna near FADs. However, despite recent advances in telemetry, no information is available on the swimming behavior of juveniles of large tuna species that normally associate with payaos. Fish aggregating devices (FADs), including payaos, play an important role in global tuna fisheries (Fonteneau et al. 1997; Dempster and Taquet 2004). In the Philippines, payaos were first introduced in the 1970s primarily to target tuna (Floyd 1986; Dickson and Natividad 1997). Aside from adult tuna, small pelagic species including juvenile tuna that aggregate near the payaos are also captured (Monteclaro 1997). However, little is known about the behavior of such small fish, and the efficiency of a payao has not been evaluated quantitatively. In this study, we present the results of a telemetry experiment on several juvenile yellowfin tuna *Thunnus albacares* conducted around a payao in the Philippines.

2 Materials and Methods

The experiments were conducted around a payao deployed approximately 10 km off the coast of Miagao in Panay Island, the Philippines. In Panay Gulf, several payaos were installed in the same network at a few kilometers distance, but the actual numbers and positions were not clarified. The depth of the water was approximately 500 m. A set receiver (VR2-DEL, Vemco Ltd., Canada) was installed on the anchor line of the payao at a depth of approximately 20 m by scuba diving on August 13, 2005. The receiver decodes the ID numbers and swimming depths of the fish implanted with transmitters within the detection zone and records the numbers and time stamp. A preliminary study was

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Table 1 Details of tagged fish

Fish ID	Fork Length (cm)	Release	End of record	Duration (h)	Transmitter
TY1	22	14:15 August 13, 2005	14:21 August 13, 2005	0	V7
TY2	22	14:18 August 13, 2005	23:39 August 17, 2005	129	V7
TY3	26	12:45 August 15, 2005	16:12 August 19, 2005	99	V9P

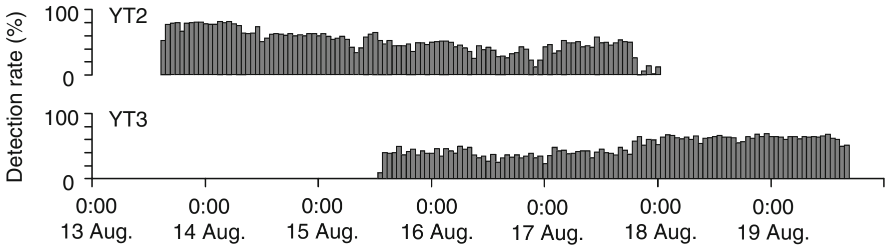

Fig. 1 Time series data of the hourly detection rates of YT2 and YT3

conducted to determine the detection zone of the receiver around the payao.

Experimental fish were captured around the payao by handline. Three yellowfin tuna (YT1–3, 22–26 cm FL) were captured from August 13–15, 2005. The details of each fish are given in Table 1. The fish were implanted with coded ultrasonic transmitters (V7-2L-R256 or V9P-2H-S256, Vemco Ltd., Canada). The V7 transmitter weighed 0.9 g in water, was 7 mm in diameter, and 20 mm long. This transmitter emitted a train of six pings for identification. The V9P transmitter, which has a pressure sensor, weighed 2.9 g in water, was 9 mm in diameter, and 46 mm long. The transmitter emitted a train of eight pings for identification and depth measurement. The surgical operations were performed just after catching the fish aboard a 10-m outrigger fishing boat and took only about 1 min for each fish. The fish were laid on their backs on a makeshift operating table. An incision of approximately 15 mm was made in the abdomen of the fish to allow the transmitter to be inserted. The wound was sutured with one stitch. The fish were released immediately after the operation. The receiver was retrieved at 16:12 on August 19, 2005, and the stored data were downloaded.

3 Results

The maximum detection distance of the receiver was approximately 700 and 500 m in radius for V9P and V7 transmitters, respectively. Figure 1 shows the time series data of the hourly detection rate of YT2 and YT3. YT2 was monitored for 129 h until the middle of the night on August 17, 2005. YT3 was monitored for 99 h until the end of the experiment. These fish were monitored continuously without interruption over 1 h until the end of each record. There was no difference in the hourly detection rate during daytime (from sunrise at 6:41 to sunset at 19:10) and nighttime (Mann–Whitney Test, $P > 0.05$).

Figure 2 shows the time series data of the swimming depths of YT3. YT3 dived occasionally over 50 m. The maximum swimming depth was 105 m at 18:59 on August 15, 2005. YT3 showed a diurnal vertical swimming pattern. YT3 frequently stayed in a relatively shallow and narrow layer between 5 and 15 m during nighttime. In the daytime, the fish swam in a deeper and wider layer between 10 and 25 m. YT3 was swimming in significantly deeper waters (19.5 ± 10.6 m) during the daytime when compared to that at nighttime (12.5 ± 8.9 m) (Welch's *t*-test, $P > 0.05$).

While fishing for more fish to tag, we recaptured YT2 2 days after release at 8:30 on August 16, 2005.

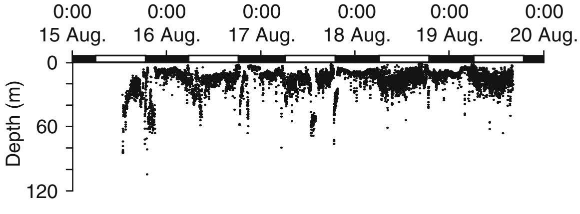

Fig. 2 Time series data of the swimming depths of YT3

This allowed us to examine the wound, and the fish was re-released immediately. Although the wound was not completely healed, it was fastened by the stitch tightly. Twelve days after release, a ringnet operating in another payao over 3 km away from the tagging site re-recaptured YT2 with YT1 on August 25, 2005. The transmitter of YT2 remained, but that of YT1 was missing. The fisherman reported that the wounds were healed.

4 Discussion

YT1 was monitored only briefly after release. When the fish was recaptured, the transmitter was missing; it had probably dropped out. The recapture of YT2 2 days after release indicates that the fish was not only alive, but also active enough to show its feeding behavior. This fact also suggests that the trauma of the operation was not serious. In addition, the re-recapture of YT2 and the recapture of YT1 indicate that the fish were active enough to swim to another payao over 3 km away. These results suggest that it is possible to conduct telemetry experiments involving juvenile yellowfin tuna.

Although the number of experimental animals was limited, the similarities between the observed swimming behaviors of payao-associated juvenile yellowfin tuna and FAD-associated adults of various tuna species are very striking. YT3 swam within a limited shallow range during nighttime and dived to deeper waters during daytime. Ohta and Kakuma (2005) reported that adult yellowfin tuna showed a similar diurnal vertical swimming pattern and dived to depths over 200 m. Dagorn et al. (2006) reported that adult yellowfin tuna stayed at depths over 1,000 m. Although the maximum swimming depth of juvenile yellowfin tuna was not as deep as that of the adults, YT3 was swimming in deep waters, maximum 105 m. The observed vertical movements during daytime were probably feeding excursions for small prey. In an echo survey near payaos also in Panay Gulf, separate clusters of small prey (5–10 cm BL) during daytime at depths between 20 and 30 m within a range of about 600 m from the payao raft were observed.

This depth range coincides with the swimming depth of YT3 during daytime.

As all the experimental fish were captured by handline and YT2 showed feeding behavior just 2 days after release around the payao, the presence of these prey organisms was an important factor in maintaining the aggregation of juvenile yellowfin tuna near these floating structures. All the fish were detected without interruption, and the hourly detection rate did not show any differences. This indicates that these fish were swimming within the detection radius of the receiver most of the time. Ohta and Kakuma (2005) revealed that adult yellowfin tuna also exhibited a similar pattern of association with anchored FADs.

YT2 remained around the payao for 129 h after tagging, but the record ended in the middle of the night on August 17, 2005. Departure from the tagging site at nighttime has been reported in adult yellowfin tuna (Holland et al. 1990). YT2 probably moved away from the payao in the horizontal plane. The reported recapture of both YT1 and YT2 provides direct evidence to support this horizontal movement. YT1 and YT2 were captured and released at almost the same time, then recaptured at the same time in another payao over 3 km away. Unfortunately, we did not know the hour of departure of YT1 because of the lack of the transmitter; these fish might be swimming together in the same school.

5 Conclusions

A telemetry experiment on juvenile yellowfin tuna Thunnus albacares was conducted around a payao in the Philippines. The similarities between the observed behaviors of payao-associated juvenile yellowfin tuna and FAD-associated adults are very striking. The results show the feasibility of undertaking telemetry studies on juvenile yellowfin tuna.

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Summary of French Artificial Reefs Immersions Since 1968, Sites, Volumes, Types and Costs

Sylvain Pioch and Jean-Pierre Doumenge

Abstract Since 1968 and the immersion of the first artificial reef next to the French Mediterranean shores in front of Palavas, only two studies have been initiated by Barnabé et al. (2002) on the Mediterranean scale, and another was directed by Lacroix et al. (2002) for the whole French coast.

The coastal area of Languedoc-Roussillon was a pioneer region for the immersion of artificial reefs (Doumenge 1968), but in terms of size, the largest one was submerged in front of Marseille (32,000 m^3). Nowadays, most of the reefs are located off the Mediterranean coast; new projects are being developed on the Atlantic side (e.g. Yeu Island or Cap Breton, etc.), the English Channel (Le Tréport), and the overseas' islands of Reunion and Martinique.

Thus, approximately 40 reef sets have been submerged in French marine waters; some of them are the result of several releases (e.g., Gruissan, Marine Park of the Blue Coast (Charbonnel et al. 2000)). These projects are most often created at the request of fishermen working close to the coasts. The development of the artificial reef was initially connected to "development and support for inshore fishing" (creation of the site in 1968 in Palavas, unpublished Lacroix D.). Currently in France, artificial reefs are linked to production and protection issues (Pary 2004; Claudet 2006).

This summary of the French experience in artificial reefs describes both the quantitative approach (sites, volume, design) and recent project costs (since 2000).

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1 French Fisheries Situation

France is one of the very few countries in Europe and the world with three major coastlines (5,500 km of coastline) and a unique maritime domain through the exercise of sovereignty over many islands in the Pacific, Atlantic, and Indian Oceans. It is the second World Exclusive Economic Zone, covering an area of 11 million km^2, or 20 times the surface of the continent (behind the United States; Japan is number sixth with 4.5 million km^2). With about 7,700 registered ships, the French fleet represents about 11% of tonnage and 15% of the power of the European fleet. The ships specializing in small-scale coastal fishing are less than 12 m long; they represent over 80% of the units comprising the French fishing fleet. However, more than half of the fleet has a vessel tonnage of over 24 meters.

France ensures approximately 10% of the fish catches in the European Union, which for the past several years has made France the fourth largest European Union country in terms of catch. In 2006, the total output of marine products amounted to 766,000 t, of which 570,000 t came from fishing (Ofimer 2007), resulting in a turnover of 1,800 million Euros, of which 1,266 million is for fishery products. In its Plan Ahead for Fishing, the Ministry of Agriculture and Fisheries said that "the fisheries sector is a key economic issue for the country" with nearly 8,000 vessels and 24,000 staff on board. However, the seafood sector occupies a

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marginal position in the French economy (0.14% of the domestic GDP).

In fact, fishing activity should be measured by more than the tonnage of the product because it also contributes to coastal management and development. With a rich history, artisanal fishing in France is imbued with cultural values and a collective identity, having a strong personality. The first submerged HAs (artificial habitats) were made in 1968 in the Mediterranean area in front of Palavas-les-Flots on the Languedoc coast. And from now, the goals are linked with two priorities:

1. Increasing fish production
2. Ensuring the protection of small coastal grounds against illegal trawling

Usually the immersed construction are made of prefabricated elements and have often been downgraded (waterpipes, electricity poles, stairs, old concrete elements) without prior reflection on their ecological integration (Pioch 2004). The aim of developers is political: to support the survival of coastal fishing. Considering the progressive thinking in the study of the target species (relationship with habitat), management objectives to be set up by the fishermen themselves, and hydrodynamic studies of the site and nearby reefs, these adjustments are favored by the main users and beneficiaries: coastal artisanal fishermen (Ducloy 2006).

Nowadays, the new projects are better integrated, and reefs are created specifically for the target species (e.g., "reefs baffles" designed by Charbonnel of PMCB realized in the framework of the "Prado" project at Marseilles). Drawing up a comprehensive map of sites and volumes is a complex process, as projects are not organized by a national organization. However, we tried to propose an overview of the location and major facilities in French artificial reefs according to the elements and bibliographic information available (Barnabé et al. 2000; Lacroix et al. 2002; Pary 2004; Charbonnel 2007). There is a predominance of complexes in the Mediterranean, but also a growing number of projects in the Atlantic. It seems that in the future, balancing projects in the Atlantic and Mediterranean will gradually take place because of communication among fishermen. The study of quantitative areas, sites, and volumes of HAs (excluding wrecks) immersed in France showed 30 complexes (on several groups of reefs) to make up an area of approximately 88,000 m^3 submerged between 1968 and 2007. Some complexes have had several immersions, always at the request of fishermen (e.g., Gruissan, 3 immersions; Agde/Marseillan, 3).

The average cost in Euros for the last six adjustments made in the Mediterranean between 1999 and 2007 of more than 900 m^3 (fisheries exploitation potential) is € 1,447 million for 7,150 m^3. But if we excluded the Marseilles area as non-standard, the average cost of projects is €436,080 for an average volume of 1,580 m^3.

2 Towards national land plan establishment and policy

Currently in France, there is no national policy concerning the immersion of artificial habitats or HAs (no longer "reefs," which is too general a term). The projects are often initiated by associations, unions, or mixed coastal communes. However, in 1985, the French government recognized the necessity to integrate HA into its coastline management scheme in collaboration with other member countries of the European Union. The Integrated Mediterranean Program, initiated in 1983, analyzes the French coast with scientific monitoring sites (provided by IFREMER in Languedoc).

– Meetings are regularly conducted between representatives of the state and those of the local authorities (PACA regional councils and LR)
– taking into account the views of professional groups (CRPMEM "Prud'homies" and local fishermen via small trades).

The project consisted of submerging 36,957 m^3 of HAs divided between the region of Languedoc-Roussillon (15,000 m^3) and Provence-Alpes-Cote d'Azur (21,932 m^3). The lack of exchange and complexity of fish data collected were the reasons for any monitoring abandonment. A major project was completed when the

AH site	Year(s)	Production and protection (m^3)	Future project (from end of 2007)
31 complexes (around 40 groups)	1968–2007	88,071 m^3 (40% for Marseille poject)	11 projects

"PRADO" was finished in 2008 in Marseilles. Between 2007 and 2008, it was planned to immerse about 32,000 m^3 of HA. With this project alone, the French coast HA volume was to be increased by one third.

The management and monitoring of this site in the fisheries logs will undoubtedly improve our objective knowledge about resource development and fisheries. More generally, given the work of S. Pioch, it seems essential to create a national approach for project organization for future development in French HAs. It must be based on standardized methodology development. This approach should help with the harmonization and evaluation of future developments in order to identify the advantages and disadvantages of integrated projects on geo-coastal systems.

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The Artificial Habitat, an Evolutionary Strategic Tool for Integrated Coastal Area Management

Sylvain Pioch, Jean-Claude Raynal, and Gérard Lasserre

Abstract The growing demand for marine fisheries resources (95 million tons in 2006, a number that should increase by 2–3% globally each year, UNESCO 2007) as well as the global outlook in terms of population growth (in 2050 the world population is expected to reach 8.9 billion persons, UNFPA 2004) represents a real threat to the worlds fish stocks. This threat deals particularly, in short and medium terms, with offshore or coastal fish stock (Worms et al. 2006; Cury 2008).

In order to address alimentary and socio-cultural food issues, to act on preserving and developing resources, the crisis manager generally focuses on two fundamental principles: (1) to act on preserving the species by managing fishing efforts and limiting catches (an example of management by the policy of "TAC" and "quotas" fishing); (2) to impact the environment by developing, for example, marine ranching and by expanding artificial coastal habitats with "artificial reefs." In light of their systematic developmental approach (FAO 1995), we prefer to use the term: "artificial habitats" (or AH). Based on the Japanese experience in the field of artificial habitats, and on a feasibility study in the town of Saint-Leu (Reunion Island), we will study the methodology and planning proposals for implementing an ICAM strategy that is essential to the success of the development of fishery facilities (Denis 2001; Henocque, Leçons et futur de la gestion intégrée des zones côtières dans le monde, VertigO – La revue en sciences de lenvironnement 2006).

Our contribution will be based on a geo-socio-systemic approach pertaining to artificial habitats in the coastal area. In order to reach this goal (to restore, maintain, or increase the fish level), it is imperative that HA implementation be considered as part of a concerted methodology using an eco-systemic and a socio-systemic approach for sustainable territory enhancement.

1 Introduction

Coastal areas are subject to multiple human-induced pressures that heavily affect submarine natural habitats and species (Denis et al. 2001). Indeed, almost two thirds of the worlds human population are concentrated on this narrow strip between the land and sea, with an occupancy level five times higher than the average density of inhabited land. By 2050, an estimated 75–80% of humanity will live near the coasts (Saunier and Laffitte 2007). The growing demand for marine fisheries resources (95 million tons landed in 2006, with expected annual increases of +2–3% globally, UNESCO 2007) augmented by the global outlook in terms of population growth (the world population is expected to reach 8.9 billion in 2050, UNFPA 2004) represents a real threat to fish stocks, both in the short

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and medium terms and both offshore or coastal fish stocks (Worm et al. 2006; Cury et al. 2008). The needs of populations, both for water and energy, will grow, thus increasing the production of pollution and waste, which also affect marine ecosystems.

Coastal areas are also threatened by the removal of marine resources by fishing. Nowadays, the sea remains the only source of protein for one quarter to one third of humanity. The outlook for the worlds fishery resources provided by the FAO is clear: "it is a fact that in the short and medium term, demand for fish and the pressure on stocks will increase as a result of population growth and the increase in incomes" (FAO 2004). But this human pressure on the marine environment will not be without consequences. According to FAO, the balance of world fisheries indicates that 77% of marine species that are of interest for fisheries are fully exploited, overexploited, or depleted (FAO 2004, 2006).

The steady increase in global demand for aquatic products is about 2% per year, which presages increasing pressure on the environment (Saunier and Laffitte 2007). For Worm, all species targeted by fishermen will disappear by 2048 if the current pressure on fish stocks and the marine environment are maintained, i.e., if no deliberate policy aimed at the recovery of stocks is created. Faced with these food and socio-cultural issues, to act to maintain and develop resources, managers generally base their decisions on two fundamental principles: (1) the influence of fishing pressure, managing fishing by limiting catches (an example of management by the policy of "TAC" and "quotas" fishing) and (2) the effects on the environment; increasing resources by extensive "mariculture," for example, enhances the number of juveniles, or popularizing management of the seabed of the coastal zone with artificial reefs (which we call "artificial habitats" in terms of their integration ecosystem, FAO 1995). Of course, in parallel to these measures acting on the environment and fishery resources (ecosystem), other actions such as regulations and technical operating fisheries across socio-maritime systems are heavily involved in practice. Fisheries are also fighting against IUU fishing and establishing control instruments, management plans, etc. (Commission Européenne 2007b). For effective coastal development that can restore, maintain, or even increase the level of fish populations, it is imperative that their implementation is part of a concerted approach to development goals balancing regional ecosystems and sociosystemic aspects.

2 The Geo-system: An Entry for the Integrated Management of Coastal Zone?

Human knowledge about marine exploitation has long been underpinned by the existence of the submarine "Eldorado." It is a vision now largely anachronistic, even though some developers continue more or less skillfully to use the exhibit. Learning to manage global maritime space, for example, by ground restoration of the coastal areas to develop their biodiversity and increase the productivity of fisheries, is part of a heritage, and therefore included in the duration of ocean space by coastal communities. It is important to remember when trying to respond positively to the needs for aquaculture products and also to the communities desires to have areas available for underwater recreation, artificial habitat development includes the issue of territory. The approach presented here is based on a geo-systemic approach, which is tantamount to a study of the relation between societies and nature (Scheibling 1994). This spatial unit, formed by more or less balanced elements of climate, soil, fauna, flora, and landscape, is what geographers used to describe the "natural environment" (Brunet et al. 1995). This definition is different from many ecologists approach because it takes into account human action, without necessarily seeing the human presence in an ecosystem as disruptive. "Nature" is being influenced by man, and therefore, we independently observe that nature changes and that companies that find places reduce the effectiveness of the analysis and scope of the proposed solutions (forgetting the development of society and therefore its acceptance).

Therefore, it is worth considering the functional and structural reality of a "geo-socio-system," i.e., the structuring the action (including no-structuring aspects) that an organized population exerts on the biophysical environment. For convenience, we will talk about the geo-system, knowing that it remains linked to human action in the various ecosystems. In our particular case, we are in a dynamic problematic stewardship of the coastal zone, knowing that the geo-system will find its place. It is based on marine space, land, and lagoon, supported by fishing, aquaculture, agriculture, urban development, industrial, and leisure activities. A geo-system also includes all past and present men in the middle (Veyret 2004). It is an essential element of foresight to know

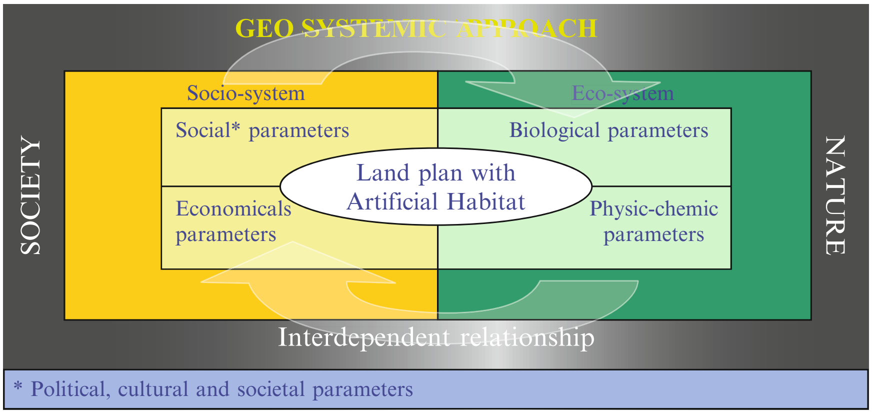

Fig. 1 Geo-systemic approach and the nature-society relation (Pioch 2008)

and control within the framework of public policies that is perfectly displayed (see diagram below).

When the geo-systemic approach focuses on coastal area management, it is a similar approach using a strategy called ICZ(or A)M, an acronym for Integrated Coastal Zone (or Area) Management (Cadoret 2006) (see Fig. 1). This is a dynamic process that brings together government and society, science and policy makers, and public and private interests in preparing and implementing a plan for protection and development of systems and therefore of coastal resources. This process is designed to maximize choice for long-term emphasis on resources and their reasoned and reasonable use (Cicin-Sain and Knecht 1998). The ICAM cannot replace sector planning, but it focuses on the relationship between the various sector activities to achieve broader objectives. This approach is based on nature, but also on society and its perceptions. The policies on maritime transport, fisheries, energy, and tourism have evolved separately, sometimes causing failures, inconsistencies, and conflicts in the use of resources and territories (Doumenge 2004; Cadoret 2006).

Coastal development is a way to reduce these conflicts by assigning rules to use a specific space. Finally, while ensuring the reconciliation of economic imperatives categorically, the overall development of communities and relative stability of ecosystems and levies by man of natural resources, including fish, is involved (Meur-Ferec 2007). In this sense, the European Parliament (Commission Européenne 2007a) promotes the implementation of ICZM strategies in Europe, which is considered in all the national legislation of EU countries (Interministerial Committee for Planning, in July 2001, September 2004).

3 The Japanese Management Experience with the Service of a Strategy Type ICAM

The Japanese experience is a successful implementation of ICZM facilitated by a "mono-actor" in regard to the use of resources. The system is very close to the actors (fishermen), and the politicians listening to the social demand for seafood have put in place tools designed by a scientific-technical sector structured in order to conserve and manage the resources and activities that depend on them. Japan is undoubtedly regarded as the world leader in the successful development of the seabed at both application (stability of coastal fisheries) and prospective (Bailly 1989; Baine 2001) levels. Extensive coastal mariculture began with the first artificial habitats made by the Japanese that are described as taking place between 1652 and 1655 (during the reign of Emperor Joo). They were made of stone to extend exploitable fishing zones near the village. The first studies on artificial habitats date from 1903 (Fukuoka Prefecture), and the most important tests began in 1952 with the first government planned "development of fisheries areas in deep water" (Simard 1985).

For Mariojouls (2004), "the originality of Japan is the integration of fisheries and aquaculture in an overall project management of the coastal strip to increase marine production. In 70 years, actions taken cover several areas: in addition to the development of the fisheries sector supported by a network of research and development organizations and a professional and highly efficient administration, operations are carried out large-scale release of juveniles of many species

(Note: 90 in 2006, NCSE1) to support fish stocks exploited, and arrangements of coastal areas, and the HA to increase capacity and productivity of fishing zones." According to Denis et al. (2001) and Bailly (1989a), this draft extensive "mariculture" (marine-ranching) is understood as "all actions aimed at increasing the production of plant or animal marine trophic species without input." Human action is limited in some cases, for example, the contribution of juveniles.

Thus, the national plan of reorganization and promotion of the sector is divided into three goals called "three pillars":

- Ensuring the supply of aquatic products.
- Facilitating the development of the fishing industry, and the quality and health safety of aquatic products.
- Driving advanced research and ensuring the development and monitoring of research and development (MAFF 2007).

This policy is based on a network of important players, such as the Fisheries Research Agency, university research centers, prefectural (departmental) deparments, and private enterprises. The Fisheries Research Agency, created in 2001, brings together the nine research institutes of the Fisheries of Japan. University research is very active. Japan currently has 34 research departments specializing in fishing or aquaculture distributed in different universities. The prefectural (departmental) centers are real interfaces between the administration, fishermen, and the general public. They are responsible for ecological monitoring of the marine environment and carry out search operations in connection with the research areas of national politics. Private companies are developing innovative models for coastal areas, and generally these companies have their own research units or act in collaboration with other actors in the network.

The management of fisheries resources in Japan constitutes a real social project. This sector is a priority, and the objective is a self-sufficiency rate of 65% by 2012. This objective is ambitious because we must consider that Japan, with a quota of 65.7/kg/year/capita, is the worlds largest importer of the product. Fishing cooperatives play a central role and have initiated projects, accompanied and guided at the national level by the Fisheries Agency (also by the Japanese Institute of Technology for Fishing Ports, Grounds, and Communities), which ensures coordination and development of coherent programs for the improvement of fishing grounds. The funding for this type of operation was significant for the period 2007–2008, when it was provided with 602 millions Euros (National Statistics of Japan Ministry of Internal Affairs 2008).

It is clear that the exclusive use of coastal areas by fishermen has enabled the effective implementation of this management, while elsewhere in the world, multi-stakeholder/multi-purpose issues hamper its development. In this context, the Japanese experience should enlighten us concerning the tools and solutions in place, but must be implemented within the other geo-systems (multi-stakeholders).

4 Artificial Habitat, an Ownership Tool for Coastal Zone Management: The Example of French Overseas Reunion Island

The case study presented in this chapter took place in the town of Saint-Leu (Reunion Island) where the demand for artificial habitats followed a space-sharing conflict. Indeed, to protect a coral reef rich in fish and with high ecological value, setting aside the areas coral heritage has been considered. This site is considered important by many, including by scientists, environmental associations, tourists, and fishermen, and the aims are contradictory, divided by considerations of the natural resources and protection of the environment and species. Fishing is a traditional activity carried out both by professionals and by boaters, and represents an important food.

The site is natural, and beyond its important local status (with limited economic significance, but traditional value) is a World Heritage supranational area [conservation of nature and corals on a world scale and in France the program "CRISP – French Development Agency" initiated in 2002 (CRISP 2008]. The activity of coastal fishermen to support their income and as a source of food has been confronted with the introduction of Marine Park Reunion (a legitimate conservation project of the World Heritage Foundations ecological program), and the town of Saint-Leu is trying to provide an answer. The electoral context (study initiated in June 2007, municipal elections in March 2008) shows that the local people, faced with the establishment of a reserve whose objectives are environmentally justified, see an imposition by the State and Europe, despite the important work of consultation and public debates prior to the project (Européenne Commission 2005). The conflict between institutional actors and stakeholders is arbitrated by the Saint-Leu municipality, which, posing as spokesperson for the fishermen, must also meet the requirements of the "Réunion Countie," the State, and Europe [political biodiversity conservation with the creation of marine protected areas up to 10% of French EEZ in 2012 (Commission Européenne 2006)]. It is in this context that the introduction of artificial habitats has emerged as a transactional tool to reestablish dialogue. The aim is to proposed a consensual solution to achieve common objectives (but expressed differently):

- Preserve the fauna and flora of the coastal zone by providing new nesting areas, food, and shelter for local species (Bolopion et al. 2000).
- Propose new areas for professional fishing and recreation for the town and for tourists.
- Increase futures fishery resources.

The creation of artificial habitats appears to be a complementary measure to the creation of the reserve and also contributes to creating habitats for economic targets species. Indeed, the sandy areas have the lowest organic marine production (Gabrié and Montaggioni 1985). The only exploitable resource in the sandy areas from 0 to 300 m is the "giraffe crab." The decline in resources, especially target species for recreational or professional fishing, is a general phenomenon resulting from exploitation and the lack of habitat necessary for the renewal of species that could settle there and reproduce (Tessier 2005). The creation of artificial habitats could help find a balance between creating new fishing areas and species management targets. For this, appropriate management measures should necessarily accompany the immersion of reefs.

These measures have been formalized by the establishment of a charter of use. Indeed, this charter emerged very early as a response and a desire on the part of individual users (the idea being a social consensus). It was also accepted in principle by the other actors and the state (including for its simplicity and authority that self underlies). These are the users who need to accept a "management contract" that involves the regulation and functioning of the reef area. Its implementation is based on creating a body bringing together a local committee and local fisherman. It is an additional guarantee to the ownership of a common area. The charter allows initiating a process of "privatization" of space through the access control system in place and thereby creating an awareness of "owner" beneficial to the management. This approach is obviously inspired by Japanese fisheries, but also the system "Prudhomies" Mediterranean holders use rights and territorial claims (Cazalet 2007). In addition to the recent establishment of the Marine Park and protection of coral areas (NMR Marine Nature Reserves), a network of artificial habitats created on the sand would provide habitat for species exported from the reserve. The "reserve effect" plays a positive role for fishermen. They understand the benefit to fish in an adjacent area created with AH from the MPA could enhance the installation of adults species. AH is an interesting complement to the MPAs fishing reserve in Reunion Marine Park. This project was rapidly appreciated because Reunion counties had already immersed AH that was interesting for fishermen. This first test of artificial habitats was carried out by Tessier in 2003 in the Bay of Saint Paul (northwest of the Reunion). Tessier (2005) shows that concentrations of fish densities are important in and around habitats, especially juveniles. Artificial Habitats are tools that can contribute to the protection and sustainable improvement of the management of coral reefs and species. They are a consensus medium for dialogue among users, environment managers (Marine Park), ICZM policy, and the State because they open a new relational field around one common objective: sustainable resources.

5 Conclusion

Artificial habitats are tools to protect and/or produce resources. They are intended to partially offset the significant decrease in certain marine resources as a result of overfishing. These artificial habitats would "catalyze" the emergence of manager conscience by appropriating an area "known" through a management unity bounded by a physical layout. Finally, they could induce reflection on exploitation management in order to sustain resources. They help to involve fishermen in decision-making by a geo-type systemic approach in a global strategy of ICAM. Indeed, such resource management as an ecosystem approach to fisheries (EAF; FAO 1995) often focuses solely on the functioning of submarine ecosystems. They do not take into account the socioeconomic (sociosystemic) dimension of fishing activities, which however are necessary for the sustainability and success of the proposed development.

Land plan tools, such as an AH project, can help managers and governments as "transactional" tools for balanced coastal development planning that includes the stakeholders (especially historic-cultural activities such as artisanal fisheries) to work toward sustainable resource exploitation in a real ICZM sharing strategy.

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Spatial and Temporal Variation of the Fish Assemblage on a Large Artificial Reef Assessed Using Multiple-Point Stationary Observations

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Abstract Over the past decade, the sizes of artificial reefs in Japan have increased, with heights reaching 30 m or more. However, there are few studies about the efficiency of the high-rise artificial reefs, and it is not clear how effectively these reefs gather and nurse fish resources. In these large artificial reefs, it is difficult for divers to view entire schools of fish. We developed a stationary observation system called FISCHOM (fish school monitoring system) to replace or complement visual censuses conducted by divers. FISCHOM consists of a stereo camera, a programmable timer, and a battery in a stainless steel pressure-resistant housing with transparent acrylic glass rated to a depth of 80 m. FISCHOM takes periodic underwater stereo images, and its maximum duration of operation is approximately 2 months when the photographing interval is set to 1 h. By using multiple FISCHOM systems around an artificial reef, researchers can obtain quantitative data on fauna and body size compositions of the reef fish assemblages and their trends. In this study, we will try to describe the possibilities offered by FISCHOM to study the artificial reef fish assemblages. Then, we will show the survey results conducted at a high-rise artificial reef in the Sea of Japan. We describe the advantages and limitations of FISCHOM for observing artificial reef fish assemblages based on the results of the survey.

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1 Introduction

In Japan, artificial reefs are installed mainly to develop fishery grounds for fishery promotions. In recent years, development has expanded from coastal waters to offshore regions. High-rise artificial reefs themselves have expanded three-dimensionally to develop fishery grounds at suitable depths for fishermen to use conveniently (Takagi et al., 2001). The biggest high-rise artificial reefs reach about 40 m in height and about 5,000 m^3 in volume. Introduction of an artificial reef is generally performed as a public work, and much tax revenue is injected into such projects. Therefore, strict evaluation of the effects of artificial reefs is required. Previously, reef fish assemblages were assessed by means of visual censuses made by scientific divers, fishing, and acoustic surveys with echo sounders. All these methods have advantages and disadvantages. For example, the visual census by scientific divers who have technical knowledge and experience is suitable for roughly determining fish fauna, fish school volume, and fish body size as well as some qualitative features such as fish behavior. However, it is difficult to obtain exact quantitative data by visual census. Moreover, the time that divers can remain submersed is limited from several minutes to several hours. Submersible time becomes a more critical factor when divers must assess fish assemblages on high-rise artificial reefs installed at larger depth. Although the fishing method can enable one to determine fish fauna and fish body size directly from samples, this information is affected by the selectivity of the fishing gear. Moreover, the fishing method is destructive and is difficult to sustain over several days. The acoustic method can be used to quickly detect the volume of the fish assemblage

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around an artificial reef. However, it is difficult to determine fish fauna (fish species composition) and fish body size, especially when two or more fish species are intermingled. Furthermore, no information can be obtained near or inside of the artificial reef because the reef itself is a strong acoustic shelter. It is also unrealistic to prolong an acoustic survey over several days.

Here we propose a new technique to enhance the assessment of fish assemblages in artificial reefs and to address many of the problems of conventional survey techniques. The technique involves multiple-point stationary observation using equipment that can photograph continuously for a long period of time. These photographs enable researchers to accurately describe how many fish of various species exist, and the stereo photography function enables the estimation of fish body size. By using two or more pieces of equipment simultaneously, researchers can adapt the system to a large artificial reef, and it becomes possible to describe the time series of the fish fauna and body size composition according to each part of the reef. In developing this technique, we created the fish school monitoring system FISCHOM (Takahashi et al. 2005) and verified its performance in several investigations. In this paper, we report the advantages and disadvantages of FISCHOM compared to conventional survey techniques.

2 FISCHOM

FISCHOM is an abbreviation for *fish school monitoring system*. FISCHOM consists of a digital still camera, a flashlight, a programmable timer unit, a battery, DC/DC converters, and a pressure-resistant housing. There are specifications for two types of cameras: a monaural camera and a stereo camera. The visual field of the stereo camera is divided in two by four optical mirrors as proposed by Gluckman and Nayar (2001). The camera and the flashlight can take photographs at an arbitrary time interval as controlled by the programmable timer unit. Although the main part of the pressure housing is stainless steel, the front side is transparent acrylic glass for observation purposes. We minimized the cost of constructing FISCHOM by using inexpensive commercial items for all components except the pressure housing. We constructed four stereo camera FISCHOMs and three monaural camera FISCHOMs. Table 1 and Fig. 1 show FISCHOM specifications and appearance, respectively.

Table 1 Specifications of FISCHOM

		Stereo camera type	Monaural camera type
	camera	EOS Kiss digital (Canon Inc.)	EOS Kiss digital N(Canon Inc.)
	light source	Speedlight 420EX (Canon Inc.)	Speedlight 430EX (Canon Inc.)
camera	resolution (dots)	3,096 × 2048	3,096 × 2048
	baseline length (cm)	18	–
	conv. angle (deg.)	0	–
	FOV (horizontal)	15	30
	FOV (vertical)	22	22
timer unit		LOGO! 12/24RC (Siemens AG)	
DC/DC converter		EDC-2000	
battery		Portalac PE12V17 (12/17Ah) (GS Yuasa Powersupply Ltd.) Lithium battery (12V/60Ah) (Kaiyo Denshi Co., Ltd.)	
	dimension (cm) (diameter × depth)	43.5 X 31.0	39.5 × 31.0
pressure housing	weight (kg) (in air/ in water)	46/12	35/8
	material	Stainless steel/acrylic glass	
	available water depth	80 m	

Fig. 1 Appearance of FISCHOM stereo camera type

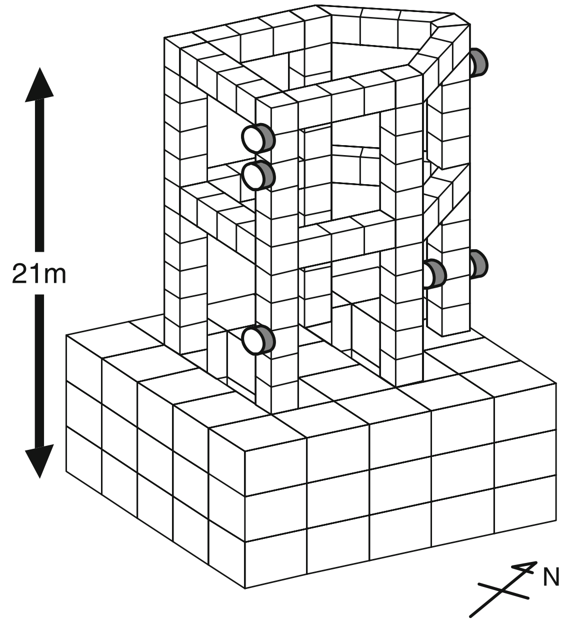


Fig. 2 Schematic of surveyed reef and installed positions of FISCHOMs

3 Test Surveys of a High-Rise Artificial Reef

To verify the performance of FISCHOM, we conducted several multiple-point stationary observations of the fish assemblages of an artificial reef. The candidate for the test survey was a large artificial reef installed off the southern shore of Sado Island at a depth of 45 m (Fig. 2). The height of the reef is 21 m, and the structure features a large vacant space in the center. The survey periods used in this research are shown in Table 2. Five FISCHOMs were installed for every survey: four on the outside of the reef (two on the top and middle of the south side, and two on the top and middle of the north side) and one on the inside to observe the vacant space in the center. The programmable timers of all FISCHOMs were set to take photographs at a constant interval of 1 h. All FISCHOMs operated normally in every survey. The survey periods were about 20, 30, and 60 days for the first, second, and third surveys, respectively.

4 Data Analysis

FISCHOM takes photographs of the space immediately in front of the camera at arbitrary intervals (see Fig. 3 for a sample photograph). If a fish is photographed, the fish fauna at the time of photographing is estimated by counting the number of fish visible for every species. The time series of the fish fauna is obtained by counting all fish in every photograph. When two or more FISCHOMs are installed simultaneously on an artificial reef, researchers can determine the difference in the space usage of every species. In other words, researchers can estimate the spatiotemporal variations of reef fish assemblages.

Table 2 Profiles of three surveys

	Survey period	Deployed FISCHOMs	Interval of photoguraph
1st survey	July 1–21, 2005	5	1 shot/hour
2nd survey	Sep. 27–Nov. 1, 2005	5	1 shot/hour
3rd survey	May 25–July 25, 2006	5	1 shot/hour

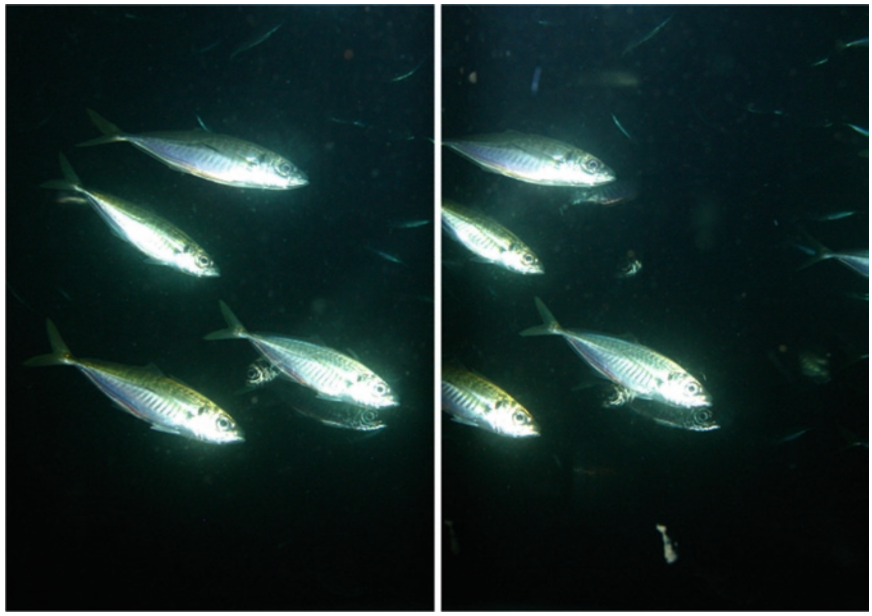


Fig. 3 Example stereo image photographed by FISCHOM stereo type

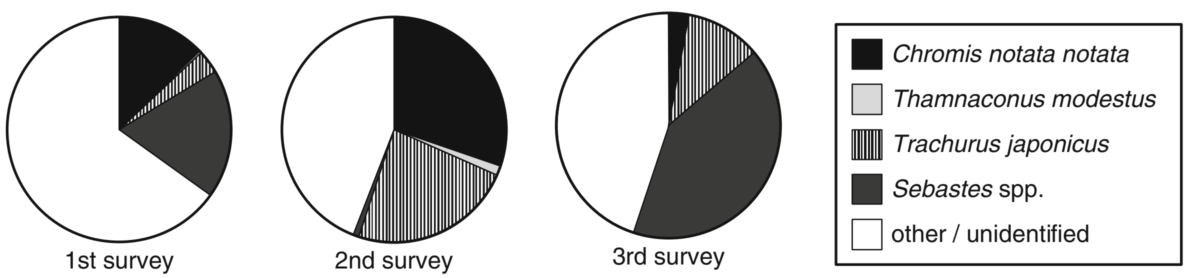


Fig. 4 Fish species compositions in three surveys

It is also possible to estimate fish body size using stereo measurement. The stereo measurement is the method of measuring the three-dimensional position of an object by the principle of triangulation from two pictures with azimuth difference. The fish body size can be estimated by measuring the distance between the three-dimensional positions of the head and the tail of a fish photographed appropriately by a stereo camera. The estimation of fish body size is carried out when the whole and straight fish body is clearly photographed in both two visual fields of the stereo camera.

Therefore, information on the spatiotemporal variation of reef fish assemblages is accompanied by data on fish body size.

5 Results

5.1 Fish Fauna (Fish Species Compositions)

Figure 4 shows the fish fauna estimated by calculating the number of all fish in all photographs taken in each survey. Three species – pearl-spot chromis (*Chromis notata notata*), Japanese jack mackerel (*Trachurus japonicus*), and rockfish (mostly *Sebastes thompsoni*) – were dominant in all surveys. Black scraper (*Thamnaconus modestus*), Temminck's surfperch (*Ditrema temmincki temmincki*), striped beakperch (*Oplegnathus fasciatus*), and others were observed occasionally. The proportions of the three dominant species changed for each survey. Rockfish was most prevalent in the surveys conducted in early summer. In the second survey, conducted in autumn, the ratio of pearl-spot chromis and Japanese jack mackerel to rockfish increased.

5.2 Time Series and Diurnal Cycle of Fish Appearance

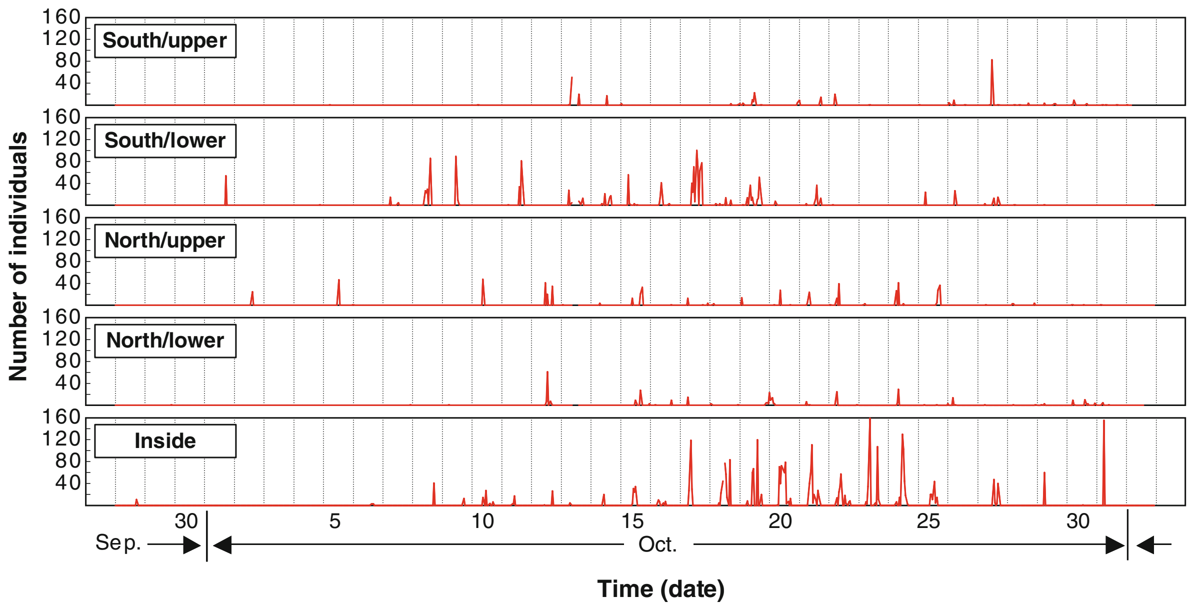
Figure 5 shows the time series of the occurrence of Japanese jack mackerel in the first survey. This graph shows when, where, and how many Japanese jack mackerel were observed. Many Japanese jack mackerels appeared on the south/middle layer and inside the reef. Japanese jack mackerel had a diurnal cycle, because they appeared only in the daytime

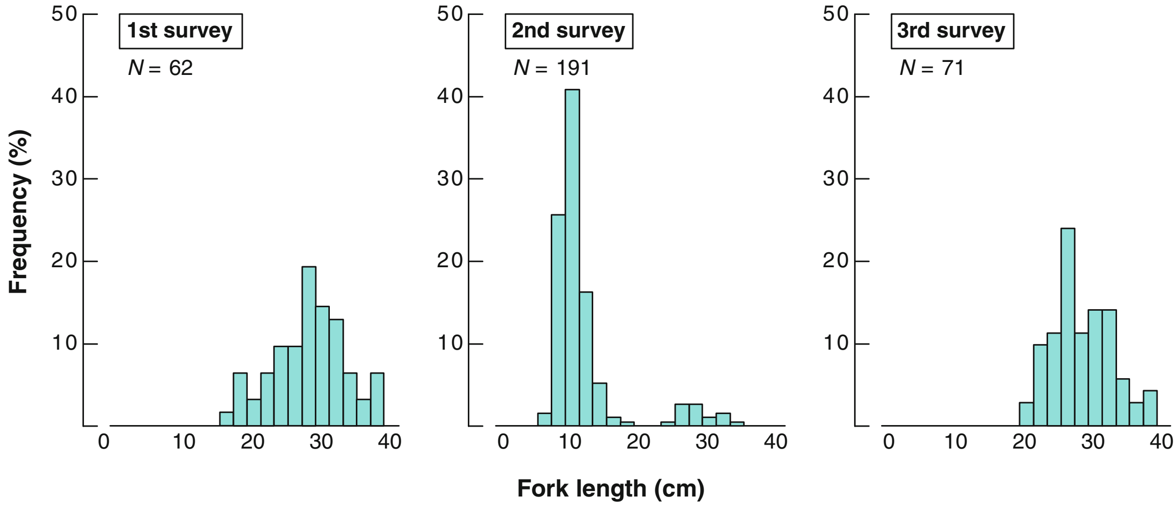
5.3 Fish Body Size

Fish body length (fork length or total length) was estimated using stereo measurement. The three-dimensional position of the snout and fork (or tail) was also estimated using stereo measurement, and fork length (or total length) was obtained by calculating the distance between these two points. Figure 6 shows the frequency distribution of fork length of the Japanese jack mackerel as estimated by stereo measurement. Juvenile fish of around 10 cm in fork length schooled on the reef in autumn, and large fish of around 30 cm in fork length schooled in the early summer.

6 Discussion

We have shown that the features of the fish assemblages on an artificial reef were quantitatively and precisely estimated by means of multiple-point stationary observation using FISCHOMs. Although we did not discuss fish density here, it can also be estimated if one defines the sampling volume (i.e., the size of the photography range) of FISCHOM. The sampling volume must be treated carefully because it can change according to water transparency and light conditions. One limitation of FISCHOM is that much time and effort are needed for image analysis. A well-trained expert must spend considerable time sorting, counting, and performing stereo measurements of fish. The time and effort for analysis will be reduced in the future by the application of the latest image-analysis techniques.





In this paper, fish assemblages on a high-rise artificial reef were the main targets of survey by FISCHOM, as it would have been difficult to assess them by visual census. FISCHOM is so compact and flexible that it is useful for long-term, on-site observation.

7 Conclusion

We developed the stationary observation system FISCHOM, which stays at an artificial fish reef for a long period of time and performs interval photography. By analyzing the pictures of fish school photographed by FISCHOM, it is possible to estimate the fish fauna (fish species composition) and the fish body size information of the reef fish assemblage. When two or more FISCHOMs are installed simultaneously on an artificial reef, researchers can determine the difference in the space usage among fish species. FISCHOM is effective especially for the assessment of high-rise artificial fish reefs in which the submersible time of a diver is restricted.

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Artificial Reefs: Perceptions and Impact on the Marine Environment

Gérard Veron

Abstract Artificial reefs have a positive image in the media and the public. However, if they soon appeared as a convenient response to fisheries depletion, there has since been a shift between this ideal perception and a real understanding of their functioning and impact.

The assessment of their actual performance is constrained by a knowledge deficit about ecosystems' natural variability, the definition of appropriate spatial and temporal scale studies, and the difficulty and cost of field surveys. In such a context, a number of questions can be raised, such as: (1) Is there an increase or redistribution of biological resources? (2) Are habitats a limiting factor for resources under heavy exploitation or limited recruitment? (3) Is ecosystem engineering enough and adapted as a response to resources degradation?, (4) What kind of control may be applied in regard to the system's possible modification?

In France, artificial reefs are mainly used for the purpose and benefit of coastal fisheries. In this context, multiple experiences may become risky in a free access system and not lead to a significant reduction in marine pollution and degradation sources. In addition, in such conditions, technological improvements could become useless.

Yet the demand is increasing and becoming more diversified. New perspectives are opening with the awareness of better control of uses at sea and the convergence of research-related issues concerning biodiversity, ecological engineering, marine protected areas, and marine areas specifically devoted to renewable energy devices. Besides, the technological improvement of monitoring tools supported by better experimental and follow-up procedures should help to achieve more significant and effective results for the benefit of coastal zone management.

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1 Introduction

In France, where the public and media have a positive image of artificial reefs, we must admit that this perception has been based on very imperfect knowledge about their functioning and a partial assessment of their direct or indirect effects on ecosystems. The reefs are sometimes presented as an obvious response to the loss of ecosystems, but the demonstration of the effects is often partial, limited to a few species and/or restricted to the immediate environment of submerged structures. The performance evaluation of these coastal developments remains constrained by the lack of knowledge of the natural variability of ecosystems and the difficulty in defining relevant spatio-temporal scales to select the reference stations not impacted. The cost and complexity of implementing the tools and means of investigation needed for the validation and qualification of additional biological and economic values generated by these devices comprise another factor explaining this situation.

2 An Inescapable Management

In France as in Europe, artificial reefs are often presented as a possible response to the loss of biological resources in the coastal zone. They aim to increase the

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productivity of the environment, usually to support or to restore coastal small-scale fishing. Probably there is a paradox to seek to correct a situation partially resulting from a lack of management with a reef deployment that requires more fishing regulations. It seems indeed that, without a coherent strategy management, reefs cannot be "the" solution, and they are even likely to induce unanticipated impacts or adverse effects on the biological resource and their uses.

Free access to groups of actors motivated by divergent goals can be a handicap to the demonstration and evaluation of the effectiveness of these devices. The control of fishing activities associated with the artificial reef is vital to assess the biological added value expected and to align catch capacity to capture surplus resources produced and control their redistribution. Without this strategy, the increase in fishing pressure usually responds to improvements in performances in the newly equipped zone and leads to a dissipation of economic benefits. Beyond the controversial question of increasing or spatial redistribution of the marine resource, which remains a crucial issue, especially when we confer to the reef an extractive vocation, we must ensure, by increasing accessibility of the fish, that the reef does not become an additional fishing gear for fleets often characterized by an overcapacity of capture.

It should also be recalled that the submerged structures are located in an open system, subject to the influence of fishing and environmental conditions. Therefore, these conditions must be included in the evaluation process. It is therefore necessary to adopt a coherent management strategy for the adjacent area and around the deployment to ensure that we measure the direct impact of reefs, the natural fluctuations of the resource, or, indirectly, the effect of development through the impact of restrictions on uses or modification of the pre-existing fishing techniques.

3 A Lack of Data and Indicators

It is very difficult, in the case of fisheries, to collect the necessary data to establish a reference state. In addition, when they are available, the production data are difficult to exploit given the spatio-temporal mesh of statistical collection, not well adapted to the small scale of artificial reefs studies. This means that the relevance of statistical data provided tends to be low. In the French Mediterranean coastal area, the zone most equipped with artificial reefs, the information on catches is notoriously inadequate in regard to the quantities produced and the precise location of fishing sites. Although the professional fisherman is required to provide regular statistical information, the number of vessels is too often the only available indicator for the manager. For example, in the Languedoc Roussillon region, after 40 years of artificial reef deployments, there has been, for vessels of less than 12 m, a decrease of almost 30% of this index over the last 10 years.

The lack of standardization of the monitoring does not allow scientists to compare results between different sites of immersion and limits the sharing of issues information learned from different experiments. This is a handicap for the definition of thresholds of natural, seasonal, and inter-annual fluctuations, for constructing impact hypotheses, and for verifying the predictions. Moreover, in the absence of results, when the effect cannot be revealed, the temptation is to increase the size of the first realization to reach a critical value over which the impact becomes noticeable. In a way, this is to ignore the assumption is that, rather than a non-measurable effect, there is no effect at all to expect from a non-functional development, not adapted to the host environment or inadequately managed. Under these conditions, the development of reefs contributes to increase the risk of unwanted or adverse effects, an inherent phenomenon in the artificialization of the natural environment.

4 A Response to a Limited Habitat and to Marine Biocenosis Devaluation?

The habitat can be considered as a restrictive factor in some cases of highly degraded coastal sites, thus justifying the establishment of restoration or compensation policies. However, the question probably does not arise in these terms in the general context of an intense and sustained exploitation of fishery resources and limited larvae and juvenile recruitment. In other words, are artificial reefs a sufficient response to the decrease in biomass observed for several years?

The artificial reefs are sometimes presented as a tool of ecological restoration, including protection and regeneration of habitats. We probably need to understand the meaning of restoration as an improvement or a return to a satisfactory situation because the return to the initial state is most likely an unattainable goal. The search for alternative or complementary solutions to reduce the degradation of coastal ecosystems is essential if we consider that artificial reefs have little impact on the major causes of loss or degradation of ecological communities,[1] which are the various sources of pollution, eutrophication, introduction of alien species, etc. However, artificial reefs are likely to interfere in partial but beneficial ways on commercial and recreational fishing, because they remove a limited area from fishing pressure or because they modify and improve, more or less intensely, pre-existing practices and uses.

5 Artificial Reefs and Bioethics

The development of the seabed by artificial structures as a tool of restoring a natural environment is sometimes presented as a suitable ecological response to the degradation of the marine environment, but also as the impossible conciliation of two principles. In other words, is the question of transformation of natural ecosystems by artificial reefs legitimated or justified, or does it reflect a true issue of environmental ethics?

The development of artificial reefs is usually in the immersion of hard material on a flexible substrate, often wrongly considered as the poorest. The positive or negative impact on biodiversity is indisputable, and therefore the amplitude depends in particular on the volume of submerged structures. Although analysis is not provided, the question of controlling the effects must be addressed at the earliest stages of the project to ensure the harmlessness of the project. Furthermore, we can wonder about the durability of the very positive perception of these artificial deployments when, at the same time, Europe is developing a political incentive to encourage the initiatives of nature conservation.

6 Multifunction Reefs

Developers have a certain propensity to combine all the beneficial properties attributed to artificial reefs in the same device. Yet some of these functions can be contradictory or antagonistic, and therefore are difficult to place on the same site. For example, the protection of juveniles is a recurrent goal for artificial reefs that aims primarily to reduce predation on the younger stages. In that case, accurate knowledge of ecosystem functioning and host interactions between species involved in the reef is a prerequisite to ensure that the shelter given to certain species from predation is not also a trophic opportunity for other species.

The increasing the efficiency of artificial reefs is often created by the complexity and specification of the form of new habitats, both in the general configuration of their organization (the occupation of the water column, creating a corridor between the different groups of reefs, ecological niches, etc.) and through the design modules (habitats more complex by the addition of materials and shapes, resting places for different species or animals of different sizes, etc.). The higher level of sophistication of the habitat is motivated by the idea of dependence of the target species in relation to the structure that provides food and shelter against predation. This option is not free of uncertainty and contradictions when it is manifested by an increased residence time of fishes when we consider that the productivity of a reef is largely based on its potential for export.

7 Multi-purpose Reef

Sometimes it is proposed to shape the dominant scheme of the artificial reef design exclusively to support the coastal fishery to move towards a multi-purpose development of these facilities (recreational fishing, tourism submarines, awareness, replacing habitats, etc.). It should be recalled here that there are few examples of coexistence of diverse activities, carried out simultaneously on the same coastal area. Moreover, when these practices are compatible, the diversity of their objectives induces increased management at sites where they are practiced, and their multiple impacts do not facilitate the study of underlying mechanisms. In fact, if the multi-purpose concept is often mentioned, particularly during the early stages of projects, it appears that whether from deliberate choice or from use, the final purpose of these deployments of artificial reef facilities is generally unique.

[1]EU Marine Strategy Framework Directive (2008/56/EC)

8 Towards New Perspectives

For several decades artificial reef projects have been developed in France, mainly on the Mediterranean coast, without a coherent framework to deal with the diversity of objectives, materials used to construct submerged structures, and heterogeneity of the monitoring protocols. Despite the lack of a conclusive demonstration of the expected effects, this trend has persisted because of the apparent relevance of the proposed solution, the strong political motivation to support small-scale fisheries, the efforts of fishermen's communities to initiative these projects, and the multiplication effect of subsidies obtained at the local, regional, and European levels.

Several elements can be advanced today to describe the current evolution of the perception of artificial reefs and the expansion of their potential applications. There is now a consensus on the essential prerequisites that must first be satisfied before implementing artificial reefs, in particular concerning the diagnosis of the situation to be corrected and the analysis of alternative solutions. In case of enforcement, awareness of necessary management and control of the concerned uses and users are also an admitted point. Regarding the monitoring procedures, the new regulations require conducting an assessment over a period of at least 5 years after immersion. They are becoming more rigorous and enriched by the sharing of information learned from various science programs conducted in France and abroad.

Furthermore, the demand is growing and diversifying. New prospects are opening up with the convergence of some research issues around biodiversity, including the need to assess the effectiveness of the establishment of marine protected areas or ecological engineering operations to compensate for environmental degradations of some coastal zones caused by the development of industrial or harbor complexes.

The artificial reefs are also presented as a possible means of mitigation to facilitate the public acceptability of the decision to reserve some coastal areas for the development of renewable marine resources. This broadening of the range of objectives is accompanied by a change of context, as in the above examples, public authority and private developers are encouraged to demonstrate the positive impact of their achievements on the ecosystems and to justify of their validity. This turning point coincides with technological advances of the measurement tools and improvement of methodologies for monitoring and assessment.

9 Conclusion

The goals and challenges for the coming years should lead to a break with the piecemeal approach to find, at least at the regional level, greater coherence between the objectives of projects. The improvement and the standardization of assessment protocols and scientific monitoring should increase the reliability of the analysis of the impacts of artificial reefs. It is also essential to encourage and assist the fishermen in the necessary changes in fishing practices and management arrangements associated with these facilities. In addition, it is very important to integrate the demand for diversification of potential applications of artificial reefs to the different ongoing or foreseen activities in the fields of recreation, sports, tourism, culture, and offshore industrial devices. Finally, we must encourage the sharing of data and outcomes obtained from various experience sites to promote the use of artificial reefs as real tools for maritime spatial planning.

Biodiversity

Characterization of Three Populations of *Phallocryptus Spinosa (Branchiopoda, Crustacea)* from North-East of Algeria

Mounia Amarouayache and Farid Derbal

Abstract *Phallocryptus spinosa (Thamnocephalidae)*, which was long considered as *Branchinella spinosa*, lives in saline lakes of America, Africa, and occidental Asia. There are very few data on its biology and ecology. It has been recently found in the Eastern Hauts Plateaux of Algeria and co-occurs with *Artemia salina* in Sebkha Ez-Zemoul. Three populations have been studied focusing on their biometrics and reproduction. During the sampling period in April 2005, only adults (more than 90%) and some pre-adults were found. Adults are short (16.14 ± 1.1–17.84 ± 2.65 mm) compared to Mediterranean populations. No differences were found between the total lengths of males and females. The latter are oviparous and begin to reproduce when they reach the length of at least 11.5 mm. They are not very fertile (15.6 ± 8.45–27.65 ± 14.49 cyst/brood) and produce cysts of a mean diameter range of 276.28 ± 18.79 to 292.18 ± 21.81 µm, according to the studied population.

1 Introduction

Phallocryptus spinosa is an Anostracan Branchiopoda that lives in Palaearctic saline water bodies. It was long considered to be *Branchinella pinosa* Milne-Edwards 1840. Rogers (2006) gave a new description of the species and classified it as *Phallocryptus* Biraben 1951. Studies on this species are very scarce (Ketmaier et al. 2008). *P. spinosa* is known in Algeria in five biotopes (Samraoui et al. 2006). It co-occurs with *Artemia salina* in the Sebkha Ez-Zemoul hypersaline lake (35°53 N, 6°30 E), where relatively large (40 mm) dead individuals were sampled in January 2003. This study is a first characterization of three populations in the northeastern part of Algeria. Data on biometrics and reproduction are presented.

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2 Materials and Methods

The study sites are the temporary lake complex located in the Eastern Hauts Plateaux (1,000 m in altitude), Ank Djemel (35°46′N–6°52′E), El-Tarf (35°42′N–7°08′E), and Guellif (35°47′N–7°E). Their salinity is around 10 g L^{-1}. Individuals of *P. spinosa* were sampled in May 2005, using a plankton net of 125-µm mesh size. They were fixed in 4% formaldehyde.

For each population, 20 males and 20 females were measured using a micrometer fixed on a microscope. One numeric (setae number) and 12 metric parameters were considered according to the method of Amat (1980 modified).

The data for the three populations were compared by one-way ANOVA test. A least significant differences test (LSD) was applied in cases where differences existed among populations.

The length at first sexual maturity was determined when 50% of females presented signs of reproduction. Ovisacs were dissected, and the cysts were counted to determine fecundity. Their mean diameter was measured.

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3 Results

In the three localities, *P. spinosa* adults dominated at more than 95% and pre-adults represented the rest, while young stages were totally absent.

The sex ratio was 1.02 for Guellif and higher for males in the other localities (2.09–3.46). There was no difference between total length of males and females (0.18 ≤ Student's t ≤ 1.07, α = 0.05). This length was between 16.14 and 17.84 mm. Table 1 shows the values of the different parameters and the LSD test.

In general, the three populations presented significantly equal values for the different parameters. The length of the furca seemed to have the most variant values.

The most common were the width of the ovisac, the eye diameter, the distance between the eyes, the length, and the width of the important abdominal spine. The setae number ranged between 98.5 and 125.2. There was a significant difference between El-Tarf and Ank Djemel.

The length at first sexual maturity was respectively estimated at 11.5, 13, and 14 mm for Ank Djemel, El Tarf, and Guellif, respectively.

Females are oviparous and carry 27.65 ± 14.49, 15.6 ± 8.45, and 23 ± 15.42 cysts/female in their ovisac, respectively. Their diameters are significantly different (12.40 ≤ Student's t ≤ 13.94, α = 0.05) at 282.84 ± 20.56 µm (n = 208), 292.18 ± 21.81 µm (n = 189), and 276.29 ± 18.79 µm (n = 61), respectively.

4 Discussion

P. spinosa adults dominated in the three sites, whereas young stages were totally absent in May. It was certainly the period of the last generation, as was the case in the saline areas of southwest of Sardinia (Mura 1993) and in the south of France (Thiéry and Puente 2002).

Adults are small in length. Alonso (1996) brought back a maximal length of 40 mm in the populations of the Spanish Iberian Peninsula compared to 30 mm in the Camargue (Defaye et al. 1998). Mura (1993) found lengths between 6 and 11 mm in the Sardinian population.

Concerning biometrics, the three populations seem to be close. Ketmaier et al. (2008) recently studied the phylogeography of the Palaearctic species and found no differences among African populations. Females reproduce starting from 11.5 mm. They are not very fertile compared to the Spanish populations, which produce thousands of offspring (Alonso 1996).

Cyst diameter of the studied populations is larger than in those of the Camargue (220–265 µm) (Defaye et al. 1998) and smaller than in the Spanish populations (300 µm) (Alonso 1996).

5 Conclusions

This work gives new data on the biology and ecology of the Branchiopod *Phallocryptus spinosa*. The biometrics and reproduction of three populations have been studied.

Table 1 Means (±S.D.) of various morphometric parameters of males and females *Phallocryptus spinosa* successively, for Guellif, El-Tarf, and Ank Djemel populations

Parameters	Guellif	El-Tarf	Ank Djemel	Parameters	Guellif	El-Tarf	Ank Djemel
A	16.35a (±1.45)	16.26a (±0.92)	16.92a (±2.67)	G	0.99a (±0.12)	0.97a (±0.22)	1.18a (±0.49)
	16.25a (±1.64)	16.14a (±1.10)	17.84b (±2.65)		0.95a (±0.15)	0.86a (±0.14)	1.17b (±0.24)
B	8.87a (±0.78)	9.31b (±0.52)	9.35ab (±1.49)	H	1.11a (±0.33)	0.90b (±0.09)	1.04a (±0.18)
	9.63a (±1.23)	9.84a (±0.94)	10.85c (±1.83)		1.09a (±0.14)	1.07a (±0.07)	1.09a (±0.19)
C	1.26a (±0.18)	1.16a (±0.15)	1.45b (±0.29)	I	2.35a (±0.44)	2.31a (±0.17)	2.45a (±0.34)
	1.06a (±0.27)	0.88b (±0.13)	1.28c (±0.31)		2.07a (±0.32)	2.05a (±0.19)	2.17a (±0.34)
D	1.80a (±0.17)	1.63b (±0.22)	1.60b (±0.35)	J	0.65a (±0.09)	0.65a (±0.08)	0.64a (±0.11)
	0.94ab (±0.22)	0.86a (±0.07)	0.97b (±0.20)		0.50a (±0.05)	0.48a (±0.06)	0.50a (±0.10)
E	3.82a (±0.34)	3.83ab (±0.37)	4.13b (±0.57)	K	0.67a (±0.19)	0.72a (±0.17)	0.63a (±0.15)
F	1.31a (±0.32)	1.16a (±0.20)	1.39a (±0.46)	L	0.41a (±0.13)	0.43a (±0.11)	0.42a (±0.10)

A, total length; B, abdominal length; C, furca length; D, length of first antenna; E, length of ovisac; F, width of ovisac; G, width of the third abdominal segment; H, width of head; I, distance between complex eyes; J, maximal diameter of complex eye; K, length of the big abdominal spine; L, width of the big abdominal spine

Significant differences were determined by ANOVA and LSD test ($P < 0.05$). Values in each line that share the same letter are not significantly different

This preliminary study must be completed by other investigations checking cyst hatching quality and population dynamics. Such data will allow us to know whether Algerian *P. spinosa* can be used for aquaculture aims or not, and it should be protected for ecological reasons.

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Biological Invasion: The Thau Lagoon, a Japanese Biological Island in the Mediterranean Sea

Charles-François Boudouresque, Judith Klein, Sandrine Ruitton, and Marc Verlaque

Abstract Shellfish aquaculture represents one of the major vectors of marine macrophyte introductions. In the 1970s through at least the 1990s, massive imports of Japanese oysters, *Crassostrea gigas*, from the Pacific (mainly from Japan) to Europe were carried out. As a result, a large number of exotic species have been introduced to European shores. In the Mediterranean Sea, the Thau Lagoon (south-western France) has become a hot-spot of introduction of marine macrophytes. A total of 58 species of macrophytes have been labeled as introduced. They currently represent 32% of the species diversity and 48–99% of the macrophyte biomass on hard substrates. Most of them are native to the north-western Pacific, including Japan. These figures do not take into account cryptogenic and cryptic introduced species and could therefore prove to be underestimates. The Thau Lagoon could therefore be the harbinger of the next century globalized world ocean.

1 Introduction

Biological invasions, together with climate change and the human-induced changes in land use, constitute the most worrying expressions of global change. Biological invasions are the result of species introduction. An introduced species (= established alien) is defined as a species that meets the four following criteria:

(1) It colonizes a new area where it did not previously occur.
(2) The extension of its range is linked, directly or indirectly, to human activity.
(3) There is geographical discontinuity between its native area and the new area.
(4) Finally, new generations of the non-native species are born in situ without human assistance, thus constituting self-sustaining populations (Boudouresque et al. 2005).

In the marine realm, the main routes of introduction are fouling on ship hulls, ballast waters, escape from aquaria, trans-oceanic canals, and aquaculture (Boudouresque 1999).

The Mediterranean is one of the areas worldwide most severely hit by biological invasions, with more than 600 introduced species (Boudouresque et al. 2005; Zenetos et al. 2008). In the eastern Mediterranean, the Suez Canal constitutes the main vector of species introduction. In contrast, in the western Mediterranean, the main vector is aquaculture. Species such as fish or mollusks are transported from one aquaculture basin to another, distant from it, with all the accompanying species (e.g., parasites and epibiota). When the recipient habitats are suitable, these species can survive and become established, resulting in unintentional introductions (Verlaque et al. 2007).

Here, we shall only consider the macrophytes, a set of multicellular photosynthetic organisms belonging to the Magnoliophytes, Chlorobionta, Rhodobionta (kingdom Plantae), and Phaeophyceae (kingdom Stramenopiles) (Boudouresque et al. 2006).

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2 The Thau Lagoon Story

The 70-km² Thau Lagoon (southern France) is by far the leading site of oyster culture in the Mediterranean Sea, with a standing stock of 25,000 t and an annual production of 12,000–13,000 t of Japanese oysters, *Crassostrea gigas* (Thunberg). As the lagoon is not suitable for oyster breeding, the farmers are wholly dependent on the importation of spat and adults from other basins.

In the early 1970s, European farmers faced a collapse of oyster stocks due to diseases, namely parasites and pathogens introduced from Japan. From the 1970s (licitly) through at least the 1990s, and perhaps up to the present (illicitly), farmers have imported huge amounts of oysters, both adults and spat (more than 5 billion small oysters), directly from British Columbia, Japan, and Korea. Officially, since 1977, the only *C. gigas* authorized in the French Mediterranean lagoons is that produced in the Atlantic (Verlaque et al. 2007).

As a result of the failure in decontamination processes and/or quarantine of these authorized and non-authorized imports, an astonishingly high number of exotic macrophytes were introduced into the Thau Lagoon. There is no doubt that other taxa, such as metazoans, are also concerned, but, in contrast to macrophytes, they have not been the subject of specific investigations.

3 The Thau Lagoon "Japanese Botanical Garden"

As many as 58 species of introduced macrophytes have been observed in the Thau Lagoon (Table 1; Verlaque 2001; Verlaque et al. 2007). Most of these species (72–88%) are of Pacific origin, generally native to an area including Japan and/or Korea.

As far are native macrophytes are concerned, a cumulative census dating back to the nineteenth century (Bory de Saint-Vincent 1822; Huber 1892) has now reached 164 species. However, a thorough exploration of the lagoon, from 1994 to 2004, failed to track down 43 of these species; they may be either misidentified or extremely rare species, or species that became extinct because of the dramatic changes suffered by the lagoon over time. This change may be related to climate warming, given that the third peak of the Little Ice Age occurred during the nineteenth century (Le Roy-Ladurie 2004; Luterbacher et al. 2004). Another possibility is that these local extinctions were the consequence of competition with introduced species.

Overall, 121 putatively native and 58 introduced species of macrophytes currently occur in the Thau Lagoon. The introduced species represent 32% of the total species diversity, a worldwide record value to be compared with the percentage of introduced species within the whole Mediterranean (5%) (Boudouresque et al. 2005). Indeed, the Thau Lagoon resembles a botanical garden exhibiting Japanese marine macrophytes. In addition to the species clearly labeled as introduced (Table 1), possible cryptogenic (sensu Carlton 1996) and cryptic introductions are worth considering.

The former are species whose extensive range area might be the result of ancient introduction events, before the first inventories in the area and whose native region (within the current area) remains unknown; they are therefore classified as native by default. The latter closely resemble a native species; identification of their possibly exotic status would require an in-depth study. Several species in Table 1 were at first considered as native until, on the basis of a genetic study, they were assigned to a sibling exotic taxon.

4 The Thau Lagoon: A Japanese Landscape?

In the Thau Lagoon, the dominance (coverage, biomass) of introduced macrophytes is overwhelming on natural and artificial hard substrates, e.g., oyster tables (Figs. 1–3). They account for 97–99% and 48–95% in spring and autumn, respectively.

In contrast, soft substrates are still dominated by putatively native species (Fig. 2), such as *Gracilaria* spp., *Zostera marina* Linnaeus, and *Z. noltii* Horneman. However, it should be pointed out that *Z. marina* occurs throughout the northern hemisphere, so that the introduction of exotic genotypes (e.g., via seeds) to the Thau Lagoon cannot be ruled out. *Zostera japonica* Ascherson *et* Graebner, a species morphologically close to *Z. noltii*, occurs in Japan; it has been introduced to the Pacific coast of North America, probably as seeds in shipments of Japanese oysters (Harrison and Bigley 1982). The possibility

Table 1 Introduced taxa recorded in the Thau Lagoon (from Verlaque et al. (2007), updated and modified)

Species	Abundance	Kingdom, phylum, and class	Probable origin	Probable vector
Agardhiella subulata (C. Agardh) Kraft *et* M.J. Wynne	C	P Rh F	At-Pa (?)	Sh-ST
Ahnfeltiopsis flabelliformis (Harvey) Masuda	CC	P Rh F	Pa	ST
Antithamnion nipponicum Yamada et Inagaki	CC	P Rh F	Pa	ST
Antithamnionella spirographidis (Schiffner) E.M. Wollaston	RR	P Rh F	Pa	Sh-ST
Asparagopsis armata Harvey (as *Falkenbergia* phase)	RR	P Rh F	Pa	Sh-ST
Ceramium sp.	CC	P Rh F	Pa	ST
Chondria coerulescens (J. Agardh) Falkenberg	CC	P Rh F	At	ST
Chondrus giganteus Yendo f. *flabellatus* Mikami	C	P Rh F	Pa	ST
Chrysymenia whrightii (Harvey) Yamada	C	P Rh F	Pa	ST
Dasya sessilis Yamada	CC	P Rh F	Pa	ST
Grateloupia asiatica Kawaguchi *et* Wang	C	P Rh F	Pa	ST
Grateloupia lanceolata (Okamura) Kawaguchi	CC	P Rh F	Pa	ST
Grateloupia minima P.L. Crouan *et* H.M. Crouan	C	P Rh F	At	ST
Grateloupa subpectinata Holmes	C	P Rh F	Pa	ST
Grateloupia patens (Okamura) Kawaguchi *et* Wang	R	P Rh F	Pa	ST
Grateloupia turuturu Yamada	CC	P Rh F	Pa	ST
Griffithsia corallinoides (Linnaeus) Batters	CC	P Rh F	At-Pa	ST
Herposiphonia parca Setchell	C	P Rh F	Pa	ST
Heterosiphonia japonica Yendo	C	P Rh F	Pa	ST
Hypnea valentiae (Turner) Montagne	C	P Rh F	Pa	ST
Laurencia okamurae Yamada	C	P Rh F	Pa	ST
Lithophyllum yessoense Foslie	C	P Rh F	Pa	ST
Lomentaria flaccida Tanaka	RR	P Rh F	Pa	ST
Lomentaria hakodatensis Yendo	CC	P Rh F	Pa	ST
Nemalion vermiculare Suringar	C	P Rh F	Pa	ST
Neosiphonia harveyi (Bailey) M.S. Kim, H.G. Choi et al.	CC	P Rh F	Pa	Sh-ST
Nitophyllum stellato-corticatum Okamura	C	P Rh F	Pa	ST
Polysiphonia atlantica Kapraun *et* J. Norris	R	P Rh F	At-Pa	Sh-ST
Polysiphonia fucoides (Hudson) Greville	RR	P Rh F	At	FB-ST
Polysiphonia morrowii Harvey	C	P Rh F	Pa	ST
Polysiphonia paniculata Montagne	RR	P Rh F	Pa	Sh
Polysiphonia stricta (Dillwyn) Greville	RR	P Rh F	At	Sh-ST
Pterosiphonia tanakae Uwai *et* Masuda	CC	P Rh F	Pa	ST
Rhodophysema georgii Batters	RR	P Rh F	At-Pa	ST
Rhodothamniella codicola (Børgesen) Bidoux *et* F. Magne	CC	P Rh F	Pa	Sh-ST
Porphyra yesoensis Ueda	CC	P Rh B	Pa	ST
Cladophora hutchinsioides Hoek *et* Womersley	C	P Chl U	Pa	ST
Codium fragile (Suringar) Hariot	CC	P Chl U	Pa	Sh-ST
Derbesia rhizophora Yamada	C	P Chl U	Pa	ST
Monostroma obscurum (Kützing) J. Agardh	CC	P Chl U	Pa	Sh-ST
Ulva fasciata Delile	C	P Chl U	Pa	Sh-ST
Ulva pertusa Kjellman	CC	P Chl U	Pa	ST
Acrothrix gracilis Kylin	R	S Chr P	At-Pa	ST
Chorda filum (Linnaeus) Stackhouse	C	S Chr P	At-Pa	ST
Cladosiphon zosterae (J. Agardh) Kylin	R	S Chr P	At	ST
Colpomenia peregrina (Sauvageau) Hamel	CC	S Chr P	Pa	Sh-ST
Desmaretia viridis O.F. Müller	CC	S Chr P	At-Pa	ST
Dictyota okamurae (E.Y. Dawson) I. Hörnig et al.	C	S Chr P	Pa	ST
Halothrix lumbricalis (Kützing) Reinke	C	S Chr P	At-Pa	ST
Leathesia difformis (Linnaeus) Areschoug	C	S Chr P	Co	ST

(continued)

Table 1 (continued)

Species	Abundance	Kingdom, phylum, and class	Probable origin	Probable vector
Microspongium tenuissimum (Hauck) A.F. Peters	R	S Chr P	At	ST
Pilayella littoralis (Linnaeus) Kjellman	CC	S Chr P	At-Pa	ST
Punctaria tenuissima (C. Agardh) Greville	C	S Chr P	At	ST
Saccharina japonica (Areschoug) Lane et al.	R	S Chr P	Pa	ST
Sargassum muticum (Yendo) Fensholt	CC	S Chr P	Pa	ST
Scytosiphon dotyi Wynne	R	S Chr P	Pa	Sh-ST
Sphaerotrichia firma (E. Gepp) Zinova	C	S Chr P	Pa	ST
Undaria pinnatifida (Harvey) Suringar	CC	S Chr P	Pa	ST

Abundance: CC, very common; C, common; R, rare; RR, very rare. Kingdom: P, Plantae; S, Stramenopiles. Phylum: Chl, Chlorobionta; Chr, Chromobionta; Rh, Rhodobionta (according to Baldauf 2003; Boudouresque et al. 2006)

Class: B, Bangiophyceae; F, Florideophyceae; P, Phaeophyceae; U, Ulvophyceae (according to Yoon et al. 2006; Brodie et al. 2007)

Origin: At, Atlantic; Co, cosmopolite; In, Indian Ocean; Pa, Pacific

Vector (primary introduction–secondary dispersal): FB, fishing baits; Sh, shipping (hull fouling, ballast water); ST, shellfish transfer

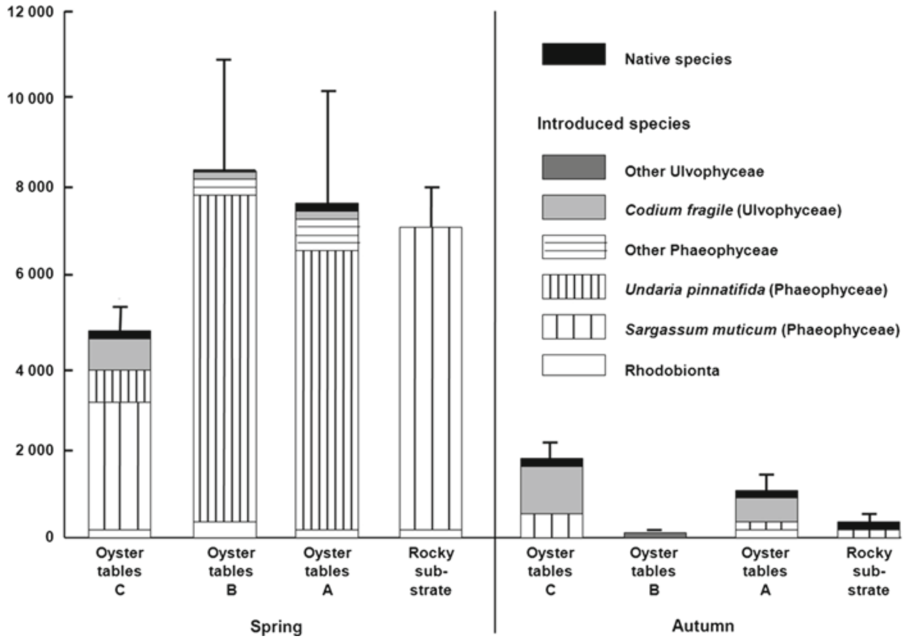

Fig. 1 Mean total macrophyte biomass (g wet weight/m² ± SE) on oyster tables (zones A, B, and C of aquaculture facilities) and on shallow rocky substrates (Bouzigues) of the Thau Lagoon, in spring and autumn

that the '*Z. noltii* stands' of Thau may actually be constituted, at least partly, by *Z. japonica*, has not been investigated. Finally, several *Gracilaria* species may be suspected of being a complex of cryptic species, so that 'native *Gracilaria*' could prove to mask a mixture of native and introduced species (a hypothesis not yet supported by genetic data; Jan Rueness, unpublished data).

5 Conclusion

The Thau Lagoon looks, from a biological diversity point of view, like a Japanese island in the very heart of the Mediterranean. In a context of global homogenization of biodiversity due to biological invasions (Clout 1998; Galil 2000), it could be the harbinger of the next century globalized world ocean.

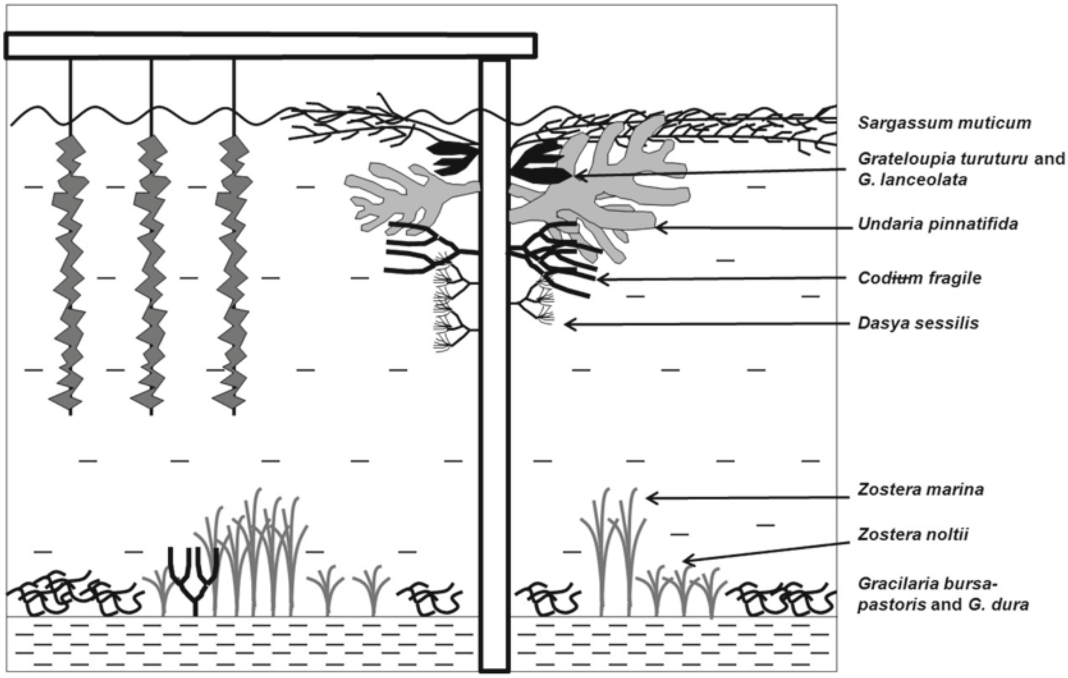

Fig. 2 A sketch of the dominant macrophytes on and beneath (4 m depth) an oyster table of the Thau Lagoon, in spring. Only *Zostera marina*, *Z. noltii*, and *Gracilaria* spp. are putatively native species (see text)

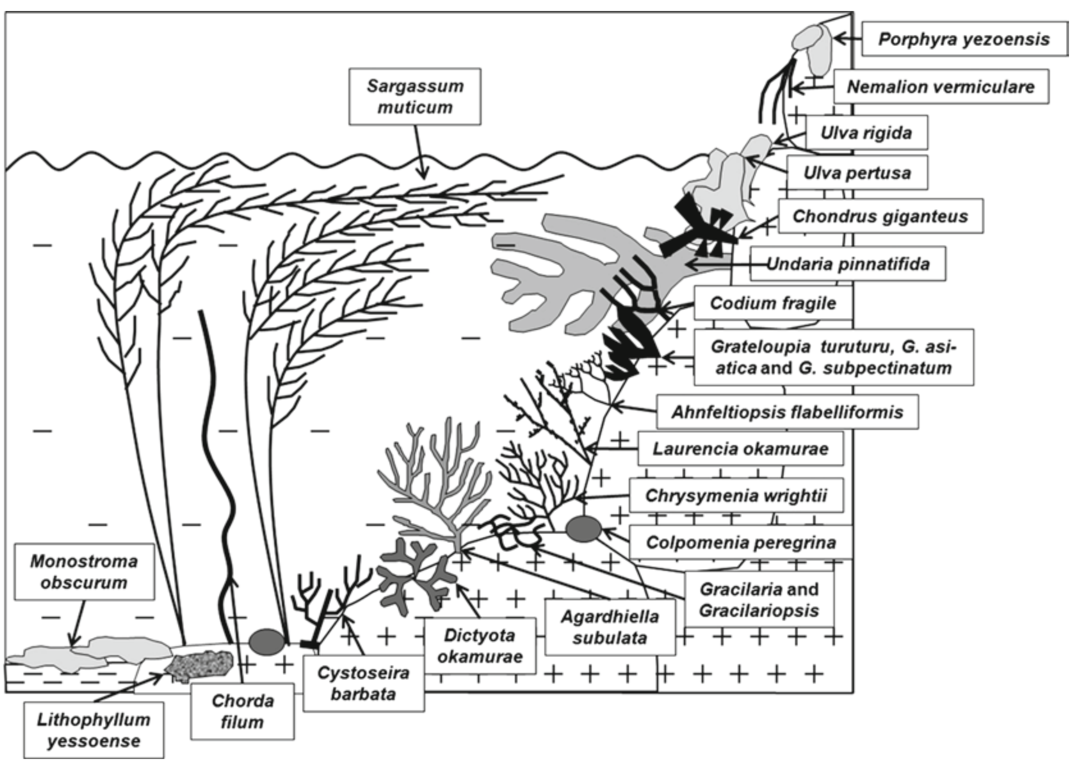

Fig. 3 A sketch of the dominant macrophytes on shallow (down to 1 m depth) rocky substrates of the Thau Lagoon in spring. Only one species, *Cystoseira barbata*, is certainly native

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Distribution of Giant Viruses in Marine Environments

Hiroyuki Ogata, Adam Monier, and Jean-Michel Claverie

Abstract Viruses are the most abundant biological entities in the sea. Infecting marine organisms from oxygen-producing phytoplankton to whales, viruses regulate the populations of many marine organisms and play important roles in global biogeochemical fluxes. Thanks to the recent improvement of sequencing technology, billions of bases of sequences from marine microbial communities are being determined by different groups of researchers.

Those environmental sequence data, or "metagenomes," now provide unprecedented opportunities to reconstruct and characterize the composition and the dynamics of mostly unculturable microorganisms in different habitats.

We recently developed a new computational method called phylogenetic mapping to obtain a comprehensive picture of the distributions of microorganisms contained in environmental samples. We used our method to investigate the distributions of large DNA viruses represented in the Sorcerer II Global Ocean Sampling Expedition microbial metagenomic data set. Using DNA polymerase genes as a taxonomic marker, we identified 811 homologous sequences of likely viral origins. Most of these sequences corresponded to phages, being consistent with the large number of phage-like particles in marine environments. Interestingly, the second largest viral group corresponded to the one containing mimivirus, the largest known virus (with 1.2 Mb-genome), which infects amoeba. Albeit only recently discovered, giant viruses of the mimivirus family appear to constitute a diverse, quantitatively important, and geographically ubiquitous component of the population of large eukaryotic DNA viruses in the sea. We also identified several DNA polymerase homologs closely related to African swine fever virus, a large virus pathogenic to domestic animals and until now limited to terrestrial animal hosts.

Finally, our approach allowed the identification of a new combination of genes in viral-like sequences. In conclusion, high throughput metagenomics is becoming a technique of choice to follow the changes of marine microbial environments induced by climatic changes and/or human activities.

1 Introduction

Viruses are the most numerous organisms on our planet, including marine environments. Several studies suggest the existence of 10^6 to 10^9 viruses in only 1 mL of sea water (Suttle 2005; Patel et al. 2007; Monier et al. 2008a). Another study suggests the existence of > ten viruses per microbe (Rohwer 2003). By infecting and killing marine organisms, including photosynthetic algal plankton, viruses significantly contribute to the dynamics of ecosystems and thus affect the global and local climate changes and biogeochemical fluxes. Recent genomics studies are revealing their tremendously large genetic diversity (Ogata and Claverie 2007). The huge viral genetic diversity is also pointed out by the fact that the majority (>60%) of viral genes

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from environmental samples lack similar sequences in the current sequence databases (Edwards and Rohwer 2005). Viral genomes thus may be an important source of new protein functions for biomedical and biotechnology developments. Of all viruses, the giant mimivirus (discovered a few years ago) has been of our particular interest. The goal of our study is to characterize and understand the ecological and evolutionary significances of mimivirus and related giant viruses. Recently, we have developed a new bioinformatics method called "phylogenetic mapping" (Monier et al. 2008a). With this method, we characterized the taxonomic distribution of large DNA viruses in the sea using a publicly available metagenomic sequence data set. In this paper, we first provide a short introduction to the recent progresses in the study of mimivirus. We then outline the results obtained by our phylogenetic mapping study.

2 Discoveries from the Giant Mimivirus

Mimivirus (*Acanthamoeba polyphaga* mimivirus, APM) is the largest known virus in both particle size and genome size. The original strain of mimivirus was discovered from the water of a hospital in Bradford, England (La Scola et al. 2003). Due to its large particle size (0.7 μm in diameter for the whole particle including surface filaments) and the gram-positive staining property, the mimivirus was initially mistaken as a bacterium. Electron microscopic studies then revealed the icosahedral form of the mimivirus capsid (0.4 μm in diameter), a common structure of many viruses. The genome sequence of mimivirus was determined in 2004 (Raoult et al. 2004). The size of the genome turned out to be 1.2 Mb (encoding >900 genes), being larger than many bacterial genomes. The analysis of this giant viral genome revealed a number of surprises. Most remarkably, the genome encoded eight genes for proteins central to the protein translation system, including four aminoacyl-tRNA synthetases (Abergel et al. 2007), a part of the hallmark enzymes for cellular organisms. The initial characterization of mimivirus has thus quantitatively and qualitatively blurred the traditional barrier between viruses and cellular organisms, and stimulated debates about the origin of viruses and their role in the emergence of eukaryotes (reviewed in [Claverie 2006; Claverie et al. 2006]). The following characterizations of mimivirus further led to many surprises. Proteomic studies by Renesto et al. (2006) showed that the mimivirus particles contain at least 114 distinct proteins, which are likely to be involved in the early phase of its replication cycle. The mimivirus infection cycle has been characterized and reported by Suzan-Monti et al. (2007). Mimivirus enters amoeba cells by phagocytosis. After infection, mimivirus establishes its own intracellular compartment called the "virus factory," the viral replication and assembly center. The mimivirus virus factory is very large, its size being comparable to the nucleus of amoeba. The group of Zauberman determined the precise structure of the mimivirus DNA injection system (Zauberman et al. 2008). To inject its genome into the host, the mimivirus opens up five triangular faces around a vertex of its icosahedral capsid. Zauberman coined this structure "the stargate." La Scola et al. very recently discovered a new strain of mimivirus (named "mamavirus") from the water of a cooling tower in Paris, France, and a smaller virus co-infecting amoeba with mamavirus (La Scola et al. 2008). The small virus named Sputnik cannot replicate within amoeba by itself.

However, the small Sputnik virus starts to replicate upon co-infection with the larger mimivirus or mamavirus. Sputnik uses the virus factory of mimivirus for its replication. The co-infection of both viruses led to the production of mimivirus virions having aberrant morphologies. Getting infected with viruses is thus not limited to cellular organisms. Viruses themselves can act as the host of other viruses and occasionally "get sick" (Ogata and Claverie 2008).

3 Existence of Mimivirus Relatives in the Sea

Mimivirus belongs to the group of viruses collectively called nucleo-cytoplasmic large DNA viruses (NCLDVs), which include five diverse viral families: mimiviruses, phycodnaviruses, iridoviruses, poxviruses, and asfarviruses (Raoult et al. 2004). Their genomes substantially vary in size across different NCLDVs (from 100 kb up to 1.2 Mb) and exhibit large genetic variation across the different virus families.

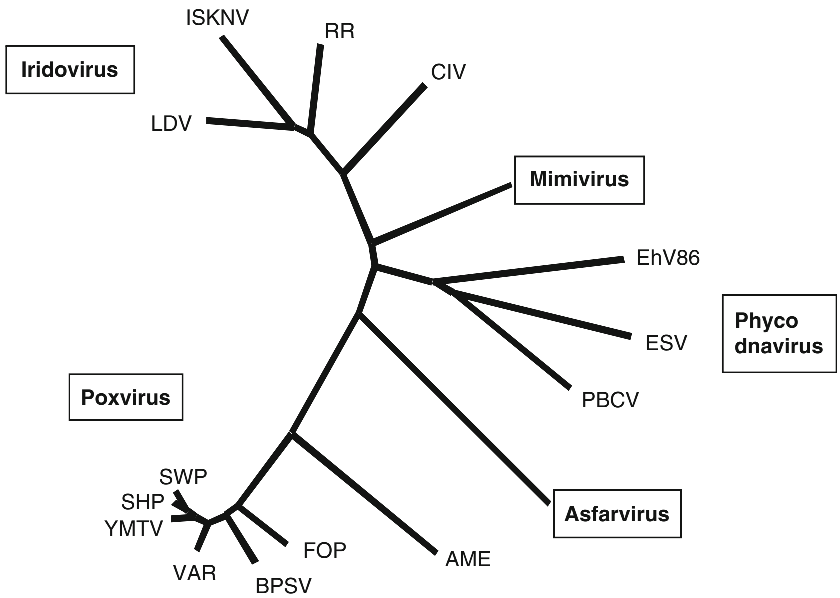

Fig. 1 A phylogentic tree of nucleo-cytoplasmic large DNA viruses (NCLDVs)

As shown in the tree (Fig. 1), the mimivirus makes its own lineage among NCLDVs. Although mimiviruses have been isolated from freshwater of anthropogenic systems, an early study by Ghedin and Claverie suggested the existence of viruses closely related to the mimivirus in marine environments (Ghedin and Claverie 2005). Ghedin and Claverie identified sequences similar to the sequence of the mimivirus genome in a marine metagenomic sequence data set. More recently, Monier et al. (2008b) provided evidence for the existence of mimivirus relatives using a larger set of marine metagenomic sequences. These results prompted us to further characterize the population of the mimivirus family in marine environments (Monier et al. 2008a).

Specifically, we asked the following question: Are the mimiviruses relatively abundant in the sea? To answer to this question, we used the publicly available large marine metagenomic sequence data set, generated by the Sorcerer II Global Ocean Sampling (GOS) expedition by the group of John Craig Venter (Rusch et al. 2007). The Sorcerer II expedition was inspired by the H.M.S. Challenger expedition, but aiming at the assessment of the global genetic diversity of marine microbial communities rather than macro-organisms. The first phase of the GOS Global Ocean Sampling expedition was finished, and the generated metagenomic sequence data sets were released in 2007. This phase of the expedition was targeted to the characterization of marine bacteria. They sequenced DNA from 0.1 to 0.8 sized fractions, which is compatible with the sizes of large viruses. The GOS sequence data set is composed of 7.7 million reads corresponding to 4.9 billion bp of assembled sequences.

4 Phylogenetic Mapping for the Classification of Metagenomic Sequences

Metagenomic sequences are usually short and "anonymous" (i.e., from unknown source species). To gain insights into species abundance represented in a metagenomic data set, we need to assign taxonomy to metagenomic sequences. A powerful approach involves the use of phylogenetic methods. However, when applying phylogenetic methods to environmental shotgun sequences, the treatment of the short sequences requires special attention. These sequences show large variation in size and possibly correspond to different parts of a selected marker gene. Piling up multiple short sequences on representative markers from known organisms does not provide an appropriate alignment. We thus developed a new phylogeny-based method. The method called "phylogenetic mapping" analyzes individual metagenomic sequences one by one and determines their phylogenetic positions using a reference multiple sequence alignment and a reference tree. As an attempt to investigate the presence, the taxonomic richness, and the relative abundance of different large DNA viruses in marine environments, we analyzed the GOS data set using PolB sequences as our reference.

5 Abundance of Mimivirus-Like PolB Sequences in the Sea

We first screened the GOS data set by using the Pfam protein profile (PF00136) corresponding to PolB. This resulted in the identification of 1,947 PolB-like sequences (from 23 to 562 amino acid residues). By our phylogenetic mapping method, we could assign the best branching position for 1,423 PolB fragments, of which 1,224 (86%) were mapped on viral branches. The best branching position was statistically supported for 869 PolB fragments, of which 811 (93%) were mapped on viral branches. The inferred taxonomic distribution of the GOS PolB fragments is shown in Fig. 2. The largest fraction of the PolB fragments was mapped on the phage group. Of 866 cases of mapping within the phage group, 633 were supported. This is consistent with the current estimate of the large number of phage-like particles and their genetic richness in marine environments. Interestingly, the second largest group was the mimivirus group showing 115 supported cases. We found two PolB fragments mapped with good support on the African swine fever virus (ASFV) branch. ASFV infects terrestrial animals and is pathogenic to domestic pigs.

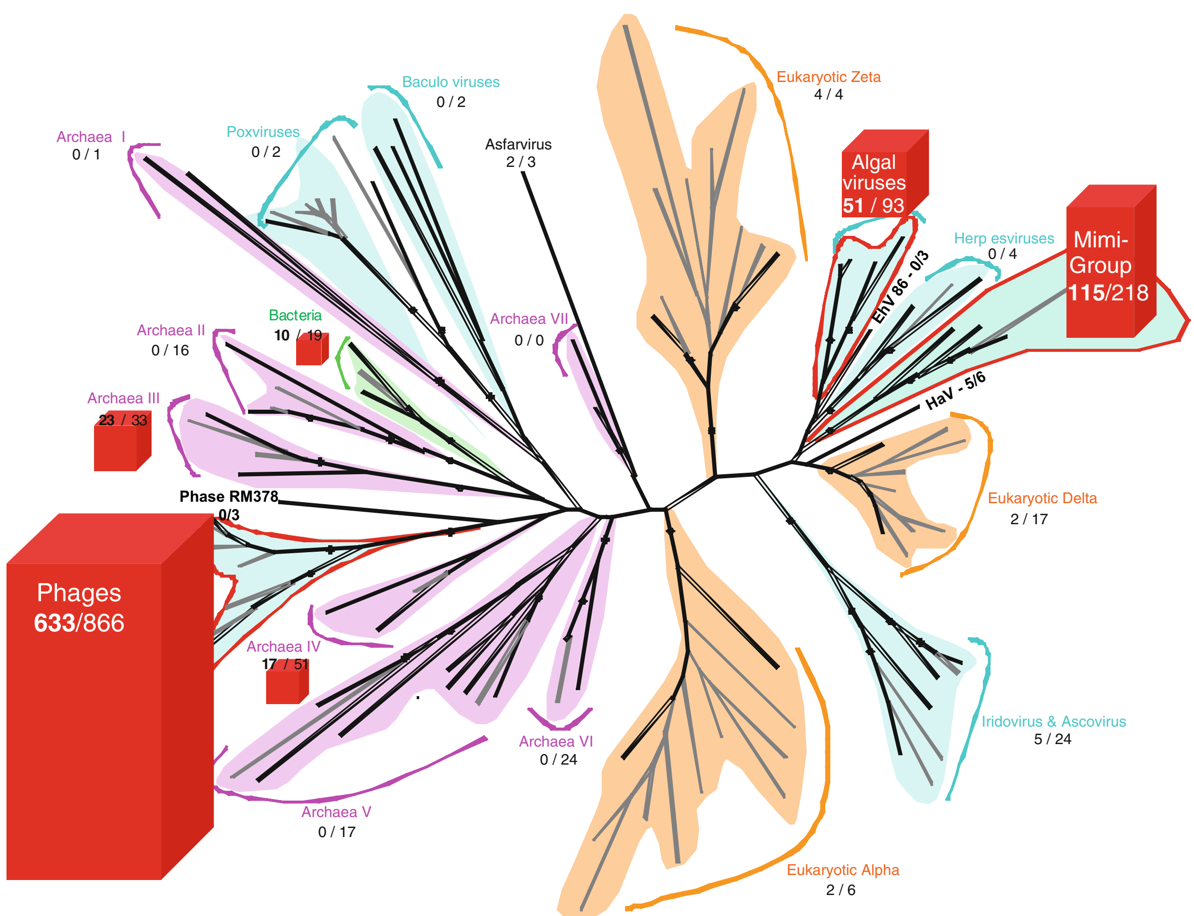

Fig. 2 Taxonomic distribution of viruses in the GOS metagenomic data (modified from Monier et al. 2008a)

6 Geographic Distribution

Figure 3 shows the relative abundance of different virus groups across different GOS sampling sites. PolB fragments classified in the phage group were found in 42 (95%) of the 44 sample sites; the two samples without phage PolB fragments were GS08 (Newport Harbor, Richmond, USA) and GS32 (mangrove). In most samples (32 sites), putative phage PolBs exhibited a higher abundance relative to putative eukaryotic viral PolBs. On the other hand, the relative abundance of eukaryotic viral PolBs was higher than that of phage PolBs in 12 sampling sites. Phage-type PolBs showed a higher relative abundance than eukaryotic viral PolBs in tropical waters (T ≥ 20°C), whereas an opposite tendency was observed in temperate water (T < 20°C). Interestingly, among eukaryotic viral PolBs, putative mimivirus-family (i.e. Mimiviridae) PolBs showed the most widespread distribution, being detected in 38 (86%) of the total sites. One of these sampling sites (mangrove located in Isabella, Ecuador) exhibits only viral PolBs classified in the Mimiviridae group. Mimiviridae PolBs were also relatively abundant in two of the three samples from a hydrostation located in the Sargasso Sea. PolB fragments grouped with chloroviruses were also widely distributed. They were detected in 16 (36%) samples.

7 Concluding Remarks

The use of a phylogenetic approach provided a comprehensive picture of the taxonomic distribution of large viruses enclosed in the GOS metagenomic data. As expected, the highest genetic richness corresponded to phages. Interestingly, our data suggest that Mimiviridae represent a major and ubiquitous component of large eukaryotic DNA viruses in diverse marine environments. High throughput metagenomics as well as the associated bioinformatics is now becoming a powerful approach to investigate species richness and their population dynamics, which might be tightly linked to global and local environmental changes.

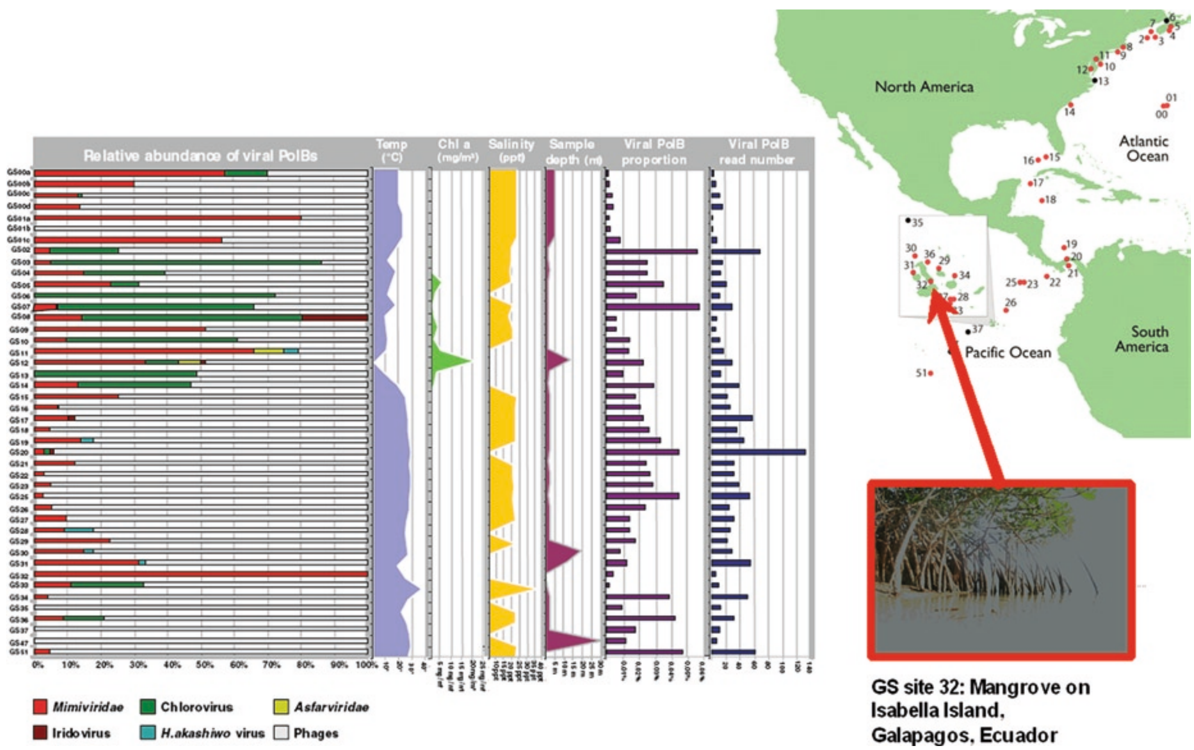

Fig. 3 Geographic distribution of viruses (modified from Rusch et al. 2007; Monier et al. 2008a)

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Catch, Bycatch of Sharks, and Incidental Catch of Sea Turtles in the Reunion-Based Longline Swordfish Fishery (Southwest Indian Ocean) Between 1997 and 2000

François Poisson

Abstract Data from voluntary logbooks (5,884 longline sets) collected between 1997 and 2000 were analyzed to assess the potential impact of the Reunion-based longline swordfish fishery (South West Indian Ocean) on sharks and on sea turtle populations. Blue shark (*Prionace glauca*) represented between 75% and 88% of the total catches of sharks studied. Bycatch discarding varied with species, ranging from low discards (2.6%) for *mako shark* (*Isurus sp.*) to high discards for blue shark (86.5%). Estimation of the total catch of sharks (retained and discarded individuals) represented in weight between 7% and 9% of the total catch of the major species caught by the fishery. Of concern is the decline of blue shark CPUE (Catch Per Unit of Effort) from 2.2 to 1.03 sharks per 1,000 hooks between 1998 and 2000. Of the 22,974 hook-timers deployed during cruises onboard commercial, 49% of the blue shark (*Prionace glauca*) and 41.2% of the oceanic whitetip shark (*Carcharhinus longimanus*) were retrieved alive. It was assumed that among the 15,250 blue sharks caught during this period, at least 7,099, released alive, could have survived.

The observed sea turtle catch rates were low compared with those reported for other longline fisheries. The fishery had 47 interactions with Leatherback (*Dermochelys coriacea*), 30 with Hawksbill turtles (*Eretmochelys imbricata*), and 16 with Green turtles (*Chelonia mydas*).

World catches of sharks grew from 600,000 to over 810,000 t between 1984 and 2004. In addition to directed shark fisheries, pelagic longline fisheries for tuna and swordfish operating on the high seas are interacting with a large number of shark species (Buencuerpo et al. 1998; Francis et al. 2001; Bonfil 1994; Ward et al. 2004). These fisheries also have been identified as a major source of sea turtle mortality (Pinedo and Polatcheck 2003; Lewison and Crowder 2007). There has been considerable concern over the conservation status and sustainability of elasmobranches and sea turtles in world fisheries during the past decade; in 1999, the FAO initiated an International Plan of Action for the Conservation and Management of Sharks (IPOA Sharks) and has strongly encouraged shark fishing countries to implement national action plans and to conduct stock assessments; (http://www.fao.org/fishery/ipoa-sharks/en) the Hawaii-based longline swordfish (*Xiphias gladius*) fishery was closed for over 2 years because of the magnitude of interactions with turtles and is now subject to strict management measures (Gilman et al. 2007). In the western Indian Ocean, attempts to establish a regional process to address the conservation and management of sea turtles started in 1995, and the relative importance of sea turtle mortality because of fisheries was assessed during a dedicated FAO workshop held in Tanzania in 2006 (http://www.ioseaturtles.org).

In the Indian Ocean, the data held by the Indian Ocean Tuna Commission (IOTC) on sharks are scarce and to a large extent incomplete. The reported catches of sharks represented typically only the sharks that were retained on board, and in many cases, these catches also refer to dressed weights (without any indication of the type of processing). Moreover, shark catches are not recorded by species. Furthermore, in the past, when only fins were retained on board, fishers rarely recorded the weights or

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numbers of sharks from which the fins were taken. Finally, in the majority of the cases, shark bycatch is simply not recorded at all (Anonymous 2007).

All these factors combine to make it difficult to estimate the total catches of sharks in the Indian Ocean. Since the implementation of the dedicated Working Party on Ecosystems and Bycatch (WPEB), the IOTC Secretariat has not received any reports from members or cooperating parties on the amounts of sea turtles caught incidentally by their tuna fisheries. Observer programs are likely to be the most important sources of data on bycatch species. The scientists of the WPEB noted that coverage by observer programs in the Indian Ocean was currently very low and encouraged all IOTC members to increase the amount of information available in the future (Anonymous 2007). In response to this recommendation, data from voluntary logbooks and observations collected between 1997 and 2000 during scientific cruises onboard Reunion Island (21°S, 55°E) swordfish longliners were revisited and analyzed to assess the potential impact of the domestic fishery on sharks and sea turtle populations. During this period, the Reunion-based longline swordfish fishery caught an average of 1,800 t of swordfish per year, which comprised 5.5% of the average total catches of 33,000 t estimated for the Indian Ocean.

In this study, the impact of the domestic longline fishery on three major species and one group of sharks and three turtles species was examined. The objectives were for sharks to estimate per species the catch rates, the survival rate, the condition when released, and the total catch; concerning sea turtles, the objectives were to report the total number of interactions and the direct mortality as well as to present the trends of the catch rates.

As a result of the cooperation of fishers, the logbook program provided a large number of observations distributed over several fishing seasons. Collection of data on the major species and especially swordfish was the priority form the beginning of the program in 1994, and data collection on bycatch, discards, and turtles incidental catches was introduced gradually. Because of possible unreported information on sharks, a part of the data for sharks was not considered in the analyses. However, the quality of the data on bycatches after 1997 is considered good. In this study, the largest percentage of bycatch was composed of sharks, mainly blue shark (*Prionace glauca*).

These results provided a clearer perspective of the magnitude of shark bycatch, species commonly caught in the pelagic longline fishery. The sharks observed are highly migratory; they are obviously subject to fishing mortality from other tuna longline fisheries in the Indian Ocean. Of concern is the indication that CPUE (Catch Per Unit of Effort) of blue shark for the boats operating close to the island has declined from 1998 to 2000 concomitant with a steady increase of the number of hooks deployed by this vessel class. Of the 22,974 hook-timers deployed during scientific cruises onboard commercial, 49% of the blue shark and 41.2% of the oceanic whitetip shark (*Carcharhinus longimanus*), were retrieved alive. It is difficult to draw conclusions with this limited temporal series, but our study provided a reference point for the status of the four species/groups studied and serves as a baseline for future shark and sea turtle surveys.

Although no solution has been found to either avoid or to mitigate shark and sea turtle bycatch, using nylon leaders (Ward et al. 2008) and releasing animals are the best mitigation policy that could be implemented even though the outcome of the released animals is still uncertain due to the lack of data on survival. The approach developed by Moyes et al. (2006) combining tagging and biochemical analyses should be extended to other non-target species to estimate the impact of this measure. The observed sea turtle catch rates were low compared with those reported for other longline fisheries. In most of the case, turtles were released alive. One Leatherback turtle (*Eretmochelys imbricata*) remained hooked for almost 14 h and was hauled alive.

It is difficult to draw conclusions with this limited temporal series, but our study provided a reference point for the status of the four species/groups studied and serve as a baseline for future shark and sea turtle surveys.

Despite the fact that no solution has been found to either to avoid or mitigate shark and sea turtle bycatch, using nylon leaders (Ward et al. 2008) and releasing animals are the best mitigation policy that could be implemented even though the outcome of the released animals is still uncertain due to the lack of data on survival. The approach developed by Moyes et al. (2006) combining tagging and biochemical analyses should be extended to other non-target species to estimate the impact of this measure.

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Biodiversity Requires Adaptations Under a Changing Climate in Northwest Europe: Planning and Coastal Wildlife, the Example of Normandy in France

Isabelle Rauss, Pascal Hacquebart, Catherine Zambettakis, Emmanuel Caillot, Emmanuel De Saint Léger, and Franck Bruchon

Abstract The Interreg IIIB BRANCH project evaluated potential impacts of climate change on coastal wildlife in Northwest Europe and aimed at identifying planning strategies in cooperation with stakeholders. A specific technical and methodological effort focused on elaborating a GIS as a support for prospective work. This paper deals with the example of the French coastal case study sites in Normandy:

1. Several *key marine and coastal habitats and species* have been studied (quantity, quality, localization, evolution) so as to integrate ecosystem *functionalities* in scenarios showing risks on marine habitats versus climate change factors (sea level rise, temperature rising): benthic fauna, flora, bird, arthropods, and hydrosedimentary dynamics.
2. A new method has been elaborated in order to build a common database integrating *pluridisciplinary information*, to elaborate an initial status assessment, to support *long-term monitoring*, and to *be flexible, to progressively integrate* new marine, social, and economical knowledge.

The results were presented and debated at regional and national workshops, which raised seven main recommendations for adaptation measures leading to a better consideration of climate change in coastal management and its effects on biodiversity:

1. *To anticipate in order to reduce impacts*: it is essential to act now
2. *To improve information and awareness of local stakeholders*: to better assess the socioeconomic issues of climate change
3. *To build up a shared vision*: among local stakeholders on challenges and planning measures to be taken
4. *To reduce the uncertainties*: in order to reduce their inhibitory effect on the decision-making processes
5. *To respect the natural processes*: which guarantee the sustainable development of the coast
6. *To integrate climate change into public policies*: a specific transversal policy on climate change is essential, from large to local scale
7. *To diversify and improve tools*: BRANCH highlights the need of considering biodiversity as dynamic, functional, and at larger spatial and time scales, in order to help wildlife adapt to climate change and to maintain biodiversity and ecosystem services

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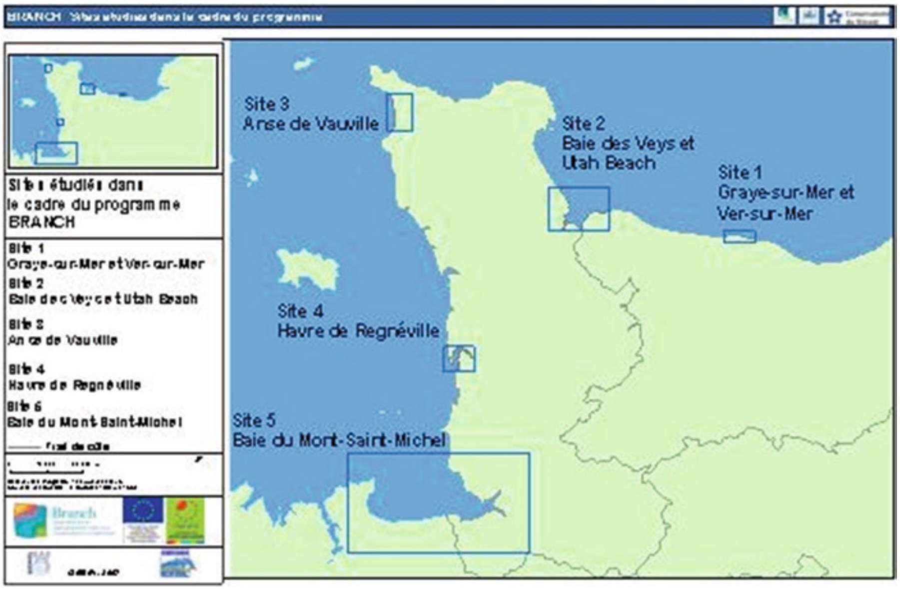


Fig. 1 The case study sites in Normandy (Graye sur Mer, Ver sur Mer, baie des Veys, Utah Beach, Dune de Vauville, Havre de Regnéville sur Mer, baie du Mont Saint Michel)

1 Introduction

The BRANCH project aimed to show how spatial planning could help biodiversity adapt to climate change. It brought together planners, policy makers, and scientists from England, France, and the Netherlands. Partners shared experience and knowledge to produce practical recommendations, based on science. The final report sets out what the project achieved and what should happen next (Branch, 2007).

Northwest Europe is a built up and economically powerful part of the world. The landscape is already fragmented, and wildlife is under pressure. For many species and habitats, climate change will intensify this pressure.

In the short term, climate change is unavoidable. We are already seeing its effects on biodiversity. Along our coasts, wildlife is being constrained between rising sea levels and flood defenses. On land, it is being forced to higher latitudes and altitudes by rising temperatures. Some habitats and species must find new places to establish themselves or they will disappear. This is a major challenge for Europe as it seeks to meet its target to "halt the loss of biodiversity by 2010" and beyond.

People are dependent on nature. It sustains us and improves our quality of life. Some natural benefits, such as raw materials, are economically valuable. Others, like well-managed natural habitats, help us to cope better with the effects of flooding and pollution. Some benefits, like places for recreation, make us healthier. All these services provided by biodiversity are threatened by climate change.

Planners can play a vital role in ensuring these natural benefits continue, despite climate change. Planners already have some mechanisms to help maintain and create landscapes that allow wildlife to adapt. But new policies are needed. This project provides guidance on how this should happen.

Several case site studies have been used as a model for terrestrial and coastal issues. We report here about the French case study, showing the results obtained in the Baie des Veys (Lower Normandy, France, Fig.1).

2 Coastal Issues: The Example of Normandy

BRANCH used seven case studies in Lower Normandy to assess how climate change may affect coastal biodiversity. The study sites included sand dunes, salt marshes, mudflats, grazing marshes, and reed beds. The study investigated the species and habitats of each site and their links with the geomorphology of the coast. It assessed how the estuary and dune areas were likely to change under rising sea levels and what risks each site might face. BRANCH-recommended tools for planning, management, and monitoring, and discussed them with stakeholders.

BRANCH models show that mudflats, dunes, wetlands, and freshwater ponds are the most vulnerable coastal habitats to climate change. Sand dunes are eroding on all the study sites. Dunes help to protect wetlands and freshwater ponds from salinization and flooding by the sea. Rises in sea level may make the erosion worse. Salt marshes are currently expanding at all the study sites, in contrast to the UK case studies, but this may be halted by sea-level rise. There are also high rates of sedimentation.

Long-term monitoring of coastal habitats, species, and geomorphology will help stakeholders to better understand how the coast works and how it might change. There are opportunities to protect intertidal habitats. Loss of habitat could be partly compensated by habitat re-creation through managed realignment. But this could lead to new conflicts between different protected habitats and with land use, particularly agriculture. BRANCH provided guidance to help stakeholders manage and plan for climate change.

This includes:

- Reports summarizing current wildlife and coastal dynamics at case study sites and considering how these are likely to change in the future.
- A new flexible mapping tool for planners to help evaluate land use decisions for the coast. This includes habitat and species datasets.

This mapping tool:

- Can show maps of vulnerability to sea level rises of 50 and 100 cm
- Forms a baseline for a long-term monitoring program at the study sites
- Takes into account current uncertainties of climate change impacts and will evolve to integrate developments in knowledge on extreme climate events

The interest of Conseil Régional de Basse-Normandie and Agence de l'Eau Seine Normandie in the BRANCH project, which they co-financed, shows that French local stakeholders are seriously considering climate change (Figs.2 and 3).

3 The Example of the Baie des Veys

The intertidal area's size in this protected estuary is almost 30 km². Salt marshes were partly destroyed by land reclamation until 1972 and are now building up again,

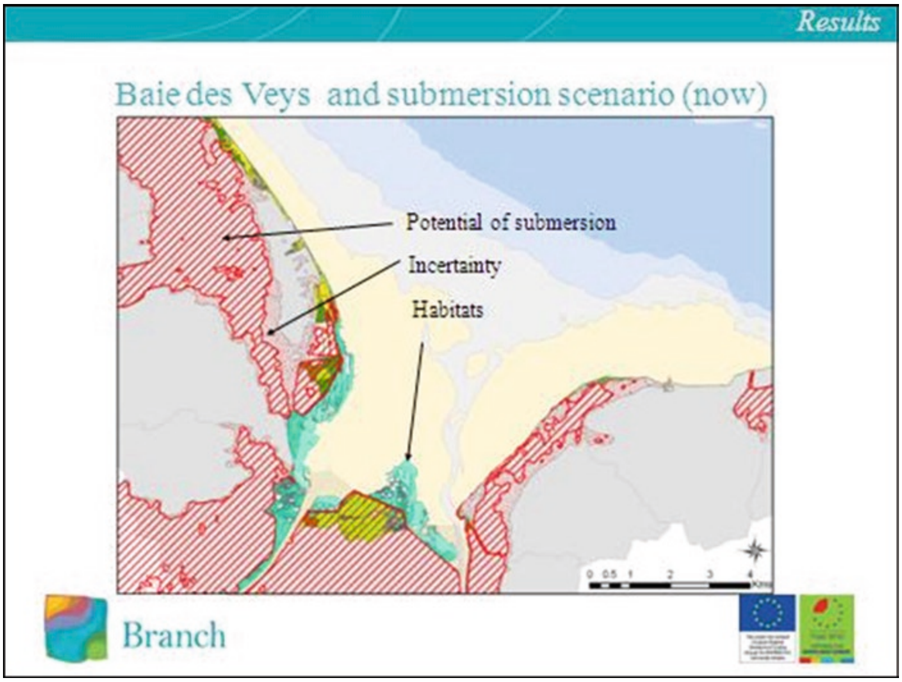


Fig. 2 Maps and risks of submersion (red) representing as well the uncertainty (dotted red) linked to the precision of data

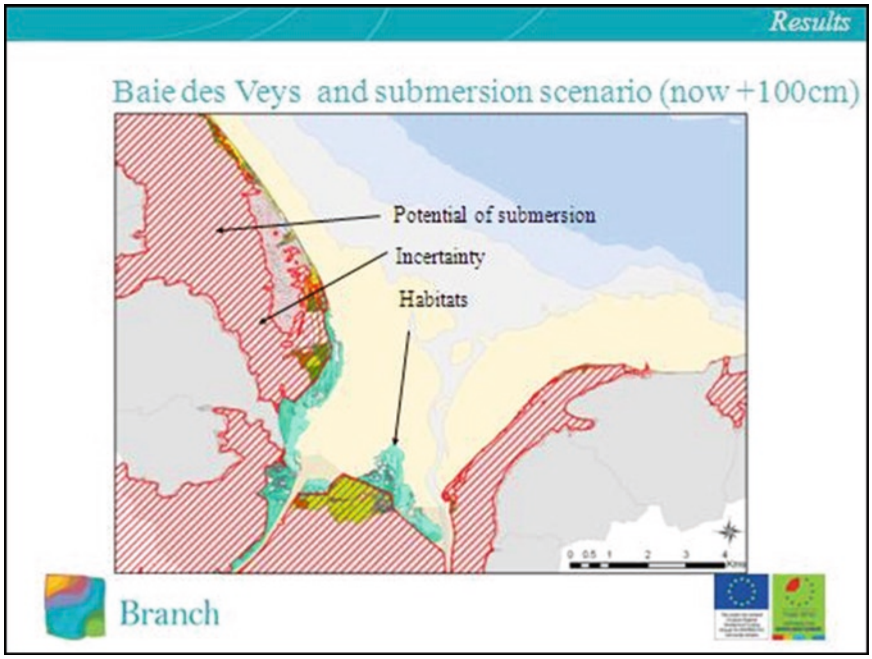


Fig. 3 Maps and risks of submersion in the future with one of the scenarios (sea level rise of 1 m)

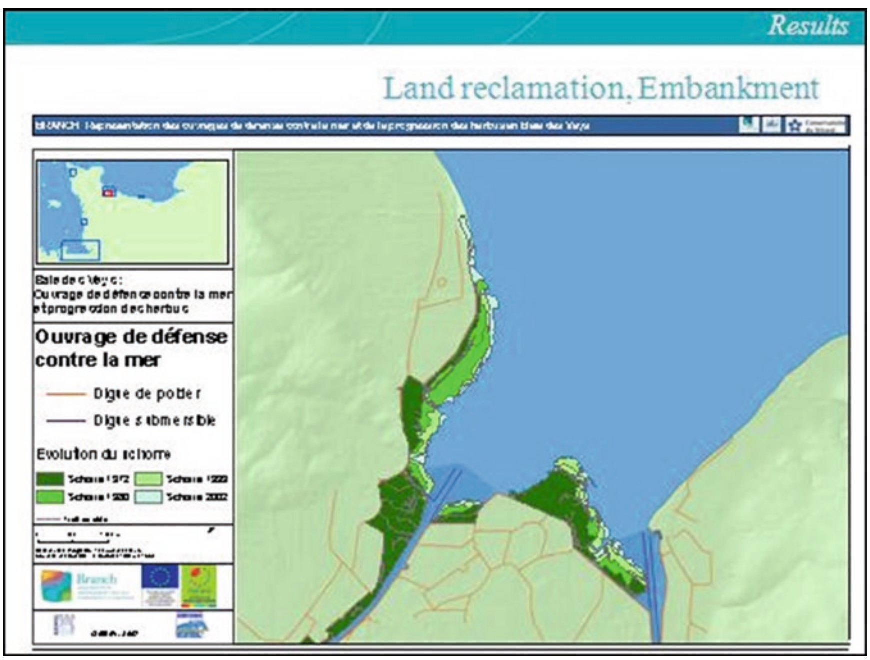
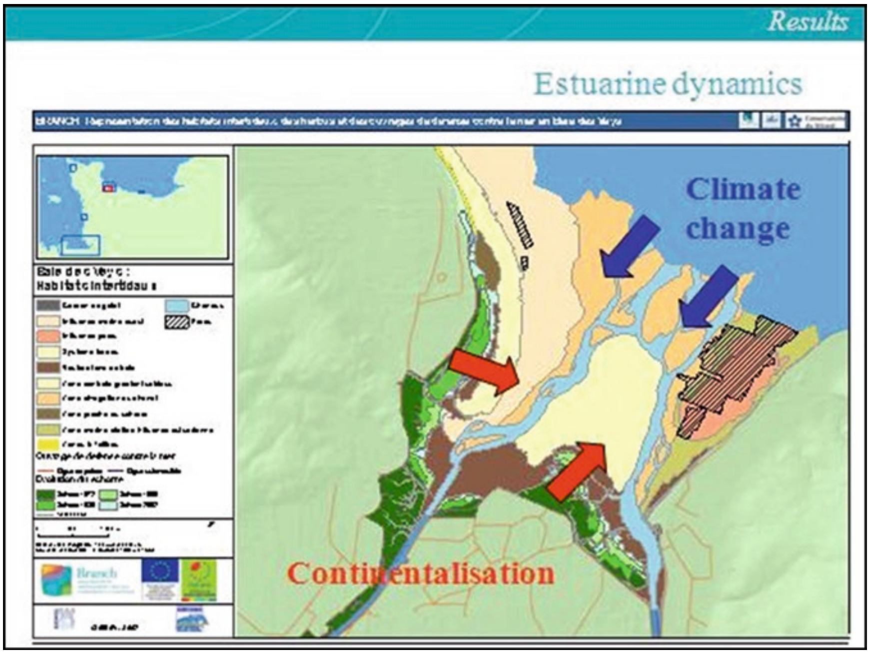


Fig. 4 (**a, b**) Maps showing the main dynamics of the site, linked to old and current processes (morphodynamics, human activities, etc.); a way to understand the whole system and talk about the future, climate change, risks, and potentiality (**b**, **c**)

expanding onto the mudflats (Fig.4a). The biodiversity of the intertidal area supports businesses, such as cockle harvesting and oyster farming.

The combination of sea-level rise, expanding salt marsh, and accumulating sediment in the estuary may reduce the area available for mudflats. This would have major consequences for intertidal seabed invertebrates and for coastal waders. Higher sea levels and changes in the prevailing winds could also alter the coastal processes in the estuary (Fig.4b).

In the Polder du Carmel (Fig.4c), sea defenses were opened recently to create new intertidal habitat.



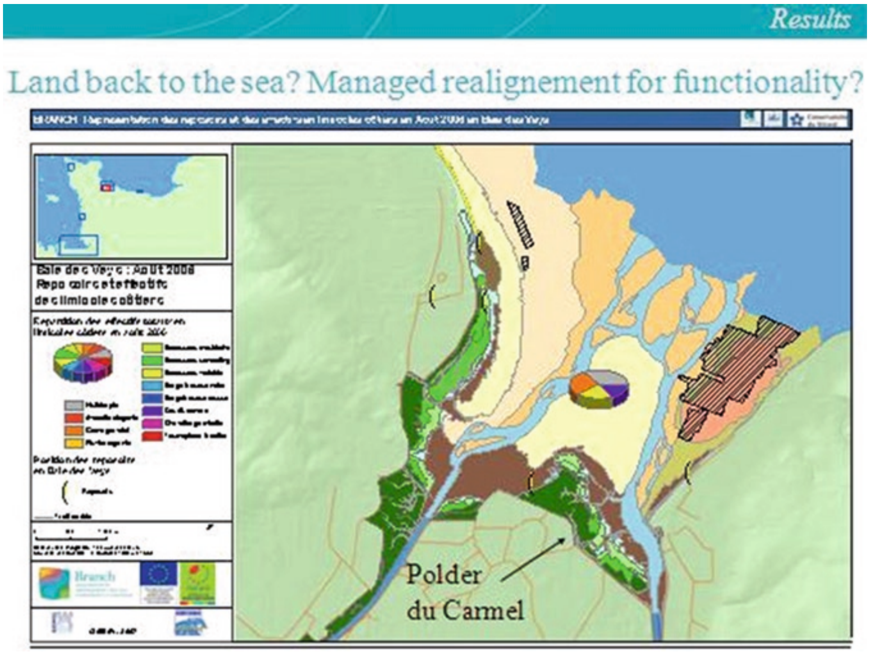


Fig. 4 (continued)

There are now large areas of saltmarsh, many wading birds, and an exceptionally rich population of crustaceans. This example shows that managed realignment can enhance coastal biodiversity and improve the resilience of the estuary to climate change.

4 Stakeholders and Climate Change

The consultation of the local stakeholders on the adaptation to climate change was one of the activities of the BRANCH program. Three seminars were convened to exchange and to discuss on this issue; they were held in Normandy (at Vains and Cantepie, 9 and 10 July 2007) and in Paris (12 July 2007). Around 80 personalities attended these three meetings, civilian servants, local stakeholders, experts, and participants in the scientific and technical activities performed within the framework of the program. The discussions were based on a series of recommendations that completed an assessment comparing the perceptions of actors on this subject, for a better integration of adaptation in the climate change strategies and policies, at all levels, international, European, national, and local (Piper 2007). These discussions started after short presentations of the models and tools developed in the program, and they aimed at detailing the territorial effects of climate change on the seacoast biodiversity in Normandy, and at giving orientations for adaptation activities concerning climate change in the future. The following developments stem from the discussions in the three seminars; they synthesize the expectations and the recommendations expressed by the participants in these meetings (Lethier 2007).

The topic has long been a subject for experts. Today, climate change has become a public and general debate. Civil services, local communities, users, private sector, and the entire society feel concerned and daily feel the effects of this global change. The reality of climate change is not avoided by anymore. In general, scientists and public stakeholders are now aware that this phenomenon is due to the modes of production and consumption. The effects of climate change are known, even it remains difficult to assess and evaluate their consequences precisely. The current knowledge makes it possible to underline that climate change is global and universal; its origins are often – at least partly – outside the area where the local stakeholders intervene. Its effects are still discreet and difficult to anticipate; in this context, it is rather complicated for the local actors to make adequate decisions and to define relevant adaptation measures.

In addition to the direct effects of climate change on species (diminution, extinction, modification of behaviors, etc.) and habitats (degradation, fragmentation, etc.), the participants insisted on the effects of climate change on the natural equilibriums and on the essential maintenance of the natural processes, sometimes less known and not well perceived, whereas they guarantee the values and the socioeconomic functions of the ecosystems. Related to social and economic issues, the participants highlighted the necessity to work towards adapting to these phenomena by preserving biodiversity services in order to guarantee human well-being and the development of the whole region. They mentioned the psychological effects of climate change on the local people who remain indecisive about the measures to be taken because of the lack of information and adequate tools to decide.

The uncertainty of the local stakeholders who have recently become aware of the climate change issue reflects their feeling of vulnerability. The participants concluded the necessity to adapt the answers to the local context; these answers should be debated, coordinated, and integrated. In their opinion, it is of paramount importance to act at the right time and at the right scale, and to take into account the ecological dynamism. It is necessary to optimize the use of the existing tools (e.g., risk prevention, urbanism, environmental assessment, and ecological networks, among others), although these tools should be reviewed and, if necessary, adapted, even supplemented by new instruments, in order to respond adequately to the situation. Because of the lack of knowledge and because BRANCH did not assess the socioeconomic aspects of the topic, the participants recommended to work further in order to reduce the uncertainty of the actors. This needs more research, especially in the fields of economy, inventory, monitoring, methodologies for adaptation, and awareness for local stakeholders and users. The governance should contribute to a better dialogue between the actors and optimization of the means.

The responses to climate change should proceed from a strategic approach, take into account the geopolitical regional context, and mix adaptation and attenuation methods. This strategy should gather all the stakeholders, public and private, and be based on a long-term vision set at the adequate geographical level; finally, it should lead to a strengthened cooperation between the actors: (1) a socioeconomic setting: the natural stocks contribute to the sustainable development, at national and regional levels. Therefore they should be considered in the public policies, notably in their territorial chapters. All uses and socioeconomic activities are concerned by biodiversity, in particular the ecosystem services; (2) improved technical instruments:

the local stakeholders lack operational tools to decide and to act; they wish to benefit from coordinated protocols and a detailed road map that guide them towards adequate measures for the short, mid, and long terms.

5 Conclusion

The debates led to seven main recommendations leading to a better consideration of climate change in land management and its effects on biodiversity, as appropriate, with adaptation measures:

- To anticipate in order not to be impacted: not dealing with the subject would quickly increase the costs; it is essential not to wait much longer before acting
- To inform and make the local stakeholders aware: the awareness of climate change is real but still very much emotional; it is based on a limited amount of information and data that need to be increased to better know and assess the socioeconomic costs of climate change
- To build up a shared vision among the stakeholders: BRANCH is the first phase of a process that should facilitate the emergence, in the long term, of a unique common vision of the local stakeholders on the land management measures to be taken, the procedure to be applied, and the means to be allocated to a global program of adaptation actions, as needed by the challenges
- To reduce the uncertainties: thanks to the progress of the improvement of knowledge and technologies, it will allow the local decisions to be taken on a topic that maintain today the local stakeholders uncertain
- To respect the natural processes: beyond the heritage value of biodiversity, which calls for relevant conservation measures, the socioeconomic values of biodiversity make essential a global approach that aims at preserving the associated complex natural processes, as well as the ecosystemic services that guarantee the sustainable development of the seacoast
- To integrate the activities within public policies: the reduction of climatic change effects needs an integration of adaptation measures in each public policy that contributes to land use management (agriculture, transport, tourism, urbanism, natural risks, etc.). However, a specific policy on climate change is essential, especially in terms of knowledge and technique improvement as well as building local capacities.
- To diversify the commands and improve the tools: the local stakeholders have strong expectations in term of various tools (technical, legal, fiscal, institutional, etc.) that allow them to set an ambitious and fair policy to reduce the climate change impacts on biodiversity.

Taking Biodiversity into Account in Territorial Planning Documents: A Methodological Approach Applied to the Marine Field

E. Seigneur and N. Mazouni

Abstract The awareness of biodiversity erosion was truly concretized in 2002 during the World Summit on Sustainable Development in Johannesburg with the signature, by 150 states, of the Convention on Biological Diversity (CBD). Its ambitious objective is to reduce the rate of biodiversity loss from 2010. In addition, strategies for biodiversity by European Community and at national and regional levels are based on the CBD.

Faced with the importance of the biodiversity stakes, it became essential to integrate this aspect within all territorial planning documents. However, if all agree on the need to preserve biological diversity, many investigations of the methodological approaches are still needed. In this context, we aimed to extract rigorous methods from the scientific world and adapt them to obtain an operational tool that can be usefulness for local managers. This work was carried out to support the implementation of the Biodiversity Regional Strategy (SRB), which was initiated by the Languedoc-Roussillon region (France).

Our work reported here started from a synthesis and an analysis of all methods used to take into account the biological richness in the current steps of territorial planning, both in the terrestrial and marine fields.

Then, specific attention was devoted to the marine domain with three regional planning documents: the maritime topic of SCOT (Scheme of Territorial Coherency), the marine ZNIEFF (Zone with Flora and Fauna Interest), and Natura 2000 in the sea. Based on theses analyses, we first established methodological suggestions for a biodiversity assessment framework.

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1 Introduction

For over a century, a gradual awareness of the consequences of human activities on the environment has led to a growing mobilization of the international community. In 1963, the "Red List" of the International Union for Conservation of Nature (IUCN) presented the beginning of the "threatened species," but the term "biodiversity" appeared only in 1992 during an international conference in Rio de Janeiro, with the adoption of the Convention on Biological Diversity or CBD (Kiss and Beurier 2000). The CBD developed an international strategy for biodiversity in 1994, a pan-European Action Plan in 2002 and, at a national scale, France developed a National Biodiversity Strategy (SNB) in 2004. The SNB itself is composed of regional strategies (SRB). One of the guidelines is to integrate "the conservation of biodiversity into all public policies of land management."

However, financial, human, and institutional resources for conservation are often limited, and facing the complexity of the biodiversity concept, the problem of setting priorities is arising. Indeed, species and protected areas are mostly the result of a hierarchy of these elements. This phenomenon is amplified in the marine

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field where, according to the Royal Society, among nearly 1.8 million species listed, only 16% are marine.

Meanwhile, governments are increasingly looking to integrate the issues of maritime territory into public policy. In May 2008, a European framework directive called the "Marine Strategy" was adopted with the objective of achieving a good ecological status in the EU by 2020, with a need for "preservation of marine biodiversity" (2008/56/CE art. 3).

The present work was done within the regional research program "Syscolag" (Mazouni et al. 2006). Our purpose is to propose a methodological framework for considering maritime issues in coastal planning. Our approach, based on an overview of all procedures currently used to assess biodiversity, aimed to support the "regional biodiversity strategy" conducted by the Regional Council of the Languedoc-Roussillon.

2 Material and Methods

The first stage of this work was to understand what was really covered by the term "preservation of biodiversity" in the context of planning documents. Two types of approaches were implemented: a documentary approach and an analysis of the actors practices.

The study of the main features led to the identification of the environmental aspects of biodiversity considered in these approaches. For this purpose, our work was focused on the analysis of existing documents: conventions, directives, and national legislation; documents of national and regional strategies for biodiversity, especially that of Languedoc-Roussillon (Biotope 2008), and methodologies used in procedures for inventory and identification of species or protected areas. This study was enriched by interviews with different actors directly involved in the process. We considered three major stakeholder groups: environmental associations (to obtain concrete information on the inventory methods); national services (to determine their role in the consideration of environmental issues in public policy); and local stakeholders (to identify sector police coordination).

Finally, interviews with local authorities in the Languedoc-Roussillon region have permitted the assessment of needs and expectations of these actors. These interviews took place around two main themes: the approach to the biodiversity concept by the actors and their willingness to implement a maritime component in some territorial planning documents.

3 Biodiversity in Planning Documents

Since 1999 (Law on Sustainable Development of the Territory), all planning documents have to take into account the environmental stakes. Since 2005, one among these is "biodiversity". Indeed, one of the French biodiversity strategy priorities is to integrate biodiversity conservation in public policies. In order to identify how the concept of biodiversity can be included in the planning document, we summarized the articulation of the main information sources that are used to build the initial state of the environment (Fig. 1).

Although often cited as an objective in itself, the "preservation of biodiversity" is generally restricted to the identification of natural areas and/or the presence of species with a protected status. Indeed, these documents should reflect a principle of "balanced and economical use of space" (art. L121, urban Code) that leads to define protected areas, as opposed to "urbanized" spaces. To offset the risk of fragmentation of these areas, planning documents (including SCOT, Scheme of Territorial Coherency) recommend preservation of the natural cuts of urbanization and definition of ecological corridors (the LOADDT[1] also recommends the establishment of a national network of biological corridors). However, these approaches are mainly based on data from land occupation, while the functional aspect of these spaces (interactions between areas and species) is not really considered.

The areas identified as "natural areas" are considered rather in descriptive terms, and "biodiversity" is associated with the presence of protected areas that are essentially based on the number of protected species.

This partly explains why the concept of "biodiversity" is often equated with species richness. In addition, attention is mainly focused on particular species listed on national or local "priority lists," usually linked to their vulnerability, their scarcity, or their symbolic nature. In terms of territorial planning, the complex

[1]Law on sustainable development of the territory no. 99–533 June 25, 1999.

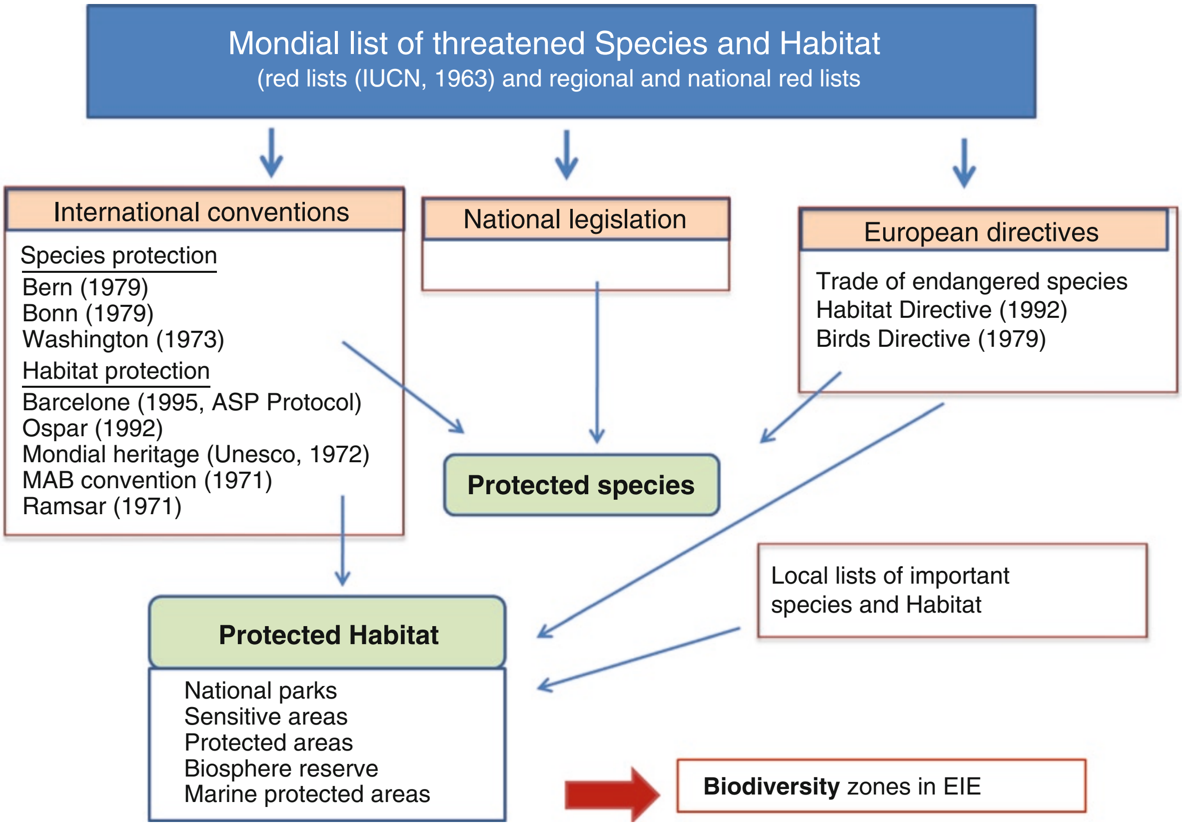

Fig. 1 Sources of information used for the elaboration of the initial state of the environment (EIE)

notion of "biodiversity" is commonly reduced to threatened or emblematic species. Knowing that biological diversity is a multi-component concept (Gaertner et al. 2008; Mérigot et al. 2007a, b; Wilsey et al. 2005), a methodological improvement of its assessment is needed. "Priority species and habitats," including Natura 2000, are in limited number. This fact is even more pronounced in the maritime field where only 10% of protected species (in France) are marine. Protected areas are developing (mainly with the Natura 2000 network), but still mainly located in coastal areas. For example, today only *8 marine habitats and 17 marine species* (belonging exclusively to the group of vertebrates) are directly involved in the creation of marine protected areas in the Natura 2000 network (Fig. 2).

However, there is often a gap between the legal status of protected areas and implementation of effective measures for their conservation. This reflects the difficulty of classifying protected areas in the territories subject to strong anthropogenic pressures such as coastlines or those historically considered as common property such as the maritime domain. In the marine environment, protection transposed from terrestrial instruments tends mainly to protect areas by wrapping it, more or less, in cotton wool. But the general principle of free movement and the issues of delimitation and monitoring of these areas make it difficult to apply a protected perimeter. Therefore, participative management practices seem generally better suited to this environment.

4 Toward a Methodological Proposal

However, especially in the context of the Marine Strategy Framework Directive 9388/2/2007 (which recommends achieving good environmental status in the Communitys marine environment by 2020), stakeholders need a way to take maritime issues into account. From the results of this study, three major lines of work have been released. They should constitute bases for the construction of a protocol to improve the consideration of the marine territory biodiversity.

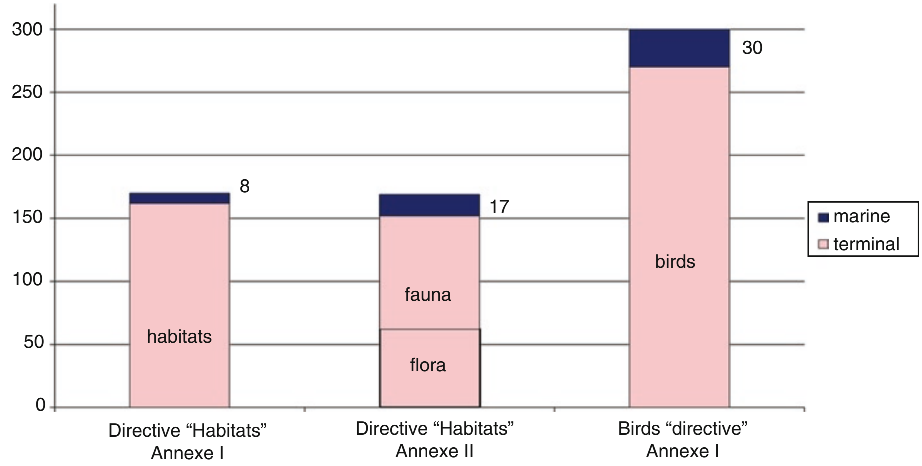

Fig. 2 Areas and species subject to protected areas of the Natura 2000 network in France

4.1 To Establish a better definition of the term Biodiversity from the beginning of the process

We have highlighted the fact that Biodiversity notion is often blurred and difficult to grasp for the local stakeholders. This is due both to the complexity of this multi-component concept and the frequent combination of this relatively new term with broader concepts, such as the preservation of the environment. The act of defining what is meant by biodiversity with the stakeholders as early as the creation of the project would thus improve their appropriation of the concept. In addition, focus on the definition of descriptors could directly prevent confusion with other concepts, such as public policy of environmental preservation. Biodiversity may also be defined in terms of issues and activities related to it or in relation to services provided by this diversity. This would therefore allow the stakeholders to see such a concept in terms of economic development for their territory.

4.2 Improve the integration of various aspects of marine biodiversity

Currently, protected marine species are few in number, and ones directly involving the creation of protected areas are even smaller. In addition, these areas are mostly coastal ones, and the objectives of the protected area (10% of the Mediterranean by 2010) or the representativeness of the Natura 2000 network are far from being achieved. To overcome this delay, several tracks should be explored:

- To increase the number of marine species in the protection texts, based on existing work of the IUCN Red List of fauna and marine flora (Boudouresque et al. 1996)
- To start out with protected areas based on species already listed (including the Berne Convention, amended in 1999)
- To expand the taxonomic groups covered by these protections (currently it is mostly mammals and birds)

In addition, criteria used to determine conservation priorities are based primarily on the vulnerability, the scarcity, or the flagship species and habitats. However, the structural and functional aspects of biodiversity are increasingly seen as having a major role in "ecosystem integrity" (Lepart 2005). It might therefore be beneficial to associate it with conventional approaches of conservation. This approach seeks to integrate groups of species and habitats less "emblematic" from a naturalistic point of view, but with a structural role or a significant commercial importance (e.g., for fisheries).

Thus, these functional aspects would be more easily integrated within the processes of territorial planning. Indeed, we have previously highlighted that "biodiversity" is mostly considered in terms of protective perimeters or a space "tank" with an intrinsic value, but disconnected from the surrounding areas.

To overcome this lack of interaction, we should move from a "naturalistic" point of view to a more dynamic one, based on the interdependence of areas, for example, thinking of natural goods in terms of uses and services related to it.

4.3 Establish a Knowledge Transfer in a Participative Way

The scientific knowledge, both of natural components and functional interaction, is incomplete, particularly in the marine field. The precaution principle would like to preserve everything, but we must take into account the territorial imperatives. The stakeholders need access to clear data in order to adopt management strategies based on the issues they face. We must therefore find a compromise between the ideal and realizable by isolating a reasonable number of indicators to answer management needs.

The protocol must also meet national and local regulations, particularly those formalized by the Marine Framework Directive. However, it should also integrate the applicability related to time and budget available. It should therefore be flexible and take into account concerns expressed by the actors to create a "dashboard," adapted to the territory, indicating the level of error associated with these actions. It appears therefore essential to involve all stakeholders in developing the protocol and to accompany them throughout its implementation so they can appropriate the method.

5 Conclusion

The concept of biodiversity, far more recent than the environmental regulations, is now mostly reduced to wrap threatened species in cotton wool. However, this approach has shown its limits: the report of the Millennium Ecosystem Assessment (2005) indicates that approximately 60% of ecosystem services that support life, such as fresh water, fisheries, or natural regulation of hazards, are degraded or unsustainably used.

Therefore, we must find other solutions, along with protected areas, to reduce ecosystem degradation. One answer is to integrate these concepts into public policies, including land use, to enable a more global view of issues and interactions between human activities and other living species that allow such activities.

Interviews conducted during this study confirmed the real desire of local stakeholders to consider the environmental aspects of their territory. But it also reflected their difficulty to deal with the term "biodiversity." Indeed, it is a complex concept issued by the scientific community, which was then used in multiple documents, but not really defined from an operational point of view. Therefore, in terms of planning, this multi-component concept of "biodiversity" is often reduced to protected species and areas. This phenomenon is even more pronounced in the marine field, for which the areas and species protected are far fewer.

This study has highlighted the lack of methodological tools for local decision makers to incorporate the concept of "biodiversity" in their coastal planning. The work done to analyze, organize, and synthesize the various approaches to take account of "biodiversity" has set up the link between the different instruments available and the needs of stakeholders to deal with legal constraints.

The present work has also emphasized the importance of a multi-disciplinary approach, involving all the stakeholders throughout the process. It should be considered as a basis for next step: the establishment of an operational protocol. This process would allow local actors to assess the marine biodiversity of their territories. For this, it will be necessary to define, with them, what the concept of "biodiversity" means in terms of the issues in ecological, functional, economic, and social ways. This will require important changes in working methods between both sides of the scientific community and local actors in order finally to elaborate an ecosystem-based approach to the management of human activities, enabling sustainable use of marine goods and services.

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Coastal managements

Temporal Changes of Benthic Macrofauna of the Mellah Lagoon (Northeast Algeria): Effects of Development Works

Brahim Draredja

Abstract The Mellah lagoon is located in the northeast of Algeria (8°20′E–36°54′N). This brackish water lagoon (865 ha) communicates with the sea by a long channel (900 m). A development operation (widening and deepening) was carried out at the end of 1980s. In order to understand the influence of this operation on benthic macrofauna, a survey of the temporal variations of the qualitative composition of the macrozoobenthos in the Mellah lagoon from 1979 to 1998 was carried out. The hierarchical analysis using the Sorensen index, taking into account samples from before and after the works on the channel, shows two periods. The first period gathers the studies done respectively in 1979, 1979–1980, and 1988, whereas the second one groups together all studies from 1991–1992 and 1998.

The difference between these two periods is in relation with the increase of the lagoon water salinity from the end of 1980, inducing a more significant specific richness.

1 Introduction

The Mellah lagoon is located in the northeast of Algeria (8°20′E–36°54′N), far from all sources of pollution, within the El-Kala national park (Fig. 1), a protected natural reserve since the adherence of Algeria in 1982 to the Ramsar Convention concerning the protection of the wetlands. This brackish water lagoon (865 ha) communicates with the sea by a long and narrow channel, which is periodically dredged, related to its filling with silt. The objective of this study is to appreciate the influence of development works on the composition of benthic macrofauna from 1979 to 1998.

2 Methodology

The first study of the Mellah macrozoobenthos was carried out by Bakalem and Romano (1979) in June 1979, this work was followed up by Semroud (1983) in 1979–1980, then by Draredja (1992) in April 1988, by Grimes (1994) in 1991–1992, and recently by Draredja (2005) in 1998. A development operation (widening and deepening) was done in the late 1980s (FAO 1987).

In order to understand the influence of this operation on benthic macrofauna, a survey of the temporal variations of the macrozoobenthos qualitative composition in the Mellah from 1979 to 1998 was carried out. The hierarchical analysis using the Sorensen index took into account samples realized before and after the work on the channel using the Excel 5 software of Clarke and Warwick (2001).

3 Results and Discussion

Semroud (1983) identified 37 species in the lagoon. During the development work on the channel in 1988, Draredja (1992) found 33 species. Grimes' (1994) inventory counted 37 species. In 1998, Draredja (2005) counted 43 species. The hierarchical analysis carried out using the Sorensen index showed two periods (Fig. 2). The first period (G1) collected the studies done respectively in 1979, 1979–1980, and 1988, whereas

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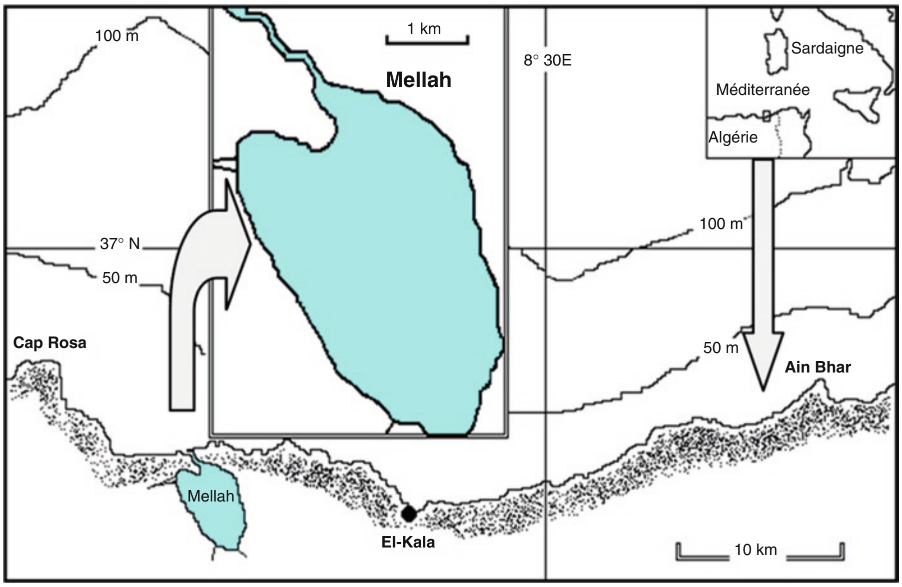

Fig. 1 Location of the Mellah lagoon

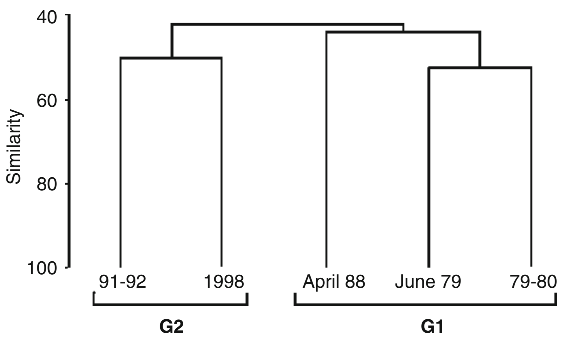

Fig. 2 Similarity (Sorensen index) between study periods of the benthic macrofauna collected in the Mellah lagoon between 1979 and 1998

the second one (G2) grouped all studies carried out in 1991–1992 and 1998. Every group presented a near or superior similarity to 50%, thus showing a very comparable qualitative composition.

4 Conclusion

It is evident that this inventory largely depends on the sampling effort, on the sampling gear used, and the study period chosen by the different authors. However, using this comparison, we want to determine if through these development works we can observe a noticeable change in the composition of the benthic macrofauna of Mellah lagoon. Two definite periods appeared: before and after the adjustment of the channel. The difference between these two periods is certainly in relation to the increase of the lagoon water salinity since the end of 1980, inducing a more important specific richness. However, these results open the way for future prospective research.

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Regional and Governmental Action Plan for Integration of Port Development and Environmental Restoration

Keita Furukawa

Abstract There is now a strong public desire to conserve and restore the natural environment of the coastline and a trend towards public participation in those activities by coastline communities. National policy initiatives in this regard have been made these last 10 years in Japan. All revised laws are clearly expressing a concern for environmental conservation and restoration. For example, the Port and Harbor Law clarified the governmental commitment to environmental conservation in the port and harbor administration, stipulating that environmental considerations must be incorporated into port and harbor development. Two case studies from Tokyo Bay and Mikawa Bay will be presented as case studies as representatives of the top-down and bottom-up approach, respectively. These studies tried to highlight the importance of role sharing with the concerned parties to find a balance of the top-down and bottom-up approaches to enable wise use of the coastal ecosystem.

1 Introduction

There is now a strong public desire to conserve and restore the natural environment of the coastline and a trend towards public participation in those activities by coastline communities. National policy initiatives in this regard include the enactment of the Basic Environment Law (1993). The Basic Environment Law states our responsibility to future generations, stipulating the framework for environmental policies that ensure the benefit of a sound and rich natural environment for both a social structural development that would reduce the load of socioeconomic activities on the environment as much as possible. Also of paramount importance is the need for international collaboration to achieve these goals. The focus is also directed towards the need for economic measures, the promotion of recycling programs, the designation of a public national holiday (Environment Day), the support for environmental education, and the importance of cooperation with private organizations. Based on this Law, the Basic Environment Plan was introduced in 1994 followed by a revision 2000. In 2001, Prime Minister Koizumi declared the implementation of "Creation of a society with harmonious coexistence of nature and humans" in his policy speech. In 2002, the National Bio-diversity Strategy of Japan was formulated. Thus, the target became clear in the first half of the 2000s.

However, implementations of such environmental targets are managed by several different organizations. After the enactment of the Basic Environment Law, revisions of the River Law (1997), the Coast Law (1999), the Port and Harbor Law (2000), and the Fisheries Basic Law (2001) were made. All revised laws clearly express a concern for environmental conservation and restoration. Followed by such revised laws, the Law for the Promotion of Nature Restoration (2002) was introduced. It gave direction for the implementation system, i.e., Adaptive Management and Public Participation.

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2 Case 1: An Adaptive Approach to the Implementation of the Tokyo Bay Restoration Plan

2.1 Environmental Situation of Tokyo Bay

Tokyo Bay is an enclosed, highly populated, and densely used bay in Japan (Furukawa and Okada 2006). The area of the bay is regarded as north of the broken line connecting the eastern and western points of Suzaki and Kenzaki (Fig.1). The bay has an open interchange with the Pacific Ocean. The Kuroshio Current in the Pacific Ocean flows near the mouth of the bay. Tokyo, the capital of Japan, is located in the catchment area of Tokyo Bay and has a population of 8 million people. Several other major cities are located in the inner bay catchment area: Yokohama, 3.5 million people; Kawasaki, 1.3 million people; Saitama, 1.0 million people; and Chiba, 0.9 million people. The inner bay catchment area contains a total population of some 25 million. The concentration of population and industry in the catchment area of Tokyo Bay has brought remarkable changes to its coastal area. Figure2 illustrates the changes in the shape of the bay from 1920 to 2002. The reclamation of tidal flat areas and shallow water areas increased rapidly from 1958 to 1976. Consequently, the surface area of Tokyo Bay decreased by 26% from 1900 to 2000. The present tidal flat area of Tokyo Bay is only 10 km^2, compared to 136 km^2 in 1900.

The area of tidal flats has decreased by about 90% over the last 100 years (Ogura 1993). A total inter-tidal zone biomass of 12.6 × 10^4 t was lost as 126 km^2 of tidal flats were reclaimed (Furota 1980). Furthermore, Kakino (1986) suggested that 3 × 10^4 t of bivalve biomass was killed by the blue tide conditions in September 1985.

The water quality of Tokyo Bay has been eutrophied even though both the residence time of sea water and the nutrient load into the bay have decreased as a result of topographical and water circulative changes. The COD (Chemical Oxygen Demand: an indicator of organic matter content) value is currently at 3–5 mg l^{-1}, while it was about 6 mg l^{-1} in the 1960s. In addition, the typical phenomena of eutrophication, such as red tides (Nomura 1998) and anoxic water (Furukawa and Ishii 2004), still occur in Tokyo Bay. The major sediment condition is sludge.

2.2 Tokyo Bay Restoration Plan

On 26 March 2003, the "Tokyo Bay Restoration Plan" was endorsed by the Council for the Promotion of Tokyo Bay Restoration, which is formed by 11 central government bodies and 7 regional government bodies (4 prefectures and 3 cities).

The Tokyo Bay Restoration Plan was initiated by a decision of the Japanese Cabinet in December 2001.

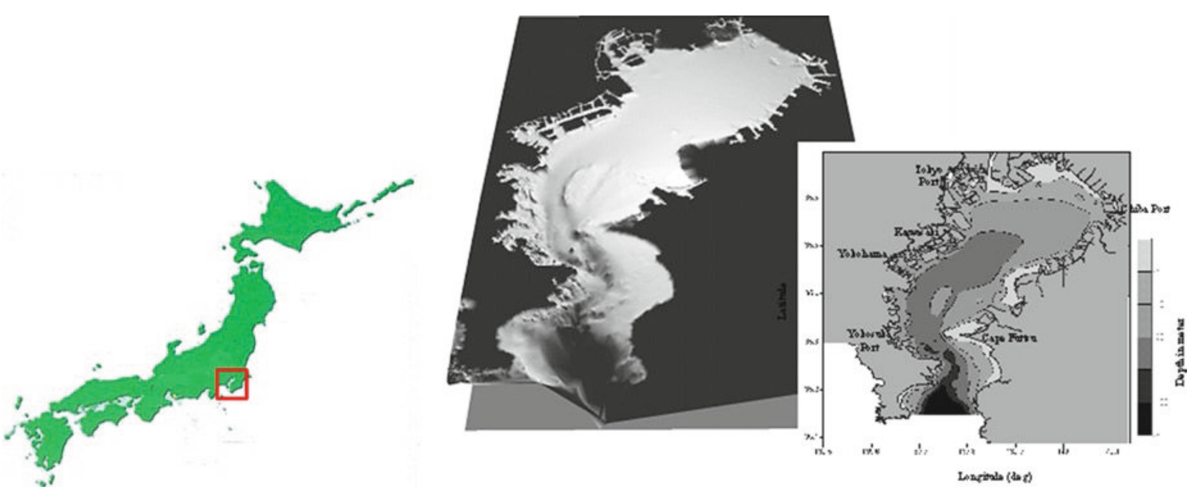

Fig.1 Location of Tokyo bay and its topography

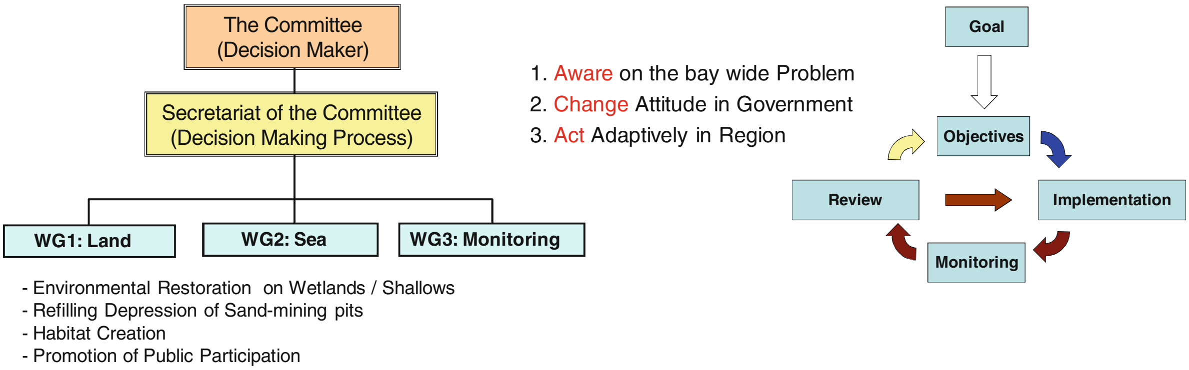

Fig.2 Structure of the Council for Promotion of Tokyo Bay restoration (*left*) and its adaptive management cycle (*right*)

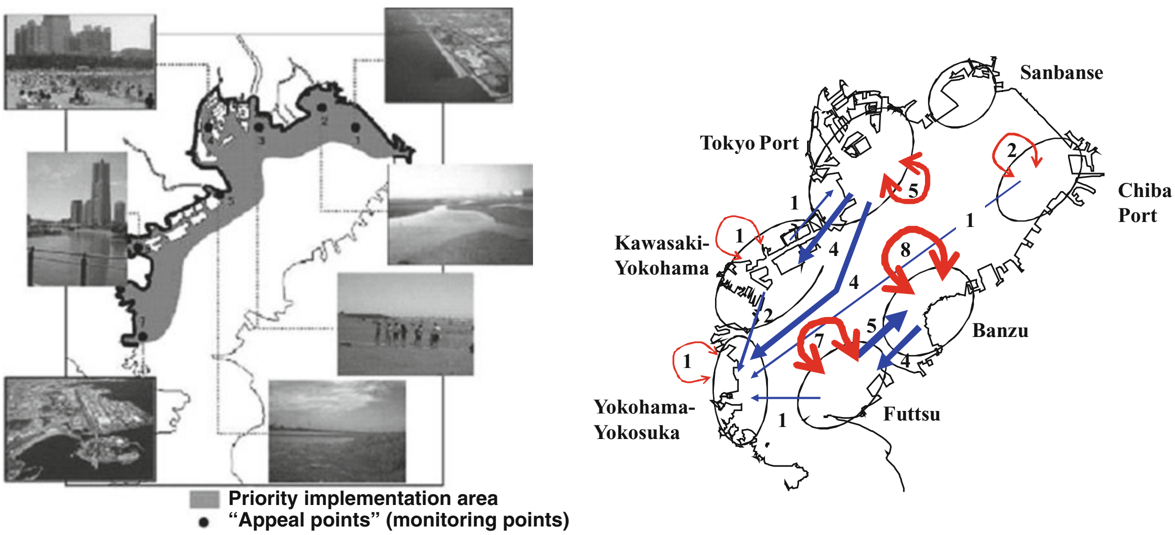

Fig.3 Zoning and monitoring points of the Tokyo Bay restoration plan (*left*) and the ecological network of the bay illustrated by the planktonic larvae link (*right*)

Seven prefectures and cities surrounding the bay and related central government ministries formed a council to promote the restoration of Tokyo Bay. The goal was set to "restore the beautiful coastal environment for enabling pleasant use and sustaining biodiversity as a wealth of capital." It is to be achieved by a collaboration among the related bodies within 10 years with an adaptive management cycle (Fig.2).

The process for assessing achievement of the goal is unique. Several monitoring points with specific targets (named "appeal points") have been set in a area of priority implementation. Each point has a specific target image of restoration, as shown in Fig.3, left. It is thus possible to assess the project in terms of specific goals. The priority implementation area is matched with the coastal line, which has a relatively weak ecological network (Hinata and Furukawa 2006: Fig.3, right). Thus, the local enhancement of ecological functions in the appeal points will contribute to the enhancement of the holistic restoration of the bay.

3 Case 2: An Ecosystem Approach in the Miakawa Port Development Plan

3.1 Mikawa Port Development Plan

Mikawa Port is located in Ise-Mikawa Bay, Japan (Fig.4). The port is now revising its original port development plan. The original port development plan

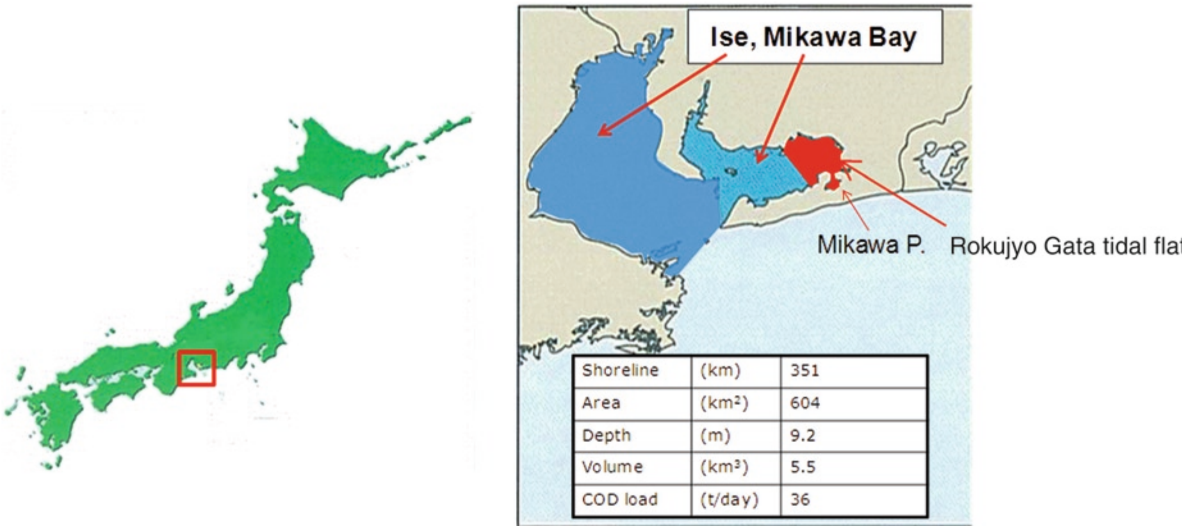

Fig.4 Location of Mikawa Port, Ise-Mikawa Bay, in Japan

for the Mikawa Port was enacted in 1995 with the Rokujo-gata reclamation. Since 2004, the revision processes have been started with the establishment of the "Mikawa Port Planning Committee." The committee members were selected from academia, port manager, local fisheries, and the public. Strategies for development, future plans, and details of zoning according to use were discussed. Finally, a revised plan that has smaller and offset reclamation of 200 ha (c.f., 500 ha in the original plan) was proposed by the committee secretariat.

Some people are worried about disturbance of the ecosystem in the rich, important natural environment of the Rokujo-Gata tidal flat because of the planned reclamation. Especially changes in the ecological network of *Asari* (*Ruditapes philippinarium*) larvae because of changes in the tidal circulation caused by reclamation have been discussed. Since *Asari* is a popular Japanese delicacy, it is an important catch for local fisheries. In April 2007, a special working party was formed including researchers and experts in the fields of the marine environment, hydrology, coastal engineering, fisheries, coastal ecology, and habitat restoration.

The working party decided to employ an ecosystem approach to assess the ecosystem impact and check the validity of mitigation countermeasures for the construction of the revised reclamation plan. In the ecosystem approach, holistic environmental conservation, assessment of the ecosystem network of *Asari* larvae, integrated mitigation countermeasures, and local water circulation were treated as major issues.

3.2 Implementation of Ecosystem Approach

To assess the impact on the Rokujo-Gata tidal flat by the reclamation, habitat suitability for a targeted species was used for testing holistic environmental conservation. *Asari* was selected as the targeted species because of its domination in the flat. Furthermore, *Asari* drew the attention of society because of its value as a fisheries resource. Mature Asari is an un-mobilized benthos, so it can be a good local environment indicator.

The suitability was quantified by the Habitat Suitability Index (HSI) method (US Fish and Wildlife Service 1981; Furukawa and Wallace 2006). The HSI is an integrated function of multiple suitability indices (SI) for physical, chemical, and biological environmental indicators that relate to the biomass of the targeted species. Thus, if we can chose suitable targeted species, the HSI can be an indicator of the holistic impact on the place we wish to assess. The HSIs were calculated for Before, After, Control, and Impact (BACI) to check the impact qualitatively.

Not only the habitat conservation, but also the ecological network was checked to assess the BACI of the reclamation by the port development plan. Field observations for determination of the detailed distribution of *Asari* lavae in the bay were conducted. Assimilated by the observation results, the numerical passive tracer transport model suggested the existence of a firm network in the bay (Fig.5). The network was not changed significantly by the reclamation.

Both local (habitat) and regional (network) effects were minimized by the revised reclamation plan, as shown in previous subsections. Nevertheless, ecosystem responses were not fully predictable. To mitigate unexpected responses in an adaptive way, integrated countermeasures were planned for keeping diverse habitat restoration and public access on shore, for example, beach construction in front of a vertical seawall, backfill of a scour hole in the bay, and installation of water/habitat quality improvement facilities on the seawall.

4 Conclusion

Both the top-down and bottom-up approach case studies showed the importance of the following points.

1. In the top-down process, sharing a good understanding of the scientific and engineering background of wetland restoration projects with stakeholders and their involvement are keys to achieving the wise use of the coastal environment.
2. In the bottom-up process, the scientifically sound ecosystem approach will contribute to the impact

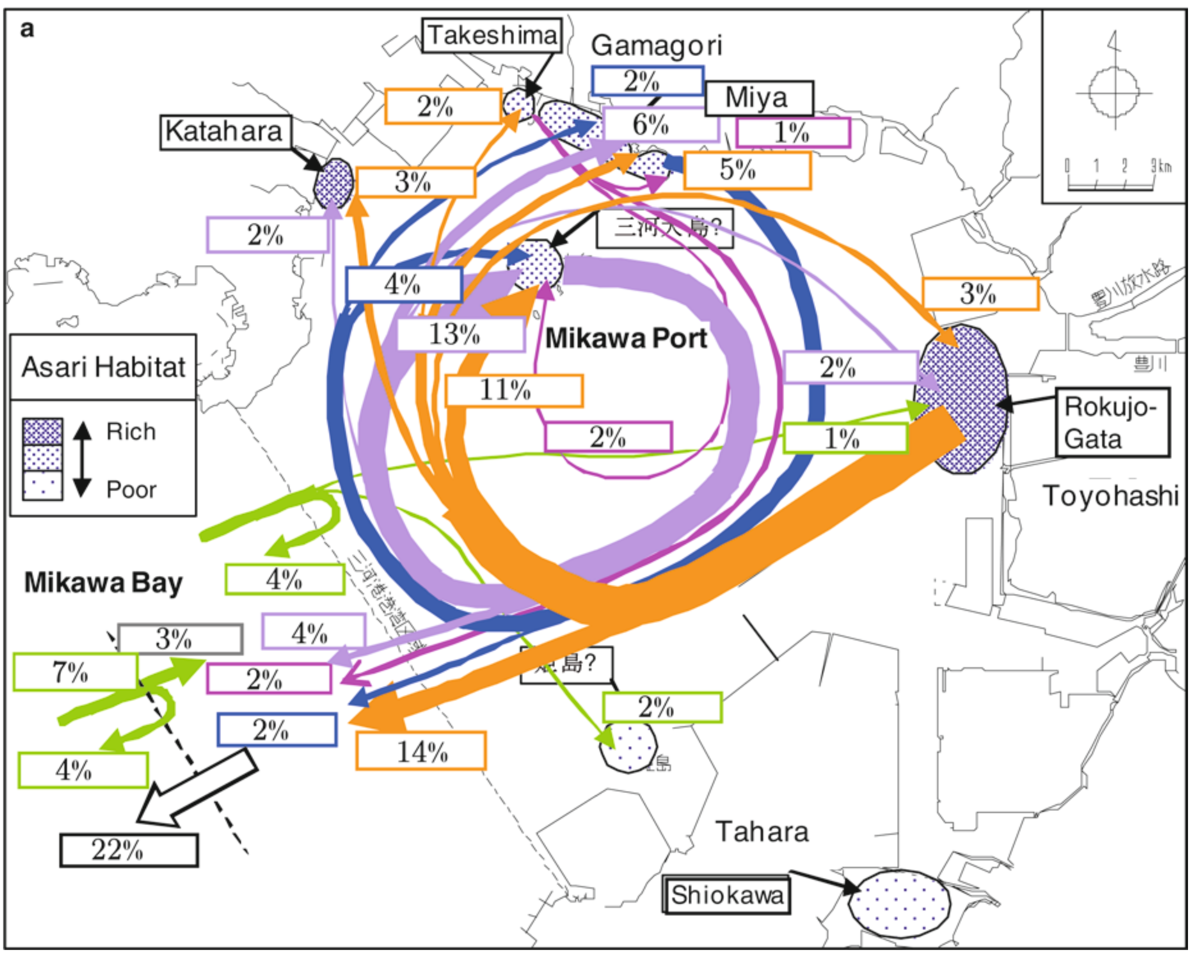

Fig.5 Schematic outline of the ecological network of *Asari* in autumn (up: no reclamation, down: with reclamation)

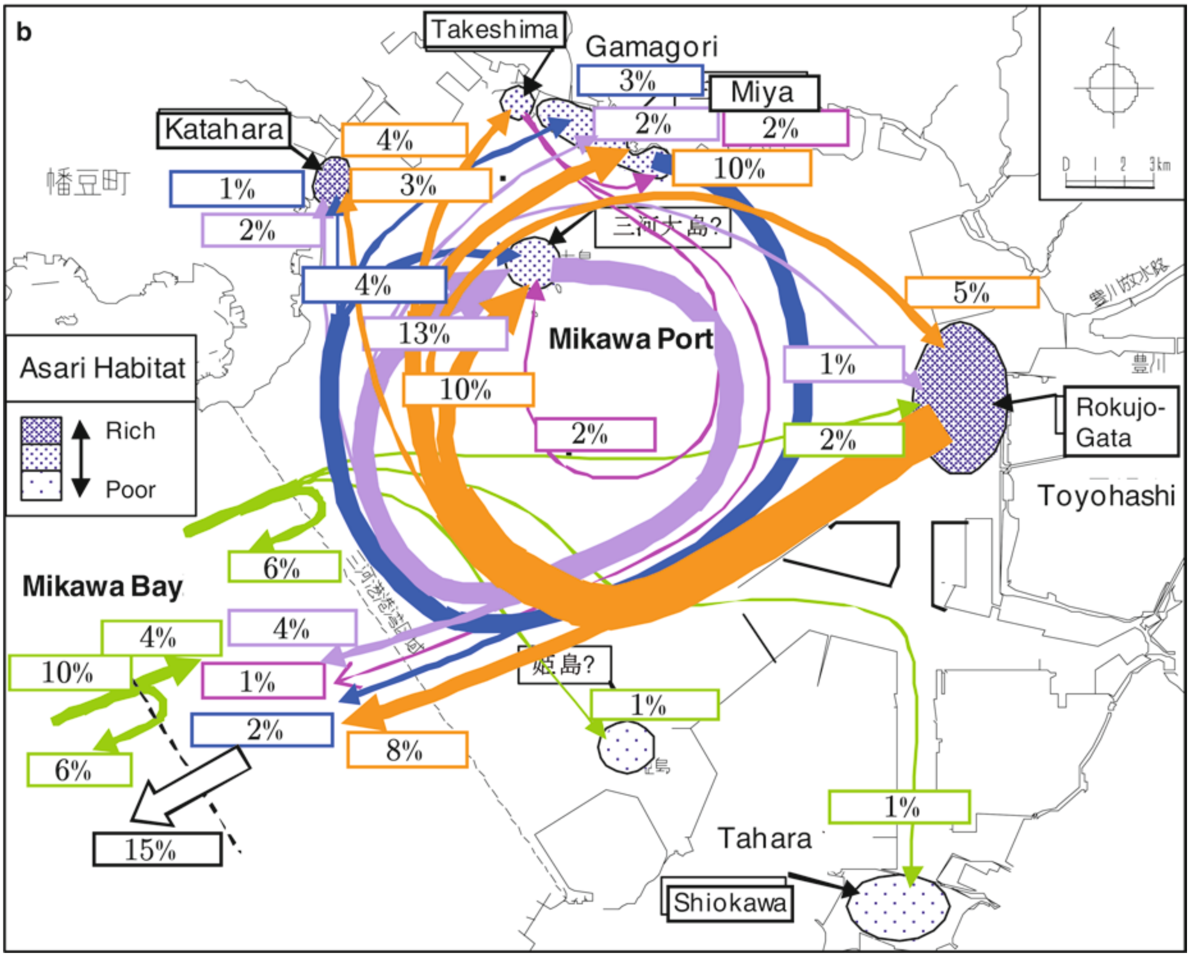

Fig.5 (continued)

mitigation plans regarding the port development of the area, promoting the sustainable use of the tidal flat ecosystem.

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Towards Integrated Coastal and Ocean Policies in France: a Parallel with Japan

Yves Henocque

Abstract Launched at the end of May 2007, the first phase of the so-called "Grenelle Environment Round Table" ended in October 2007. This large national consultation started a process for an overall environmental review and a shift in strategy in regard to issues like fighting climate change, protecting and managing biodiversity and natural environments, protecting health and the environment, while promoting economic growth, and developing an ecologically responsible democracy.

Following the screening and gathering of more than 200 proposals in all domains, a second building phase started in December 2007 in order to specify these proposals as regards their technical and financial feasibility and to elaborate the content of a first Grenelle Act, which was submitted to the Parliament in July 2008 and adopted in June 2009.

Among the 32 steering committees, one was devoted to the integrated management of the seas and the coasts under national jurisdiction (COMOP 12). This committee was chaired by a Member of Parliament and composed of about 15 members representing a number of public agencies, socio-professional organizations, and regional governments. It is comprised of the working groups covering the topics of governance and integrated management, fisheries, and impact of land-based pollutions.

Very early, the committee members agreed that there was an urgent need to fill the current void in regard to coastal and marine planning by addressing the overall governance framework through which the users and uses of oceans and seas could be regulated.

In doing so they were in line with the current European policy expressed in the blue paper on Maritime Policy and its environmental component, the Marine Strategy Framework Directive. Both integrate the need for a dual EU/regional approach, the setting up of Marine Eco-Regions as management units, and the need for cooperation among Member States for devising marine strategies, notably through the use of existing instruments deriving from national and international agreements.

Taking into consideration the European framework, the committee built up an overall governance and planning scheme, the details of which are given in its final report (COMOP 12, 2008). Most of its recommendations have since then been included into the second Grenelle Environment Act that was to be enacted by the end of 2009. The proposals were as follows:

– *Strategic planning*: management requires the definition of a vision, principles, and objectives for planning at an appropriate scale. Maritime strategic planning is used as a benchmark for maritime activity development and environmental protection. The national ecosystem-based vision and framework should be developed as a National Maritime Strategy for the Coast and the Sea, which then should allow more specific regional strategic plans to be prepared by local stakeholders and decision-makers in collaboration with the State.

Governance: management of the coast and the sea, a common good should concern all stakeholders, public and private; new forms of governance should be set up at appropriate scales from the national to local

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level: at the national level there should be a National Coastal and Marine Board (following the existing National Coastal Board); at the inter-regional level, there should be Regional Coastal and Marine Boards for each of the selected marine eco-regions or maritime basins.

– *Funding*: beyond the need to maintain the existing resources for the management of the sea, the COMOP 12 emphasized the need for a funding instrument and mechanism devoted to the maritime activities and related programs or projects. This funding would be sourced from maritime activities taxes (licenses) as well as water taxes through corresponding water agencies and would take the name of National Fund for Management of the Coast and the Sea.

Since then, these principles and goals, soon to be confirmed by the law, have been further developed in the frame of the Grenelle of the Sea, which will last till the end of 2009.

1 Introduction

Like in most European countries, the institutional framework for coastal zone management in France is comprised of two elements: a land use planning system administered by territorial authorities at one or more sub-national territorial levels and sectoral laws, which are usually reflected in sectoral administrations within the national government.

Since 2007, land use planning, environmental protection,[1] and nature conservation have been, for the first time, regrouped under the same ministry: the *Ministry for Ecology, Energy, Sustainable Development, Land-use, and Regional Planning (MEEDDAT)*. The *Ministry of Agriculture and Fisheries* is responsible for fisheries and aquaculture. The *General Secretariat for the Sea* is under the Prime Minister's authority and in charge of coordinating the State policy and actions at sea. As a major coordinating instrument, the *Inter-ministerial Committee for the Sea*, the secretariat of which is entrusted to the *General Secretariat for the Sea*, is supposed to meet once a year for deliberating about the government's marine policy at the national and international levels in all kind of maritime activities. Under the *Ministry of Interior*, the *Ministry of State with responsibility for Overseas France* is in charge of the overseas territories the Exclusive Economic Zone (EEZ), which represent more than 90% of the total French maritime territory.

At the level of the *Département*,the State is represented by the *Prefet* assisted by the numerous decentralized administrations for Equipment, Agriculture, and Fisheries, Maritime Affairs, Environment, Health, Research, etc. Since 1993, all activities related to water as a resource are coordinated for each water basin by a *Water Inter-service Mission (MISE).*

The decentralization of power to local governments started only at the beginning of the 1980s through the Decentralization Act enacted in 1982, creating a new regional authority in between the central government, its *Départements*, and the municipalities: the Regions and their regional council with still limited power regarding the development and protection of the coast though they recently were given the responsibility of ports. At the municipality level, the most important planning tool in the hands of the Municipal Council is the *Urban Local Plan* (*PLU*), for which it has the power of issuing permits for uses included in the PLU, including the seashore.

2 Recent Elements of Reflections and Decisions

2.1 In Europe

In 2002, the European Parliament and Council adopted a Recommendation on implementing Integrated Coastal Zone Management in Europe, which asked Member States to undertake a national inventory of coastal legislation, institutions and stakeholders. Based on this information, Member States were also asked to develop national strategies, following eight key principles, to implement an integrated approach to management of costal areas and to report progress to the European Commission by February 2006.

[1] Relevant legislation on water quality includes the Water Act (1992) which provides for two types of integrated water basin management plan: *Schéma Directeur d'Aménagement et de Gestion des Eaux* (*SDAGE*) and *Schéma local d'Aménagement et de Gestion des Eaux* (*SAGE*).

2.2 In France

In early October 2005, the Strategic Analysis Center and the General Secretary of the Sea were asked to jointly engage in strategic reflection in order to nourish France's contribution to the definition of an integrated maritime policy for the European Union as well as to develop the constituent elements of a national maritime policy. In December 2006, the so-called *Poseidon* working group created for this purpose came up with a report proposing a vision, including (1) the boosting of safety and security in maritime activities; (2) improvement of knowledge of the marine system; (3) contribution of the maritime sector to the economy, growth, and employment, in particular through innovation and competitiveness; and (4) protection and exploitation of France's EEZ resources.

The working group then recommended the following measures:

Systematic consideration of public policies' maritime dimension throughout the parliamentary and governmental decision-making process.

Setting up complementary maritime and land policies, considered a condition of success where integration relates as much to "vertical integration" (synergy between actors at different levels of governance) as to "horizontal integration," including sectoral and territorial policies.

Mobilization of all actors, which include the central and local governments, the private sector and civil society, as well as supranational institutions like international courts, regional seas conventions or interregional networks.

Strengthening of local cooperation and responsibility, which means strengthening the decentralization of decision-making and encouraging new forms of governance.

Align with and use the European framework as an incentive, where France seeks, together with the other Member States and its partners, to contribute effectively to turning Europe towards the sustainable development of its coasts and regional seas.

While the demand for a more bottom-up approach has continued to grow, it is in such a context that the *Grenelle of the Environment* and later the *Grenelle of the Sea* were launched, both meant to stir up a national debate that would lead to recommendations and State commitments legally framed by the two successive *Grenelle Environmental Acts*, including, but not exclusively, the coast and the sea.

2.2.1 The European Framework

A "Blue paper" has been used by the EU since 2007 as a tool for its integrated maritime policy. This concerns the surveillance activities, the maritime spatial planning, and the Integrated Coastal Management (ICM), the European Marine Observation and data network.

The final target is to reach a sustainable use of the oceans and the seas, to develop the knowledge and innovation base, and to increase the quality of life in coastal regions. At the same time, Europe wishes to maintain some leadership in international maritime affairs and to increase its maritime visibility.

In the framework of Integrated Maritime Strategy Directive, underpinning the European Union maritime policy, the choice was an ecosystem-based approach to management of human activities impacting the marine environment, including:

Monitoring and assessment issues

Particular challenge of hazardous substances

Two important deliverables from the consultation process (2002–2004)

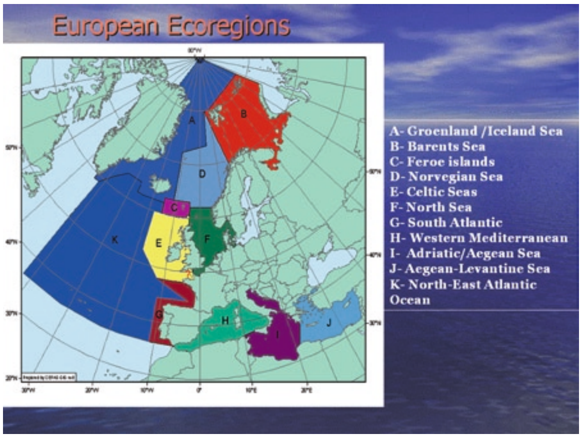
A guidance document on the application of the ecosystem-based approach to the marine environment

Study on the identification of European Marine Regions on the basis of hydrological, oceanographic, and biogeographical features

2.2.2 Marine Strategy Directive

A framework for the development of marine strategies has been established in order to achieve good environmental status in the marine environment by 2021 at the latest. Member States marine strategies have defined the following points:

- Preparation (2009–2015): Initial assessment
- Determination of good environmental status
- Environmental targets, monitoring program
- Programs of measures operated by 2018 at the latest
- Member states' progress report every 3 years
- Marine strategies review every 6 years



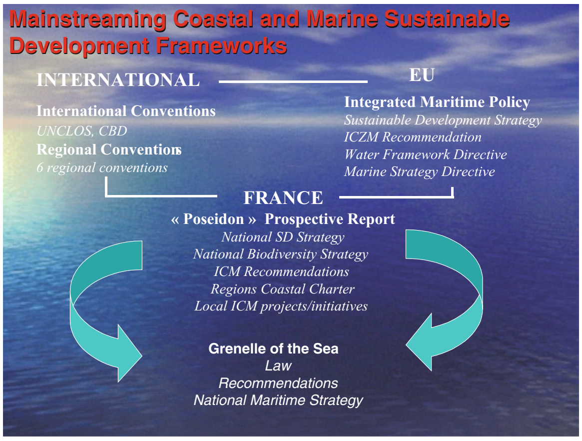
European ecoregions are

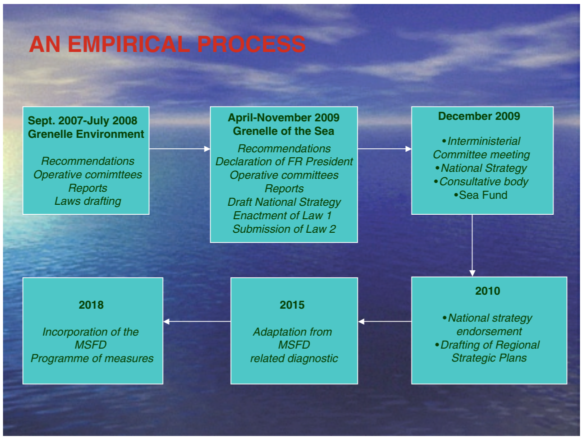
The case of France as a Europe Union member state.

In France, there was a national meeting to define the future directions of the environmental policy. Different parts of the society, such as scientists, politicians, associations, syndicates, etc., have been gathered to collect their advice as a basis for a national decision.

Like the preceding meeting that took place on Grenelle street (rue de Grenelle), this last meeting also was called "Grenelle de l'environnement." Another meeting devoted to marine and maritime problems was also called "Grenelle de la mer."

Several relationships have been established between the different levels:



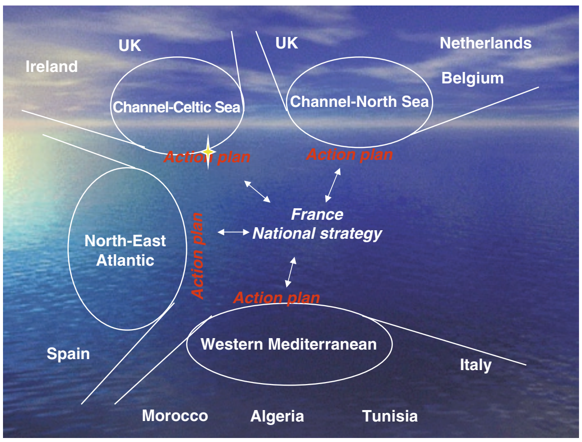


Content of "Grenelle Environment Act 2":

1. Climate change mitigation
   - Reduction of building energy consumption
   - Urbanization measures
   - Transport measures
   - Research for sustainable development
2. Biodiversity and natural habitats
   - Loss of biodiversity countermeasures
   - Water ecological good quality
   - Diversification of agriculture and forestry
   - Integrated coastal and ocean management
3. Environmental and health risks prevention
4. State stewardship (exemplary)
5. Governance (role of local governments, information, conflicts, consumption, education, etc.)
6. Specific measures regarding the overseas territories
   - Contents of the sea-related chapter
   - Three governance level strategies

1. National
   - Government: drafting a national Maritime strategy
   - Stakeholders: National Coastal and Ocean Board give advice
2. Regional (maritime basins)
   - Central/local governments co-chair each
   - Regional Coastal and Ocean Board composed of representatives of all stakeholders
   - Prepares an action
3. Local
   - Strategic plans are adapted by each coastal region and implemented at the local level
   - Challenges

– Looking the other way around:
– From ocean to coast and water basin
– From large marine ecosystems to effective local management
– Developing the ecosystem-based approach
– Making planning instruments compatible
– Building on the existing system while promoting local stakeholders' participation
– Behavioral changes



2.2.3 France and Japan National Contexts: Fertile Grounds for Further Collaboration

France and Japan are two very different countries from the geographical, environmental, socioeconomical, and cultural points of view, but they are both going through the development of their own maritime policies.

France is part of the European Union, at the edge of the European continent, bordered by three oceans or semi-enclosed sea bodies, the Atlantic ocean, the Channel/North Sea, and the Mediterranean Sea.

Japan is part of the Pacific Ocean countries community, as an archipelago located offshore of the Asian continent and thus surrounded by ocean and semi-enclosed sea bodies, mainly the Pacific Ocean, the Okhotsk Sea, the Japan Sea, and the East China Sea in the south.

In Japan, the "Bill for the Basic Act on Ocean Policy" took effect in July 2007. The contents of this new law "define the basic principles of Japan on the ocean, clarify responsibilities of the national government, local governments, business operators, and citizens, formulate the Basic Plan on Ocean Policy, specify the basic items concerning measures on the ocean, and stipulate the establishment of the Headquarters for Comprehensive Ocean Policy for the purpose of promoting these measures in a comprehensive and systematic manner, aiming at realizing a new ocean-oriented nation" (T. Kuribayashi 2008[2]). As the basis of the Basic Ocean Plan, 12 basic measures have already been agreed upon. They include the promotion of development and conservation of the marine environment within the EEZ and other areas, securing the safety and security of the oceans, including maritime transport, promotion of ocean science and technology, ocean industries, and their international competitiveness, integrated coastal management, and enhancement of citizen's understanding of the oceans, conservation of remote islands, and international cooperation.

On the French side, the ocean-related legal framework will be part of an overall environment-related law leading to the making of a national maritime strategy founded on a number of recommendations issued from a national debate that at least should last till the end of 2009.

The core of these recommendations is expected to be more specifically reflected into regional strategic plans within the framework of the national maritime strategy and along the European maritime policy guidelines and incentives.

In these respective national contexts, both countries have numerous ongoing local ICM initiatives taking place in specific regional and local context. The question is therefore how to progressively build up, from

[2] Tadao Kuribayashi 2008. Thinking About the Basic Ocean Law; Enactment of the Basic Ocean Law and its Significance. Ship & Ocean Newsletter no 10, September 2008.

national to local and vice-versa an adequate, a viable and well-supported management process that can help each nation to consolidate its national framework, hence promoting their ocean-state position in their respective maritime region.

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Pôle de compétitivité Pôle Mer PACA: Maritime Cluster in Provence–French Riviera Region

Patrick Baraona and Guy Herrouin

Abstract What is a competitiveness cluster?
This is the combination of companies, training centers, and public and private research centers, engaged in a partnership in a geographical area (Provence-Alpes-Riviera Region for Pole Mer PACA).

The Maritime Cluster in Provence-Alpes-Riviera is one of the 17 French clusters with international objectives.

To create synergies around common innovative maritime projects, this R&D projects will meet four main requirements:

To create new wealth with high added value and qualified jobs.

To be able to position itself on world markets that are characterized by a high potential for growth.

To define objectives and means of an efficient strategy for economic development and innovation.

1 Pôle Mer PACA and Pôle Mer Bretagne: twin clusters

These two French marine competitiveness clusters, are coordinated across two major coastal regions: Brittany and Provence-Alpes-Côted'Azur. These two regions have significant potential for industrial growth in the marine sector (700 companies), and 75% of French research in the field is concentrated here. The singular history and the specific nature of the marine environment of each area encompass many different yet complementary problems.

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2 Key Figures in Provence-Alpes-Riviera Region

2.1 Figures:

More than 1,000 public researchers in this region
60,000 jobs directly in the maritime sector
Marseille, the biggest French harbor and third oil harbor worldwide
Toulon, the largest French military maintenance harbor
One of most important areas in the world for nautism

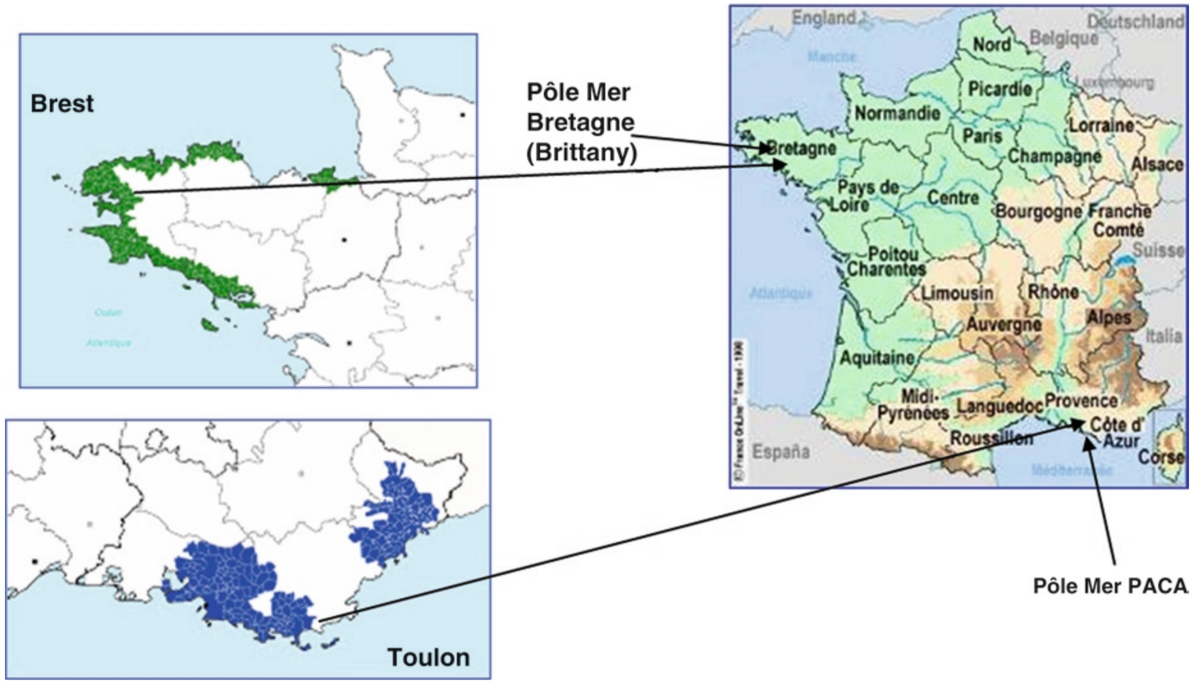
2.2 Pôle Mer PACA:

More than 250 members (2/3 companies and 1/3 research centers), more than 60 labeled joint projects, €220 million quoted in initial R&D projects
Granted by the PACA Region Council

2.3 Objectives within 5 years:

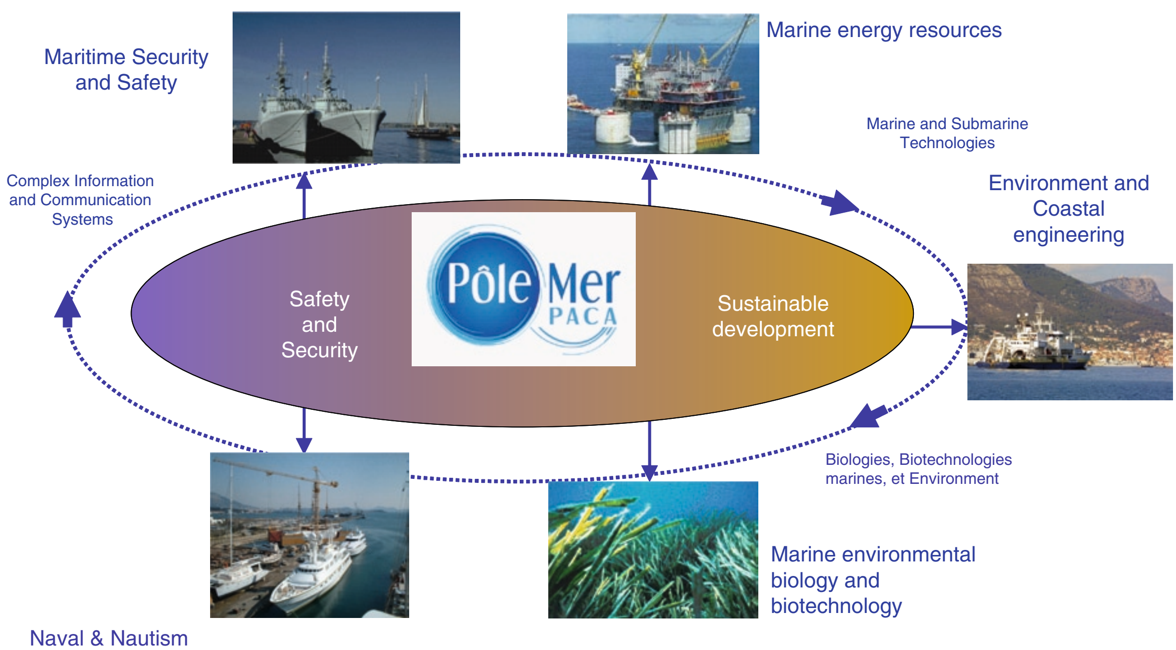
To be recognized at a world level as a centre of expertise in
The knowledge and monitoring of the maritime environment
To develop services in coastal engineering and environment
To develop a global offering of products and innovative services for shipping, offshore projects, renewable energies, and for "homeland security"

H.-J. Ceccaldi et al. (eds.), *Global Change: Mankind-Marine Environment Interactions, Proceedings of the 13th French–Japanese Oceanography Symposium*,
DOI 10.1007/978-90-481-8630-3_35, © Springer Science+Business Media B.V. 2011



2.4 Key technologies covered by the cluster:

Electromagnetics and acoustics
Software engineering
Onboard electronics
Underwater technologies
Signal processing
Materials
Biotechnologies
Robotics and automation
Ship engineering



2.4.1 Some examples of projects awarded by Pôle Mer PACA selected in environmental, energy, and marine biological resources

Pole Mer is particularly deeply involved in the coastal environment, physical–chemical–biological processes, aquaculture, biodiversity, and observation and monitoring technologies.

3 Environmental and coastal management

The group plays a key role in environmental challenges linked to the sea, combining valorization of human and economic activities, all the while safeguarding resources some projects:

GIRAC: Integrated management of coastal sewer network discharges

✓ GIRAC project aims at improving bathing water by developing operational tools enabling to know in real-time sanitation discharges and their evolution with regard to meteorological conditions.

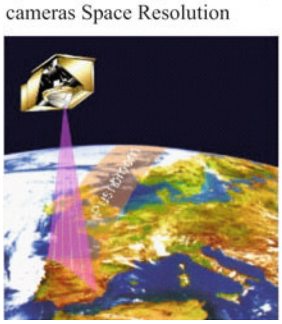
✓ Leader: Veolia Environment, partners: SME's, lab's: Ifremer,…

RENOUV 'EAU : a desalination plant with a renewable energy source

- Feasibility study for coupling renewable energies with managed water ressources
- The aim of this project is to perform a feasibility study comparing the whole coupling possibilities between renewable energy sources and processes for sea water desalination in coastal areas.
- Veolia Eau: leader, SME's, lab's partners: Institut Paul Ricard, Ifremer,…

REGICOLOR : Exploitation of the satellite data of color of water

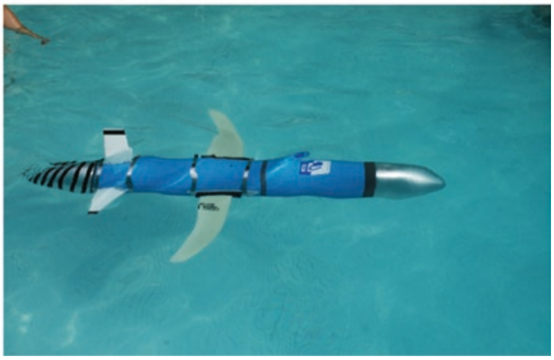
Spectrometer imagor MERIS (EU) with sweeping 15 spectral bands, distributed on 5 cameras Space Resolution



- Leader: ACRI (SME)
- Partners: Alcatel Alenia Space, Oceanographical Observatory of Villefranche, Ifremer
- REGICOLOR Application : Follow-up of coastal water : turbidity, eutrophication

SEA EXPLORER: Submarine exploration systems

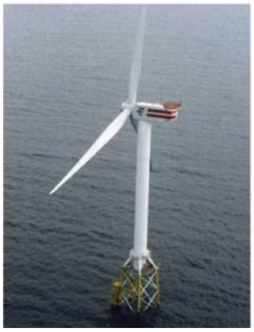
- physical (current, temperature) and biogeochemical (salinity, concentrations of components, etc.…)
- SME'S: ACSA leader, ACRI partner, lab's (Ifremer, COM, OOV… partners)



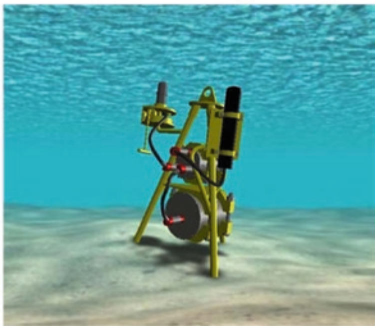
Concerted development of coastal areas

- The Mediterranean coasts are faced to important pressures: demography, tourism, climate risks, pollution and management of water resource etc.
- But also a vital potential for natural resources and biodiversity
- Pôle Mer is seeking to contribute to the valorization of coastal zones with ambitious settlement projects which take a long-term approach to environmental challenges,

- **Renewable energies**



- **Deep offshore engineering**



SHAMASH:Production of lipidic biofuel from microalgae

- The goal of this project is to test a new procedure for high-yield production of microalgae for use in the production of a new biofuel.
- High productivity: 30 times agrofuel, 80% lipidic content,…
- Research project: lab's:INRIA, Ifremer, OOV, CEA,…

ECIMAR: Indicators for biodiversity and valorization

- Better knowledge of marine biodiversity and characterization of chemical markers for the purpose of environmental indicators for perturbations from natural or anthropic origins,
- Valorization of marine biodiversity

- Project leader: CNRS Inserm
- Research partners COM/Dimar, USTV, MNHM, Centre Phytophar, Inserm, CEAB Blanes (Spain), University of Barcelona, and Oceanographic Center of Crete

First results of Pôle Mer PACA 2005–2007

	Environment and coastal engineering	Marine biological resources	Maritime energy resources	Naval and nautism	Marine safety and security
Number of studied projects: 134	44	17	18	29	26
Number of financed projects: 42	16	5	3	10	8
Total cost of projects: 213M€	59 M€	17 M€	5 M€	37 M€	95 M€

3.1 GIRAC: Integrated management of coastal sewer network discharges

- GIRAC project aims at improving swimming water by developing operational tools enabling know in real-time the extent of sanitation discharges and their development with regard to meteorological conditions.
- Leader: Veolia Environment, partners: SME's, laboratories, Ifremer, etc.

3.2 RENOUV EAU: a desalination plant with a renewable energy source

- Feasibility study for coupling renewable energies with managed water resources
- The aim of this project is to perform a feasibility study comparing the whole coupling possibilities between renewable energy sources and processes for sea water desalination in coastal areas.
- Veolia Eau: leader, SME's, laboratory partners: Institut Paul Ricard, Ifremer, etc.

3.3 REGICOLOR: Exploitation of the satellite data of the color of water

Spectrometer imager MERIS (EU) with 15 sweeping spectral bands, distributed on five Space Resolution cameras

- Leader: ACRI (SME)
- Partners: Alcatel Alenia Space, Oceanographical Observatory of Villefranche, Ifremer
- REGICOLOR Application: Follow-up of coastal water: turbidiy, entruophication

3.4 SEA EXPLORER: Submarine exploration systems

- Physical (current, temperature) and biogeochemical (salinity, concentrations of components, etc.)
- SME'S ACSA leader, ACRI partner, laboratories (Ifremer, Com OOV, and partners)

3.5 Concerted development of coastal areas

- The Mediterranean coasts are faced with important pressures: demography, tourism, climate risks, pollution, management of water resources, etc.
- But also a vital potential for natural resources and biodiversity.
- Pôle Mer is seeking to contribute to the valorization of coastal zones with ambitious settlement projects that take a long-term approach to environmental challenges.

4 Marine Energy Resources

Concentrate global competences and know-how in offshore engineering for marine energies: petrol, renewable energies.

5 Biology, Marine Biotechnology

5.1 SHAMASH: Production of lipid biofuel from microalgae

- The goal of this project is to test a new procedure for high-yield production of a new biofuel.
- High productivity: 30 times agrofuel, 80% lipidic content, etc.
- Research project, laboratories: INRIA, Ifremer, OOV, CEA, etc.

5.2 ECIMAR: Indicators for biodiversity and valorization

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- Valorization of marine biodiversity
- Project leader: CNRS Inserm
- Research partners COM/Dimar, USTV, MNHM, Centre Phytophar, Inserm, CEAB Blanes (Spain), University of Barcelona, and Oceanographic Center of Crete

Funding:

- Public: National Interministry Fund, Agencies: OSEO (SMEs) ANR (Research), regional funds for cooperative research "projects PRIDES label"
- Private: private investors (venture capital)

Depending on type of project

5.3 Pôle Mer PACA: Main international objectives

To create partnerships with foreign companies and research centers

Interested for studying partnership with Japanese clusters

Business – Innovating to increase competitiveness and gain control of world markets

Accumulation of Bromoform, a Chlorination Byproduct, by Japanese Flounder, *Paralichthys olivaceus*

Toshio Iibuchi, Takeya Hara, Shuji Tsuchida, Seiji Kobayashi, Ichiro Katuyama, Tsutomu Kobayashi, and Michiyasu Kiyono

Abstract Bioconcentration of bromoform by Japanese flounder *Paralichthys olivaceus* was studied. In the experiments, chlorine was produced by electorolysis of seawater, and bromoform was generated slowly in the flowing water until residual oxidants were treated with thiosulfate. The uptake rate constants were 1.3–1.7 h^{-1} in the muscle and 17.1–28.7 h^{-1} in the liver.

The biological half-lives in the muscle and liver were 3.4–5.1 h and 3.1–4.3 h, respectively. Thus, considering this lower persistence of bromoform in marine organisms, we concluded that continuous chlorination at lower concentrations would be appropriate for an anti-biofouling method at power plants in terms of environmental preservation.

1 Introduction

Power plants using seawater in their cooling water circuits usually suffer from biofouling. Chlorine is often used to lessen biofouling fixation and damages. When chlorine is added to seawater, it oxidizes the bromide ions, yielding hypobromous acid (HOBr). Hypobromous acid reacts with organic matter, and then various organohalogens are generated (Fig. 1). Among them, bromoform is considered the major component (Jenner et al. 1998). The acute toxicity of bromoform was reported to be weak (Taylor 2006). If fish excrete bromoform easily, we do not need to worry about the chronic toxicity of bromoform. The aim of this study was to clarify the bioconcentration of bromoform in Japanese flounder *Paralichthys olivaceus*, which is one of the main fish species reared at some fish culture farms utilizing heated seawater from power plants in Japan.

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2 Methods

The experiments were performed under the flow-through conditions. Chlorine was produced by electorolysis of seawater, and bromoform was generated slowly in the flowing water. The residual oxidants in seawater were decomposed by thiosulfate treatment before it was put in the fish exposure tank (Fig. 2). Fish (30–36 cm in TL) were exposed to bromoform at two different concentrations, 28×10^{-6} (the first experiment) and 9.7×10^{-6} g/l (the second and the third experiment) respectively. After the uptake experiment (41 h for the first experiment and 24 h for the second), the fish were transferred to clean water. The fish were reared for 16 h for the first experiment and 8 h for the second, and the excretion of bioconcentrated bromoform was studied. During the experiments, the temperature of the seawater was maintained at 19.5–20.0°C.

The concentrations of bromoform in seawater were determined using gas chromatography equipped with mass spectrometry (GC/MS) after purge and trap extraction.

H.-J. Ceccaldi et al. (eds.), *Global Change: Mankind-Marine Environment Interactions, Proceedings of the 13th French–Japanese Oceanography Symposium*,
DOI 10.1007/978-90-481-8630-3_36, © Springer Science+Business Media B.V. 2011

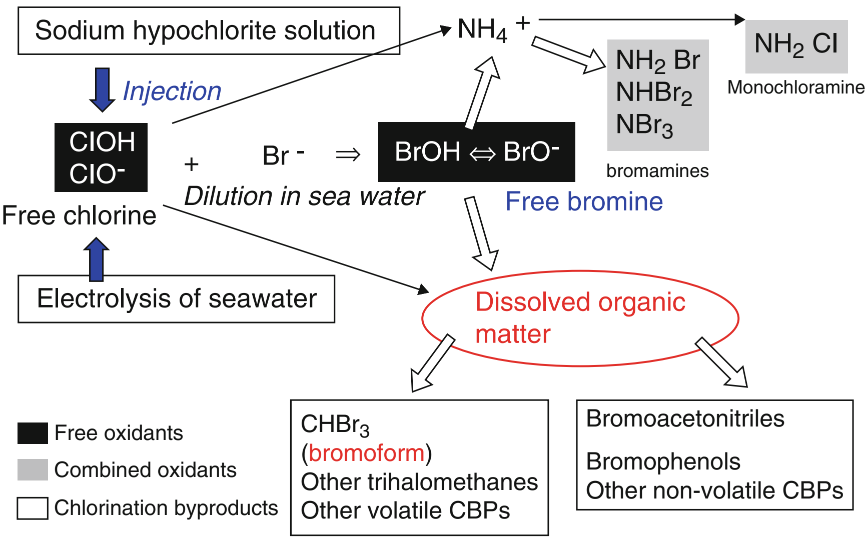

Fig. 1 The formation of the chlorination byproducts (CBPs) in chlorinated seawater. Redrawn from Khalanski (2002).
In seawater, hypochlorous acid oxidizes the bromide ions, yielding hypobromous acid. It reacts with dissolved organic matter, yielding various kinds of halogenated organic materials, including bromoform

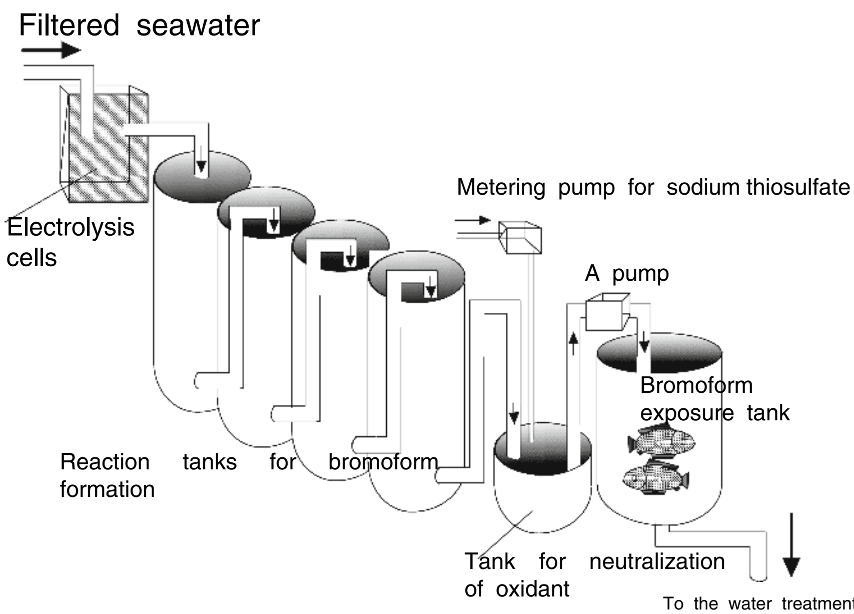

Fig. 2 Schematic arrangement of the tanks for the bromoform exposure experiments on Japanese flounder, *Paralichthys olivaceus*. The experiments were performed under flow-through conditions. Seawater was electrolyzed yielding bromine. It reacted slowly with dissolved organic materials in the reaction tanks. After the oxidant was neutralized, seawater was introduced into the exposure tank

Chemical analysis of fish samples was conducted at the Japan Food Research Laboratories (JFRL) with their professional technique. In brief, the muscle (2–4 g) or liver (1 g) tissue was homogenized in the solution containing 4% trichloroacetic acid solution (20 ml) and hexane (4 ml) in the centrifuge tube. NaCl (8 g) was added to the tube, and then it was shaken and centrifuged. Then 2 µl of the hexane layer were taken for the determination of bromoform using GC/MS. Lipid contents were also determined by the Soxhlet extracting method. In the preliminary study, using the fish bought from a firm utilizing heated sea water from a power plant for raising them, we clarified that the concentrations of bromoform were affected by lipid contents in the muscle of fish (Fig. 3). Therefore, the bromoform concentrations in the muscle and liver were corrected by the lipid content in these tissues and organs.

For the first experiment, the method for data correction was that the overall mean of the lipid contents was multiplied by each of the lipid basis concentrations of bromoform. For the second and the third

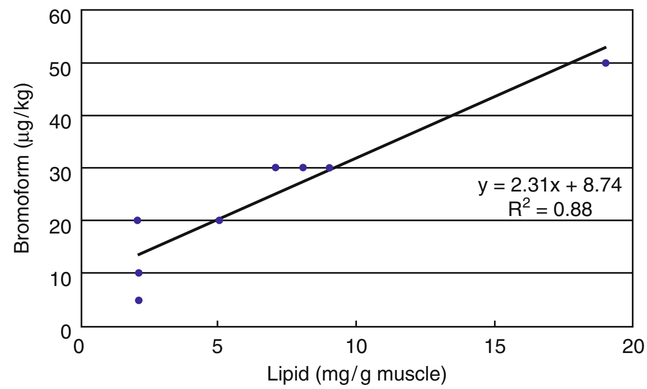

Fig. 3 The relationship between lipid contents and concentrations of bromoform in the muscle of fish raised in the water from a power plant in Japan. The concentration of bromoform was affected by the lipid content. All animals were raised in the same conditions

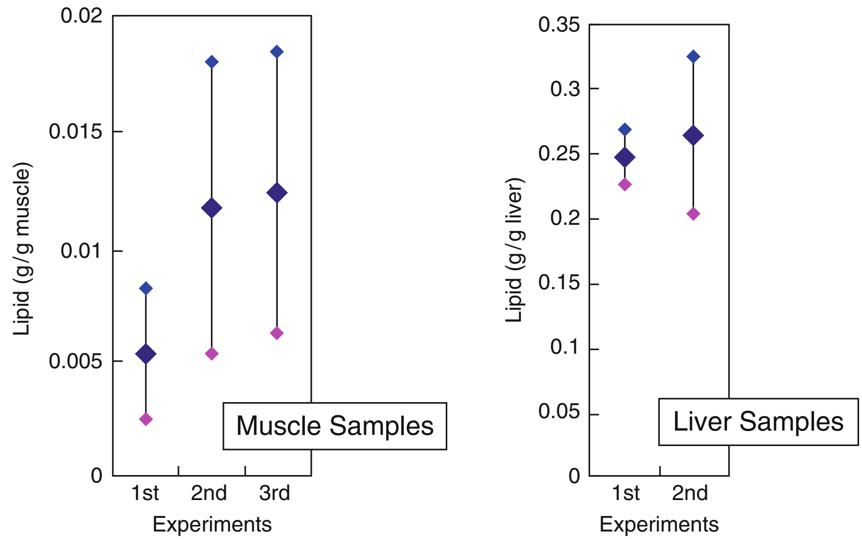

Fig. 4 Concentration of lipid of the samples in the bioconcentration experiments. In the second and the third experiment, there was a large variation in the data for lipid content

experiments, the standard deviations of the lipid content were so large (Fig. 4) that another method for data correction had to be applied. As the conditions of the second and the third experiments were the same, the data were combined (i.e., $n = 5$ for each sampling time).

The relationship between lipid and bromoform was expressed as a mathematical model (Fig. 5) composed of lines that have the same slope and different intercepts ($R^2 = 0.87$). This mathematical model was applied to calculate the corrected concentration of bromoform for each sampling time, where the lipid content would be the same as the mean value. The corrected data were analyzed by applying the compartment model to calculate the parameters relating to uptake and elimination (OECD 1996).

3 Results and Discussion

Figure 6 indicates the results of the first experiment. Figure 7 demonstrates the results derived from the data of the second and the third experiments. Fast uptake and fast excretion of bromoform by Japanese flounder were shown. The parameters relating to uptake and elimination of bromoform are shown in Table 1.

The bioconcentration factors for liver were higher than those for muscle, but even those were 130. Usually this value is regarded as low enough compared to other hydrophobic organic chemicals. As for the depuration rate constant and the biological half-life value, this study is probably the first to clarify them in fish species. The biological half-life values were about 5 h.

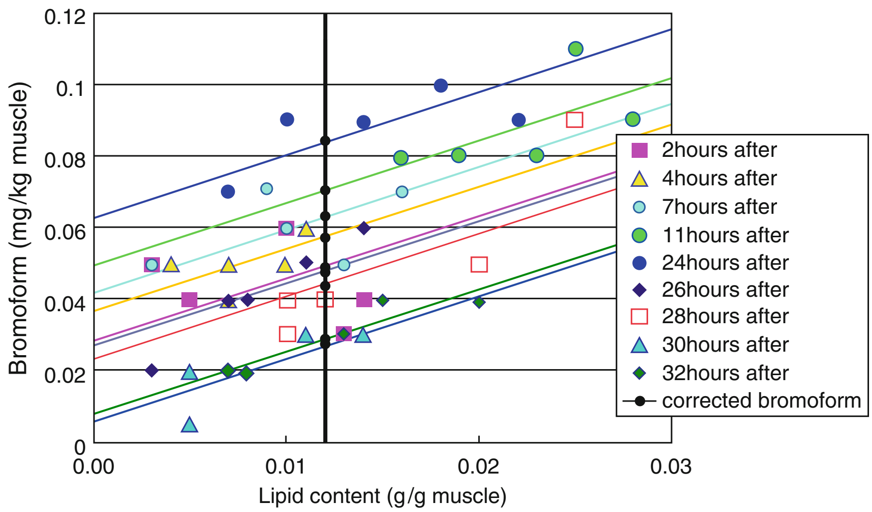

Fig. 5 The method to correct the concentration of bromoform for variation in tissue lipid content for the combined data of the second and third experiments. The parallel lines were from the regression of the bromoform concentration on the lipid content for each sampling time. The values of Y of this model when the lipid content = the mean value (*the vertical line*) were calculated as the values of the bromoform concentrations representing each sampling time

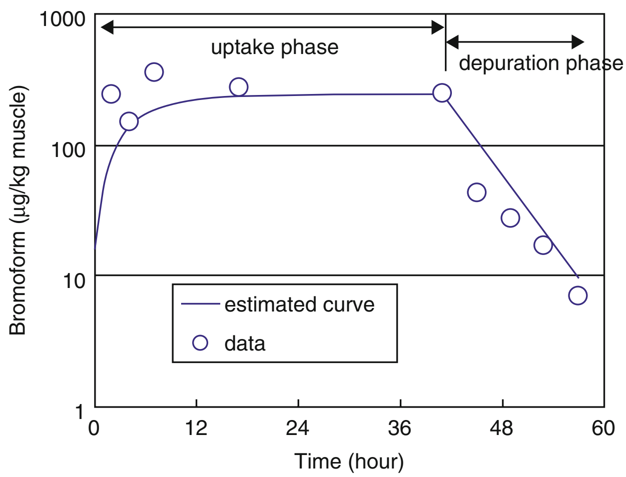

Fig. 6 Concentrations of bromoform in the muscle of fish (the results of the first experiment). The biological half-life in the muscle was 3.4 h

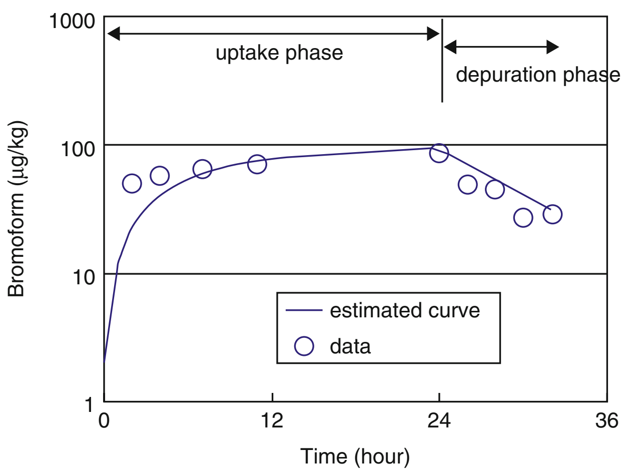

Fig. 7 Concentrations of bromoform in the muscle of fish (the results of the second and the third experiments). The biological half-life in the muscle was 5.1 h

Table 1 The parameters relating to uptake and elimination of bromoform in Japanese flounder, *Paralichthys olivaceus*

Organ	Experiment	Concentration in the water (μg/l)	Depulation rate (h^{-1})	Uptake rate (h^{-1})	Biological half-life (h)	BCF (k_1/k_2)
Muscle	First	28	0.20	1.7	3.4	8.5
Muscle	Second + third	9.7	0.13	1.3	5.1	10.0
Liver	First	28	0.16	17.1	4.3	106.9
Liver	Second + third	9.7	0.22	28.7	3.1	130.5

Those were short enough. It means, even if fish accumulate bromoform in their bodies, the fish can excrete bromoform easily when the fish migrate to the area where the concentration of bromoform in water is low. In the past, BCFs only were studied in fish species. In common carp, BCFs were reported to be 7–21 (NITE 2008). In Atlantic menhaden, BCFs were 3–50 (Gibson et al. 1979). BCFs of Japanese flounder (present study) were comparable to such data in the literature.

According to the other literature, 96 h LC50 (7.1–29 mg/l) was reported in fish species (NITE 2008); it is supposed that bromoform has a weak hazardous effect on fish. Considering the results of this study, there seems no need to worry about the toxicity of bromoform from power plants on marine organisms. Therefore, we concluded that continuous chlorination at lower concentrations would be appropriate for an anti-biofouling method at power plants available today in terms of environmental preservation.

Acknowledgement This research program was carried out under contract with Tokyo Electric Power Company, Japan.

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Results of the Implementation of Integrated Coastal Zone Management (I.C.Z.M) in Provence-Alpes-Côte-d'Azur (P.A.C.A) and Outlook for the Mediterranean Context

Pascale Janny and Philippe Lassalle

Abstract This chapter first sets the institutional context. It gives impetus to the structural elements that now make up the integrated coastal zone management (I.C.Z.M) projects. Every project has its particular characteristics. But there is a shared I.C.Z.M approach: principles of action, engineering and communication instruments, and relations and mediation between the stakeholders involved in coastal management.

Concerning the regional outcome of I.C.Z.M in Provence-Alpes-Côte d'Azur (south of France political region) in terms of methodological knowledge, suggestions for deploying the approach can be extended to the maritime context. The Mediterranean dimension is relevant for a future common framework for action in the field of I.C.Z.M, enabling the implementation of a new system of governance.

It could be useful also for the realization of joint projects, committed to by the various stakeholders and founders in the face of current coastal issues concerning the role of the anthropic pressure being exerted and by the extreme vulnerability of these territories.

It is up to us, the public stakeholders, scientists, socio-professional organizations, and users, all of us, to take up the issue and develop a shared and sustainable future vision of this coast.

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1 Logical Commitment from the National to Local Level and Vice Versa

The European I.C.Z.M recommendation of the 30 May 2002 is the base of reference for current I.C.Z.M projects. This recommendation was the result of an experience that was iterated in many projects in order to reach completion. Therefore, it is the feasible starting point for defining the various territorial levels (coastal, regional, and facade). The eight key points of a strategic approach that it identifies and the ten key principles of an I.C.Z.M approach that it sets out are the basis for this process.

Since the 1992 Rio Earth Summit, a common I.C.Z.M language has (been) developed, an international, Mediterranean, European, national language, a common language that encompasses experiences, links them, and puts them together in a way that describes the realities and conceptualizes them. The I.C.Z.M definitions adopted by I.C.Z.M operators reveal this common language.

These definitions call for another way of dealing with the coast and coastal sea, a new system of governance that starts in the field and is part of an open, human and forward-looking geography. The report produced by the Délégation à l'Aménagement du Territoire (DATAR; French state agency for national planning – Délégation Interministérielle à l'Aménagement et à la Compétitivité des Territoires, DIACT since 2005): "Construire ensemble un développement équilibré du littoral" ("Toward a balanced coastal development") (2004), already revealed this "new way of acting" beyond action that incorporates the various public coastal zone policies.

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The report's three key proposals are still relevant:

Adopt an objectives approach, then a contracting approach, beyond spatial planning.

Develop knowledge of coastal zone ecosystems and uses.

Focus the projects' approach on action, adapting to environmental changes and the long term.

The experience acquired confirms the "warning message" issued by the Conseil National d'Aménagement et de Développement du Territoire (CNADT; French regional planning and development council) in 2003. This message recognized the limited effectiveness of centralized state action. It stated that decentralization is appropriate. There is now clearly "commitment and responsibility from local authorities in dealing with the coast and coastal sea."

2 P.A.C.A's "Family" of I.C.Z.M Projects

The nine Mediterranean facade projects that won the I.C.Z.M DATAR/Secrétariat Général de la Mer 2005 call for projects and, more particularly, the five Provence-Alpes-Côte d'Azur projects (which cover more than 35% of the regional coast) represent another way of dealing with the coast and an original integrated coastal zone management practice.

The Camargue (submitted by the Parc Naturel Régional de Camargue):

Long-term operational management of the coastal and marine zone.

The City of Marseille [jointly submitted by the Ville de Marseille and the Communauté Urbaine de Marseille Provence Métropole (MPM is a gathering of Marseille city and the smaller towns belonging to neighborhood) with the backing of the town planning agency]:

Search for consistency in and an operational and forward-looking system of governance for the city's coastal development and upgrade projects and integrated coastal management (AIZC) projects, particularly Marseille harbor projects.

And willingness, involvement of stakeholders in the development of each project, including the design, implementation, follow-up, and development phases, through spatial and multi-partnership contracting methods.

The Massif des Maures (submitted by the Syndicat Intercommunal à Vocation Multiple SIVOM littoral des Maures):

A regional authority (five communes between Hyères and Saint Tropez) leads the operational expertise in the field of I.C.Z.M (the influence of a marine science observatory) combined with expertise at the Maures coastal region level (territorial and cultural engineering).

The Communauté d'Agglomération Nice Côte d'Azur (CANCA):

Mobilization of the coastal and marine environment technological platforms and expertise of the Sophia Antipolis science and technology Park to develop an observation, forecasting, and decision-making assistance tool in the fields of oceanography for the I.C.Z.M project

The Cross-border Riviera-Roya (submitted by the Communauté d'Agglomération de la Rivera française - Menton):A process of operational exchanges, cooperation to join forces in the event of accidental marine and coastal pollution, as well as two additional sections: the realization of the water management "bay contract" study and the introduction of a GIS-type system for sharing geographical information.

It should be noted that previous initiatives (Parc Marin de la Côte Bleue) or those stemming from current bay contracts (Toulon, Nice, Golfe de Lérins) extend this initial network toward a network of projects.

Promoting initiatives in neighboring territories would allow effective regional I.C.Z.M networking, representative of the issues regarding the Provence-Alpes Côte d'Azur coast and the Mediterranean facade.

3 Geographic Scale

The territorial scale of the PACA/I.C.Z.M projects falls within the scope of the department (administrative division).

Therefore, from the west to the east of the region, there is:

An emblematic territory (the Camargue), a patchwork of natural environments that form a fragile coast (a "delta")

A coastal, port, and maritime city (Marseille), with exceptional natural areas that is seeking a sustainable and operational system of governance for its facade

A "mountainous" and "Mediterranean" coastal region (Les Maures)

A city (Nice Côte d'Azur) seeking innovative initia ers regarding sustainable coastal development, and the consideration of natural risks

A multi-urban area (the French Riviera, Menton), comprising a cross-border Riviera area, a tri-national "population catchment area" seeking to develop institutional cooperation between the French Riviera, Monaco, and the Italian towns and authorities that make up a single coast

These geographic areas constitute an "appropriate area for introducing I.C.Z.M. policies."

The PACA project areas are the first I.C.Z.M "geographic unit." All of them pose challenges; each is a project area, a coastal cultural area, and a land-sea area.

There is a link in a comprehensive, planned approach that is being introduced across the coast of the PACA region. Wider discussion should be initiated at the regional level (such as the PACA coast charter project, adopted by the regional council on the 29 June 2007).

4 Implementation

Implementing the projects brought five significant advances:

– Project management methods
– Engineering and communication tools
– Examples of good practices and innovative solutions
– A process of exchange with the other projects
– Suggestions for deploying the I.C.Z.M approach across the area and, eventually, across the Mediterranean coast

Overall, the result is a strong integrated territory management capacity, in other words, the introduction of a territory approach (by stakeholder commitment area, commitment toward projects) and the development of management tools appropriate to the problems in each of these areas.

5 Methodological Knowledge

Effective methodological knowledge afforded by the I.C.Z.M initiatives in the PACA region is apparent:

– Joint processes that bring together institutions and businesses, scientists and decision-makers, public and private interests, for the collective implementation of reasoned coastal system and resource upgrade projects
– Territorial processes of territory-wide and human-scale consensus building and maintenance, a place of governance, making it possible to manage the complexity of interrelations and to go in search of stake-holders
– Proposal for management and citizen involvement initiatives that are adaptable and reviewable, preferably for costly and permanent installations and developments
– Proposal for negotiated and reviewable charters, involving the various sea and coastal management stakeholders in the life of the projects, rather than the introduction of imposed and badly received regulations
– Evolving projects, accompanied by long-term follow-up and evaluation, so as to enable their continuous improvement and bind them to environmental, economic, and social developments
– The emergence of confidence, action, and consensus in the joint development of consistent, joint projects

The effective involvement of scientists in I.C.Z.M initiatives is also evident.

The results concern:

Studies conducted by the Parc Naturel Régional de Camargue (study regarding sandy coast conservation issues, inventory of benthic macrofauna in the Golfe de Beauduc, comprehensive study of the wedge shell)

Development of the "Plan de Gestion de la rade de Marseille" (Marseille harbor management plan): multidisciplinary cooperation regarding the activities, uses, and quality of the environment

The creation of scientific tools to assist the integrated management of the Communauté d'Agglomération of the Nice coast

The diagnostic study of the environmental issues of the Riviera (prioritizing issues, diagnosis of the ecological pressures and issues, assessment of environmental vulnerability and ecological risks)

A frame of mind for I.C.Z.M on the part of oceanologists (shared by the Villefranche sur Mer, Toulon, and Marseille oceanology centers). Oceanologists play an educational and communication role in developing the coastal area and marine environment management vision, given their insight on future issues (biodiversity, global warming) and regional planning (metropolitan roles, environmental management of major ports, management of natural areas of significance, aquaculture, marine habitats, etc.).

Work concerning long-term issues (climate change, fragility of biodiversity, carrying capacity of natural areas, increased anthropic pressure on the coast, aquaculture, species and environmental management, risk management, etc.) will develop oceanologists' invaluable capacity to anticipate future events.

The Mediterranean coastal area must increase the sphere of influence of its oceanography.

Research teams must work together.

"We must develop a European oceanology" (Ceccaldi 2006).

Oceanology is important for the development of integrated coastal zone management.

Businesses are also awaiting this initiative, which is needed to firmly establish projects in coastal areas.

The "Pôle Mer PACA" ("PACA Sea Cluster"), a globally oriented competitiveness cluster, has a role to play in this development. Concerning I.C.Z.M project holders, we can see that the territories have developed a variety of engineering tools: database creation, studies, stakeholder audits, data sheets, and GIS mapping.

6 Outlook for I.C.Z.M Deployment

6.1 Toward Strategic Planning

The recommended objective is to draw up a framework for strategic action. This involves outlining the European I.C.Z.M recommendation of 2002. The framework for action will be based on three appropriate geographic levels: national, regional and facade, then extended Mediterranean wide.

The I.C.Z.M approach is based on two key elements:

The implementation of a governance approach (project management method, success indicators, an adaptive dialogue and mediation system)

The dissemination of scientific knowledge (observe, describe, understand, plan, and anticipate to decide on actions)

How can we bring the French approach to the fore?

6.2 Toward an Appropriate Framework of Action

By using the three geographic levels recommended by the "Coast" CIADT (Comité Interministériel d'Aménagement et de Développement du Territoire; Interdepartmental Regional Planning and Development Committee) of September 2004 (these are not levels that are simply superimposed, but levels that exchange, "subsidiaries," and "interlap").

First outlook: prospects of the projects, implementation prospects (objective: to promote a "national I.C.Z.M label" shared with the regions)

Second outlook: regional prospects, decentralization prospects (the issue of a coast charter with all of the stakeholders)

Third outlook: prospect of a French strategy in Europe and with Europe (propose a common framework for action for the projects)

The prospect of a Mediterranean basin-wide strategy

The prospect of a Mediterranean basin-wide strategy for integrated coastal zone management is a *fourth outlook.*

On 21 January 2008, following the meeting between the contracting parties at the Barcelona Convention, the new integrated coastal zone management protocol was adopted in Madrid.

Fourteen of the 22 countries have already signed it, including France, who played an active role in developing the protocol. The protocol sets out a common framework for commitment. It links the coast with land-sea interactions and capitalizes on the advances of neighboring countries to sustainably manage coastal zones.

The initiative, begun by the Conservatoire du Littoral to protect the "value" (not just the biodiversity, but also the culture) of small Mediterranean islands ("the islands are miniature continents," *Puri Canals, Vice-President of the IUCN*), demonstrates a willingness to commit to safeguarding the common Mediterranean heritage. The "Petites Iles de la Méditerranée" (PIM) initiative is now a full part of the conservation of coastal zones.

The I.C.Z.M colloquium, organized as part of the French Presidency of the European Union and held in Nice on 18 and 19 December 2008, aimed toward biodiversity conservation in the Mediterranean basin. More precisely, this colloquium will be dedicated to the coastal zone at its land-sea interface.

It will contribute toward bringing the I.C.Z.M protocol of the Barcelona Convention into force.

This colloquium is a priority topic of the Union for the Mediterranean (UpM; Union pour la Méditerranée).

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Ceccaldi HJ (2006) L'Europe a besoin d'une océanologie forte sur la Méditerranée. Ann Inst Méditerr Transports Marit 20:283–300

Outline of Ongoing Research Activities of the Marine Ecology Research Institute, Mainly Regarding Thermal Issues in Japan

Michiyasu Kiyono and Katsutoshi Kido

Abstract The Marine Ecology Research Institute (MERI) was established in 1975 by an agreement among three government agencies, which were then known as the Fisheries Agency, Energy Agency, and Environmental Agency.

MERI is the only research organization in Japan specialized in research to elucidate the effects of thermal power plant cooling systems on coastal fishery resources and marine organisms.

Most fossil fuel and all nuclear power plants in Japan are located at the seaside and employ the once-through cooling system. The water pollution control law of Japan lists "heat discharge" as one of its regulation targets. However, no law, regulation, or guideline for heat discharge has been enacted so far in Japan. Issues concerning the temperature rise of water between the intake and discharge at power plants and the structural design of intake and discharge facilities have been settled by an agreement between power companies and local governments under the guidance of national agencies, including the Nuclear and Industrial Safety Agency.

No serious damage to local fishery resources because of power plant operations has been reported so far.

Recent public concern has focused on the impacts of these power plants on the marine environment and on preserving the ecosystem. Impacts of chemical substances including anti-biofouling substances, are also of concerned. With the possible global warming, thermal impacts on marine ecosystem have again become a public issue. In addition to the elucidation of power plant impacts on fishery resources, MERI has also given higher priority to the following:

- Technological development of impact assessment on the coastal ecosystem, including macrophyte beds,
- Application of intake and discharge facilities for new biotopes of marine organisms living around power plants,
- Exploitation of water currents created by cooling water intake and discharge for water quality improvement in and near harbor areas,
- Impact assessment of anti-biofouling substances
- Investigation of the effects of ocean temperature rises as well as ocean acidification and CO_2 sequestration,

MERI will deal with any coastal environment issues in cooperation with national agencies, the power industry, and the fishery industry.

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1 What is MERI

The Marine Ecology Research Institute (MERI) is the only research organization in Japan specialized in research to elucidate the effects of thermal power plant cooling systems on coastal fishery resources and marine organisms.

MERI is an independent and non-profit research organization established in 1975 by an agreement among three government agencies: the Fisheries Agency and what were then the Energy Agency and Environmental Agency. Ninety staff members, including 50 scientists, belong to MERI and work in field surveys and laboratory experiments. Most of the MERI projects are commissioned works with the national agencies and the electric power companies.

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MERI manages two marine laboratories. The Central Laboratory was established in 1979 in Onjuku Town, Chiba Prefecture, on the Pacific Ocean, as the major research facility for the institute, where clean seawater is available for experimental works. The Demonstration Laboratory was constructed in 1984 in Niigata Prefecture on the Japan Sea, adjacent to Kashiwazaki Nuclear Power Plant, Tokyo Electric Power Co., Ltd. Actual thermal effluent from the nuclear power plant is available for experiments at the Demonstration Laboratory. At both laboratories, well-designed rearing facilities are installed to prepare marine organisms for experiments. Several tens of fish and invertebrate species have been reared and supplied for experiments done in and out of MERI. Using these facilities, some marine and freshwater endangered fish species, such as a sillagofish, *Sillago parvisquami*, an endangered marine fish, and Japanese bitterling, *Tanakia tanago*, a national treasure species, also have been propagated.

2 Marine Thermal Issues in Which MERI Has Been Involved

Most fossil fuel and all nuclear power plants in Japan are located at the seaside and employ the once-through cooling system. Impacts of thermal discharge (intake and discharge of cooling water) have been a public concern, and several studies, therefore, were conducted to elucidate power plant impacts on marine fishery resources and marine organisms in the 1970s–1990s (Kiyono and Shinshima 1982; Kinoshita 1985; Yamamoto et al. 1991; Marumo et al. 1992; Tsuchida 1995). Since no serious damage to local fishery resources because of power plant operations has been observed or reported so far, recent public concern has focused on impacts on the marine ecosystem and its preservation. With the possible global warming, however, thermal impacts on fishery resources again have become one of the major public issues. The following are the marine thermal and power plant operation issues and the relating ongoing research projects to which MERI has given higher priority:

- Elucidation of impacts of power plant cooling system operation on marine organisms and local fishery resources
- Demonstration of power plant impact assessment on the coastal ecosystem
- Management of the coastal ecosystem in the vicinity of power plants
- Offshore environmental radioactivity monitoring
- Larger scale environment changes
- Impact assessment of seawater temperature rise in the larger scale area
- Investigation of impacts of CO_2 ocean sequestration, storage, and ocean acidification

3 Macrophyte bed survey throughout Japan

3.1 *Intensive surveys around power plants*

According to the Japan Meteorology Agency, the average surface seawater temperatures of the western and southern ocean areas of Japan have increased by about 1°C in the past 100 years. Under these conditions of rising ambient sea water temperatures, MERI has been carrying out intensive field surveys around the thermal plume areas of large nuclear power plants (blue circles in the Fig. 1) to determine more precisely the impacts of temperature change on marine macrophyte and

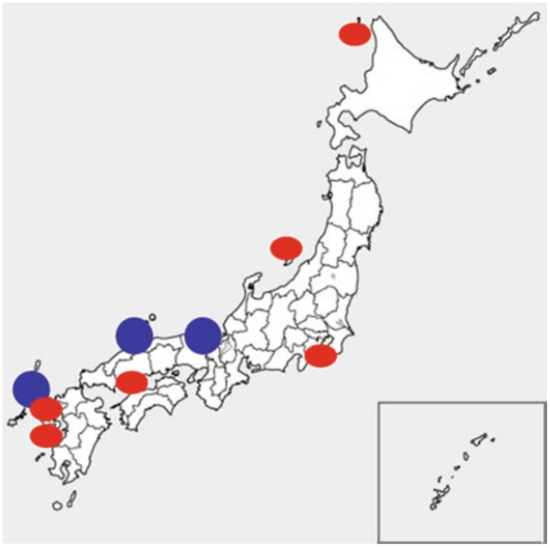

Fig. 1 Survey area

non-moving invertebrates. We believe that the data obtained from these field surveys are also very useful to predict what may happen under the possible higher seawater temperature conditions in the future.

3.2 *Macrophyte Data Compiling*

Declines in standing crops and areas of macrophyte beds have caused much concern. To clarify the state of the art of macrophyte bed distribution along the coast of Japan, MERI has collected almost all existing survey data available and also conducted some supplementary field surveys at places indicated by the red ovals in the Fig. 1. These data are being integrated on the GIS (Geographic Information System) map. This GIS map will be very useful to examine the increasing or decreasing tendency of macrophyte beds in local areas, because data from macrophyte observations conducted for the last 20–30 years have been compiled in this survey.

The field surveys around power plants and the compiling of macrophyte data were conducted under contracts with the Nuclear and Industrial Safety Agency and the Fisheries Agency, respectively.

Fig. 2 Areas where macrophyte damage by rabbit fish was reported

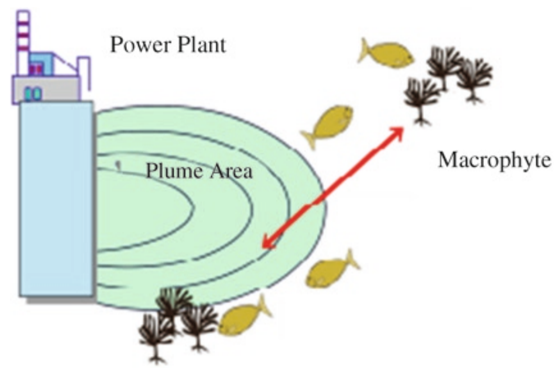

Fig. 3 A conceptual figure of rabbit fish living in the plume area of a power plant

4 Surveys on Macrophyte Bed Damage by_Rabbit Fish, *Siganus fuscescens*, Feeding

Feeding by fish may be one of the major causes of seaweed decline, and the rabbit fish, *Siganus fuscescens*, an omnivorous subtropical species, is the one of most concerns. This species is now observed in the northern area with the increase in seawater temperature. The red ovals in Fig. 2 show the area where macrophyte bed damage possibly by rabbit fish feeding was reported recently.

Rabbit fish may settle first in the plume area or stay there during the winter season and feed on macrophytes near the power stations (Fig. 3).

MERI has been conducting a project to elucidate rabbit fish feeding and temperature preferences to examine the possibility of rabbit fish settlement in the plume areas and macrophyte damage caused by rabbit fish feeding. The survey was conducted under contract with the Nuclear and Industrial Safety Agency.

5 Demonstration Survey of Ecological Impact Assessment Using a Virtual Power Plant

The Japanese environmental impact assessment law requires the assessment of power plant impacts on ecosystems. Assessments of the terrestrial ecosystem have been conducted focusing some important or typical species in the target ecosystem. The marine ecosystem, however, has not been included in the standard procedure of power plant impact assessments so far, mainly because assessment technologies applied to the marine ecosystem are not considered to be well developed. Therefore, MERI has been conducting development and systematization of the marine ecosystem survey technologies and has been demonstrating power plant impact assessments, considering a virtual power plant. In the MERI survey, typical fields of the coastal ocean, i.e., macrophyte beds and tidal flats, have been selected as target areas for the assessment. In this survey, field surveys, integration of field data on the GIS map, mathematical simulation of thermal discharge, and the impact assessment on marine organisms using the habitat suitability indexes established by field data and experimental data were undertaken (Figs. 4 and 5).

Fig. 4 Distribution of macrophyte bed area considering a virtual power plant. Red zone: kelp-type macrophyte bed area, green zone: eel grass bed area

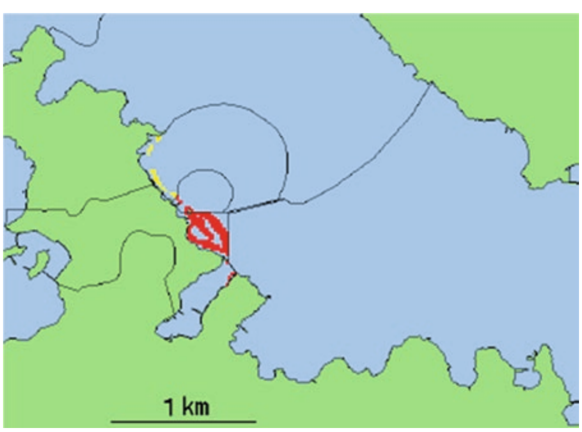

Fig. 5 Distribution of a temporal prediction of the thermal plume area and kelp type macrophyte areas possibly affected by the reclamation (*red area*) and by thermal discharge (*yellow area*)

This project was conducted under contract with the Nuclear and Industrial Safety Agency.

6 Management of the Coastal Ecosystem Around the Power Plant

The thermal plume, intake and discharge currents, and coastal facilities of power plants may be potentially useful for fishery activities and coastal environmental management. MERI has reported catching more sea bass around the thermal plume area of a power plant in the northern part of Japan and studied the behavior of sea urchins (Dotsu et al. 2000) and bigeye trevally (Miura and Yamamoto 2008), *Caranx sexfasciatus*, a subtropical game fish species, around power plants (Fig. 6). MERI has also been conducting the following projects on coastal environment management: the application of discharge currents to restore the lower DO condition of bottom layers often observed in the enclosed coastal sea areas during the summer season and the application of underwater reefs to form new biotopes for macrophytes. These projects have been conducted under contracts with the Fisheries Agency and the Nuclear and Industrial Safety Agency.

7 Offshore Environment Radioactivity

Since 1983, offshore of all nuclear power plants and nuclear fuel reprocessing facilities, radioactive monitoring surveys of marine organism, seawater, and sediments have been conducted (Fig. 7). Supplemental analyses of variations affecting radioactivity, such as scavenging activities, fish feeding habits, growth stages, etc., are also undertaken. (Kasamatsu and Ishikawa 1997; Kasamatsu and Inatomi 1998; Iibuchi et al. 2002)

8 Projects Relating to CO_2 Ocean Sequestration

MERI also has been involved in CO_2-related projects, such as CO_2 ocean storage and sequestration. In these projects, our institute was in charge of the preliminary

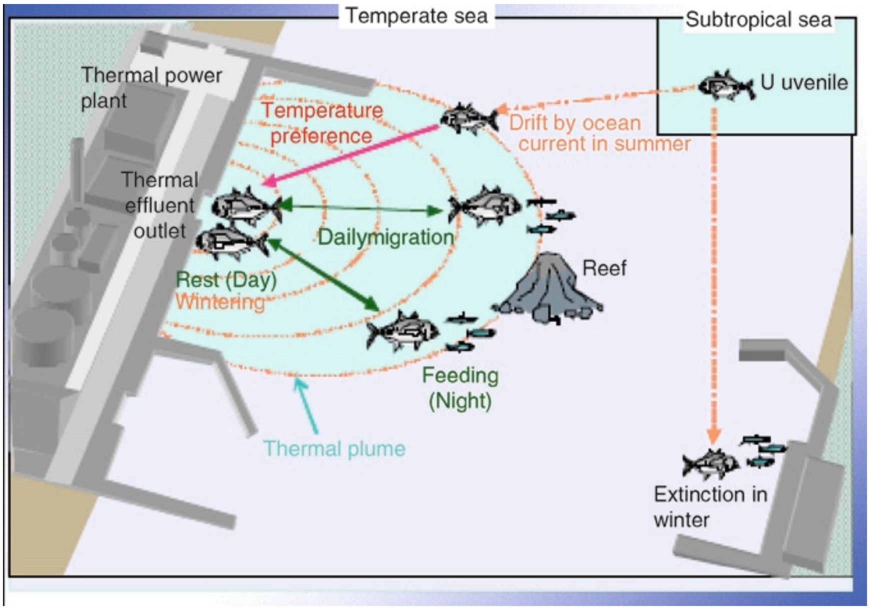

Fig. 6 A conceptual figure of a new fish biotope near a power plant

Fig. 7 Monitoring areas

examination of impacts of higher CO_2 condition on the survival and growth of some marine animals. Species used for experiments have been surface species so far, although deep sea species should be much desirable for test species. In our preliminary experiments, the 24-h median lethal concentration of several marine species and CO_2 effects on the growth of juvenile Japanese sillago, *Sillago japonica*, after 5 months exposure (Fig. 8) were examined. Impact examination of seawater acidification and carbon storages under the sea bottom area also has been conducted (Kita et al. 2003; Kikkawa et al. 2003a, b, 2006; Ishimatsu et al. 2004).

9 Other Main Ongoing Projects of MERI

9.1 Mussel Fouling and Jellyfish Invasion

Demonstration experiments to examine the impacts of chlorination have been carried out by MERI. Some results are presented in the paper submitted by Mr. Iibuchi et al. in the proceedings of 13th Japan–France Oceanography Symposium. Regard-ing a jellyfish species, *Aurelia aurita*, habitat areas and population of their polyp in Ise Bay, Japan, have been elucidated. Comparison of invasion quantity with polyp populations in Ise Bay is the next target of this project. These projects have been conducted under contracts with electric power companies.

Fig. 8 Devices for long-term exposure

9.2 Bioassey Technology Development

Monitoring of chemical concentrations in coastal areas, examination of the dose-response of marine fish and shellfish, and technological development of bioassays for reproductive success have been conducted using facilities of the Demonstration Laboratory, one of the largest scale facilities for exposure experiments in Japan. These projects were conducted under contract with the Fisheries Agency (Hotta et al. 2001, 2003).

10 Conclusion

No serious damage to local fishery resources because of power plant operations has been observed in Japan so far. With the rise in surface seawater temperature around Japan, however, the thermal impact on the fishery resources and the marine ecosystem is still a major public issue. Intensive field surveys around thermal plume areas of power plants are needed to determine more precisely the temperature impact on fishery resources and the marine ecosystem. The data obtained from these surveys will contribute to predicting what may happen with possibly higher seawater temperatures in the future in Japan. Impact examination of seawater acidification and carbon storage under the sea bottom is also required.

MERI will deal with any coastal environment issues, including the thermal issues, in cooperation with national agencies, the power industry, and the fishery industry.

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Mass Mortality of a Coral Community in Ishigaki Island, Okinawa, Japan, Caused by the Discharge of Terrigenous Fine Particles

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Abstract A mass mortality of coral communities dominated by *Porites* spp. was observed in the reef flats off the east coast of Ishigaki Island, Okinawa, Japan. Mortality was concentrated around the mouth of the Todoroki River, from which a large amount of terrigenous fine particles had been discharged during a heavy rain. Mortality was observed over an area of ca. 30 ha in early June 2001. Damaged corals included massive *Porites* and *Favia*, branching *Montipora*, and the blue coral *Heliopora coerulea*. The most conspicuous damage was evident in large colonies of *Porites* because of the abundance of this species in the area. Fine particles <0.063 mm in diameter dominated (ca. 90%) the sediment collected by traps in June 2001; coarser particles, 0.125–1 mm in diameter, were collected 1–2 months after the event. The total nitrogen content of collected sediment was higher (>0.5%) just after the mass-mortality event and decreased over time. This mass mortality was apparently caused by several simultaneously occurring factors, including low tide, unusually strong north winds, and heavy precipitation that caused extensive sediment runoff.

1 Introduction

A mass-mortality event in a coral community dominated by *Porites* spp. occurred on an Okinawan coral reef off the east coast of Ishigaki Island, Japan, in June of 2001. During a period of heavy rain, a large amount of terrigenous fine particles was discharged onto the reef from the Todoroki, a small river that flows onto the reef. Many dead corals covered with fine particles were observed over an area covering ca. 30 ha, particularly around the southern part of the river mouth.

Mass mortality of marine invertebrates caused by unusually high or low temperatures has been reported repeatedly in various habitats, and the effects of such mass mortalities on community structure have been discussed (e.g., Crisp 1964; Glynn 1968; Tsuchiya 1983; Tsuchiya et al. 1987). The mass mortality of corals may affect community structure and dynamics of coral reef ecosystems. Disturbances to coral reefs in Japan have several causes, including bleaching events, invasion by the crown-of-thorns starfish (*Acanthaster planci*), and sedimentation input of terrigenous fine particles (reddish clay; Nishihira 1987; Yamaguchi 1987; Ismail et al. 2005).

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Sediment runoff has been a serious environmental problem in the coral reefs of Okinawa since the 1950s (Yamazato 1987). However, quantitative analyses of the impacts of this sediment loading have been lacking, because disturbance events occur at different time scales, and the tolerance of marine organisms to sediment has not been well studied. Here, we report a mass-mortality event observed in a coral community and provide information on the characteristics of the sediment that was deposited on the reef.

Given that massive *Porites* spp. show high tolerance to severe environmental disturbances such as sedimentation and low salinity (Chou and Teo 1985; Potts et al. 1985; Sakai et al. 1986; Omija 1996), this event is considered to be abnormal, exceeding the tolerance of even these coral species.

2 Study Area and Methods

A general outline of the coral communities in the study area (Fig. 1) was presented by Takahashi et al. (1985), and the community dynamics related to a mass bleaching event of 1998 were documented by Kayanne et al. (2002). The study area supported a dense population of the blue coral *Heliopora coerulea*, and branching *Montipora* and *Acropora aspera* were the dominant coral groups on the reef crest. Massive *Porites* dominated the area around the mouth of the Todoroki River. Manta-tow surveys (English et al. 1997) were adopted for this investigation, and aerial photographs were used to determine the routes of the tows (Fig. 1). The tows were conducted at regular intervals. The observer was towed along the surface of the water behind a small boat and recorded the species composition and coverage of corals. The path of the tow was simultaneously recorded using a GPS.

A spot-check method for coral reef monitoring (Nomura 2004) was also adopted for the survey after the study area had been selected. Spot checks showed the differing levels of impact of the disturbance on the coral communities. Two snorkeling observers surveyed a given area for 15 min and recorded the condition of each coral species or colony, e.g., the degree of bleaching and mortality, and the sedimentation status of fine particles on the coral.

The relationship between the mass-mortality event and meteorological data, which were recorded by the Ishigaki Island Meteorological Observatory, was analyzed to determine the causes of the mass mortality.

Fig. 1 Aerial map of the study area with census lines by the Manta Tow method and points studied by spot-checking method (*circles*) for the mortality of corals and localities where sediment tarps were set (Station N, M, and S). The results are also shown

Sediment traps were constructed using PVC pipes that were 12.5 cm long and had an internal diameter of 5 cm. The pipes were set along 15-m transects perpendicular to the shoreline. Two sets of pipes were fixed into the seabed, 30 cm above the bottom, at the beginning of each transect, and two additional sets were fixed at the end of each transect. Each set consisted of 3 tubes, for a total of 12 tubes at each station. The tops of the tubes were covered with baffles to prevent large animals from getting inside. The traps were set at three stations around the mouth of the Todoroki River (Stations N, M, and S; Fig. 1). The traps were positioned according to the methods described by English et al. (1997). Grain size composition of the trapped sediment along with its carbon and nitrogen content was analyzed using a CN analyzer (Sumigraph 90, Shimadzu Co Ltd). Calcium carbonate content was determined using 1M HCl.

3 Results

3.1 Mass Mortality in the Coral Community

Mass mortality in the coral community was observed over a wide area extending about 30 ha around the river mouth (Fig. 2). Over 90% of the mortality was observed in an area 500–1,000 m south of the river mouth. Disturbed areas in which mortality levels of 50–75% and 25–50% were recorded covered about 8 and 27 ha, respectively (Fig. 1).

A general survey of the condition of the coral communities around Ishigaki Island was also conducted, and we found that the mass mortality of corals was restricted to the mouth of the Todoroki River. No corals affected by the runoff of fine particles were seen in other reefs in Ishigaki.

The damaged corals included massive *Porites* and *Favia*, branching *Montipora*, and the blue coral *Heliopora coerulea*. The most conspicuous damage was evident in *Porites* because of its high abundance (Ministry of Environment, Japan 2002), although the runoff seemed to affect most of the coral species in the area, rather than selectively affecting *Porites*.

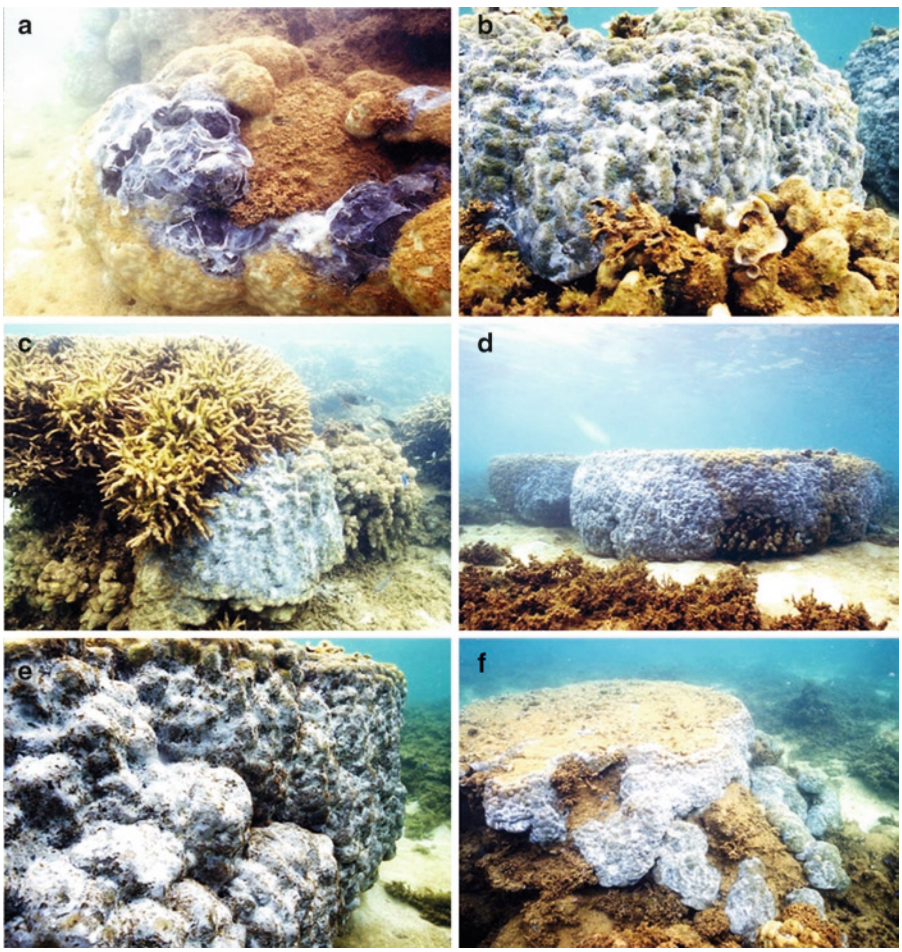

Fig. 2 Mass mortality of corals around the mouth of Todoroki River, Ishigaki Island, Okinawa, Japan. Dead large colonies of the massive *Porites* (**a**)–(**f**) were common, and the branching coral *Montipora degitata* was also seen (**c**) (photo by T. Kobayashi)

The accumulation of fine particles on both dead corals and the benthic surface was widely observed, and it was obvious that the runoff of fine particles had affected the corals and resulted in their mass mortality. Large dead colonies of *Porites*, 1–2 m in diameter, were frequently seen in the disturbed area (Fig. 2).

3.2 Causes of Mass Mortality

Data on precipitation, wind direction and velocity, atmospheric pressure, and tidal behavior (Fig. 3) were analyzed to determine possible causes of the mass mortality. From 0200 h on 31 May 2001, when a low-pressure zone moved into the Ishigaki area, heavy rain was recorded (238 mm in 24 h). When the peak of the precipitation was recorded between 2000 and 2200 on 31 May, strong winds of 11–12 m/s were recorded from the north-northeast. Low tide occurred during the period of heavy rainfall.

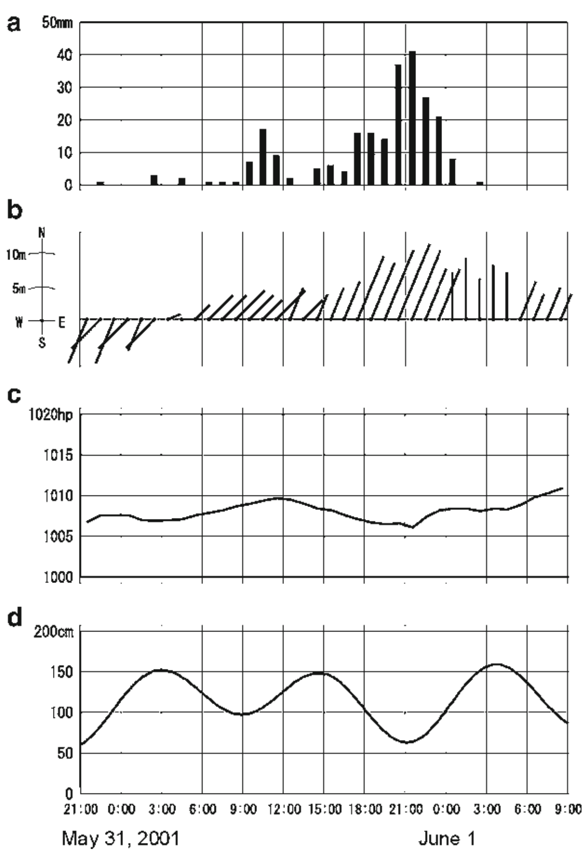

Fig. 3 Meteorological data, (**a**) precipitation, (**b**) wind direction and strength, (**c**) atmospheric pressure, and (**d**) tidal behavior from May 30 to June 1, 2001

According to meteorological records, heavy rains are occasionally recorded for this area, so it is debatable whether the sediment deposited by the heavy rain was the sole cause of this mass mortality. The low tide may have exacerbated the effects of the low salinity and increased sedimentation on the coral community.

The reef flat topography in this area is probably also an important factor that influenced the mortality. The topography is shallow and enclosed between the shoreline and the reef edge, with no channels connecting it to the open sea. One channel is located about 1,600 m north of the river mouth. A very shallow area, with a maximum depth of ca. 2 m at low tide, extends 1,000 m south from the river mouth, and a northward-flowing water current is influenced by the dominant south wind. The water level over this section of reef was low during the period of heavy rain, when a strong north or northeast wind may have carried fine particles to the southern area of the enclosure, which was dominated by massive *Porites* spp.

3.3 Sedimentation Rate and Grain Size Composition

Sediment traps were deployed in this area from September 2000 to September 2001 to study the sedimentation patterns at two stations 1 km north (Station N) and south (Station S) of the river mouth. Sediment was collected every month to analyze the monthly sedimentation rate (Ismail et al. 2005). Additional traps were positioned around the river mouth (Station M) on 31 May 2001. Shortly after these traps were set up, the coral mass mortality occurred.

From July to September 2001, a large amount of sediment was collected both in the river mouth (M), where coral mortality was very high, and at the north station (N). Normal sedimentation rates were recorded from the south station (S; Fig. 4), where low coral mortality (<5%) was recorded. The typical amount of sediment recorded at stations N and S was <2 kg/30 days/m^2. This level has also been recorded at other stations around Ishigaki and Iriomote Islands (Ismail et al. 2005). Sedimentation rates during the mass-mortality event were comparatively high at stations N and M.

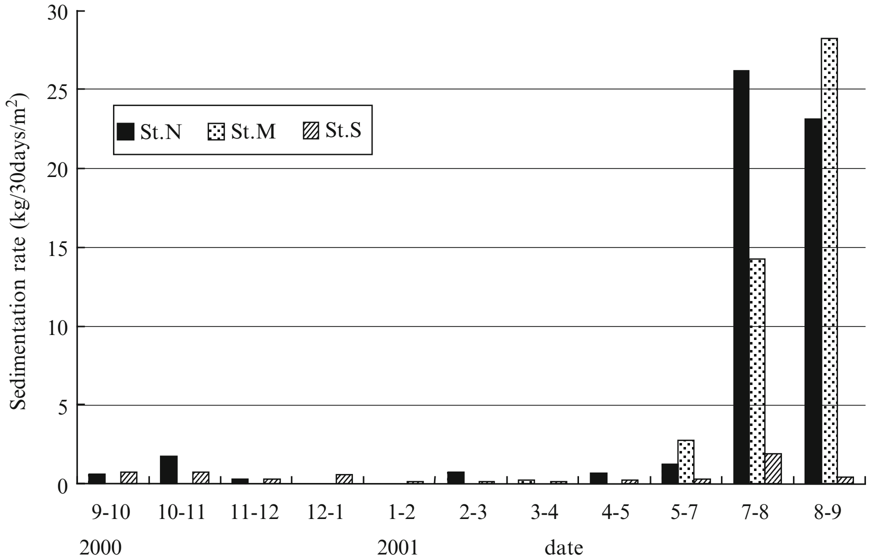

Fig. 4 Sedimentation rate at three stations, Stations N, M, and S, set around the mouth of Todoroki River from September 2000 to September 2001

The grain size composition of the sediment differed during different collection periods. Under the unusual weather conditions from late May to June 2001, fine particles smaller than 0.063 mm in diameter dominated the sediment (ca. 90%; Fig. 5a). However, the content of larger particles (the sand fraction) increased thereafter, while the proportion of fine particles (<0.063 mm) decreased to 10–30% in the sediments collected in August and September (Fig. 5b, c). A similar trend was also observed at the north and south stations (Ismail et al. 2005).

3.4 Content of Calcium Carbonate, Total Nitrogen, and Carbon in the Sediments

Sediment collected between May and July contained a large amount of calcium carbonate (>60%), but this content decreased over time (Fig. 6a). About 10% of the sediment was calcium carbonate in samples collected during August and September. However, larger amounts of calcium carbonate were recorded in July–August and August–September samples than in May–July samples.

Total nitrogen content (Fig. 6b) showed the reverse trend to that of total carbon. Nitrogen levels were higher in May–July (>0.4%) than in July–August and August–September. Total nitrogen content was fairly low (<0.05%) in the sediment collected in August–September.

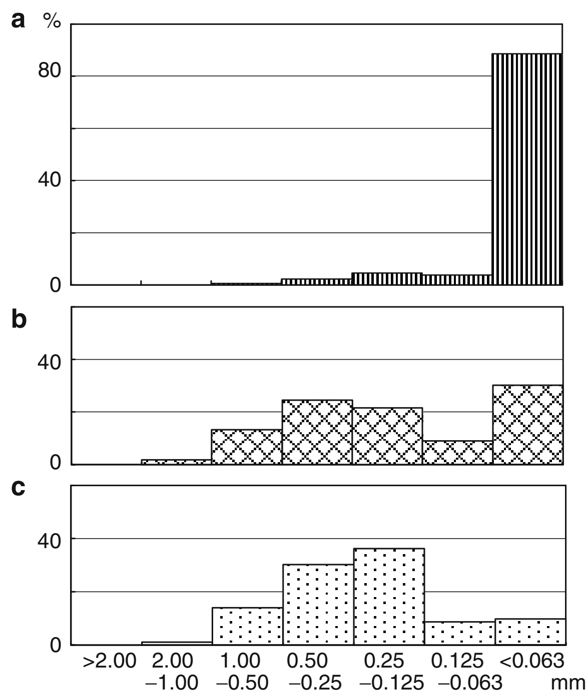

Fig. 5 Size distribution of sediments collected at three different periods, (**a**) May 30–July 1, (**b**) July 1–August 12, and (**c**) August 12 to September 8, around the mouth area of Todoroki River (Station M)

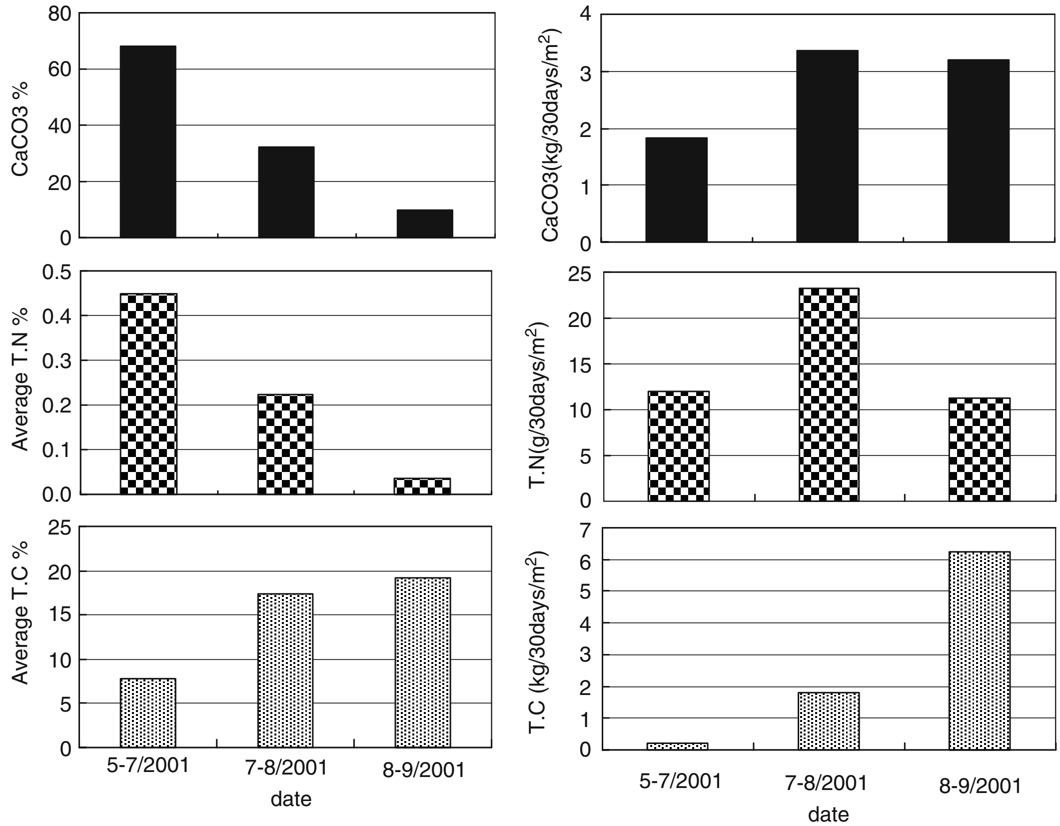

Fig. 6 Contents of (**a**) calcium carbonate, (**b**) organic nitrogen, and (**c**) total carbon in the sediments collected at Station M

The total carbon content of the sediment collected between May and July was less than 10%, but increased in the later sampling periods, when values of >15% were recorded (Fig. 6c). Increases in the amount of total carbon were also conspicuous after the event.

Seasonal changes in the total carbon and nitrogen content of the sediment collected at stations N and S are reported elsewhere (Ismail et al. 2005). In brief, large amounts of total carbon (2,456.4 $mg/m^2/day$) and nitrogen (2.02 $mg/m^2/day$) were detected at Station N in July–August 2001, but levels were low during other periods. Compared to these results, the nitrogen content at Station M fluctuated greatly during the study period.

4 Discussion

Partially or completely dead coral colonies have been observed frequently over small areas in many locations. For example, during rainy days at low tides in the winter, the upper parts of some coral colonies can be exposed to low air temperatures and subsequently die, resulting in a decrease in live coral coverage. When unsuitable environmental conditions such as low salinity, high siltation rates, and extremely low tides arise simultaneously, mass mortality of coral colonies can occur (Anthony and Kerswell 2007).

One of the most important findings of this study was that many large colonies of massive *Porites*, >2 m in diameter, died. This indicates that the confluence of unfavorable environmental conditions is a very infrequent occurrence. Events similar to those described in this study have rarely been documented in this area, although corals have been affected by current stress, bleaching, and/or normal levels of sedimentation.

When very small amounts of sediment are transported from terrestrial and river systems, the water over coral reefs is clear. In the rainy season, however, the Todoroki River discharges onto a reef on the east coast of Ishigaki Island and plays an important, but detrimental, role in the ecosystem dynamics of this area.

The amount of suspended material carried by the Todoroki River has been reported to be <10 mg/l and 90.8 mg/l (maximum: 1,290 mg/l) in usual and rainy conditions, respectively (Nakasone et al. 2001). This material is usually carried to the northern area of the river mouth and is deposited on the sandy beach or in shallow water (Tsuchiya et al. 1999; Mitsumoto et al. 2000). Mitsumoto et al. (2000) also reported the effects of this sediment on coral coverage, which was reduced in the area of the river mouth compared to the southern area. Omija et al. (1998) suggested that suspended material might be carried in a southerly direction by the strong and unusual north wind. This may be one cause of the mass mortality. The sedimentation rate was not high at station S during late May to June, when the mass mortality occurred, and the sediment was characterized by very fine particles, with coarser particles collected later. Very fine particles were initially held in suspension over the study area, but these were swept away quickly by the water current. Coarser particles may have then been transported by wave action and collected by the sediment traps.

Mass mortalities have been reported when several unfavorable environmental conditions occurred unexpectedly and simultaneously.

Cases have been reported in both mussels (Tsuchiya 1983) and sea urchins (Tsuchiya et al. 1987). In our study, heavy rain, a strong north wind, and low tide are considered to be possible causes, as all of these factors could contribute to the onset of mass mortality in corals.

Terrestrial runoff, via the inflow of agricultural fertilizers or accelerated soil erosion, is the largest source of anthropogenically derived nutrients. Several attempts have been made to determine the amount of nitrogen and phosphorous entering coral reef ecosystems (Mitchell et al. 1997; Furnas and Mitchell 2001), and the amount of sediment carried by river water has been estimated for the Great Barrier Reef (e.g., Rogers 1990). However, analyses of the chemical properties of the fine particles entering Okinawan coral reefs are still lacking, although knowledge of these properties is important to analyze in detail their effects on corals and coral reef organisms. Recently, the nitrogen input to coral reef areas was reported using nitrogen isotopes (Umezawa et al. 2002), and the importance of groundwater discharge as a nutrient source to coral reef ecosystems was discussed. Since the nutrients carried by river systems also play important roles in coral reef dynamics, it is necessary to analyze the biogeochemical cycle with reference to runoff of terrigenous fine particles.

Numerous papers have discussed the effects of sedimentation on coral reef organisms. For example, Hubbard (1986) reported that patterns of sedimentation vary in locations with different topographic characteristics. In the presence of large amounts of suspended material and high rates of sedimentation, the photosynthetic activity of zooxanthellae is affected, and increased mucus production by corals is observed (Riegel and Branch 1995). Although corals can remove particles that are sprinkled on them (Rogers 1990; Stafford-Smith and Ormond 1992), different coral species have different tolerance levels for sedimentation (Stafford 1993). Coral growth rates are also affected by sedimentation (Rice and Hunter 1992). The dynamics of fine particles is one of the important factors controlling the dynamics of coral reef ecosystems (Fortes 2001). However, the mass mortality of a coral community as reported here is serious and highlights the relationships between coral ecology and environmental factors. Our responsibility is to describe this event and, as far as possible, to try to determine its causes. If the unfavorable conditions described in this study coincide again in the future, another mass-mortality event may occur. To prevent the future collapse of this important reef ecosystem, anthropogenic effects should be minimized.

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Observation of marine environment

Alister – Rapid Environment Assessment AUV (Autonomous Underwater Vehicle)

Thierry Copros and Daniel Scourzic

Abstract ECA is mainly known worldwide for its successful PAP family (mine disposal systems). In the late 1970s, ECA designed the first operational AUV, named EPAULARD for IFREMER.

Using its know-how gained over the years in designing and building various types of free swimming and inspection underwater vehicles, ECA decided in spring 2003 to develop the A3000, the first inspection AUV with hovering capabilities. This system was successfully tested in deep water by BP in the Gulf of Mexico in July 2006.

All the experience gained during the development and trials of the A3000 have been used to develop and manufacture a Rapid Environment Assessment AUV that was ordered by the French MoD, the end user being the French Navy Hydrographic Service (SHOM).

The first part of the presentation will detail the advantages of using AUVs:

Level of equipment and tonnage of the vessel needed

Reduced manpower

Operating footprint unlimited, since an AUV is free swimming, whereas ROV or towed systems have limitations because of their umbilical cables

The surface ship can carry out parallel tasks

Operation duration is reduced, because the vehicle is not influenced by the state of the sea and the ship's movements

High quality of the data retrieved: this is due to the fact that an AUV operates without an umbilical cable

A brief description of the A3000 inspection AUV will then be presented, showing that detailed inspection and identification of underwater objects can already be achieved using AUVs.

The paper will then focus on the ALISTER REA AUV with a detailed presentation of the system including its description, performance, suite of sensors (SSS, MBE, SBP, etc.), and results of trials at sea that were conducted in 2007 and 2008.

1 Introduction

ECA is mainly known worldwide for its successful PAP family (PAP Mark 3, PAP Mark 5, PAP Plus, etc.), but during the 1960s, ECA also designed many free models of submarines, at scales of 1/10 or 1/20 used for hydrodynamic studies. In late 1970, ECA designed the first operational AUV, named EPAULARD for CNEXO (the former name of IFREMER).

Using its skills and know-how gained over the years designing and building various types of free swimming and inspection underwater vehicles, ECA decided in spring 2003 to develop ALISTAR 3000, the first inspection AUV, which carried out its first trials at sea 1 year later. This system was successfully tested by BP in the Gulf of Mexico in July 2006 in a real operational oil field with 1,400 msw. All the experience gained during the development of ALISTAR 3000 as well as the numerous trials at sea has been used to develop and manufacture a

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Rapid Environmental Assessment AUV that has recently been delivered to the Hydrographic Institute of the French Ministry of Defence (Service Hydrographique et Océanographique de la Marine, or SHOM).

2 Interest in AUVs

The interest in AUVs can be easily understood considering the ways in which customers need to mobilize today for carrying out typical missions. The advantages start, but do not end, with the level of equipment and the tonnage of the vessel needed. Using an AUV will allow operators to reduce the manpower needed, which also means that personnel logistics are reduced dramatically.

Furthermore, pilots or supervisors are not needed for over 24 h, and the online team can be reduced dramatically (QA/AC control only). The AUV's operating footprint is unlimited, since it is free swimming, whereas ROVs or towed systems are limited by their cumbersome umbilical cables. An AUV also enables the surface ship to carry out parallel tasks. When using an AUV, the operation duration is reduced because the vehicle is not influenced by the state of the sea and the ship's movements, and the dependency on surface weather is reduced, which can lead to retrieving a vehicle onboard and waiting for better weather conditions. Last but not least, the total weight of the AUV spread is much lower than a tethered vehicle spread (no umbilical cable to store, no winch, no huge power requirement). These reasons are sufficient to raise great interest in AUVs in the industrial and naval communities.

However, there are other advantages: one of them, often cited by current AUV users, is the high quality of the data retrieved. This is due to the fact that an AUV operates without umbilical cables. As a consequence, the vehicle is neither disturbed by the ship's movements transferring to the vehicle through the umbilical nor by the umbilical cable's own vibrations.

3 ALISTAR 3000 System

All the features and functions performed by ALISTAR 3000 can easily be implemented for a lower depth version, which will lead to a smaller and lighter vehicle. The aim of this chapter is to illustrate what can already be achieved using AUV's for inspection tasks.

3.1 *Description of the Alistar 3000 System*

The main characteristics of ALISTAR 3000 (Fig. 1) are:

Length: 5 m long
Maximum body diameter: 1.2 m
Overall height: 1.45 m
Total weight: 2,100 kg including up to 200 kg payload
Operating depth: 0–3,000 msw

It is equipped with four longitudinal, two lateral, and two vertical thrusters.

The actuator layout provides ALISTAR vehicles with a high maneuverability, especially at zero speed. ALISTAR vehicles therefore have the ability to hover. The vehicle has an excellent pitch and roll stability because of its mechanical architecture, reducing the energy consumption for stabilizing the vehicle, which is of paramount importance for an AUV (Fig. 2).

The ALISTAR vehicle is able to operate at 0 knots while hovering and can achieve a maximum speed over 4 knots through water

Fig. 1 ALISTAR 3000 vehicle

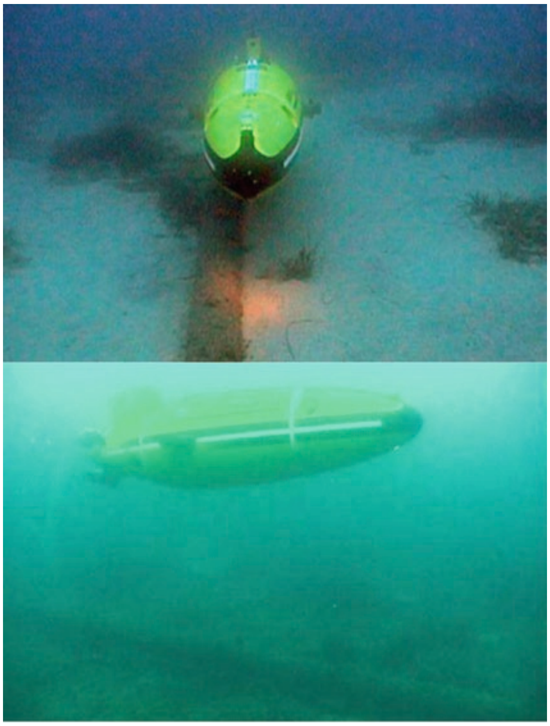

Fig. 2 ALISTAR 3000 tracking a pipe

3.2 Missions

Due to its high stability, maneuverability, and hovering capability, the ALISTAR system is primarily designed for carrying out inspection of underwater structures, such as pipes, risers, mooring lines, wellheads, manifolds, etc., but the system is also able to perform various types of surveys missions (general site survey, pre- and post-lay survey, etc.).

3.3 Inspection Capabilities

The difficulty concerning pipeline inspection with an AUV is mainly because the as-built reports, giving the position of these pipelines, are not accurate enough to be able to pre-program a trajectory with waypoints above the pipeline to be inspected (approximately 5–10 m). Proper video inspection of a pipeline also means that the AUV is directly above the pipeline with a tolerance of about 50 cm on either side and at about 1 m above it. The suite of sensors used for finding and tracking the pipes is a set of profilers and magnetometer.

This suite of sensors enables ALISTAR 3000 to:

Automatically relocate, identify, and 'lock' onto the pipe

Follow the pipe, even when buried, 1 and 2 m above it

Detect cathodic protection anodes

Record video and sonar images of the pipe

During the mission, the absolute position of the vehicle is also recorded, enabling giving the position of the pipe and comparing it to its theoretical position. When the vehicle is recovered onboard the vessel, all data and images recorded are then uploaded in order to be post-processed and presented to the customer using modern techniques such as mozaïcing (Fig. 3).

In addition to the pipe inspection, ECA is also conducting underwater structure inspection development. The aim of this work is for ALISTAR 3000 to be able to fly around an underwater structure in order to record video and sonar images of the structure (Fig. 4) that will then be analyzed onboard the vessel once the ALISTAR vehicle has been recovered and the data retrieved.

4 ALISTER AUV

The ALISTER AUV is based on the DAURADE system developed for the French Ministry of Defense for the purpose of Rapid Environmental Assessment. DAURADE development has been carried out following a contract for ECA by GESMA, the end user being the French Navy Hydrographic Service (SHOM/CMO).

4.1 Description of the ALISTER System

ALISTER's (Fig. 5) main characteristics are:

Length: 5 m long

Maximum body diameter: 0.7 m

Total weight: 950 kg including the complete suite of sensors described below

Operating depth: 0–300 m

It is equipped with two longitudinal thrusters, a set of vertical and horizontal fins aft of the vehicle and a horizontal fin at the front.

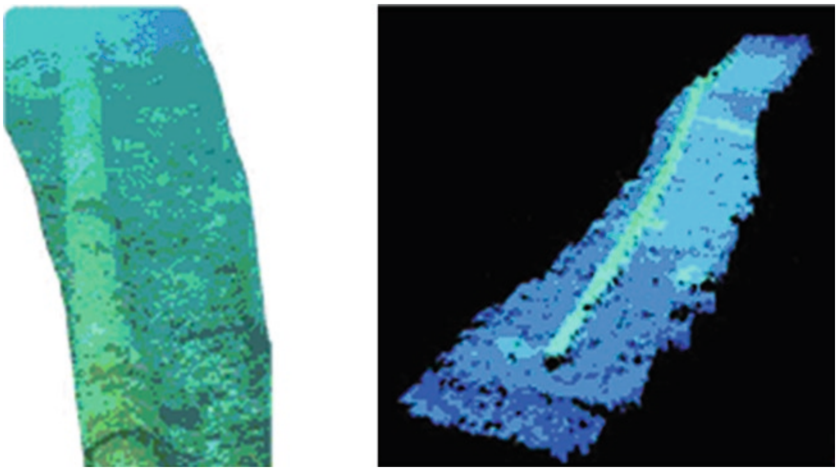

Fig. 3 Reconstructed pipe mosaic (video and sonar images)

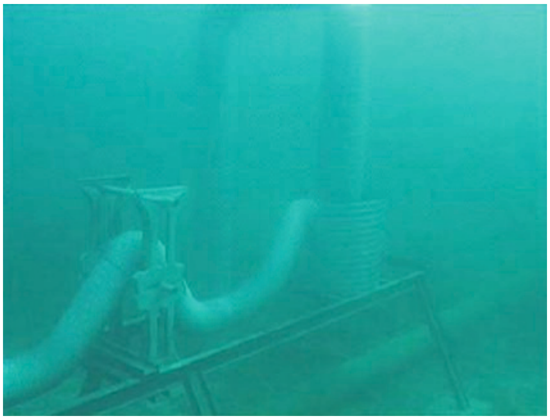

Fig. 4 Underwater structure inspection

Fig. 5 ALISTER vehicle

The actuator layout enables ALISTER REA vehicles to perform terrain following, and the two horizontal thrusters provide improved vehicle safety in case of failure of one of the main thrusters.

The ALISTER vehicle can achieve a maximum speed of 8 knots through the water.

A removable lithium ion secondary battery, housed in a pressure hull, offers 22 kWh of embarked energy with a maximum recharge time of 5 h. This battery provides 10 h endurance at 4 knots, with all sensors working. A computer-based battery management system ensures the monitoring of the battery status.

Accurate positioning, tracking, and monitoring of the vehicle are assured by the following equipment: navigation sensors such as the Inertial Navigation System (INS), Doppler Velocity Log (DVL), Kalman filter, high accuracy depth sensor, altimeter, obstacle avoidance system, GPS, and acoustic localization transponder.

ALISTER is fitted with different communication systems: a radio Ethernet link, an underwater acoustic communication system, and a fiber optic link.

The fiber optic link is used when the vehicle is onboard the ship to download the mission before launching the vehicle and to retrieve the data at the end of the mission. This link can also be used in operation in order to get data in real time, such as video and sonar images.

The radio link is used when the ALISTER vehicle is on surface to transfer data between the vehicle and the surface ship and to operate it either from the main console or from a portable remote console during launch and recovery operations.

The underwater acoustic communication system is composed of a bidirectional low data rate link used to monitor and to send orders to the vehicle.

Safety of the vehicle is provided by a health monitoring system, integrated safety weights, a safety beacon, radio and ARGOS beacons, flasher and water ingress sensors.

4.2 ALISTER Suite of Sensors

ALISTER is designed to receive the following suite of sensors (Fig. 6):

KLEIN side scan sonar
RESON multibeam echo sounder
EDGETECH sub bottom profiler
DIDSON sonar
Echo sounder
Video camera and associated lights

4.3 ALISTER Mission Management System

ALISTER Mission Management System (Figs. 7 and 8) is a part of the overall Autonomy *mission Planning* A Management System, which provides the operating team with a complete set of functions for mission planning, mission monitoring, and mission data post-processing through its two main exploitation interfaces: mission screen and Data Vision screen.

Mission planning: The mission screen offers improved creation of easy-to-use interactive tools for the creation of basic behaviors for AUV (based on predefined models). A mission model is graphically defined, composed of "phases" (associated with behaviors) and "chaining links" that specify the way mission phases are chained together. Security parameters are also defined to complete mission data (working area, intended duration of mission, thresholds, etc.). Models are managed through a database system to make mission data persistent. Integrated simulation function permits verifying the coherence and feasibility of the programmed mission.

Mission monitoring: Mission data can be uploaded from the database to the designated AUV through the Ethernet radio link in the widely used XML format. The embedded part of the Mission Management System is now able to allow the AUV to carry out the mission as specified. A Data Vision screen shows video/sonar data from AUV cameras and navigational parameters to help the operator manually control the robot (in order to get it into the right location for autonomous operations to be activated, and further to recover it at the end of the mission). The mission is monitored by means of a USBL system and, when available, by data received from the AUV through acoustic transmission. Optionally, operations can be supervised by sending single orders to the AUV according to the programmed mission or for security reasons.

Mission Post-processing: After downloading the onboard logged data, the recorder payload data are processed and analyzed by the manufacturer's provided tools.

Recorded navigational data can be re-played (Mission screen) and synchronized with recorded video/sonar data (Data Vision screen) by the operator in a "video player" manner. They also can be specifically processed for transfer to an external analyzing system.

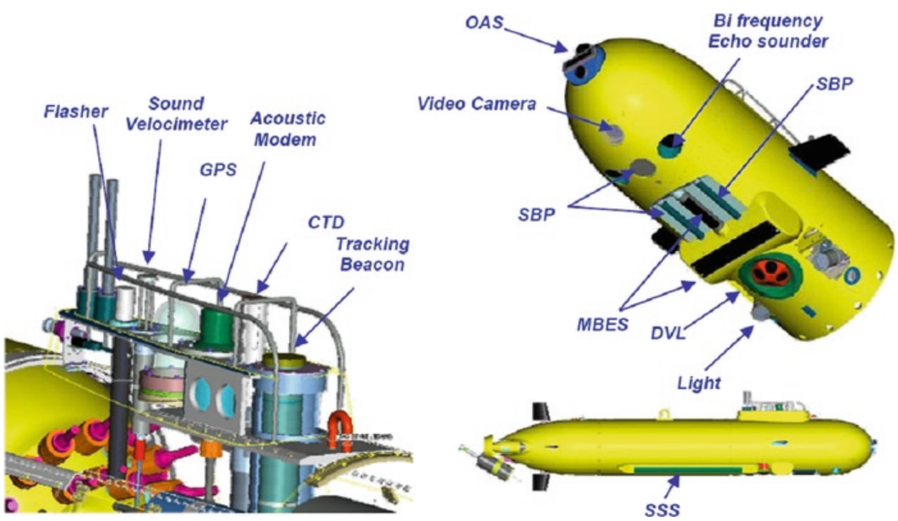

Fig. 6 ALISTER complete suitability

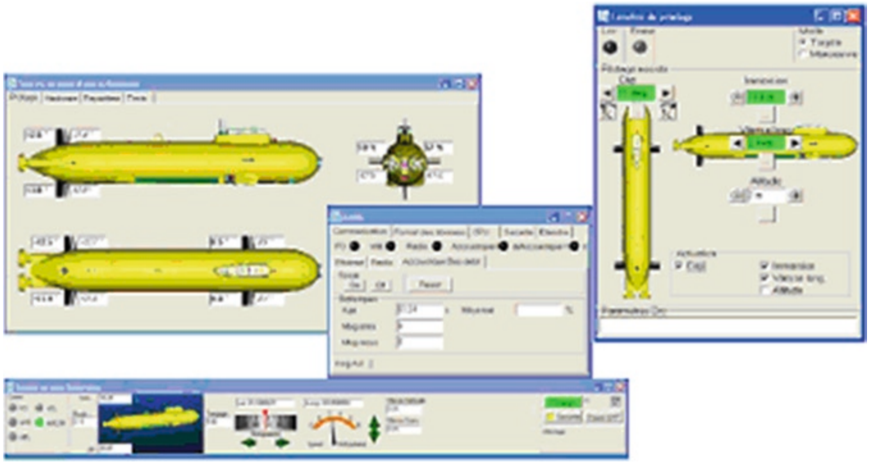

Fig. 7 Vehicle Mission Management Supervision Display

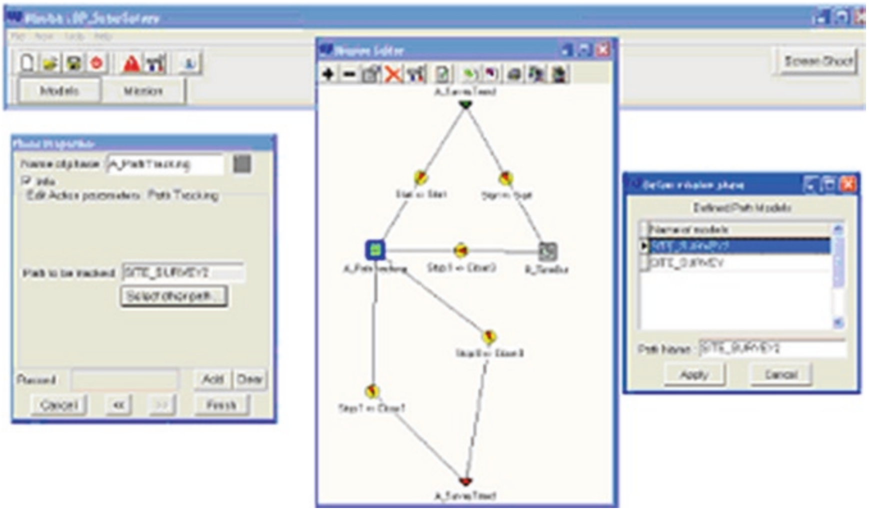

Fig. 8 Mission Management Display

The distributed software architecture of the system based on the "publish-subscribe" data server gives it large scalability and enrichment capacities for AUV behaviors according to the customer's needs.

5 Conclusion: Toward the Future

Several developments are currently being carried out in many countries to integrate Synthetic Aperture Sonars onto AUVs. These developments, combined with the increasing computing power available, will nonetheless enable better performance of underwater object detection because of the better resolution provided by these sensors, and also enable automatic detection and classification of underwater objects.

This, combined with the architecture of a vehicle such as ALISTAR 3000 with hovering capability, will provide the automatic detection and classification of underwater objects, but will also enable their identification by using a set of sensors such as acoustic cameras or low light video cameras.

Bathyscaphs, a Mediterranean Adventure in Marine Dialogues Between France and Japan

Henri–Germain Delauze, Jean–Claude Cayol, and Hubert Jean Ceccaldi

Abstract In the history of deep diving in the oceans, bathyscaphs have played an eminent role. For the first time in the exploration of the earth, mankind constructed machines with the capability to reach depths of several thousand meters and the capacity to carry observers in order to collect information on an unknown world: the deepest parts of the seas.

Such an unprecedented adventure was born mainly along the coasts of the Mediterranean Sea and attracted almost worldwide attention, especially of the maritime countries and particularly Japan.

This paper relates the first steps in deep diving bathyscaphs, the difficulties and the successes of these dives in different parts of the oceanic world and in Japan.

It also relates the great impact of the dives in the seas surrounding Japan on the scientific research community and also on Japan's population. An indirect consequence at that time was the creation of the Société franco-japonaise d'Océanographie of Japan (Nichi-Futsu Kaiyo Gakkaishi) by several Japanese oceanography professors.

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1 Introduction

Since the ancient times, men have dreamed of traveling deep into the sea to discover new worlds. With the bathyscaph, their dreams became reality. First person to use a man-made machine to explore underwater was the Swiss professor Auguste Piccard. He constructed a steel sphere to be immersed at a depth of several thousand meters (4,000 m), fixed to a float full of gasoline. With the help of an engineer from Belgium, Max Cosyns, they developed new ways to realize this kind of machine operation with the FNRS 2 (Fonds National de la Recherche Scientifique of Belgium).

The preceding project of Professor Piccard, the FNRS 1, was an aluminum alloy sphere fixed to a huge balloon filled with hydrogen to reach the highest altitude in the stratosphere. In 1932, he officially attained an altitude of 16,201 m!

To dive into the sea, bathyscaphs have a similar appearance to a submarine, but it is a manned machine constituted of a hollow sphere that is waterproof and pressure-resistant attached to a float full of a liquid lighter than seawater. It was designed to reach and explore the deepest depths of the sea. The steel sphere was heavy and could transport two or three persons: a pilot and one or two passenger(s). The float was filled with gasoline. For the dives, ballast composed of iron shot held in place by an electro-magnet was kept in the bathyscaph. At the end of the mission, the electric current was cut and the iron shot left on the seabed. Then, the bathyscaph could return to the surface.

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2 Some Remarkable Points of History

Several very fruitful exchanges in technology and industry between Japan and France had already taken place by the end of nineteenth century. During the period of *bakufu*, or shogunate Tokugawa, the first cooperation with France was established (Omato and Macouin 1990; Sabouret 2008). In the wave of the modernization of Japan in the Meiji era – which began officially in 1868 – one of the main targets of Japan was to build a national navy. Japanese authorities bought modern ships from western countries, for instance, Great Britain.

However, the Emperor Mutsuhito and the Japanese government asked the French government to help them put together the first national Japanese fleet: it was constructed in direct cooperation with a group of French specialists and a number of Japanese engineers.

One of the most significant examples of cooperation was the presence of François-Léonce Verny (1837–1908), hired first by the Tokugawa shogunate and then by the Emperor Meiji. From 1864 and 1875, more than 40 other French engineers and specialists constructed the first arsenal and dockyards in Yokosuka for the Japanese Navy.

Then, they also constructed other facilities in Kure near Hiroshima and in Sasebo near Nagasaki. Verny also constructed several lighthouses after those of Kanonzaki and Shinagawa, and created a school of naval architecture and engineering as well as the first foundry: his statue still stands in an important location in the port of Yokosuka.

From 1886–1890, the French marine engineer Louis-Emile Bertin (1840–1924) went to Japan as a special adviser and helped to create the first modern fleet constructed in Japan. War cruisers, the warships designed by Bertin and constructed by a Franco-Japanese team, were very efficient, especially the cruisers of 4,300 t, forming the nucleus of the Japanese Navy. They won the battle of Yalu against the Chinese fleet on September 17, 1894. During the war against Russia, they defeated the fleet of that country at the battle of Tsushima on May 27 and 28, 1905.

The First and the Second World Wars decreased the exchanges between the two countries, even if the areas of art and culture have shown a high level of dialogue. In this respect, the foundation of an important institution, the "Maison franco-japonaise" (Nichi-Futsu Kaikan), by the Viscount Shibusawa Eiichi and Ambassador Paul Claudel in 1925–1926 has played an eminent role in these exchanges. It is worth noting that the head office of the "Société franco-japonaise d'Océanographie" of Japan (Nichi-Futsu Kaiyo Gakkaishi) is located in Tokyo in the "Maison franco-japonaise."

After the Second World War, exchanges between France and Japan were less important, but continuous, mainly oriented to the area of physical oceanography, aquaculture, marine biology, artificial reefs, algology, and marine biotechnology, for instance.

In France, in 1950, the French National Navy (Marine Nationale) established a plan to come into that adventure with the construction of the bathyscaph FNRS III.

Jacques Piccard, a Swiss scientist, began a cooperation with engineers of the French Navy in Toulon. After several modifications of the original plan, he left his project and began another one in Trieste with an Italian shipyard in order to construct another bathyscaph, the "Trieste."

In Spring 1954, Gaston Dupouy, Director General of the National Center of Scientific Research (Centre national de la Recherche scientifique, CNRS) decided that the research administration he led would give an important grant for the scientific exploitation of the bathyscaph FNRS III. However, they also give an important grant to support scientific missions in the open sea of the research vessel of Commandant Jacques-Yves Cousteau, the "Calypso."

A special Committee for bathyscaphs was created, and Professor Louis Fage was the Chairman. This committee functioned from 1954 to 1968.

Professor Jean-Marie Peres, who was also the founder of the Centre d'Océanologie de Marseille, was the second chairman of that bathyscaph committee.

By 1955, Captain de Vaisseau Georges Houot and the Chief Engineer of Génie Maritime Pierre Willm planned to construct a new bathyscaph able to reach a depth of 11,000 m, and they called it "ARCHIMEDE." Its design and the management of its construction were managed by the French National Navy in the dockyard of Toulon.

It was a strange machine: 22.1 m long; 5 m wide; weight (when empty) 58 t; 21 t of ballast; 120 t of gasoline (full load); total weight (full load): 200 t; sphere for pilots and passengers: 2.10 m in internal diameter; weight 19 t; of stainless steel (nickel/chrome/molybdene); float of 171,000 l; 3 plexiglass portholes.

In 1961, the "Laboratoire des Bathyscaphes" (Bathyscaph Laboratory) was founded in Marseille by the National Center of Scientific Research (CNRS) in cooperation with the French National Navy. One of us (H.-G. D.) led this laboratory and was asked to participate in the future diving missions of the "ARCHIMEDE."

Commander (Captain de Vaisseau) Georges Houot was the head of the "Bathyscaph Group," including the support ship "Marcel Le BIHAN" and all the maintenance logistics, etc., based in the Toulon dockyard. He drove "ARCHIMEDE" 64 times! Three other pilots were asked to drive this machine: Captain Gérard Huet de Froberville, Lieutenant Philippe de Guillebon, and Captain Gabriel O'Byrne.

Immersed in the sea near Toulon on July 28, 1961, the "ARCHIMEDE," after some experimental dives in the harbor and the bay of Toulon between July 1961 and March 1962, began to carry out numerous dive campaigns in various locations throughout the world:

In Japan, in Kouriles Island trench between May and October 1962 (9,545 m)
In the Puerto-Rican trench between May and August 1964 (8,300 m)
In Greece at Matapan Trench between July and October 1965 (5,110 m)
In Portugal, close to Madeira Island between May and August 1966 (4,390 m)
In Japan, in west Honshu Trench between May and August 1967 (9,260 m)
In Portugal, at the Azores Islands between June and September 1969 (3,400 m)
In the Tyrrhenian Sea in October 1971 (3,250 m)
Near Portugal, Madeira Islands in July 1972 (4,330 m)
In the mid-Atlantic for the operation FAMOUS (French American Mid Atlantic Underwater Survey) in August 1973 (2,800 m) and July (and August?) 1974 (2,800 m), in connection with the USA

On July 22, 1974, the bathyscaph "ARCHIMEDE" dived for the last time. In 9 years, it carried out 139 dives.

Since June 2001, ARCHIMEDE has been displayed in "Cité de la Mer" (City of the Sea) at Cherbourg in Normandy (France).

3 Campaigns in Japan

Professor Sasaki Tadayoshi, from Tokyo University of Fisheries (Tokyo Suisan Daigaku), went to France during a trip to Europe and visited the bathyscaph. He was one of the main organizers of the bathyscaph dives in the seas in Japan. Professor Sasaki founded a Japanese national committee for bathyscaphs. One of the targets of these dives was to reach a depth deeper than the 10,916 m that had previously been reached by the bathyscaph "Trieste" in the Marianne Trench.

During the summer of 1958, the bathyscaph FNRS 3 was transported from Toulon to Kobe by the Japanese freighter "Atsuta Maru." Then, from the cities of Onagawa and Uraga, a first series of dives was carried out. Professor Jean-Marie Peres and Professor Sasaki observed the biological characteristics of specimens living in different water masses. Professor Sasaki measured surface and deep currents and practically saw the two types of waters belonging to Kuroshio and Oyashio. Professor Kumagori observed the rocky seabed and, during one dive, the bathyscaph bumped into an underwater cliff, fortunately without damage.

In August, after 2 months in Japan and nine successful dives, the bathyscaph FNRS 3 came back to France, carried on a cargoship. The campaign was supported financially by the newspaper *Asahi Shimbun*. Some journalists were able to dive in the bathyscaph.

A second series of dives occurred in 1962. In March, in Toulon, the ARCHIMEDE was put on the bridge of the ship Maori to be transported to Japan. It arrived in Yokohama harbor on April 4, 1962, and its support ship, Marcel Le Bihan, was ready to begin its mission on May 8.

Different dives were planned and carried out in the summer of 1962. The crew arrived in Onagawa in May, and the first dive took place in Onagawa on May 22 and in Tokyo on July 3. On July 10, they arrived in Hokkaido, in Kushiro harbor, where they had the help of the Japanese research vessel Umitaka Maru and the frigate Matsu.

During the campaign, several well-known Japanese personalities and scientists went in the bathyscaph ARCHIMEDE: Mr. Husaka from the newspaper *Asahi Shimbun* on June 14, Professor Sasaki and Professor Niino on July 18, Professor Kumagori on July 30, Mr. Hayashida, a photographer, on August 3, Professor Kubo on August 7, and Professor Chiba, on August 11.

The deepest dive on record, on July 25, 1962 in northeast Japan in the Kourile Trench, was under the direction of the pilot, Captain O'Byrne, the marine officer, Professor Sasaki, and of one of us (H.-G. D.), chief of the bathyscaph laboratories from 1961 to 1968. This dive reached a depth of 9,545 m.

During the dives in Japan, new observations of living fauna were observed: siphonophores, medusae, ctenophorans in plankton, worms, and numerous traces of animals, often unknown (Picard J, 2009, personal communication), several new species of fishes, worms, crustaceans, and cephalopods. Pennatulas were visible at 9,500 m depth, as were several benthic species. Dr. Lucien Laubier participated in these dives and observed the deep sea fauna. He also helped the technical team to enhance the quality of the observations with new equipment such as celerimeters, current meters, pH meters, etc.

Planctonologists wished to descend very slowly to observe thoroughly mainly the upper layers of the sea. Benthologists preferred to descend to the bottom rather quickly to observe it for a longer time (Picard 2009). Surprisingly, deep, slow currents exist near the bottom at these exceptional depths. ARCHIMEDE has the capacity to move slowly with small propellers when near the bottom.

A third series of nine dives was carried out between May 11 and July 22, 1967. The aim of the first three dives was geological surveys, physical observations for the next two and biological studies for the last four dives. The pilots were Captains Froberville and Houot. The navigators were Jean Jarry, one of us (H.-G. D.), and Philippe de Guillebon. The observers were Gilbert Bellaiche, Jean Jarry, Charles Bornheim, and the Professors Drach, Peres, and Abe observing colored holothurians, shrimps, and several new and unknown species for the first time.

All the dives were deep, between 4,510 m and 9,260 m; Professor Abe reached the depth of 7,190 m.

4 The Role of Bathyscaphs Within the France–Japan Relationship in Oceanography

The Japanese public is immensely curious about these new deep submarine activities.

This interest can be explained by the unique possibility presented by this new apparatus to observe the bottom of the oceans in the colliding boundaries of the tectonic plates where the earthquakes originate. It could perhaps also be explained by the marine biological studies of H.M. the Emperor Showa (Hirohito) and his family.

The newspaper *Asahi Shimbun* was very interested in publishing original papers on the subject. Its management asked the readers to participate on a financial level and organized an important collection of funds at the national level. Everybody in Japan has heard of the French bathyscaph.

Professor Jean-Marie Peres, Pierre Drach, and one of us (H.-G. D.) have been received officially by H.M. the Emperor Showa (Hirohito) for a private reception.

It is interesting to note that the dockyard of Yokosuka has been the headquarters of Jamstec for years. In this dockyard, the deep diving system "Shinkai 6500" and the vessel-support "Yokosuka" are usually located. We noticed with pleasure that Professor Imawaki Shiro, current President of the Société franco-japonaise d'Océanographie of Japan, is actually one of the main actors in the headquarters of Jamstec in Yokosuka.

In the wake of the bathyscaph campaigns in Japan, cooperation between the French Navy and the Japanese Navy has attained the highest level. In the civilian domain, cooperation between Ifremer and other national institutions had been developed and is well organized.

Since 1984, direct exchanges and cooperation between scientists by the channel of the two Sociétés franco-japonaises d'Océanographie (Nichi-Futsu Kaiyo Gakkaishi) have been friendly and efficient. Some cooperation between French and Japanese cities and ports, such as Le Havre and Osaka, Lyon and Yokohama, Nantes and Niigata, and Marseille and Kobé, are also in progress.

More recently, Dr. Claude Lorius received a very important Japanese prize, the "Blue Planet" Prize, granted by the Japanese Foundation Asahi Glass, for his fundamental work on ice of the polar zones and fine analysis of their gas composition.

5 Role of the Mediterranean

In all parts of the organization and execution of these deep dives using the bathyscaphs FNRS III and Arcimedes, numerous scientists and technicians from the Mediterranean coasts have participated. They were from Marseille, where the teams of oceanographers developed their research, mainly in Marseille: Henri Germain-Delauze (32 dives), Professor Jean-Marie Peres (13 dives), Jacques Picard (3 dives), Philippe Taillez, Jacques-Yves Cousteau, Jean Jarry, and Gilbert Bellaiche. They were also from Toulon, where the bathyscaph ARCHIMEDE was constructed, maintained, and tested: Captain Georges Houot and Chief-Engineer Pierre Willm. Others were from Villefranche-sur-mer: Gregoire Tregouboff (12 dives), and even from North Africa: Francis Bernard and Robert Dieuzeide.

The development of bathyscaphs is really a Mediterranean adventure.

In an excellent memoir, Jarry (2003) gives a series of nice personal descriptions related to almost all of his dives in various parts of the world.

6 Conclusions

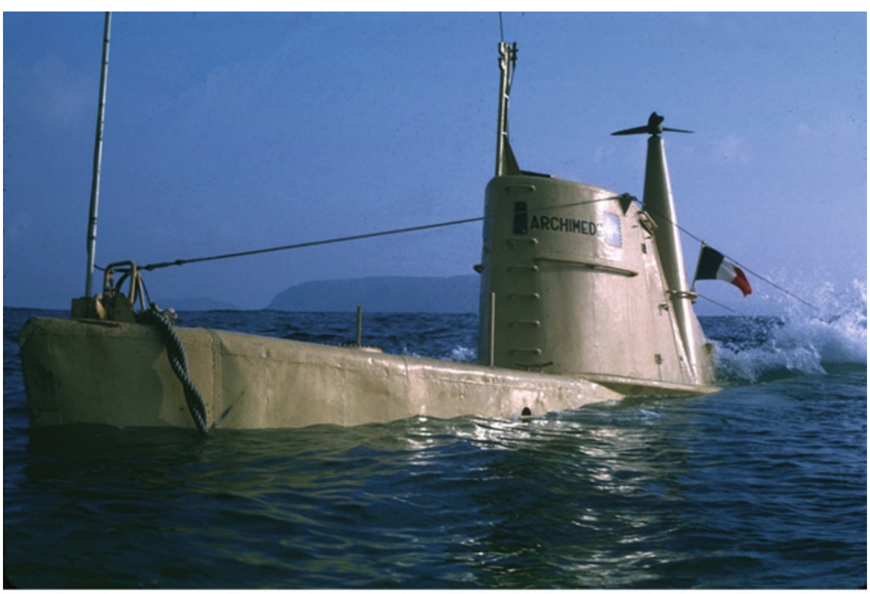
The relationships between France and Japan in the field of marine sciences and technology have a long history, since the nineteenth century. His Excellency Mr. Iimura Yutaka, Ambassador of Japan in France, pointed out, on September 12, 2008, during our meeting in the "Maison de la Culture du Japon" (Paris Nihon Bunka Kaikan) in Paris, that this scientific meeting is included in the official documentss of the 150th anniversary of the "Traité de Paix, d'Amitié et de Commerce" (Treaty of Peace, of Friendship, and Trade) between the two countries, established on September 3, 1858.

After the cooperation in the construction of a naval force of dockyards, military navy, and warships, deep diving using bathyscaphs opened a new field of cooperation.

Since these important beginnings, the cooperation has shown numerous diversifications in almost all chapters of marine science and technology. There was and still are numerous fields of common interest between France and Japan in the fields of marine sciences and technologies. One of the central points of convergence is the exploration of oceans, not only from its surface and/or from space, but at its deepest depths.

Both countries have constructed very evolved "diving machines," such as the Shikaï 2000 and Shinkai 6500 with its support vessel Yokosuka by Jamstec, which dove to depths of 6,527 m off Sanriku, Iwate Prefecture; in France, the Victor 6000, teleoperated, and CYANA, and for the USA, ALVIN and its catamaran support Lulu.

The true successor of the bathyscaphs was the deep submarine Nautile, constructed with a manned sphere of titanium, 8 m long, 19 t in weight, with a capability



Bathyscaphe ARCHIMEDE

Henri-Germain DELAUZE - ARCHIMEDE

of 3 h of observations and 120 h of security. Its support is the research vessel "Pourquoi Pas?" Russia has constructed the deep submarine MIR I and MIR II, and the USA, the SEA CLIFF.

There are several new submarine apparatuses, such as Yoyo profilers gliders (or glisseurs), Autonomous Underwater Vehicles (AUV) such as Epaulard, Alister Rea. IDef, ASTER, and several others.

Several new directions have been opened by these deep dives: new submarine networks of observations, as in the Ligurian Sea or the European ESONET Network.

Once more, the Mediterranean is opening the way, as a new European Center of Submarine Technology was inaugurated near Toulon, in La Seyne sur Mer, on October 2009. The Mediterranean marine cluster and IFREMER will be the main actors in developing the new technologies.

In the near future, the *SeaOrbiter* project will open a new path in the exploration and direct observation of the sea.

The direction in which we are moving today was opened by the great forerunners, Léonce Verny, Louis-Emile Bertin, Paul Claudel, the Viscount Shibuzawa Eiichi, Georges Houot, Pierre Willm, Jean-Marie Peres, and many others, and it is a bright way. This symposium is another example of that movement, and it opens again new fields of fruitful cooperation.

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Applied High-Temperature Superconductor Bulks and Wires to Rotating Machines for Marine Propulsion

Brice Felder, Motohiro Miki, Yosuke Kimura, Keita Tsuzuki, Ryona Taguchi, Shiliang Yuan, Yan Xu, Tetsuya Ida, and Mitsuru Izumi

Abstract High-temperature superconductors (HTS) allow providing to high-torque density rotating machines a compact, efficient, and excellent operation. Field poles that act as magnets, providing a magnetic field of more than 1.5 T around the armature structure, were initially designed for an axial-gap-type rotating machine, i.e., with the flux parallel to the rotor axis. Melt-growth Gd-123 bulks as well as Bi-2223 wire windings have been successfully assembled on the rotor plate. No iron core was used, though being an auxiliary flux control as found in most HTS motors. Both bulk and wire types have accomplished a practical motor operation within a limited output range. For bulks, a 15 kW, 720 rpm, synchronous motor was designed and tested in the group of Tokyo University of Marine Science and Technology (TUMSAT), Kitano Seiki, and University of Fukui. A bulk field pole was cooled down by liquid nitrogen and was magnetized in the motor. To enhance the torque to more than 30 kW, we developed a thermosyphon system using condensed neon. Another kind of field pole composed of HTS wire for large-scale marine propulsion is also discussed on a 100 kW, 230 rpm tested machine. A closed-cycle condensed-neon thermosyphon associated with thermal insulation is reported.

1 Introduction

Recently, several kinds of high-temperature superconductor (HTS) applications have emerged, such as transducers for underwater sonar, magneto-hydrodynamic oil separation systems, or rotating machines for ship propulsion motors and tidal turbine generators (Nick et al. 2002; Kalsi and Karon 2003; Kwon et al. 2008). The conventional design of the latter machines has a radial-gap-type structure. Nevertheless, our group has been developing two different kinds of axial-gap-type synchronous HTS rotating machines, such as a ship propulsion motor, since 2001. The distinctive design of our machine is shown in Fig. 1.

The rotor and the stator are interlaminated along the direction of the shaft inside the machine. The advantage of an axial-gap-type motor is that it can considerably miniaturize the machine size, since increasing the number of rotor plates as well as the stator units enables us to efficiently use the machine dimension and to increase the torque as well as the output.

A small-size synchronous machine with Gd-123 bulk-HTS pole-field magnets, which was aiming at the sub-MW class of ship propulsion motors, was constructed in 2002. The evaluation test was conducted in 2003, and a 3.1 kW output with 720 rpm was reported (Matsuzaki et al. 2004).

On the other hands, the HTS winding coil is convenient to increase the magnetic flux as a function of applied DC current. We have constructed an axial-gap-type

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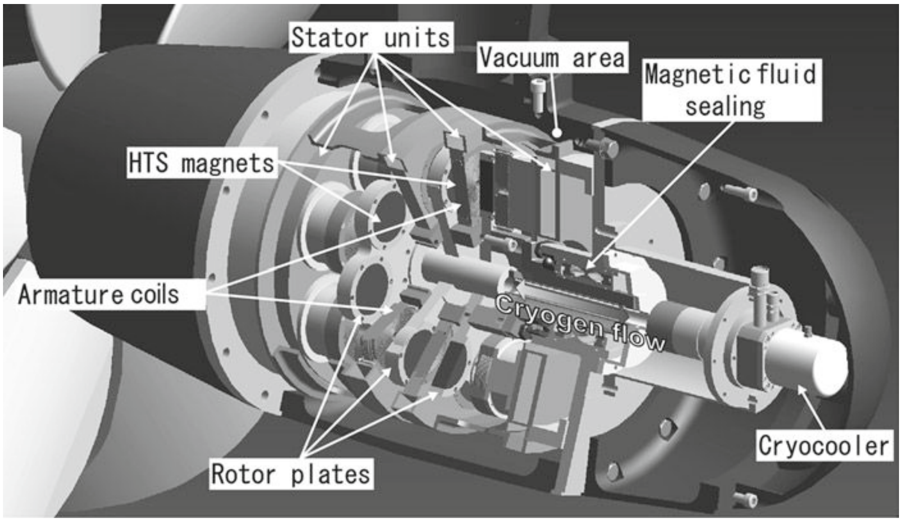

Fig. 1 Conceptual diagram of an axial-gap-type HTS (high-temperature superconductor) motor with podded-propulsion system

synchronous motor based on coupled double-pancake coils (DPC) of Bi-2223 superconducting wire.

The cooling and rotating tests were performed in 2008 (Sugyo et al. 2008).

2 Design Concept of the Motor with Gd-123 Bulk HTS Pole-Field Magnets

This distinctive machine was designed as a propulsion motor for small ships below 500 t. We have applied melt-textured $GdBa_2Cu_3O$ (Gd-123) HTS bulks as pole-field magnets to the synchronous machine. These bulk HTS can provide a high magnetic field over 3 T at 77 K. The present motor has eight bulk HTS whose sizes are 60 mm in diameter and 20 mm in thickness, fixed on a single rotor plate. They were cooled down below T_c by using a circulation of liquid nitrogen. The structure of our rotor plate, bulk HTS magnets being cooled by a lateral flow of cryogen, benefits from the larger thermal conductivity of the bulk-HTS crystals along the crystallographic *a–b* plane. In addition, for thermal insulation, the output shaft connects the rotor plate through the intermediary of GFRP (Glass Fiber Reinforced Plastics) parts, the whole being called the torque tube. Thanks to this structure, we succeeded in reducing the conductive heat invasion below 13 W. The rotor and stator units were kept in vacuum to prevent convection heat exchange with the outside. Six pairs of armature copper coils have been built up and fixed on the armature cooling base. Each armature coil was 84 mm in diameter and 20 mm thick, composed of ten layers of 20 turns of a 2-mm copper wire and cooled by a circulating coolant such as liquid nitrogen and/or liquid fluorine. Figure 2a shows the cooling structure and the geometries of the rotor and stator.

There are two different methods for magnetizing bulk-HTS magnets. The field cooled magnetization (FCM) method is the most conventional way to magnetize bulk HTS. Another one is the Pulsed Field Magnetization (PFM) method, which we proposed for bulk HTS using a pair of copper vortex-type coils (Sugimoto et al. 2004; Morita et al. 2006). The FCM method does not only provide a high trapped field, but also a homogeneously distributed field. In contrast, the PFM method applies a nonuniform magnetic field, and the maximum trapped field is lower than the FCM method.

Yet, in this study, we chose the PFM method to minimize the dimension of the motor. We adopted the vortex-type armature copper coil windings for a dual purpose: the armature coil function shared with the magnetizing coil's. The average trapped magnetic field inside the motor with our PFM method reached over 0.6 T on the surface of the armature coil. The trapped magnetic flux provided the motor with a performance of 10 kW at 720 rpm as shown in Fig. 2b.

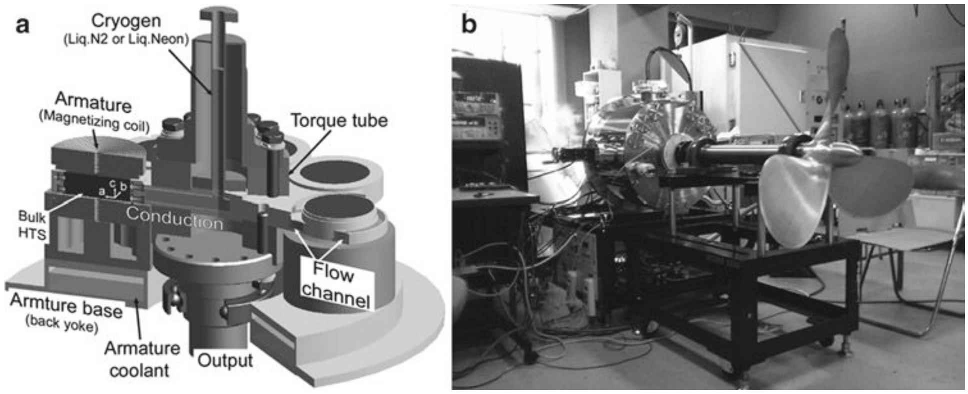

Fig. 2 (**a**) Structure of the bulk-HTS motor. (**b**) Single-rotor-type bulk-HTS motor with 720 rpm rotating test

Table 1 Design specifications of the bulk-HTS synchronous motor

Motor type		Axial-gap synchronous motor	
Diameter (mm)	500	Torque (N · m)	398
Length (mm)	660	Field-pole type	Gd-123 bulk HTS
Weight (kg)	200	Number of poles	8
Rotating speed (min^{-1})	720	Armature type	Copper winding
Design output (kW)	30	Number of armature coils	6

The design specifications of the bulk-HTS motor are shown in Table 1.

3 Specifications of the Motor with Bi-2223 HTS Wire Winding Coils

The prototype of the motor was aimed at the MW class of ship propulsion systems. The vertical-arranged motor (Fig. 3b) is applicable to conventional podded-propulsion systems as well as azimuth thrusters. A coreless field-pole coil of Bi-2223 HTS wire provides a larger output power than the bulk HTS because the total magnetic flux is also larger.

A double-pancake coil (DPS)-shaped field-pole coil is 160 mm in diameter and 26 mm thick, including the cooling plates, and is wound by 420 m of Bi-2223 tape. Sixteen HTS DPCs, forming eight split-type field poles on the rotor plate, are cooled down by using the thermal conduction from the evaporator. The closed-cycle thermosyphon cooling system using condensed neon cooled by a GM-cryocooler has been designed for the results given in this study, but was also recently greatly enhanced by the addition of optimized oxygen-free copper fins inside the condensation chamber and the replacement of the heat exchange plate. Armature coils are made of copper wire in association with a Stycast impregnation.

Figure 3a shows the inner structure of the Bi-2223 wire motor, which has the same design as the bulk-HTS motor. To reduce the heat invasion from the electrodes to the cryogenic field poles, we developed a crucial product made of bulk-HTS material. The developed HTS bulk-current-lead (BCL) has enabled the application of a large current up to 300 A without any heat leak (Tsuzuki et al. 2008).

Table 2 shows the design specification of the HTS synchronous motor with Bi-2223 HTS wire winding field-pole magnets.

4 Cooling and Magnetizing Properties of the Bulk-HTS Motor

In this section, we compare the cooling properties of each motor. For the bulk-HTS motor, we adopted liquid nitrogen as the circulating cryogen. The rotary

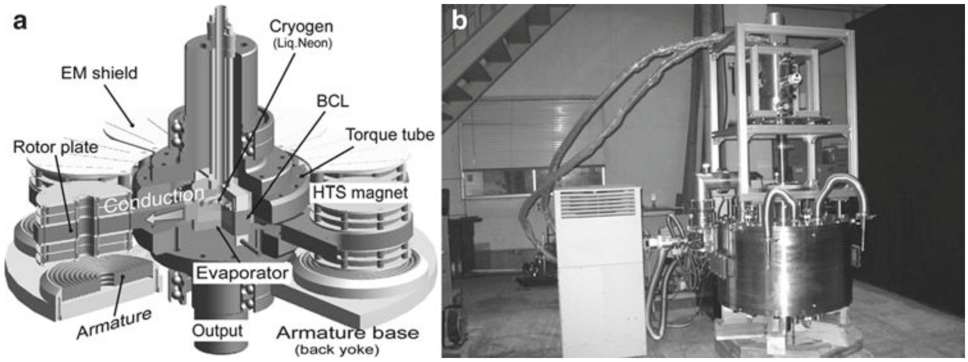

Fig. 3 (**a**) Structure of the Bi-2223 HTS wire motor. (**b**) Experimental "vertical-arranged" condition with a GM-cryocooler

Table 2 Design specification of the motor with the Bi-2223 HTS windings

Motor type		Axial-gap synchronous motor	
Diameter (mm)	850	Torque (N · m)	1326
Length (mm)	507	Field-pole type	Bi-2223 HTS winding
Weight [kg]	1044	Number of poles	8
Rotating speed (min^{-1})	230	Armature type	Copper winding
Design output (kW)	100	Number of armature coils	6

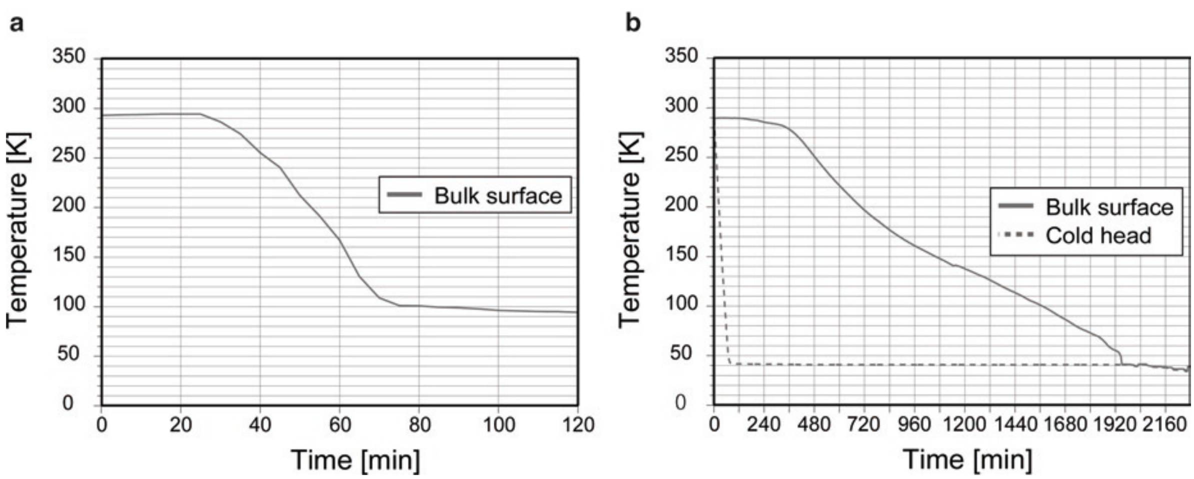

Fig. 4 (**a**) The cooling results of the bulk-HTS motor with liquid nitrogen. (**b**) The cooling results of the bulk-HTS motor with condensed neon

joint, whose structure was simple, connected the rotating shaft to the liquid nitrogen container in order to supply the coolant continuously. The temperature of the center of the bulk HTS reached 94.5 K for a cooling time below 2 h (Fig. 4a). Thanks to our PFM method using vortex-type coils, the trapped field reached 1.04 T (Miki et al. 2006).

These results led to the conclusion that the characteristic cooling structure of the rotor plate is a successful design for bulk-HTS magnets. A general property of the bulk HTS is that their critical current J*c* increases when temperature decreases. Cooling down the bulk HTS to lower temperatures is a shortcut to increase the output. So, we thought about employing condensed neon, whose temperature is lower than that of liquid nitrogen. We used the closed-cycle thermosyphon cooling system shared with the Bi-2223 wire HTS motor. Thanks to this modification, the temperature of the center of the bulk HTS reached 35 K (Sano et al. 2008). This result is shown in Fig. 4b. After cooling, we tried to magnetize them with the PFM method. The result of the magnetization was that we obtained over 1.5 T on the surface of the bulk HTS.

With the new-type thermosyphon designed in 2009, whose load test proved that it can keep a rotor plate at the temperature of 30 K with up to 45 W of heat load, we can hope to cut the cooling time by three, reach lower temperatures than so far, and then trap a still higher magnetic field.

5 Cooling and Magnetizing Properties of the Wire-HTS Motor

The Bi-2223 HTS wire motor with a single rotor plate was constructed and tested from 2007 to 2008. According to the early design specifications, we adopted a closed-cycle thermosyphon cooling system in which we employed the condensed neon gas. However, there were serious problems on the rotary joint when using the condensed neon gas. Thus, in order to check both cryogenic and vacuum specifications, we had to employ liquid nitrogen and liquid helium.

Figure 5 shows the temperature transient of a pole-field coil made from Bi-2223 HTS wire winding, during the cooling test of the wire-HTS motor. In this experiment, the motor was first cooled by liquid nitrogen for 410 min so that the temperature of the pole-field coil reached 100 K. Then, we exchanged liquid nitrogen for liquid helium. Finally, we succeeded in cooling down the whole pole-field coil below 20 K within 780 min. Figure 5a shows the evolution of the cooling.

This result shows that the thermal insulation structures, including the BCL, were satisfactory on this motor. The excitation test was conducted after cooling. The HTS-wire winding coils generated over 1.4 T with a 200 A DC current as shown in Fig. 5b. In this situation, total heat generation from the field-pole coils was calculated as being below 4 W. The operating test for this motor was conducted and the maximum synchronized rotating speed was 3 min^{-1} due to a problem coming from the test facility.

6 Conclusion

The bulk-HTS motor, including no collector ring and no current lead to excite the field-pole magnets, provides high redundancy. And the simple inner structure makes it smaller, lighter, and easier to maintain. Large bulk-HTS magnets, whose dimensions are up to 140 mm in diameter, were synthesized by the ISTEC group in Japan. The PFM method for these bulks has improved, and 0.6 T were trapped at 77 K (Kimura et al. 2008; Yamaguchi et al. 2008). The total magnetic flux of a large bulk-HTS magnet, three to four times larger than a small one, indicates the capability of

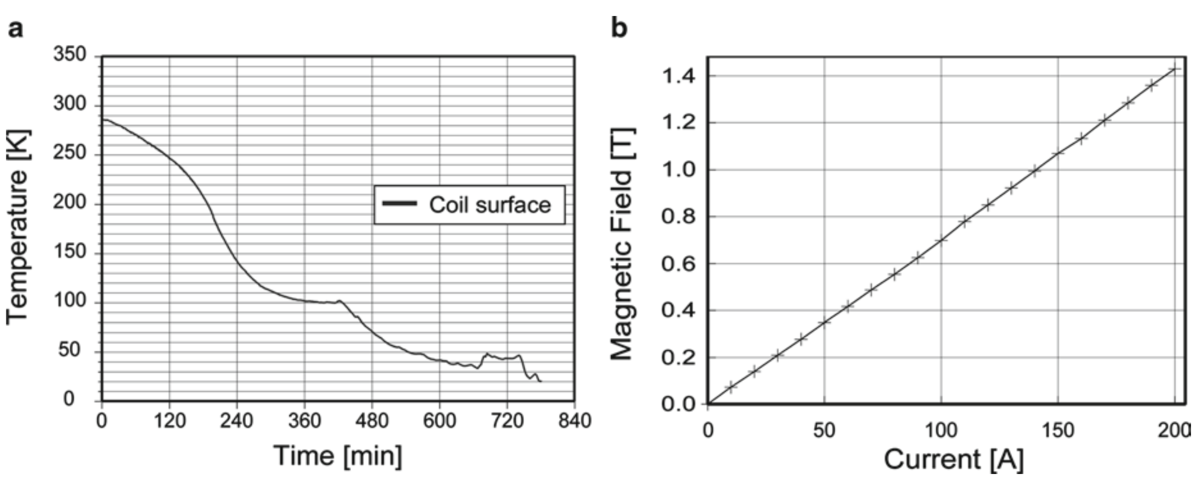

Fig. 5 (**a**) The cooling results of the Bi-2223 wire-HTS motor with liquid nitrogen and liquid helium. (**b**) The magnetic flux density of the field-pole coil as a function of the applied DC current

large-scale applications of bulk-HTS motors. As for the Bi-2223 wire-HTS motor, the technology for a vertical arrangement was confirmed. The new-type BCL, which allows a DC current up to 300 A without any heat invasion from the electrodes, was considered for use in a narrow space. Axial-gap-type HTS motors with coreless Bi-2223 HTS DPC field poles have already become the basis of numerous applications such as tidal turbines, while isolated windings can be used as transducers for underwater sonar or magnetic oil separation systems for contaminated water. Now, we have already developed an innovative small-sized rotary joint for the cryogen, as well as deeply upgraded the condensed-neon thermosyphon, thanks to an optimized design. We tried to apply the present features to both bulk-HTS and wire-HTS motors. More details about these motors cooled by condensed neon gas with a GM-cryocooler will be announced in the near future.

Acknowledgments The work on the bulk-HTS motor was partially supported by the Technology Development Fund of the Ship & Ocean Foundation Japan, and the fund was instituted by donations from the Nippon Foundation derived from revenues from motorboat racing. The work on the wire-HTS motor was partially supported by the Japan Railway Construction, Transport and Technology Agency (JRTT).

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Oceanographic Real-Time Measurement on Buoyancy Beacon Feedback in the Rhône Delta and Gulf of Fos Fruce France

Pierre Gaufrès, B. Andres, and F. Dufois

Abstract The instrumentation implanted since 2003 on two buoyancy beacons (BFI) in the Gulf of Fos-sur-Mer and in the delta margin provides local oceanographic and meteorological real-time data. The device is based on Doppler technology sensors fixed on the beacon pipe. Meteorological sensors, acquisition, and data transmission systems are clamped to the emerged structure close to Aide to Navigation (AtoN) and their energy systems. The monitoring of this device and analysis of recorded observations for nearly 4 years allows specifying uncertainties linked to this kind of measurement and provides a reliable feedback on the exploitation of the observatory.

A database of simultaneous measurements of beacons structure behavior (tilting measurement) and oceanographic and meteorological parameters has been used to design new BFIs for future projects (Lavezzi in Bonifacio Strait, Corsica, Port of Marseilles Authority, etc.).

Analysis of the current measurement enables one to characterize the current profiles on the water column located above sensors, and to distinguish flow stratification effects and wind-current correlations.

Although originally designed to monitor the structure behavior and to supply wave climate to marine pilots, the system has developed different time applications. For example, sediment dredge bottom discharge has been optimized in the Gulf of Fos and frequency analysis of sea level has been compared to coastal tide gauge data for the historical Rhône flood in December 2003.

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1 Introduction

The origin of the instrumentation of two BFIs in the Gulf of Fos came from different needs from two partners. On the one hand, the CETMEF (French Institute for Maritime and Inland Waterways) wanted to improve in situ knowledge of BFI's structural behavior. However, Port Authority of Marseilles, owner of a wavemeter (Datawell type) in the Gulf of Fos, often stormed by ships, wanted to secure this. In 2001, the Port Authority of Marseilles, CETMEF, and Maritime Service of Bouches-du-Rhône (SM13) agreed to build a metocean observatory on two BFIs (Fig. 1) located in the Golf of Fos (France).

2 Instrumentation Description

The current profiler is an Awac type from Nortek. It uses Doppler technology, based on a phase difference calculation between the emitted and received frequencies. Phase difference is linked to particle speed in the water (plankton, sediments, etc.). Those particles are driven by the current and permit its measurement. The instrument also measures sea level (pressure sensors), sea temperature, and eventually wave parameters. The Awac is clamped on supporting arms perpendicular to the beacon's main pipe (Fig. 2).

The arms were calculated so as to install ADCP 3.5 m away from the main pipe so that Awac beams

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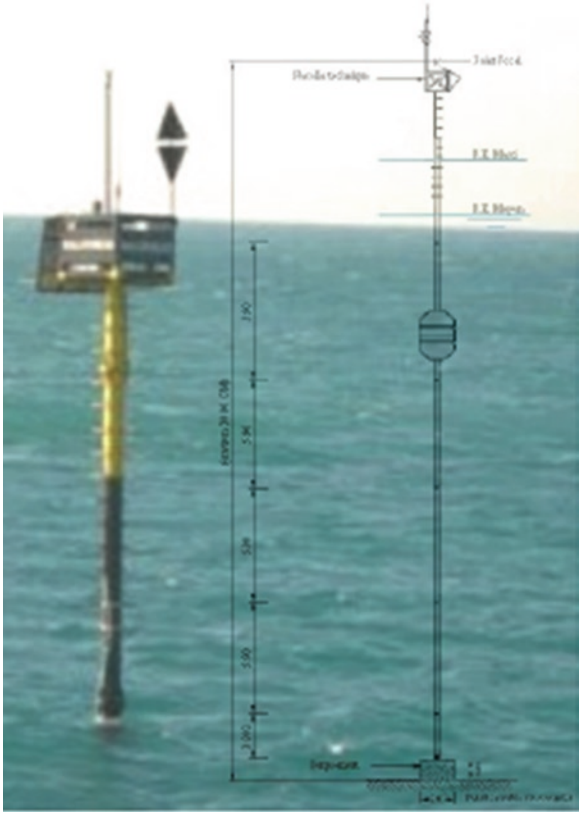

Fig. 1 BFI in its environment (*left*) and structure (*right*)

Fig. 2 ADCP on the perpendicular arm during an inspection

are not disturbed by BFI structures. Depths are 15.5 m for the Fos no. 2 ADCP and 14.5 m for the Balancelle one along the Rhône deltaic margin. The ultrasonic anemometer is a Vaisala WS425 type. It was fixed on a metal bar on the BFI nacelle, 7.0 m above average sea level.

3 Operational Use and Research

For the Marseilles Port Authority navigation needs, data are displayed on control screens of the Integrated Regulation Center (CRI) located in Port-de-Bouc and can be communicated to approaching ships. Data are treated by CETMEF for applied research: structural behavior, marine hydraulics, statistic analysis, met-ocean parameters, treatment, etc. Correlations were produced among wave height, wave direction, wind intensity, wave height, wind direction, and current direction. The surface current and wind are well correlated, especially the mistral and SW storm winds (135° and 315°) (see Fig. 3); shear current on the subsurface may be observed during some episodes (Fig. 4).

4 Conclusions

Years of real-time data experience provided by profiler technology fixed on BFIs shows the interest in coupling marine observations to AtoN buoys: direct energy supply (solar or wind), localization, safety of equipment, etc. However, we advise keeping the instrument in its original configuration, including magnetic North, independently positioned on sea bottom on a tripod and connected to a buoy by a short umbilical wire to avoid trawling. Many prospects are foreseen with spatial altimetry or coastal oceanic modeling for integrating those observations as boundary conditions.

Current projects with actual and future MESURHO instrumentation are projected in order to study flow structure in the Rhône river mouth, hydrosedimentary

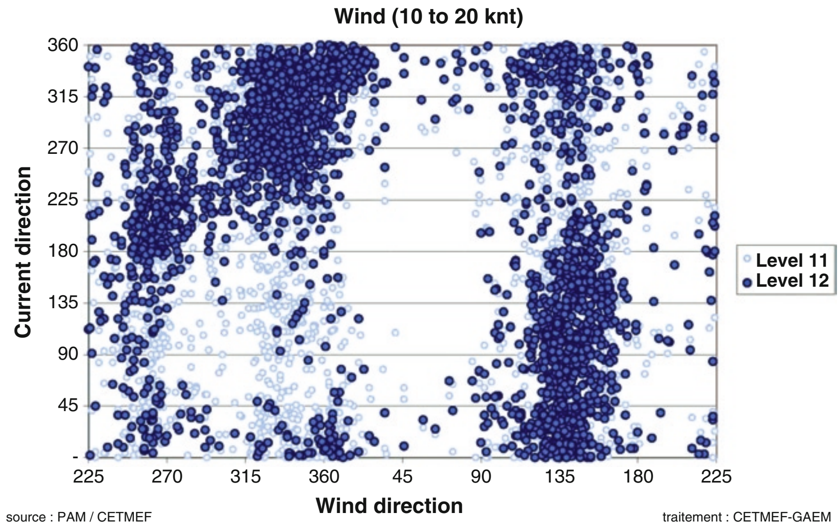

Fig. 3 Wind/current directions correlation (idem)

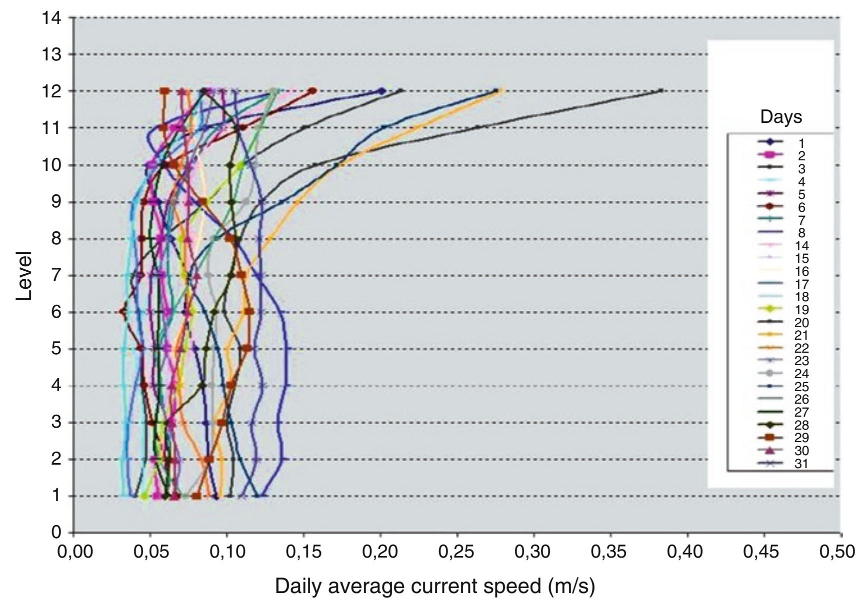

Fig. 4 Daily average current profile (idem)

processes off the coast of the Gracieuse They, or North Occidental Mediterranean Sea meso-scale dynamics of vortexes identified in the 3D Symphonie model.

Acknowledgments G. Cunty, H. Barreda (CETMEF), C. Roblin (SM13), B. Vion (CETE Méditerranée), J. Moisan, and F. Denivet (Port Authority of Marseilles).

Analysis of Phosphatase Activity from Aquatic Heterotrophic Bacteria at the Single Cell Level by Flow Cytometry: Example of a Development Achieved in the Regional Flow Cytometry Platform for Microbiology (Precym) Hosted by the Oceanology Center of Marseille

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Abstract Marine microbes play a very important role in biogeochemical cycles. Because of their biodiversity, the diversity of their metabolisms, and their physiological heterogeneity (live/dead, active/inactive cells), it is necessary to study them at the single cell level.

Flow cytometry has become a technique of choice to reach this goal, providing a fast, reliable, and multiparametric analysis of individual cells. However, the cost of such a technique makes flow cytometry out of reach for small institutes or laboratory units. It also requires a qualified and well-trained operator.

To make this technique available to all the microbiologists of the "Provence Alpes Côte d'Azur" Region, a Regional Flow Cytometry Platform (PRECYM), hosted by the Oceanology Centre of Marseille, was created by a consortium of six laboratories. Its goal is to provide researchers with the possibility to consider new approaches and to guarantee optimal technical, economic, and scientific exploitations of these instruments.

An example of such a technical development is described in this chapter. The ELF97 phosphatase substrate has been shown to be a performing tool to assess the phosphorus status of phytoplankton populations.

Recently, this technique has been successfully applied to marine heterotrophic bacteria in culture samples in which ELF-labeling was observed by microscopy.

In this study, we describe a new protocol for ELF-labeling of heterotrophic bacteria that allow detection by flow cytometry. ELF-labeled samples were stored in liquid nitrogen for up to 4 months before analysis without any significant loss of total or ELF-labeled cell abundance and of relative ELF fluorescence intensity.

This method enables studying ectoenzymatic alkaline phosphatase activity of heterotrophic bacteria at the single cell level in environments even with low cell abundance. It has been successfully applied in both fresh and marine samples.

This approach is set to improve our understanding of the physiological response of heterotrophic bacteria to phosphorus limitation.

1 Introduction

The oceans cover more than 70% of the earth (Lalli and Parsons 1997). Most of the marine organisms are actually microscopic and unicellular. Aquatic microorganisms present a wide range of size from 0.2 to 100 µm and make up to 50% of the total biomass of the planet. They play crucial roles in the functioning of the Earth's biosphere and dominate the marine ecosystem in terms of biomass, with a high rate of turnover (Azam et al. 1983). They are responsible for:

(i) CO_2 uptake for organic matter production (about half of our planet's annual primary production)
(ii) Most of the organic matter (OM) mineralization in the oceanic water column (CO_2 release)
(iii) Playing an essential role in regulating the climate (contribution to the atmospheric CO_2 sequestration

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in the deep ocean; producing chemically active biogases)

(iv) Ecosystem and public health concerns linked to the toxicity of certain species

Because of their biodiversity (including algae, protozoa, bacteria, archaea), their important biomass, and their large panel of metabolisms that make them ubiquist, they represent a key value in aquatic systems, and in particular in the cycle of chemical elements (carbon, nitrogen, phosphorus). To understand and apprehend the relationships between these microorganisms and their ecosystem, it is of prime importance to identify (at the species level), quantify (abundances), and characterize (cell activities) these microorganisms, which are often present in different physiological states (live, active, inactive, damaged, and dead). These studies imply analyses at the single cell level, and flow cytometry, together with fluorochromes, is the technique of choice to get fast and accurate data representative of the whole populations identified within a heterogeneous natural sample (Burkill 1987).

The need for such a technology led to the creation of the Regional Flow Cytometry Core Facility PRECYM (Plateforme Régionale de Cytométrie pour la Microbiologie) created by the synergy of several laboratories (for more details, see http://precym.com.univ-mrs.fr). The goal of this facility is to provide laboratories and scientists with the most advanced technology and protocols, and thus offer them the possibility to consider new approaches in their studies and opportunities in their work they could not afford without these instruments.

2 Flow Cytometry Principle

During the last decade, flow cytometry has become a very powerful technique and has impacted a wide range of fields from basic cell biology to genetics, immunology, molecular biology, microbiology, and environmental science. The principle of flow cytometry can easily be understood from its definition: measurement of the optical properties of cells (cyto-) flowing at the very center of a liquid sheath as they are individually intercepted by a light source focused at a very small volume. Particles (i.e., cells) are examined in a very short time (2–10 μs), during which several optical signals are detected and collected, mainly light scatter and fluorescence emissions in the visible spectrum.

Signals coming from the cells are photons either from the laser and scattered by the cells, or produced by the cell fluorescence. Several optical filters are used to separate the different photons according to their wavelengths and drive them to the photodetectors (most often photodiodes for light scatter and photomultiplier tubes for fluorescences). Data are then displayed and interpreted to discriminate clusters of particles based on statistical analyses of the variables collected.

3 Why Is Flow Cytometry So Popular Among Microbiologists?

Flow cytometry analyses are very fast (up to several thousand cells per second). A very large number of cells can thus be analyzed per sample (up to 10 millions!), and the statistical results are obviously representative of the population. This also allows analyzing more samples, a key issue in spatial or temporal studies. Using these statistical analyses, it is possible to electronically separate these populations and identify them using multivariate analytical techniques. Flow cytometers can nowadays readily analyze single cells at rates up to 30,000 per second. Flow cytometry provides multiparametric analyses at the single cell level (several scatters and fluorescences). The data are quantitative data and can be correlated to other biochemical data. Moreover, data are acquired in real time.

The size class distribution of microorganisms can be assessed from the scattered light, and the analysis also provides the cell abundance. The presence of unique identification markers in the cells allows separating cell clusters. These markers can be natural in the case of auto-fluorescent cells (example: natural fluorescent pigments) or induced by adding fluorescent dyes (fluorochromes) that target particular cell components or activity.

The last, but not least, is that some flow cytometers, called "sorters," are able to physically separate the cells of interest. Upon sorting, cells can be collected in a tube, a Petri dish, a 96-well plate, etc. In most cases, sorting does not affect viability, and post-cultures are possible. Sorting is also very valuable for post-analyses (molecular biology).

Unfortunately, flow cytometers are very expensive instruments, very often out of reach for laboratories. Moreover, they require highly trained people. This is why the "Plateforme Régionale de Cytométrie pour la Microbiologie" PRECYM was created in 2005.

4 The Regional Flow Cytometry Platform for Microbiology (PRECYM)

Aware of the potential of this tool, the three laboratories of the COM (LMGEM UMR 6117, LOB UMR 6535, and DIMAR UMR 6540), as well as two Research Units of the IRD (UR 99 Cyroco and UR 103 CAMELIA), and the Laboratoire de Chimie Bactérienne (LCB UPR CNRS 9043) organized a consortium with the aim of acquiring a flow cytometer cell sorter (a MoFlo from DAKO, Dk). The Regional Flow Cytometry Platform PRECYM was created in 2005 and centralizes this equipment and know-how and makes them available to other units/institutes. PRECYM also allows the optimal technical, economic, and scientific exploitations of the instruments. PRECYM is also involved in the R&D and in the technological survey, as well as the research valorization.

4.1 Example of development achieved in PRECYM: Detection of phosphatase activity of heterotrophic prokaryotes characterized at the single cell level by flow cytometry

The analysis of heterotrophic prokaryotes at the single cell level provides a very valuable insight in ecological studies. Archaea and bacteria, hereafter referred to as "bacteria" in the text, are indeed the major actors in the mineralization of the organic matter in the ocean. They are responsible for most of the aerobic respiration in the ocean. This respiration leads to the production of metabolic CO_2, a greenhouse effect gas of special concern in the context of global warming. Most of the species (or strains) are still unknown, which makes bacteria the major source of unknown biodiversity. Moreover, bacteria can be present in different physiological states ranging from live and active, to live and inactive, and dead. When inorganic phosphorus (Pi) becomes a limiting element in the environment, some bacteria are able to break down exogenous organic phosphate compounds to utilizable inorganic forms (Pi) through alkaline phosphatase activity (APA). This phosphorus could then become available for the rest of the ecosystem. The fluorochrome ELF (Enzyme Labeled Fluorescence) from Molecular Probes (Eugene, Oregon, USA) targets APA at the single cell level. Such a method has been developed on phytoplankton, but studies on bacteria are scarce, and were made by microscopy.

ELF-P is a colorless substrate (i.e., not fluorescent). When ELF-P is hydrolyzed by alkaline phosphatases, it is transformed into a green fluorescent insoluble product (ELF alcohol ELF-A), which precipitates at the site of the enzymes, inducing a bright green fluorescence within the cells when they are excited by a UV light. We have thus developed a protocol to detect phosphatase activity of heterotrophic bacteria characterized at the single cell level by flow cytometry. The method was first developed on bacteria in cultures (Duhamel et al. 2008b). It was then applied to natural fresh and marine samples (Duhamel et al. 2008a, b). By simultaneously incubating the cells with a fluorescent nucleic acid dye (such as DAPI or PI), it is then possible to detect all the bacteria, and the fraction responsible for the phosphatase activity (Fig. 1).

The protocol has been specially designed for freshwater and marine natural samples. It takes into account the need for cell concentration (when the abundance is too low) and the disaggregation step mandatory to get individual bacteria. This protocol is complementary to what is observed by microscopy. Microscopy analyses are qualitative (a cell expresses or not the fluorescence). To be quantitative (how much does a cell express the activity), microscopy must be coupled with advanced image analysis, and it then becomes highly time-consuming. Flow cytometry is instead quantitative and much faster.

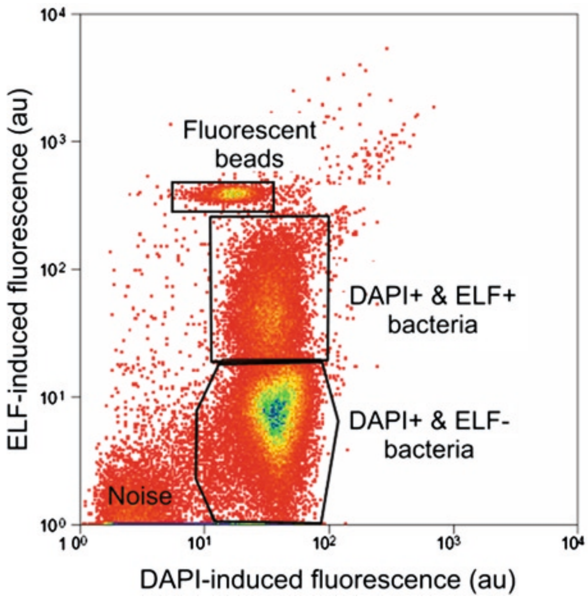

Fig. 1 A double staining by ELF and DAPI makes it possible to discriminate by flow cytometry between bacteria expressing (DAPI positive and ELF positive) or not (DAPI positive and ELF negative) alkaline phosphatase activity. DAPI is a blue fluorescent nucleic acid dye that is used to stain all the bacteria and discriminate them from the debris (noise). ELF is a substrate, which is fixed and transformed by the alkaline phosphatase into a bright green fluorescent crystal. Each dot on the figure represents a single particle (i.e., a bacterium). The color codes for the abundance. Fluorescent beads (2 μm diameter) are added in the sample as internal control to monitor the instrument during the analysis and standardize the intensities between samples

5 Conclusion

The goal of this work is to apply this approach in various natural conditions. Combining the detection and quantification of APA at the single cell level for both phytoplankton and bacteria will bring new insights to the comprehension of algal–bacterial competition for bioavailable organic phosphorus when inorganic phosphorus is a limiting factor. Combining this ELF labeling protocol with molecular techniques such as fluorescent in situ hybridization (FISH) will be very valuable for identifying the phylogenic groups responsible for APA.

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Shadows by IXSEA: An Example of a Sonar Using the Latest Technologies in Acoustics, Positioning, Informatics, and Web Techniques

Frédéric Jean

Abstract This study provides a new system for imaging the sea ground using the newest technologies in acoustics, sonar theory, positioning, informatics, and web techniques. The new side scan sonar called SHADOWS is the first commercially available sonar using the Synthetic Aperture Theory. This principle, introduced in radar since the 1970s, consists of rebuilding a long antenna by adding different contributions along time.

In radar, the EM wave speed compared with the speed of the plane or satellites permits one to use a relatively simple imaging algorithm. It is much more difficult to rebuild the long antenna in the underwater acoustic environment. This is due to a very different order of magnitude and movement interferences of the towed sonar. We thus use a very accurate and synchronized Inertial Navigation Unit also developed by IXSEA. The accuracy of the georeferenced position can be increased using the IXSEA USBL called GAPS. In our design, this method allows having a constant resolution of 15 cm along the 600-m swath. The sides scan sonar images the sea ground from 20 to 300 m on each side of the path. To avoid the blind zone at the nadir of the side scan sonar, forward looking sonar has been added to the system. It uses a newly patented imaging technique on different sectors at higher frequency. The image resolution obtained is comparable with the side scan images. The system developed uses a fiber-optic cable to collect the very important amount of data at 15 Mo/s. The data are logged and processed in real time. It provides a georeferenced mosaic displayed on a web GIS via a map server. In a post-processing mode, we use the web service technology to display and manage the images. We are currently developing new filtering and detection methods to complete the sonar system.

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Synthetic Aperture Sonar:
Sum of successive pings
Constant resolution along the swath: 15 × 15 cm^2
Length (~number of pings) adapted to range (maximum 300 m)
Real-Time Signal Processing

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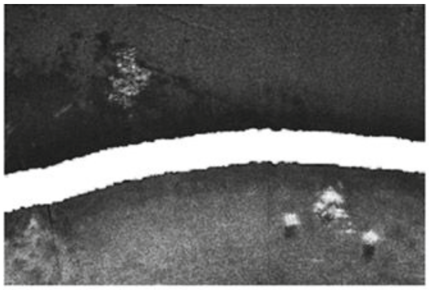
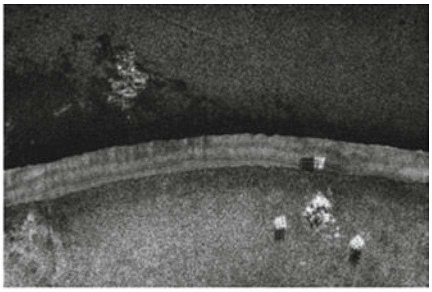
Gap Filler Concept:
Comparable resolution (40 cm)
Filling the gap at the nadir
Sectorized emission patented principle

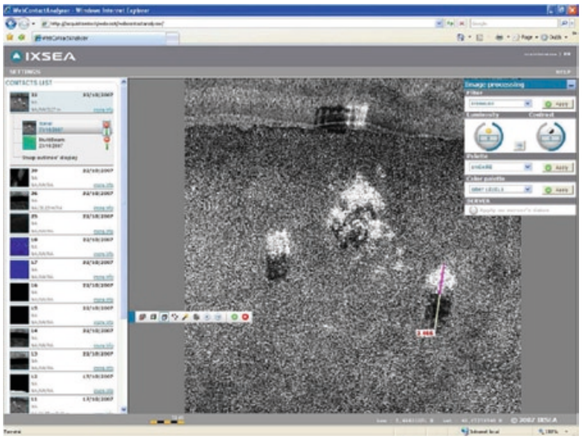


Without Gap Filler:
Missing a big target! (5mx5m)



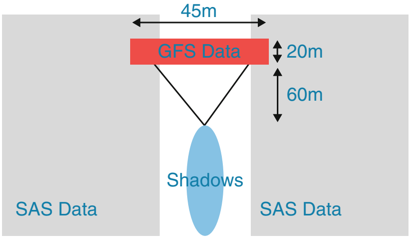
With Gap Filler:
No need for a second Line

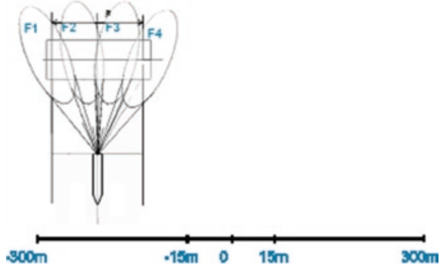
Data Handling and Graphical User Interface:
Data base
Web control command
Web data analyzer
OpenGis architecture



Performances:
Six hundred-meter swaths and 15×15 cm^2 constant resolution at 5 knots
No gap at the nadir

Real-time georeferenced mosaic
Georeference: perfect superposition between two lines, even when the sonar is turning





Relation Between Body Tilt Angle and Tail Beat Acceleration of a Small Fish, *Parapristipoma trilineatum* (Threeline Grunt), During Mobile and Immobile Periods Measured with a Micro Data Logger

Teruhisa Komatsu, Hideaki Tanoue, Natheer Mohammad, Kyoto Watariguchi, Tarik Osswald, David Hill, and Nobuyuki Miyazaki

Abstract Recently, data loggers that can measure acceleration of two- or three-dimensional axes have been downsized to 5 cm length and 1 cm width.

Thus, behavior of small fish can be studied with a micro data logger. We used a small data logger (M190L-D2GT, Little Leonardo Co. Ltd., Tokyo) to measure the behavior of a bentho-pelagic fish, the threeline grunt (*Parapristipoma trilineatum*). The logger adjusted to neutral buoyancy was attached to three individuals (TL=29.8 ± 3.2 cm) in a transparent tank with grids on the bottom for 30 min. Video camera observation distinguished immobile and mobile periods of the fish.

This observation permitted us to classify amplitudes of tail beat acceleration of the fish obtained with the data logger into those of two periods with a boundary value of 0.23 m s^{-2}.

After the observation, the fish were released into a pond (20 × 30 × 4 m^3). Acceleration data were recorded for about 71 h. The results showed that the body tilt angle was closely connected to the movement of the fish, which was related to tail beat. Mean tilt angle of *P. trilineatum* during the mobile period, 4.1° ± 15.9° (n = 116,321), was significantly different from that during the immobile period – 16.5° ± 9.2° (n = 93830) (t-test, $p < 0.01$).

When the fish decreased amplitudes of tail beat acceleration under 0.23 m s^{-2}, the body tilt angles were nearly parallel to the swim bladder axis of the fish, about 19° along their body axis. This position of the fish is energy saving.

In this way, the micro data logger of acceleration will give us a new perspective of fish behavior studies.

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1 Introduction

Numerous behavior studies on marine mammals, large fish, or sea birds have been conducted using a data logger (e.g., Yoda et al. 2001). These studies revealed their incredible adaptations to marine environments. However, the behavior of small fish has not been investigated owing to sizes of the data logger that were attached to animals. Recently, data loggers that can measure acceleration of two- or three-dimensional axes are downsized to 5 cm length and 1 cm width. Thus, we started studies on the behavior of small fish, which has not been measured with a bigger logger, with these new instruments.

Body attack angle and tail beat frequency of fish are very important factors that relate to swimming of fish (Svendsen et al. 2005). Body tilt angle is an important parameter influencing acoustic backscatters of fish, which are used for estimation of fish biomass with an echosounder survey (e.g., Love 1971). Therefore, it is very important to understand the relation between the body tilt angle and the behavior of fish. Then, we applied a micro data logger to measure the body tilt angle and tail beat acceleration of a bentho-pelagic small fish, threeline grunt (*Parapristipoma trilineatum*).

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2 Material and Methods

Three individuals of threeline grunt were used for the experiments conducted at Fisheries Technology Center of Kanagawa Prefecture, Japan, from 21 to 24 July 2007. Their mean total length was 29.8 ± 3.2 cm. They were acclimated in a round tank, measuring 1.4 m diameter and 0.7 m water depth, filled with seawater for 3 days before the experiment.

A micro-acceleration data logger (M190L-D2GT, Little Leonardo Co. Ltd., Tokyo) was used to measure the behavior of a fish. Its diameter and length were 15 mm and 55 mm, respectively (Fig. 1). It can record depth and temperature at 1 Hz, and accelerations along two orthogonal axes measuring swaying and heaving behaviors at 16 Hz for 3 days. Since the weight of the logger is 18 g in air and 6 g in water, its attachment to fish gives negative buoyancy to the fish. Therefore, we wrapped a float made of copolymer foam (Nichiyu Giken Kogyo Co., Japan) around the data logger to keep the logger (including the attachment system) neutral in seawater. After release in a large artificial pond supplied with seawater through a pipe connected to the sea, it is very difficult to capture the fish attached with the data logger. We therefore used an automatic time-scheduled release system (Watanabe et al. 2004) that allows the loggers to be detached from the fish and float on the sea surface in the pond. Thus, we searched for and retrieved the loggers after the release of the loggers floating on the surface of the pond.

Fig. 1 Picture of the micro-acceleration data logger. Arrows along the cross-sectional and the longitudinal directions represent swaying and surging directions, respectively. Gray arrow and white line show the direction of the fish head, 2 cm long

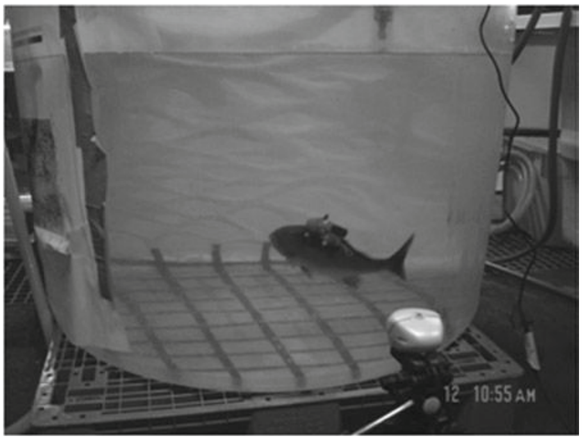

Fig. 2 Picture of the experiment to distinguish mobile and immobile periods of threeline grunt in a transparent plastic tank. Video cameras set on the upper side and along the lateral side of the tank recorded behavior of fish attached to the micro data logger. The grids were drawn with vinyl tape at 10-cm intervals

After attaching the logger to an individual fish with plastic cables, it was released into a transparent cylindrical tank (1.4 m of diameter and 0.7 m of water depth) with grids of black vinyl tape at 10-cm intervals on the bottom (Fig. 2). We observed displacement of the individual with video cameras set on the upper and lateral sides of the tank for 30 min (Fig. 2). When the fish stayed inside a grid, it was regarded as immobile. We classified movements of fish into immobile and mobile periods by this criterion. Amplitudes of tail beat acceleration measured with the logger were related to the mobile and immobile period. After the observation, the individuals were released into a pond (20 × 30 × 4 m^3) in the Fisheries Technology Center of Kanagawa Prefecture (Fig. 3) for 3 days. Accelerations of the individuals were recorded for 71 h there.

Power spectral density (PSD) was calculated from swaying acceleration records of the logger to determine the dominant stroke cycle frequency, namely that of tail beats, using Fast Fourier Transformation supplied by the software (Igor Pro, Wave Metrics) according to Ropert-Coudert et al. (2006). Tail beat frequency was obtained from the predominant frequency of swaying acceleration data (Fig. 4). Then, surging acceleration lower than the predominant frequency of tail beats was regarded as a component related to body tilt angle as A in the following equation (1). Body tilt angle, θ, was defined as:

$$\theta = a\sin\left(A/g\right) - \alpha, \quad (46.1)$$

where g and α were gravity and attachment angle of the logger to body axis of fish. Body tilt angle is positive and negative when the body axis is above and beneath the horizontal position of the fish, respectively.

Fig. 3 Picture of the pond (20 × 30 × 5 m^3) where three three-line grunts were released at the Fisheries Technology Center of Kanagawa Prefecture, Japan

3 Results and Discussion

Immobile and mobile periods of the individuals in the transparent tank were distinguished by camera observation. Amplitudes of tail beat accelerations during the immobile periods were pooled. Mean and standard deviation were 0.107 and 0.0636 m s^{-2}, respectively (n = 207). When normal distribution is assumed, 95% of the distribution of amplitude of tail beat acceleration during the immobile period was 0.23 m s^{-2} in the positive side of the distribution. Then, we defined 0.23 m s^{-2} as a boundary acceleration value of immobile and mobile periods. Body tilt angles of the fish in the tank were calculated based on time series data of accelerations. Using amplitudes of tail beat acceleration, body tilt angles were grouped at 0.02 m s^{-2} intervals of amplitude. Body tilt angles of the fish during the mobile period were greater than those during the immobile period (Fig. 5). Figure 5 shows that the body tilt angle was roughly proportional to the amplitude of swaying acceleration under 0.23 m s^{-2}, while the body tilt angle was nearly constant above 0.23 m s^{-2}.

Mean body tilt angle of three *P. trilineatum* individuals during the mobile period, −4.1° ± 15.9° (n = 116,323), was significantly different from that during the immobile period – 16.5° ± 9.2° (t-test, $p < 0.01$).

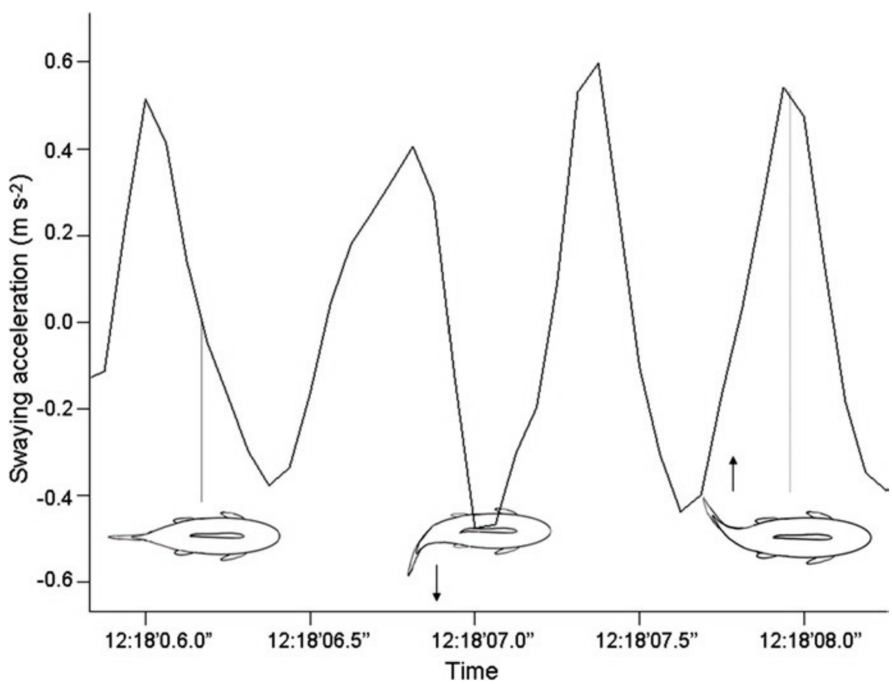

Fig. 4 Example of swaying acceleration data of an individual measured with the micro data logger. Top views of fish were drawn on the acceleration data corresponding to the positions of tail beat

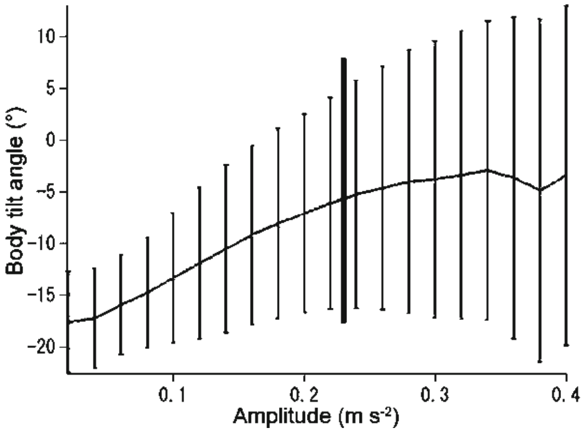

Fig. 5 Relation between amplitude of tail beat acceleration and body tilt angle of threeline grunt observed in a transparent tank. Lines passing the center of narrow vertical lines, narrow vertical lines and fat vertical lines are line connecting the mean value of each dataset grouped at amplitudes of 0.02 m s^{-2} intervals, its standard deviation, and a value of 0.23 m s^{-2} dividing immobile and mobile behavior of the fish, respectively

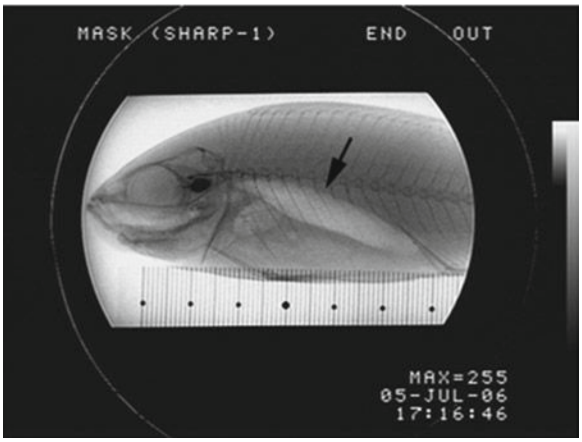

Fig. 6 X-ray picture of threeline grunt. Arrow shows the swim bladder. Angle between longitudinal axis of swim bladder and body axis of the fish is about 19°

When the individuals stopped beating their tails, the mean body angle was – 19.5° ± 7.5° (n = 4,189).

Tanoue (2009) examined the form of swim bladders of anesthetized threeline grunt using an x-ray system (Fig. 6). He reported that the mean angle between the body axis and longitudinal axis of the swim bladder of threeline grunt was 19.0° ± 1.4° (n = 31). Therefore, the body axis approaches the angle that the longitudinal axis of the swim bladder is at the horizontal position when the threeline grunt does not beat its tail actively. This body position of fish is energy-saving because there is less necessity to maintain their body at this position. Thus, it is suggested that we can estimate the resting period of the fish from body tilt angles. In this way, the micro data logger can present new perspectives of small fish behavior. These results are used for estimation of abundance and also understanding the ecology of small fish.

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Marine Observation Using a Hybrid Glider

Yann G. Le Page

Abstract Observations of marine physical (chemical) or biological phenomena have been evolving considerably within the last 20 years. Advances in underwater robotics have made it possible to take measurements in situ automatically. These techniques include Yoyo profilers (automatic vertical profile generation using ballast), autonomous underwater vehicles (AUVs, short-terms missions using an autonomous submarine), and more recently underwater gliders (long-range buoyancy driven vehicles).

The above-mentioned solutions have advantages and drawbacks that can be considered to create a more optimized vehicle. AUVs allow precise navigation in shallow coastal areas, bottom following, and horizontal transits, but is limited to a few days of endurance. Contrarily, the glider's endurance extends to months, but its maneuverability, efficiency in shallow waters, and payload volume are limited, and its wings are fragile.

In addition, most existing vehicles rely on hazardous surface position fixes and/or very expensive inertial navigation systems with poor accuracy after hours of a mission. In order to avoid collisions, an acoustic system allowing underwater position fixes using a single buoy may be used.

This paper describes how the advantages of the two types of platforms can be merged into an "hybrid glider," capable of gliding but also of being propelled at low speed and high efficiency.

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1 Hybrid Propulsion

A glider uses only variations of its buoyancy as a source of propulsion. The idea is to convert this vertical force (alternatively positive and negative) into a horizontal one, making the vehicle move forward. This conversion is done by orientating the vehicle with an optimal pitch angle: the vertical force projected in the vehicle coordinates, then has a longitudinal component that will produce speed, and a transversal component producing a non-desired transversal speed (Fig. 1). Use of adequate hydrodynamic surfaces and/or vehicle body shape reduces this non-desired transversal speed to a minimum, making the vehicle glide. The vehicle hence follows a tooth saw trajectory in the vertical plane.

These simple principles offer the following advantages compared to conventional propeller techniques:

Slightly higher efficiency (volume variation efficiency: 50% compared to a propeller: 40%) at deep depths (i.e., >300 m)

No motor used during the dive except at the tooth saw extremities, resulting in completely silent and vibration-free displacements

But contrary to what is believed, most of the gain of energy compared to regular AUVs is not from the type of propulsion, but from the limited speed. Indeed, as explained previously, the efficiency difference between a propeller and ballast is "only" a few percents.

The energy required to move the vehicle at a given speed V and for a distance D is a function of its speed to the square:

$$E = FVT = kV^3 \frac{D}{V} = kDV^2$$

Typical AUVs run at a mean speed of 4 knots, whereas a glider is designed to run at a maximum

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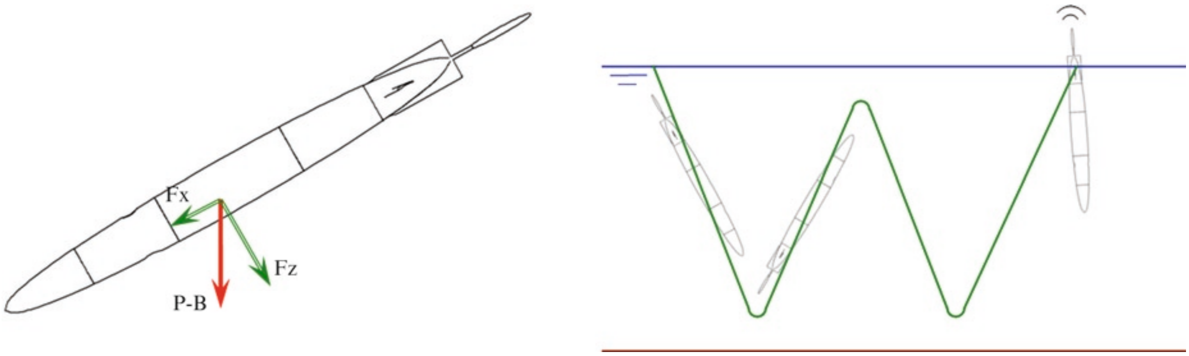

Fig. 1 Principle of operation

1 knot speed. The gain in energy to fly the same distance is then:

$$E_{1\text{Knot}} = E_{4\text{Knot}}\left(\frac{1}{4}\right)^2 = 0.0625 \times E_{4\text{Knot}}$$

The energy required is thus 6% of the one needed to cover the same distance at 4 knots. In other words, for the same amount of energy (batteries), an AUV running at glider speed would travel a distance 16 times higher in the range of glider performance.

Hence, in contrary to what is often believed, the high endurance of a glider is not due to the use of ballast, but to its low speed and low-power electronics.

Designing a propelled vehicle optimized for such low speeds benefits from two technologies:

High endurance at deep and shallow depths

Optimized water column scanning (i.e., vertical tooth saw patterns from the surface to deep waters) in glider mode

Optimized bottom/surface following and horizontal flights (e.g., horizontal lawn-mower patterns) in propelled mode.

2 Underwater Positioning

Underwater vehicles used for marine observations need to know their position with good accuracy, since marine observations done in an autonomous way by the vehicle have to be time-stamped and geo-located. Indeed, even if very accurate sensors are used, but the exact location of the measure is not known with precision, the lack of good knowledge of the location inherently reduces the measure accuracy.

Underwater gliders rely on basic dead-reckoning using low-cost and low-power instruments, and GPS fixes are used to reset the resulting error. This technique has two major drawbacks: Poor positioning estimation and high risk of collision in coastal areas with surface vessels.

ACSA underwater GPS has developed an innovative technique allowing underwater position fixes by use of a single acoustic beacon. The Synthetic Long Base Line Techniques developed for such vehicles allow precise relocalization by multiple distance measurements from the vehicle and a GPS-Intelligent Buoy (GIB™), knowing the approximate vehicle heading and speed. Such techniques allow long-term missions with no surfacing, making coastal and under-ice operation now possible with good positioning accuracy.

3 Conclusion

Marine data acquisition remains costly, mainly due to time spent at sea by people to actually sample water or deploy autonomous underwater vehicles (AUVs).

Underwater gliders have recently shown that the cost of data could be dramatically reduced by using long-range, low-cost vehicles, providing extensive datasets with no human intervention for months, even if such a platform is not adapted to coastal areas.

Hybrid platforms will now provide the same cost of data, but will allow water sampling in a much larger panel of areas of operation.

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A New Method to Measure Prokaryote Respiration at the Single Cell Level by Flow Cytometry

Lounis Mebarek, Michel Denis, and Gérald Grégori

Abstract In the ocean, microbial respiration is considered the major process representative of organic matter's biological oxidation. The corresponding metabolic CO_2 production was estimated to reach about 22 Pg C year^{-1} (1 Pg = 10^{15} g).

The balance between primary production and respiration determines if the biological system (i.e., the biological pump) acts as a source or a sink of carbon, a very important topic in the framework of the global warming. However, there is still some uncertainty about the importance of the respiration because the in situ respiration rate is generally too low (by several orders of magnitude) to be accessible to the available direct measurement methods. Some indirect methods were therefore applied, such as the measure of O_2 consumption by the Winkler chemical method or the CO_2 metabolic production by coulometry. There is also an enzymatic method (Electron Transport System test from Packard) based on the enzymatic activity of dehydrogenases, but it addresses only a potential activity.

All these methods are global and thus address the community level. Therefore, they do not take into consideration the heterogeneity of microbial assemblages.

Some fluorescent dyes, such as $DiOC_6(3)$ (Molecular Probes, USA), have been shown to be very sensitive to changes in the proton electrochemical potential difference ($\Delta\mu_{H+}$), characterizing mitochondrial and plasmic membranes bearing the cell respiratory system in eukaryotic and prokaryotic cells, respectively. In mitochondria, $\Delta\mu_{H+}$ is linked to the flux of oxygen uptake by a linear relationship. To our knowledge, no such relationship has been established in the case of whole marine cells. Our team addressed the dark respiration rate of the Chlorophyceae *Dunaliella tertiolecta* (Butcher) in axenic cultures, both directly by using a highly sensitive oxygraph (Oroboros) and by staining cells with a fluorescent dye called $DiOC_6(3)$.

The accumulation of the dye is sensitive to the membrane potential of the mitochondria. We found and standardized a linear relationship between oxygen uptake by *D. tertiolecta* and its green fluorescence induced by $DiOC_6$, enabling the determination by flow cytometry of the respiration rate of *D. tertiolecta*.

After this promising result, we are trying to extend the method to other microorganisms representative of the microbial assemblages. This study aims to continue this work and to apply it to aerobic heterotrophic prokaryotes, which are the main mineralizers of the organic matter in the ocean.

1 Introduction

In the ocean, microbial respiration is considered the major process representative of the biological oxidation of organic matter. The balance between primary production and respiration at the global scale determines if the system (i.e. the biologic pump) can be considered as a source or a sink of carbon. However, uncertainties remain about the importance of respiration because the in situ respiration rates are generally too low to be directly accessible by available methods. This is particularly true in the meso- and bathy-pelagic domains where cells are less abundant.

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Thus, indirect methods have therefore been developed, such as the measurement of O_2 consumption (by the Winkler method) or CO_2 production (by coulometry), or the ETS (Electron Transport System) enzymatic test developed by T. Packard (1971). All these methods are global and thus address the community level. They do not take into account the heterogeneity of microbial assemblages in terms of biodiversity or physiological states (living active or inactive cells, dead cells). These considerations naturally lead to the need for analyses at the single cell level.

The accumulation of some fluorescent dyes such as $DiOC_6(3)$ (Molecular Probes, USA) has been shown to be sensitive to the proton electrochemical potential difference ($\Delta\mu_{H+}$). $\Delta\mu_{H+}$ characterizes mitochondrial and plasmic membranes that bear the respiratory chain in eukaryotic and prokaryotic cells, respectively (Mitchell 1961). In a former work, the dark respiration rate of the Chlorophyceae *Dunaliella tertiolecta* (Butcher) was investigated, both directly by using a highly sensitive oxygraph (Oroboros) and by incubating cells with $DiOC_6(3)$ before flow cytometry (FCM) analysis. A linear relationship was evidenced between the cell fluorescence intensity induced by $DiOC_6(3)$ and the O_2 uptake, enabling the determination by flow cytometry of the respiration rate at the single cell level (Grégori et al. 2002). The goal of this study is to extend this method to heterotrophic prokaryotes, which are the major organisms responsible for organic matter (OM) mineralization in the ocean.

2 Assay on the Marine Bacteria *Pseudomonas Nautica* sp. 617

The method for measuring respiration by flow cytometry was developed on a mono-specific culture of *Pseudomonas nautica* sp. 617.

The culture is grown at 20°C, under agitation, in an acetate medium. This medium is adequate for FCM analysis as it does not emit any fluorescent background signal that could overlap the signal from the bacteria. The development of the method implies working on homogeneous cells (i.e., same physiological state), and therefore cells were processed when in the exponential growth phase (EGP) as determined from the growth curve (OD = 0.8 au).

3 *P. Nautica* Staining with $DiOC_6(3)$

The incubation of *P. nautica* with $DiOC_6(3)$ does not show any effect on the respiration rate. The labeling kinetics by $DiOC_6(3)$ assessed by flow cytometry suggests working with low concentrations (<100 nM) and an incubation time of 30–60 min prior flow cytometric analysis.

4 Stimulation of *P. Nautica* Respiration

In order to establish the relationship between the cell fluorescence and the oxygen uptake rate, it is necessary to get a range of respiration rates. Acting as an uncoupler, carbonyl-cyanide-m-chlorophenyl-hydrazone (CCCP) tends to abolish the membrane potential of the cells and consequently decreases ATP synthesis. To compensate the lack of ATP, cells oxidize more organic matter and thus increase their respiration rate.

Unlike what was expected, an inhibition of respiration by CCCP was observed on cells in EGP. The stimulation was however efficient on cells at the beginning of the stationary phase. Consequently, cells in EGP were centrifuged and re-suspended in a minimum medium (no C or N source) for 5 h. By providing bacteria with several substrates at various concentrations and using CCCP, it was possible to stimulate the respiration (Fig. 1).

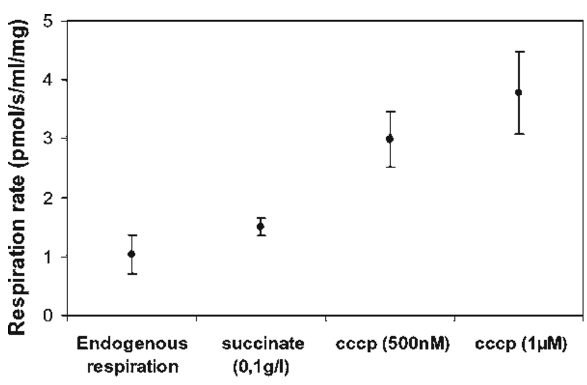

Fig. 1 Example of respiration stimulation with succinate and CCCP. Compared to the endogenous respiration intensity, the addition of succinate or CCCP induces an increase of the oxygen uptake rate (i.e., of the respiration rate)

With acetate, the respiration increased up to five times the basal respiration. With succinate and alanine, the acceleration was up to about 2.5 times. Experiments have also shown a good correlation between the mean fluorescence intensity per cell induced by $DiOC_6(3)$ and the substrate concentrations.

5 Conclusion

These preliminary works demonstrate the feasibility of the method for measuring respiration by flow cytometry. The culture conditions, $DiOC_6(3)$ concentration, and incubation time have been determined. The stimulation of the respiration seems possible by using several substrates and the uncoupler CCCP.

The last step consists now in coupling the measure of the respiration intensity (Oxygraph) with the mean cell fluorescence intensity measured by flow cytometry; then the method will be applied to other strains.

However, if there is no theoretical argument to preclude the extension of the present approach, experimental obstacles may obstruct the way ahead. For instance, the stain must reach the bioenergetic membrane, and some species containing glycoproteins are known to actively pump out some molecular probes. If staining of mammalian cells is well documented, the situation is different for bacteria. Gram negative bacteria, for instance, have an additional cell wall that acts as a strong barrier towards some lipophilic dyes. This obstacle can be solved by pretreatments with Tris and EDTA in order to permeabilize the external membrane.

Whatever the difficulties ahead, this study opens the way to a new, elegant, and efficient way to quantify cell respiration. This is a major challenge in oceanography, particularly in the aphotic zone were the abundance and activity level of microorganisms are too low to allow direct measurements of the respiration.

At least, the method described in this article is fast (less than 1 h). This makes this method also valuable for monitoring the environment at high frequency (spatial or temporal monitoring). It could become an efficient tool to investigate the impact of the anthropic activities on the dynamics of the marine microorganisms.

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Rapid Enzymatic Method for the Enumeration of Fecal Enterococci in Seawater

Marion Peirache, N. Patel, Y. Martin, and J.-L. Bonnefont, S. Mounier

Abstract The new European Directive of 15 February 2006 sought active management of the water quality in swimming areas by 2015. This new guideline requires more restrictions concerning fecal bacteria and particularly *enterococci*.

Our aim is to improve a technique for enumeration of fecal pollution indicator bacteria by recording a specific enzymatic activity (ß-D-glucosidase) using fluorimetry. This method is fast, and the real-time monitoring (1 h) is crucial in case of emergency.

The validation of our work will come only after the identification and suppression of interferences related to fluorimetry measurements.

The second aspect of our research deals with the behavior of fecal indicator bacteria according to chemical and physical parameters of the environment through the study of their viability in marine waters.

This important step aims to create estimation models.

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1 Introduction

The new European Directive 2006/7/EEC of the 15 February 2006 sought active management of water quality in swimming areas by 2015 and thus overruled the criteria of 1976. This new guideline requires more restrictive criteria concerning two fecal indicator bacteria, *Escherichia coli* and particularly fecal enterococci. Our aim is to implement an efficient, sure, and fast tool (real-time monitoring within 1 h) in order to quantify these bacteria to avoid sanitary risks and the related economic impacts. A commercial method is already available, named "Coliplage" (Veolia) or "MER" (Lyonnaise des Eaux), targeting *Escherichia coli*. Moreover, Caruso et al. (2002) used this method on the Calabrian and Sicilian coasts to assess the microbiological quality of seawater. We would like to adapt this technique (Lebaron et al. 2005) to enterococci.

2 Materials and Methods

The principle is based on an indirect enumeration through the measure of a specific enzymatic activity using fluorimetry. Methylumbelliferone (MUF*), a quantifiable fluorescent component (λex = 362 nm and λem = 445 nm), is incorporated into the specific substrate. The speed of hydrolysis expressed in picomol/min of MUF released during 30 minutes, is correlated to the enumeration of bacteria assumed by a standard method (membrane filtration, results in 24–48 h). This correlation curve will be used as a reference model for determining the number of bacteria in a sample, thanks to an enzymatic activity value (obtained in less than 1 h). ß-D-glucosidase is the enzyme used to enumerate the enterococci.

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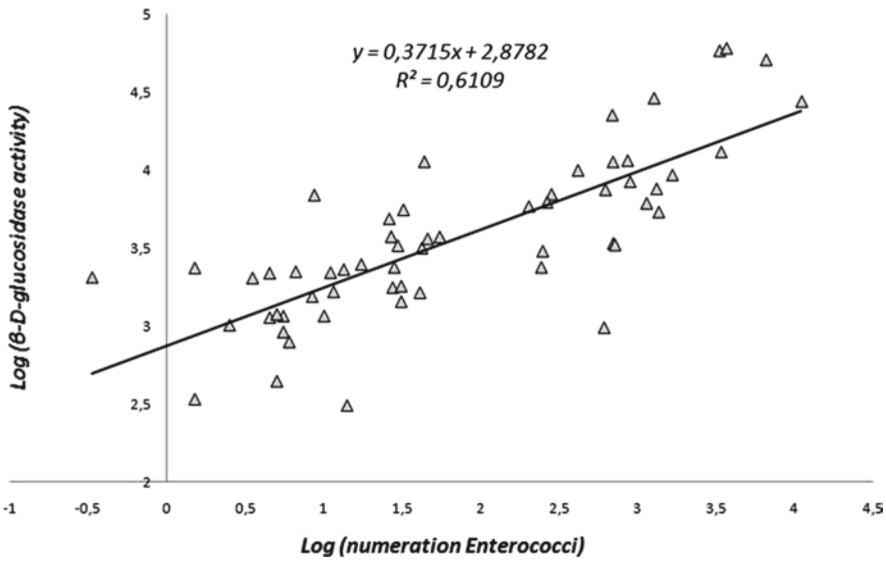

Fig. 1 Optimized Log-log correlation curve between ß-D-glucosidase activity, and enteroccci enumeration by membrane filtration ($n = 58$)

3 Results

In order to obtain a good correlation curve (Fig. 1), we have to eliminate some pairs of values (when activity or the number of bacteria is null).

We present two cases:

The first reveals enzymatic activity without bacterial enumeration. We can deduce two explanations: the viable but nonculturable bacteria (VBNC), a known phenomenon, according to which stressed bacteria cannot grow on media but keep some metabolic activities (Davies et al. 1995; Pommepuy et al. 1996). Another possibility is the enzymatic activity from other microorganisms [Davies et al. (1994) report on algal interferences].

The second reveals the presence of bacteria, although no activity is measured. The "past" of bacteria submitted to environmental and sewage treatment conditions could be responsible for a loss of hydrolysis capacity.

We tested differential filtrations through 12-µm porosity membranes; this operation permits eliminating some phytoplankton without affecting bacteria. We measured a decrease in activity, which confirms the capacity of lysis of the susbtrate by phytoplankton. Then, we tested a brown algae strain (*Skeletonema spp. = a common diatom in Mediterranean sea*) and observed positive activity.

To prove the participation of the bacteria VBNC, we carried out T90 (time necessary to eliminate 90% of target organisms) assays with a solar radiation stress. At $T + 24$ h, the activity increases drastically (Fig. 2), while no bacteria are detected. In fact, this phenomenon is due to a multiplication of *Vibrio spp*. (*V. alginolyticus* – C.I. 95%). Indeed, natural isolated strains show the ability to lyse the substrate intended to enterococci. To reach our primary goal, we have to reproduce this experiment in another season less favorable to *Vibrio spp*.

4 Conclusion and Perspectives

After these first results, we would determine with more precision the contribution of each interference, microorganisms, and/or elements of the sample. Using a 3D fluorimeter (PROTEE laboratory), we could have a better "visualization" of the interferences. Morover, the interaction between organic matter and bacteria not only has an impact on the enzymatic activity, but also on the survival rate of the bacteria. This is why we look

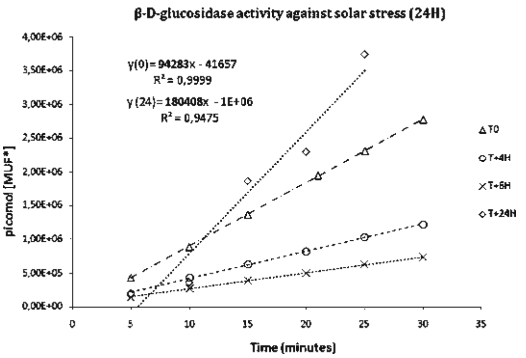

Fig. 2 T90 measurements of a sample through its ß-D-glucosidase activity. The slopes of the curves represent the rapidity of the substrate hydrolysis. We can observe a decrease in time except for *T* + 24 h, after a night in darkness, where we obtain a more intense enzyme activity than the first one measured

for the physical and chemical parameters of the environment, which could allow us to realize estimation models of survival rate.

This work is totally integrated in a larger project for better management of upstream water. In such a project, rapid monitoring methods are necessary to enhance our reaction capacity and can participate once again in the improvement of water quality for sustainable development.

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Oxygen Distribution Heterogeneity Related to Bioturbation Quantified by Planar Optode Imaging

Laura Pischedda, Jean-Christophe Poggiale, Philippe Cuny, and Franck Gilbert

Abstract Oxygen plays a key role in benthic microbial ecology. Until recently, oxygen concentration in sediments was measured with oxygen microsensors along a vertical profile (one dimension) from the surface until a few centimeters into the sediment. With this approach, however, it is a tedious job to describe or overcome the heterogeneity of oxygen distribution in environments such as bioturbated environments. Recently, a new technique has been introduced that allows the investigation of two-dimensional oxygen distribution and dynamics at a high resolution in the upper sediment column. This non-destructive technique takes advantage of an oxygen-quenchable fluorophore, which is cast into a thin sheet, the planar optode. The latter may be introduced in sediments and is used in situ or in laboratory experimentation, coupled with an optical system allowing the oxygen quantification. Oxygen optodes were used in experimentation dedicated to the study of oxygen heterogeneity induced by macrofaunal bioturbation. Oxygen images of sediments inhabited by a biodiffusor, the gastropod *Cyclope neritea*, and two gallery diffusors, the annelids *Nereis virens* and *Nereis diversicolor*, were used to compare the impact of these organisms on oxygen distribution in sediments. Diffusive oxygen flux and a heterogeneity index were quantified based on oxygen images. Results showed that all species increased oxygen distribution heterogeneity, and that this heterogeneity increased with increasing total diffusive fluxes.

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1 Introduction

Physical and chemical habitat heterogeneity is critical to the functioning of ecosystems. Ecologists have only recently begun to explore how changes in physical and chemical habitat heterogeneity may influence ecosystem functioning by affecting important processes, such as primary production, decomposition, or nutrient cycling (Aller 1994). Moreover, there is increasing evidence that changes in ecosystem functioning can unequivocally be attributed to habitat variability, that is, spatial variation of physical and/or chemical properties (Cardinale et al. 2002).

In some environments, habitat is strongly influenced by biotic activities. This is the case for aquatic sediments where the bioturbation process (i.e., particle reworking and bioirrigation), which results from the various faunal activities (e.g., feeding, gallery construction, ventilation; e.g., Rhoads 1974; Pearson 2001), creates a complex mosaic of micro- and macroenvironments (Aller 2001). The role of organisms in sedimentary particle redistribution depends on the organisms considered, which can be classified in five different functional groups (for review, see Gérino et al. 2003). Benthic organisms can construct more or less complex structures in the

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sediment column, presenting various shapes and sizes and being permanent or not (Dufour et al. 2005). Moreover, in order to renew oxygen and evacuate metabolites, these structures may be actively ventilated by their inhabitants in constant or intermittent ways (e.g., Foster-Smith 1978; Aller and Aller 1998). Because of the bioturbation process, organic material and electron acceptors available for bacteria are redistributed in the sediment column (Aller 1982), strongly influencing the pathways, rates, and extent of organic matter mineralization during early diagenesis (e.g., Kristensen 1985; Gilbert et al. 2003).

As highlighted by Cadenasso et al. (2006), ecologists often describe spatial heterogeneity as patches, which correspond to discrete homogeneous areas that differ in structure, composition, or function. The complexity of this spatial heterogeneity may increase from simple cases where patch type and number of each type are described, to more complex cases where spatial configuration and temporal changes in the mosaic are monitored (Li and Reynolds 1995). The quantification of environmental heterogeneity has long been an objective in ecology (e.g., Patil et al. 1971; Freestone and Inouye 2006). Attempts have thus been made to develop methods to quantify the spatial heterogeneity, in particular for landscape studies (e.g., O'Neill et al. 1988; Garrigues et al. 2006). Landscape ecology principles, however, have not been widely applied to marine systems, in part because of technical limitations. For instance, in the field of aquatic sediment biogeochemistry, it was difficult, until recently, to quantify oxygen spatial heterogeneity exhibited in the upper sediment column. The use of techniques, such as microsensors producing one-dimensional vertical oxygen profiles, was the only way to quantify oxygen distribution patterns within sediments (Revsbech et al. 1980). Only a few destructive measurements can be performed, which makes this technique inappropriate in systems such as bioturbated sediments and ventilated worm burrows. Recently, a new tool has been developed, the planar optode, which makes possible high-resolution measurement of two-dimensional distribution and spatial heterogeneity of solutes in the sedimentary column (Glud et al. 1996; Hulth et al. 2002; Zhu et al. 2006). In the frame of our study, this is oxygen, the most important electron acceptor for organic matter decomposition (Revsbech et al. 1981), which was quantified using planar optodes.

2 Oxygen Planar Optode

The two-dimensional oxygen concentration in bioturbated sediments and overlying water was quantified with semi-transparent oxygen planar optodes. Oxygen concentration measurement was based on the dynamic quenching of oxygen on an immobilized fluorophore. The optical sensor was composed of two thin layers, a transparent polyester support foil (~150 µm thick), and a sensing layer where the oxygen-quenchable fluorophore of platinum (II) mesotetra (pentafluorophenyl) porphyrin (Pt-PFPP) was embedded in a polystyrene matrix (~20 µm). The sensing layer mixture was composed of 3 mg of Pt-PFPP dissolved in 3 mL of toluene and 0.65 g (5%) of polystyrene pellets dissolved in 15 mL of toluene. The two solutions were mixed and spread on the polyester support foil (300 cm²). The solvent was left to evaporate slowly until the membrane became completely dry. The planar optode may also be used in situ with Sediment Profile Imagery (SPI, Glud et al. 2001), but in the frame of this work, only a laboratory design is presented. During the laboratory experiment, the high-resolution measurement of oxygen concentration is carried out by using an optical system combined with the oxygen optode (Fig. 1). For full details of the experimental setup, see Pischedda et al. (2008). In brief, the optode, inserted between the sediment column and the transparent tank face, was excited by a Xenon lamp light passing through a shutter and an excitation glass filter (405 ± 10 nm) mounted on a first filter wheel. Light emitted by the optode sensing membrane was collected through a macro lens and an emission glass filter (654 ± 24 nm) mounted on a second filter wheel. The fluorescence signal was then detected by a CCD camera. Images were captured in darkness with an integration time of 30 s for oxygen and 1 s for sediment structure (without any filter). The light shutter, excitation and emission filter wheels, and camera were computer-controlled using the Image Pro Plus-Scope Pro package installed on a Pentium 4 computer. The digital TIFF images were then stored in a 12-bit grey scale (0 to 4,095).

The acquisition and storage of oxygen images were automated with a custom-made script. Oxygen optodes were calibrated by a three-point calibration method. For the two intermediate calibration points (90%, air bubbling and 50%, nitrogen bubbling), oxygen concentration was first measured just behind the optode with an oxygen probe, immediately followed by capture of the

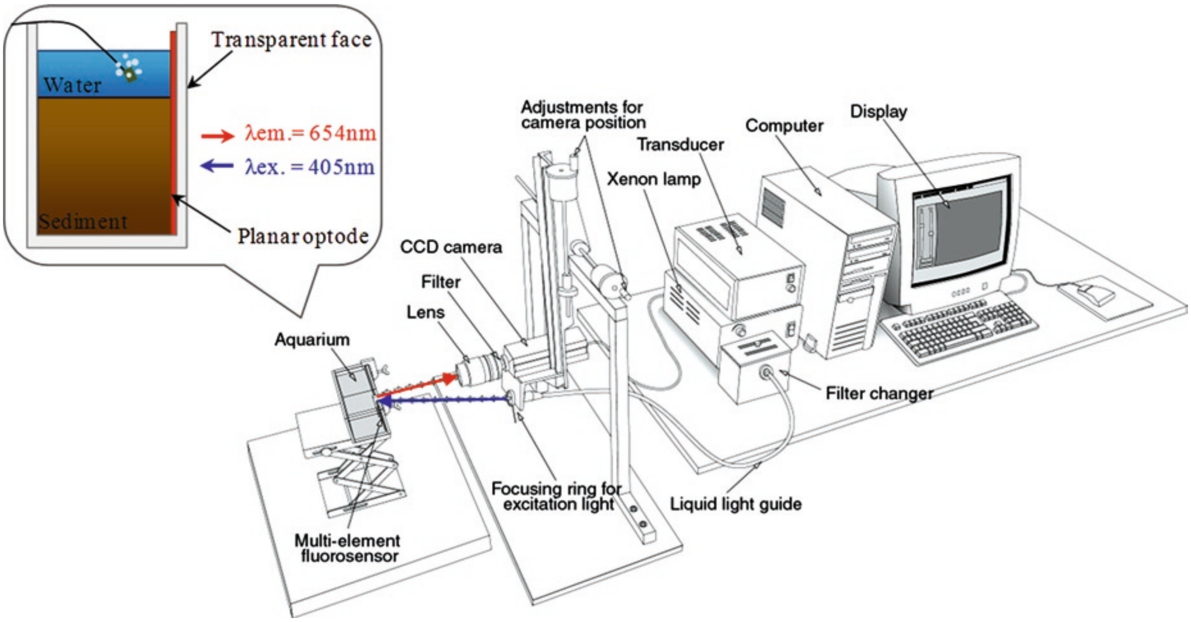

Fig. 1 Example of an optical system for two-dimensional quantification of the element (oxygen, pH, etc.) distribution patterns in aquatic sediments (adapted from Hulth et al. 2002)

oxygen image. No further calibration was performed in the water environment after the experiment began because it was difficult to remove sediments without damaging the optodes. However, for long temporal series, data can be corrected for drift, which was quantified based on temporal evolution of oxygen from the anoxic sediment. A non-linear relationship slightly modified from the Stern-Volmer equation (Klimant et al. 1995) allowed pixel intensity (arbitrary unit) to be converted into oxygen concentration:

$$I = I_0\left[\alpha + (1-\alpha)\cdot\left(1/\left(1+K_{sv}\cdot C\right)\right)\right]$$

where I_0 is the fluorescence intensity in the absence of oxygen, C is the oxygen concentration (μmol/L), K_{sv} is the quenching constant expressing the quenching efficiency (M^{-1}), and α is the non-quenchable fraction of the luminescence including scattered stray light. The constants α and K_{sv} were determined from I_0 and the two intermediate calibration points, with oxygen concentrations C_1 and C_2 corresponding to intensities I_1 and I_2 respectively, and integrated into the following equations:

$$K_{sv} = \left[I_0.\left(C_2 - C_1\right) - \left(I_1.C_2 - I_2.C_1\right)\right] / \left[\left(I_1 - I_2\right).C_1.C_2\right]$$

$$\alpha = \left[I_1\left(1 + K_{sv}.C_1\right) - I_0\right] / \left(I_0.K_{sv}.C_1\right)$$

The constants α and K_{sv}, were averaged for each zone studied taking the closest anoxic zone. Having estimated α, K_{sv}, and I_0, oxygen concentration was obtained by rearranging the first equation:

$$C = \left(I_0 - I\right) / \left(K_{sv}.(I - I_0.\alpha\right)$$

2.1 Example of Application of the Oxygen Planar Optode

This application example came from the recently published article of Pischedda et al. (2008). The aim of this study was to investigate the influence of sediment heterogeneity, induced by the bioturbation process on diffusive oxygen flux. For this purpose, oxygen planar optodes were used during laboratory experiments carried out using three macrobenthic species presenting different bioturbation behavior patterns: the gallery-builder polychaetes *Nereis diversicolor* and *Nereis virens*, which actively ventilate their burrows, and the burrowing gastropod *Cyclope neritea*, which does not construct any galleries in sediments.

Images of oxygen distribution were taken before the introduction of the organisms (control) and at intervals that differ for each organism after their introduction.

Fifteen oxygen images, with their corresponding sediment structure images, were chosen within the overall data set in order to illustrate, for each species, different patterns of biogenic structures created by the organisms. For *N. diversicolor*, the three selected images, with their respective controls, came from three different faces of the tank and were taken 19, 43 and 68 h after the organism introduction.

The two oxygen images of *N. virens* biogenic structures were taken after 12 and 35 h on two different tank faces (respective controls not available). For *C. neritea*, five oxygen images with their respective controls came from two faces of the tank and were taken 10 min, 8, 9, 22, and 28 h after the organism introduction. Starting from an oxygen image of a bioturbated sediment column, the heterogeneity index was calculated based on the horizontal variability because it is mainly due to macrofaunal reworking activities, whereas the variability along the vertical axis is mainly due to the oxygen diffusing at the water-sediment interface from the overlaying water. This distinction was taken into account in our variability index. This index is a variation coefficient obtained as follows: $f(x,z)$ is the distribution of the oxygen concentration as a function of the horizontal position x and of depth z. For each fixed value of x, the function $z \rightarrow f(x,z)$ is called the oxygen vertical profile at position x. The mean vertical profile of the image is defined by:

$$f_m(Z) = 1/1L_H \int_{x_m}^{x_M} f\left(x,z\right)dx$$

where L_H is the horizontal size of the image, xm is the minimal abscissa, and xM is the maximum abscissa in the image. For instance, we may choose $x_m = 0$ and $x_M = L_H$.

The horizontal variability of the oxygen distribution can then be expressed by:

$$V_f(Z) = 1/1L_H \int_{x_m}^{x_M} f\left(f\left(x,z\right) - f_m(Z)\right)^2 dx$$

The dimension of the last expression is the square of the dimension of the oxygen concentration. The ratio $CV_f(z)=\sqrt{(V_f(z))}/(f(z))$ is a variation coefficient. It corresponds to a normalized standard deviation of profile that is the standard deviation divided by the mean. If the previous function is null, then all the vertical profiles are the same, and we can conclude that there is no horizontal variability, whereas, if the function is not null, then some vertical profiles differ, and we can conclude that there is a horizontal variability, which is as high as the function value. The horizontal variability is measured by the previous function. In order to get a number, which expresses this variability, the mean of the previous function along the depth is taken:

$$I = 1/1L_V \int_{x_m}^{x_M} f\,CV_f(Z)dz$$

where LV is the vertical size of the image, z_m is the minimum depth, and z_M is the maximum depth in the image. It is standard to consider the water-sediment interface at depth 0, thus typical images have negative z_m and positive z_M.

Vertical oxygen profiles extracted from images make the determination of diffusive oxygen fluxes ($J_{(z)}$) possible, which were calculated from Fick's first law of diffusion (Berner 1980; Jorgensen and Revsbech 1985; Rasmussen and Jorgensen 1992):

$$J_{(z)} = -\Phi D_s \; dC_{(z)}/dz$$

where Φ is the porosity, D_s is the oxygen diffusion coefficient in sediments ($cm^2.d^{-1}$), C is the oxygen concentration ($\mu mol.L^{-1}$), z is the depth (cm), and $dC(z)/dz$ is the oxygen gradient. This approach is based on the assumption that molecular diffusion is the main oxygen transport mechanism. The sediment-water interface was localized on the sediment structure images.

Calculations were performed using the PROFILE software (Berg et al. 1998), which assumes steady-state conditions. In this study, we used the total oxygen diffusive flux (J_t), which was quantified for all images in burrowed and non-burrowed areas and was obtained as follows:

$$J_t = \left[\left(J_{fs} \times W_s\right) + \left(J_{fb} \times W_b\right)\right] / W_i$$

where J_{fs} and J_{fb} were the mean fluxes at the sediment surface and in the biogenic structure, respectively, Ws, Wb, and Wi were the widths of the sediment surface, the biogenic structure, and the image, respectively.

The gallery-builder organisms, *N. diversicolor* and *N. virens*, led to an oxygen distribution pattern, which was more spatially heterogeneous compared to controls and the burrowing organism *C. neritea*, which was close to the controls (Fig. 2). Concerning the total oxygen diffusive flux, they were three- to seven-fold enhanced for *N. diversicolor* in comparison with their respective controls; in contrast, for *C. neritea*, there was only a weak increase of about 1.2-fold of the total flux, and it seems that the total flux increase is mainly linked to the

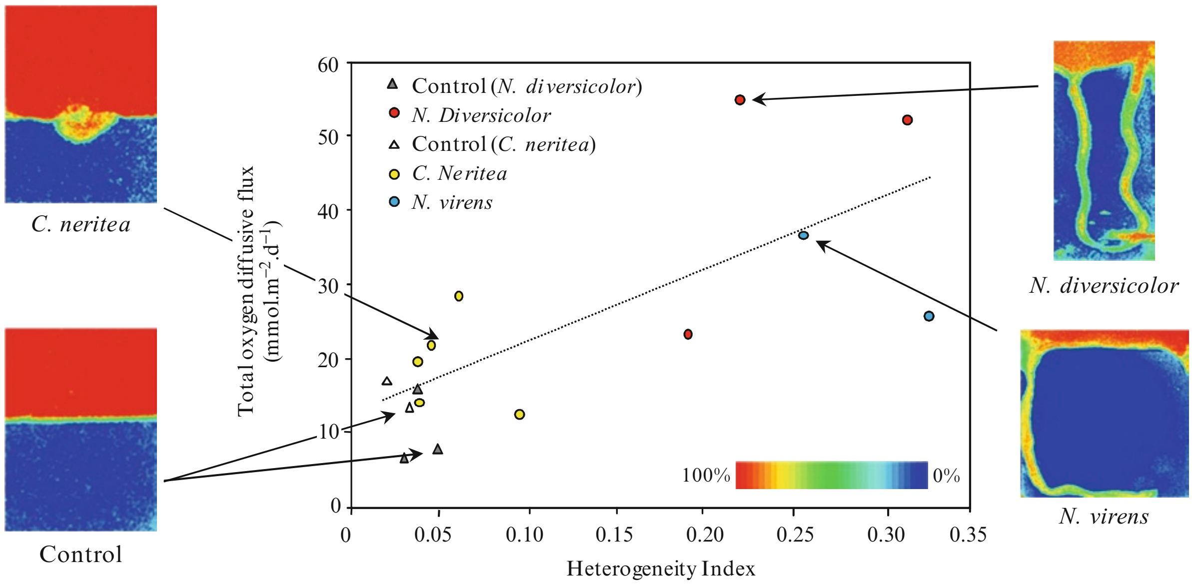

Fig. 2 Integrated diffusive oxygen flux versus variability index for the different species and tendency curve (t test, $p < 0.0005$, $n = 15$)

sediment water interface length enhancement (Fig. 2). Differences between the gastropod and the polychaetes may be explained by their bioturbation behavior. *C. neritea*, when buried, did not affect more than the first centimeter and led to the production of a simple subsurface patch in the oxygen distribution. However, the polychaete worms constructed burrows in the sediment, which strongly enhanced the interface length (two- to ninefold). This, associated with the intermittent and active ventilation of their structure, increased the degree of complexity of oxygen distribution in the sediment column. As shown by the positive relationship between the total diffusive oxygen flux and the variability index (Fig. 2), it seems that integrated flux increased with spatial oxygen heterogeneity. Indeed, sediments inhabited by *C. neritea* presented a low variability index and oxygen flux values that were close to those of controls, whereas, in the presence of the polychaetes, both values were higher. When buried, it did not affect more than the first centimeter and led to the production of a simple subsurface patch in oxygen distribution. However, the polychaete worms constructed burrows in the sediment that strongly enhanced the interface length (two to ninefold). This, associated with the intermittent and active ventilation of their structure, increased the degree of complexity of oxygen distribution in the sediment column. As shown by the positive relationship between the total diffusive oxygen flux and the variability index (Fig. 2), it seems that integrated flux increased with spatial oxygen heterogeneity. Indeed, sediments inhabited by *C. neritea* presented a low variability index and oxygen flux values that were close to those of controls, whereas, in the presence of the polychaetes, both values were higher.

Based on the use of oxygen planar optode quantification, the present work highlighted the influence of organisms, presenting distinct bioturbation behaviors, on oxygen distribution heterogeneity in aquatic sediments. Moreover, this study has demonstrated the effect of oxygen distribution heterogeneity on diffusive oxygen flux, with gallery-builders producing greater spatial oxygen heterogeneity and higher fluxes due to their complex ventilated structures compared to the burrowing species. This work showed that heterogeneity is a feature that plays an important role in the control of ecosystem functioning.

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Using a New Fluorescent Probe of Silicification to Measure Species-Specific Activities of Diatoms Under Varying Environmental Conditions

Bernard Quéguiner, Karine Leblanc, Véronique Cornet-Barthaux, Leanne Armand, F. Fripiat, and D. Cardinal

Abstract A new method is presented that enables distinguishing between active and non-active cells with regard to biogenic silica deposition during frustule formation in natural communities of siliceous phytoplankton. The PDMPO method is based on the fluorescence of biogenic silica after incubation with the probe. Only those cells that have been depositing silica (by adjunction of intercalary plates during the cell cycle or by depositing a new frustule valve upon cell division) exhibit a typical fluorescence that is proportional to the amount of biogenic silica deposited. This new method has several advantages; it is easy to use at sea, very sensitive, and samples can be conserved for several months without major loss of fluorescence. This method offers new possibilities of investigation of ecophysiological controls within the natural diatom community and will also bring more information to the new generation of sophisticated multi-element multi-species biogeochemical models.

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1 Introduction

The unveiling of plankton successions marked the pioneer studies of biological oceanography. Incidentally, it allowed Sverdrup (1953) to elaborate the now classical model of spring bloom determinism in relation to the opposite deepening of the critical depth and shallowing of the surface mixed layer. The role played by marine organisms in controlling the distribution of chemicals, hence the chemical composition of seawater, was rapidly perceived, bringing along the emergence of the concept of Redfield et al. (1963) ratios. However, the exact role of nutrients in intimately structuring the phytoplankton communities at the species, even at the class, levels was only superficially addressed. Such was also the case for the role of phytoplankton successions in intimately structuring the zooplankton communities, hence, the export fluxes inside the oceans.

Since the late 1970s to early 1980s, there has not been much evolution in our understanding of marine plankton successions. Within the framework of a global atmospheric CO_2 increase, the research focused on quantification of global biogeochemical cycles under the umbrella of the "Joint Global Ocean Flux Study" (JGOFS) with consideration of the large biogenic fluxes and stocks. Large-scale, globally orientated, JGOFS-type studies led to an absence of consideration of the dynamics of plankton successions,

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a topic that was also “neglected” by GLOBEC-type studies focused on a few key species related to upper trophic levels and orientated towards marine biological resources. Fundamental objectives of JGOFS resulted in (1) developing a systematically global approach in the elaboration of biogeochemical models of the pelagic ecosystem; such an approach did not fully consider the regional variability of ecosystems, although it was evidenced by the variability in community structures; and (2) considering the most significant processes regarding the control of carbon fluxes; such an approach was restricted to the direct relationships between primary production and the availability of associated biogenic elements, leaving only a small interest for the community structure restricted to the top-down control of the fluxes of matter.

The dominance of diatoms in a phytoplankton assemblage is of major importance for the biological pump, since they tend to export C to depth and eventually to the seafloor (although this is still a matter of debate) more efficiently than non-siliceous (flagellates) species. In the last decade, several studies have reported an increasing dystrophy in coastal waters, especially when submitted to the influence of riverine discharges (Smayda 1990). A consequent decrease in the Si:N ratio appears to be a general trend in various coastal areas, including European Atlantic and North Sea coasts (Billen et al. 1991; Le Pape et al. 1996; Del Amo et al. 1997a, b) as well as Mediterranean systems like the Adriatic Sea (Carlsson and Granéli 1999; Granéli et al. 1999). While a decrease in the Si:N ratio would lead to a predominance of non-siliceous and sometimes toxic species over diatoms, resulting in undesirable eutrophication effects such as anoxic conditions, fish and shellfish mortality, and harmful algal blooms, some studies also reveal that the effect of nutrient dystrophy could rather lead to a shift within the diatom community, favoring species with high affinity constants for $Si(OH)_4$ rather than a shift towards non-siliceous species, with potential implications for the food web structure (Del Amo et al. 1997a, b). Studies on causes of diatom shifts have been limited hitherto by the inability to provide species-specific parameterizations of nutrient uptake within natural communities. While some studies have enlightened the potential of silicic acid availability as a controlling factor of diatom dominance, only few experiments have been able to address that problem in natural communities because of the requirement of species isolation or of the opportunistic use of almost monospecific diatom bloom occurrences, which sometimes characterize perturbed ecosystems. Using this opportunistic approach, Del Amo (1996) was able to evaluate the different kinetic parameters of three diatom species occurring at different periods in the Bay of Brest coastal ecosystem: *Guinardia delicatula* (V_{max} = 0.040 ± 0.004 h^{-1}, K_S = 3.3 ± 0.7 μM), *Chaetoceros debile* (V_{max} = 0.042 ± 0.005 h^{-1}, K_S = 1.6 ± 0.3 μM), and *Chaetoceros sociale* (V_{max} = 0.044 ± 0.005 h^{-1}, K_S = 2.2 ± 0.5 μM).

Here we present the outlines of a novel method based on a combination of isotope quantification and fluorescent labeling of diatoms, which should prove useful for measuring the silica deposition activity and the kinetic parameters of silicic acid uptake by individual diatom cells within a natural community. We will first review the existing methods, present the method based on the [2-(4-pyridyl)-5{[4-dimethylaminoethylamino-carbamoyl)-methoxy]phenyl}oxazole] (PDMPO) marker, and show some preliminary results obtained in the Southern Ocean.

2 Methods for Labeling Si Deposition at the Cell Level

Tracking the uptake of any nutrient at the individual cell level can be made possible by labeling the nutrient itself or associating a marker which has the same behavior.

The first method requires the use of a radiotracer that can be revealed by micro-autoradiography. Regarding silicon, no radioactive tracer was readily available before 1991, the only available radioactive isotope being ^{31}Si, whose half-life period is 2.62 h (β^-, 1.49 MeV). Tréguer et al. (1991) introduced the use of ^{32}Si, whose half-life period is 134 years (β^-, 0.22 MeV).

This radioisotope produced by the Los Alamos US DoE facility until 2005 proved very useful in addressing the question of biogenic silica production in oligotrophic regions while enabling more easy experimental determination of uptake kinetic parameters. ^{32}Si has also been used in autoradiography studies by Shipe and Brzezinski (1999), and the technique allowed visualization of where biogenic silica was deposited in the frustule and thus inference of species-specific new biogenic silica deposition. However, this technique is

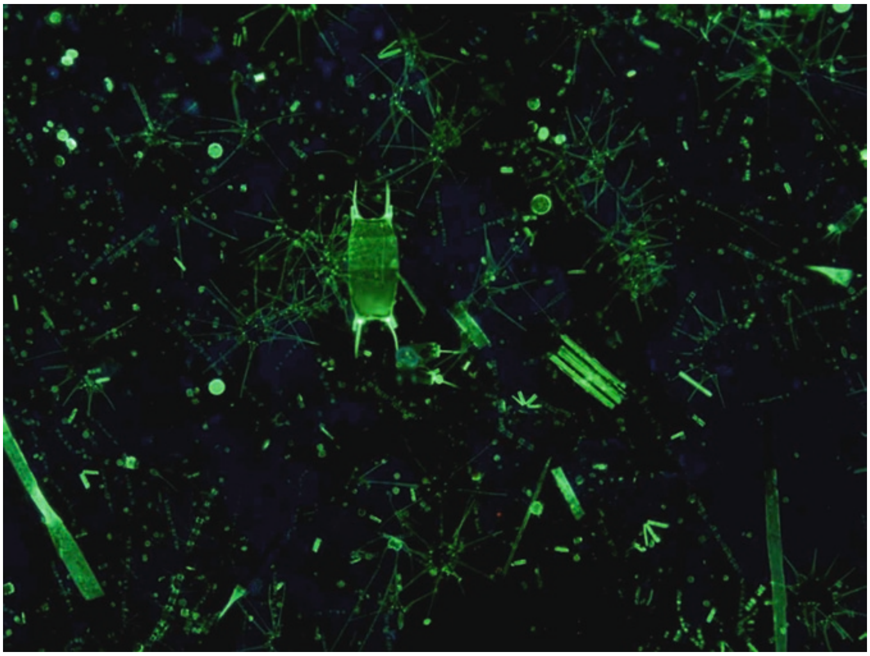

Fig. 1 PDMPO fluorescent staining of a whole diatom community

not easily performed because it requires between 4 months to a year of exposure time of the samples to a photographic emulsion (Leblanc and Hutchins 2005).

The second method requires the use of a probe that can be associated to the nutrient to track. Regarding silicic acid uptake, a first attempt has been made by Brzezinski and Conley (1994) by using rhodamine 123, a tracer of cell membrane formation. As silicic acid deposition occurs within a silica deposit vesicle (SDV) analogous to the fragmoplast of higher plants, some rhodamine 123 is incorporated into the silica frustule during the deposit phase (for more details, see Brzezinski and Conley 1994). Brzezinski and Conley (1994) got interesting results from *Thalassiosira weissflogii* cultures, but the method proved very difficult to perform because of the interference of membrane staining requiring a treatment step to separate it from incorporation into the biogenic silica.

The fluorescent probe PDMPO is also named LysoSensorTM Yellow/Blue DND-160, and can be obtained from Molecular Probe (L-7545) in a 1-mM dimethylsulfoxide matrix. It is a pH probe reacting to acidic intracellular environments (Diwu et al. 1999) and therefore prone to enter the diatom SDV and to be incorporated in the biogenic silica (Fig. 1). The feasibility of use of PDMPO for silica tracing was first demonstrated by Shimizu et al. (2001), while the precise protocol was defined later by Leblanc and Hutchins (2005), who first used the molecule in the study of natural diatom communities of the Delaware estuary. Briefly, the method is characterized by its facility of use (it only requires incubation of the natural community with PDMPO, which can be done in parallel to incubation by an isotopic tracer if quantification is needed), a slow fading under the UV light, and the long-term stability of filter samples (several years, see Fig. 2).

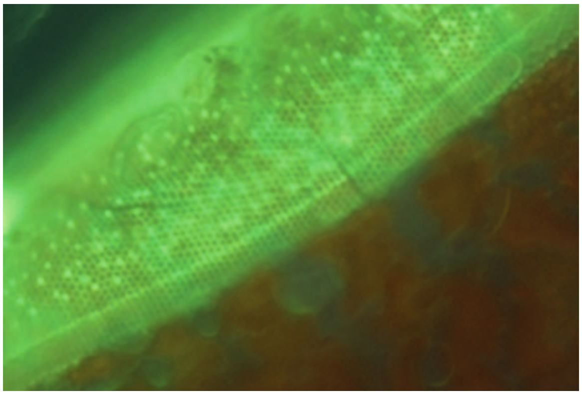

Fig. 2 Details of a diatom frustule observed 2 years after staining

3 A Case Study: Preliminary Results on the Biogenic Silica Deposition Activity in a Diatom Community of the Polar Front Zone in the Australasian Sector of the Southern Ocean

From 17 January to 20 February 2007, a 32-day oceanographic voyage onboard Australia's ice-breaker Aurora Australis was undertaken to examine microbial ecosystem structure and biogeochemical processes in the Sub-Antarctic Zone (SAZ) waters west and east of Tasmania, and also in the Polar Front Zone (PFZ) south of the SAZ, as part of the SAZ-SENSE Project (study of the sensitivity of SAZ waters to global change).

During the cruise several experiments were performed in order to study the activity of the diatom communities. The preliminary results obtained at the PFZ process station are presented in Fig. 3.

There is a clear difference between the contribution of the different diatom taxa to the overall biomass (expressed here as cell counts) and the activity of biogenic silica deposition, as evidenced by quantification of PDMPO fluorescence by microscopic image analysis. The diatom community is characterized by a high biodiversity with several different groups contributing more or less to same extent, while the biogenic silica deposition activity is almost restricted to *Rhizosolenia* sp., which contributes 91% of the total activity. These results are preliminary and are presently being processed to get a more precise estimation of the biomasses. However, it is evident that the overall picture will remain unchanged. Such a result means that some elements of the diatom community are probably undergoing different strategies of development and/or are

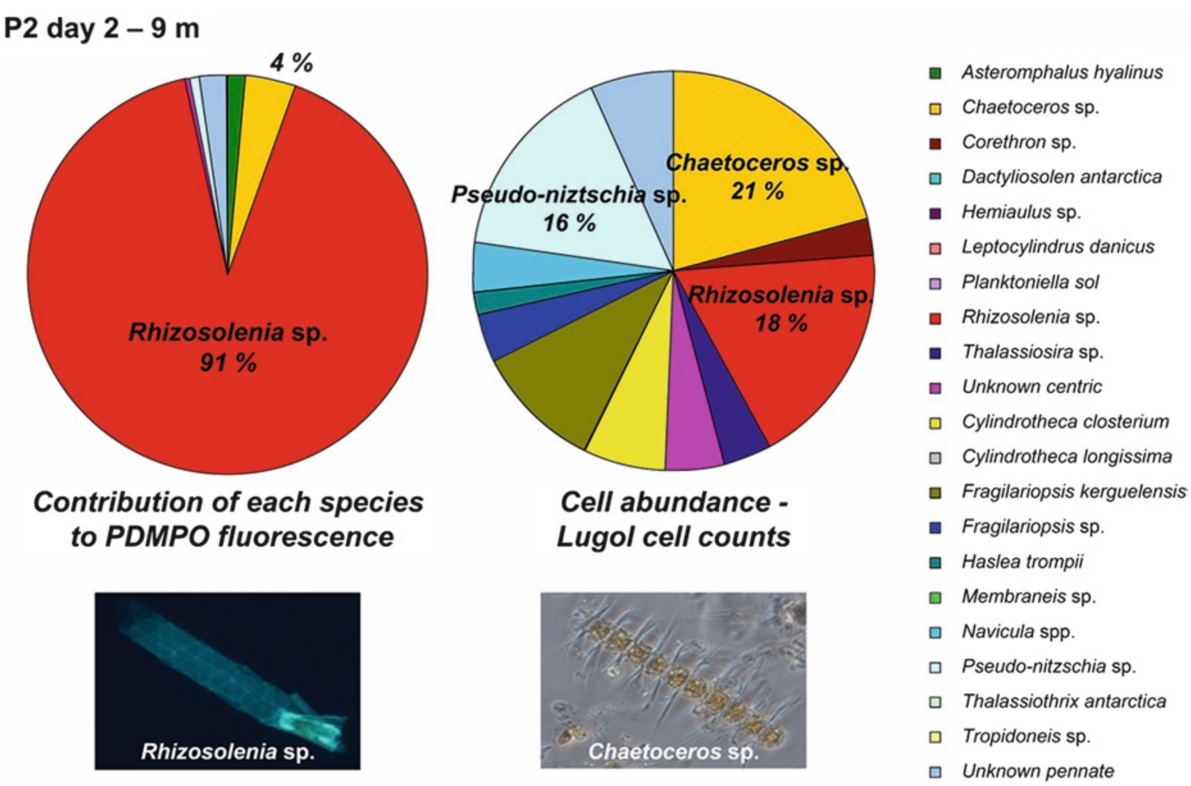

Fig. 3 Comparison between the silica deposition activity as evidenced by the fluorescence of the different diatom groups after incubation of the natural community with PDMPO for 24 h (*left*) and the contribution of each diatom taxon identified by regular cell counts of a Lugol preserved sample of the same community (*middle*). The diatom checklist is also indicated (*right*)

controlled by different factors, such as nutrient availability versus resistance to grazing.

4 Conclusion

With the increasing refinement of biogeochemical models to take biodiversity into accounty, there is an increasing need to obtain parameterization and validation data at the species level. The PDMPO, in association with isotopic tracing methods using radioactive (^{32}Si) or stable (^{30}Si) isotopes, is a very promising tool because it should enable getting individual kinetic parameters characterizing the different components of the siliceous plankton community. This method also offers the possibility of segregating the different actors of the siliceous community and will certainly give new insights into the control of the activity by different ecological factors and should help deciphering the complex interactions between species that are responsible for the successions of dominant species in the natural ecosystems. In the coming years, other methods derived from molecular biology will certainly be available to quantify key enzymatic processes by gene expression (nitrate reductase, alkaline phosphatase, nitrogenase, etc.), which will provide information on other major nutrient utilizations.

The silicon metabolism is a good choice to start this new approach at the cell level because the incorporation of the PDMPO probe is proportional to the degree of silicification (Leblanc and Hutchins 2005), which is itself directly related to the silicon uptake by the active cells (Hildebrand 2000).

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Utilization of a Submersible Ultra-Violet Fluorometer for Monitoring Anthropogenic Inputs in the Mediterranean Coasts

Marc Tedetti, Catherine Guigue, and Madeleine Goutx

Abstract The Mediterranean Sea coasts are subject to a strong anthropogenic pressure because of an intensification of urban, industrial, agricultural, and tourist activities. This anthropogenic pressure is reflected in the direct (from coastal sources) and indirect (via rivers) discharge of terrigenous materials and pollutants. These contributions promote transformations of ecosystems and associated biogeochemical cycles.

In order to assess environmental changes, discrete measurements at fixed observation sites are useful, but remain insufficient to describe the synoptic extension and the impact of these released terrigenous inputs.

An objective of the laboratory is to develop and use in situ UV fluorometers for the quantification at high spatial and temporal frequency of terrigenous and anthropogenic tracers such as polycyclic aromatic hydrocarbons (PAHs, index of chemical contaminations) and "tryptophan-like" material (rough index of bacterial contaminations).

An in situ UV fluorometer (EnviroFlu-HC, TriOS Optical Sensors) was deployed in the coastal area of Marseilles from January to July 2008. The fluorescence signal of this probe (Ex/Em: 254 ± 12.5/360 ± 25 nm) was compared to the spectrofluorimetric excitation emission matrices (EEMs) of discrete samples taken from the same area. The probe signal, due to its relative large excitation and emission bands, covers the EEM signature of several groups of compounds: PAHs, tryptophan-like material, and humic substances.

Based on our in situ observations, and in collaboration with a French company specialized in optical sensors, we propose solutions for improving the optical properties of the EnviroFlu-HC fluorometer to better target compounds of interest and miniaturize the probe. Ultimately, these miniaturized UV fluorometers will be integrated to autonomous mobile platforms (gliders).

This study is part of a laboratory program that aims to develop observation tools for operational oceanography and urban marine zone management: The project SEA EXPLORER (leader ACSA underwater GPS, partnerships: ACRI-IN, ACRI-ST, IFREMER, LMGEM-COM, LOV) funded by the Ministry of Industry (Fond de Compétitivité des Entreprises, FCE). This work also received the financial support of the Conseil Général des Bouches-du-Rhône (CG 13) and Agence de l'Eau Rhône-Méditerranée.

1 Introduction

The Mediterranean Sea coasts are subject to a strong anthropogenic pressure because of an intensification of urban, industrial, agricultural, and tourist activities. This anthropogenic pressure is reflected in the direct (from coastal sources) and indirect (via rivers) discharge of terrigenous materials and pollutants (Tolosa et al. 1996). These contributions promote transformations of ecosystems and associated biogeochemical cycles.

Recently, a submersible ultraviolet (UV) fluorometer (EnviroFlu-HC, TriOS Optical Sensors) has been designed and constructed in order to measure in real-time polycyclic aromatic hydrocarbons (PAHs) in the aquatic media.

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PAHs are highly toxic, mutagenic, and carcinogenic compounds (Samanta et al. 2002), and are thus listed as priority pollutants by both the European Union (EU) and the United States Environmental Protection Agency (US-EPA). Because of their aromatic structure, PAHs have inherent fluorescent properties in the UV spectral domain (200–400 nm) (Booksh et al. 1996; Dabestani and Ivanov 1999).

We propose in this study to evaluate, for the first time, the performances of the EnviroFlu-HC submersible UV fluorometer for its routine utilization in coastal marine waters subjected to sporadic or regular anthropogenic impacts. This evaluation was carried out through two approaches: (1) laboratory experiments in which the probe was calibrated with different fluorophores (PAHs, tryptophan and humic acid) and (2) in situ measurements at different contrasted sites in the coastal area of Marseilles (Northwestern Mediterranean Sea, France).

2 Material and Methods

Calibration experiments were performed in the laboratory under controlled environmental conditions (dark, 20°C) to assess the potential response of the Enviro Flu-HC to different aquatic fluorophores chosen according to their fluorescence spectral domains: PAHs, tryptophan, and humic acid. Field measurements were conducted from January to July 2008 in the coastal area of Marseilles aboard the R/V Antedon II.

Several contrasted sites were sampled many times over the course of the year: "SOFCOM" (43°14.3′N, 05°17.3′E, 60 m depth), the observation station of the Oceanology Center of Marseilles located near the Frioul Islands; "Riou" (43°09.0′N, 05°24.0′E, 90 m depth), a remote station in the Southern part of the Bay; "Cortiou" (43°12.8′N, 05°24.0′E, 12 m depth), the discharge area of treated wastewaters of Marseilles City located in the Cortiou creek; "Saumaty" (43°21.7′N, 05°18.7′E, 4 m depth), the harbor of Saumaty in the northern part of the Bay.

3 Results and Discussion

The EnviroFlu-HC fluorometer was able to detect several groups of aromatic compounds, but was much more efficient in the quantification of PAHs than tryptophan and humic substances at the μg level (Table 1).

The sensor signal showed great spatial and temporal variations in clean (SOFCOM, Riou) and polluted (Saumaty, Cortiou) sites, with likely a high contribution of PAHs in the harbor, and a high contribution of tryptophan-like and humic-like materials in the wastewaters (Fig. 1).

4 Conclusions

The EnviroFlu-HC fluorometer is a good tool for monitoring anthropogenic inputs in the Mediterranean coast, although supplementary fluorescence measurements would allow improving the information about the nature of the aromatic compounds detected. Based on our observations, and in collaboration with a French company specialized in optical sensors, we propose solutions to reach a better identification of the compounds of interest and to miniaturize fluorometers dedicated to the detection of marine pollutants.

Ultimately, two miniaturized UV fluorometers will be integrated into an autonomous mobile platform

Table 1 Characteristics of the EnviroFlu-HC UV fluorometer for measurements of the three types of aromatic compounds: PAHs, tryptophan, and humic acid

	PAHs	Tryptophan	Humic acid
Linearity	0.3–6 μg l^{-1}	0.5–100 μg l^{-1}	170–3,600 μg l^{-1}
Saturation	From 30 μg l^{-1}	No	No
Detection limit	0.3 μg l^{-1}	1 μg l^{-1}	250 μg l^{-1}
Quantification limit	0.45 μg l^{-1}	3.8 μg l^{-1}	1,700 μg l^{-1}
Sensitivity	240 raw counts/μg l^{-1}	30 raw counts/μg l^{-1}	0.07 raw counts/μg l^{-1}
Precision	±24 at 300 raw counts	±17 at 500 raw counts	±12 at 49 raw counts
Accuracy	Relative RMSE = 4.6%	Relative RMSE = 29%	Relative RMSE = 33%

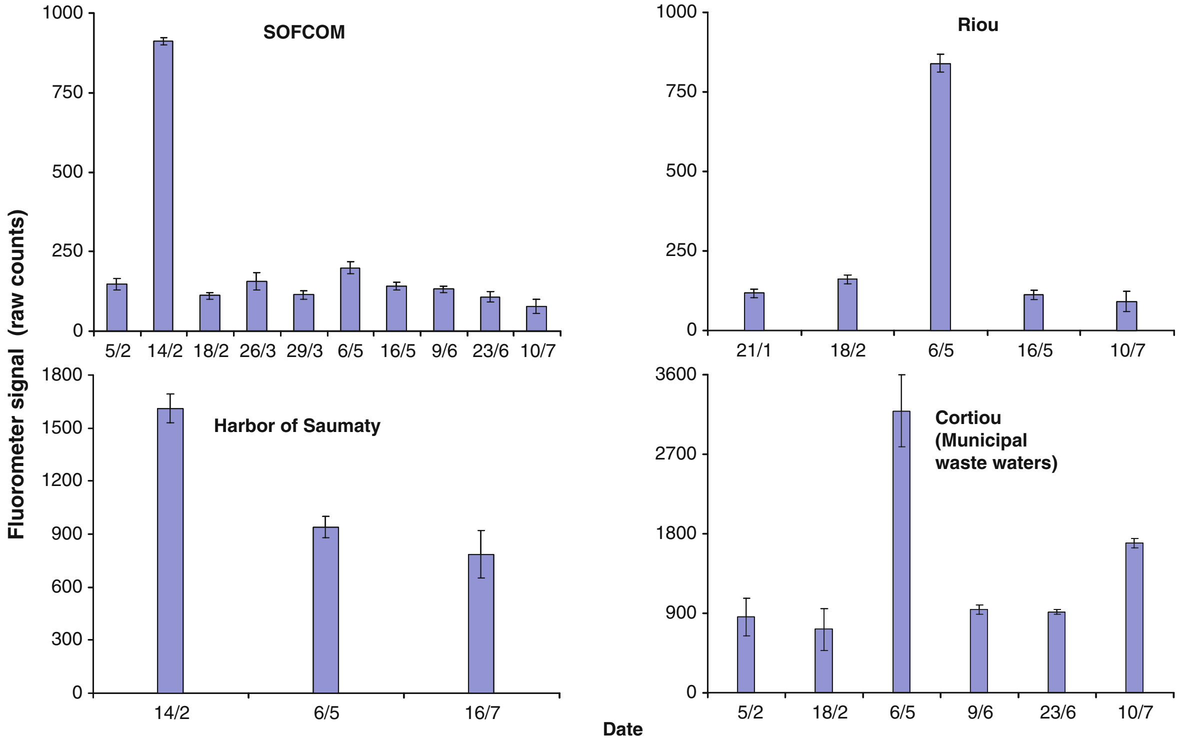

Fig. 1 Fluorescence signal recorded by the EnviroFlu-HC UV fluorometer in the surface waters (0.5 m depth) of the Bay of Marseilles from January to July 2008

(underwater glider) in the framework of the SEA EXPLORER project.

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Temporal and Spatial High-Frequency Monitoring of Phytoplankton by Automated Flow Cytometry and Pulse-Shape Analysis

Melilotus Thyssen and Michel Denis

Abstract Phytoplankton were investigated with automated high frequency flow cytometry to address their patchiness and short-term variability.

To document this, we deployed a submersible flow cytometer (Cytosub, www.cytobuoy.com) in the Bay of Marseille (North Mediterranean) at 2 m depth.

This instrument involves pulse shape analysis and can analyze cells (1–1000 µm) and even chains at a flow rate of 8 $mm^3.s^{-1}$. During experiments, which took place in summer 2005, phytoplankton were monitored *in situ* every 30 min. The seven clusters (cells sharing the same optical properties of scatter and fluorescence) resolved in the size range 1–50 µm and behaved as independent entities, suggesting that they could be considered as functional groups.

The spatial heterogeneity of oceanic phytoplankton surface distribution was addressed by running the CytoSub on board a 33-m schooner (Fetia Ura, www.seanergies.com) between the Azores and French Brittany in April 2007. The flow cytometric analysis was triggered every 15 min (spatial resolution of 2.8 km). Five clusters were resolved in the pumped surface water, and specific relationships were determined between their distributions within the different water masses sampled during the cruise.

The evidenced variabilities are critical to explain the impact of intrinsic and extrinsic factors on phytoplankton spatial and temporal distributions. Automated *in situ* flow cytometry appears to be a powerful tool to investigate phytoplankton assemblages at high frequency and at the single cell level. Undergoing technological developments are extending this capacity to the whole microbial ecosystem and aim at real-time information of stakeholders.

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1 Introduction

The importance of phytoplankton stems from its ubiquity at the surface of the ocean, which covers about 70% of the earth. More than its presence with an overall biomass of the order of four 10^{12} kg C (Nee 2004), its role in the functioning of the marine ecosystem is fundamental. Indeed, it contributes to about 50% of the annual earth primary production (Field et al. 1998). Its energy source, sunlight, appears unlimited, as does its carbon source, dissolved ça peut aussi être du CO_2 produit par respiration? CO_2.

Phytoplankton work at the entry point of the biological pump (Longhurst and Harrison 1989; Bishop 1989) that leads to the sequestration by the ocean of a significant amount of the atmospheric CO_2 generated by human activity and inducing an increase in the greenhouse effect. Phytoplankton thus play an essential role in the regulation of the earth's climate, not only with respect to the carbon cycle, but also in producing gases (i.e., DMS, dimethyl sulphide), leading to cloud condensation nuclei (Charlson et al. 1987; Malin et al. 1992).

Improving our knowledge on phytoplankton assemblages and their functioning in the ocean ecosystem is of major importance, but it is far from easy. The difficulties have many aspects. Phytoplankton cells make a highly diversified phylogenetic group with more than 15,000 species. This very large diversity accompanies an extreme variability in morphology as well as in physiology. Cell abundances are spread over a concentration

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range of about six orders of magnitude (1–10^6 cells cm^{-3}); the larger they are, the less abundant they are. Cell size is also another source of variability with the smallest, 0.5–0.7 μm for *Prochlorococcus*, up to 1–2 mm for the largest cells or chains. Growth rate is also species dependent with some species undergoing up to two divisions per day (Alpine and Cloern 1988; Furnas 1991). As single cell organisms with fast growth rates, phytoplankton react promptly to any environmental change, whether natural or of anthropogenic origin, which makes its distribution highly variable with time and heterogeneous with space both at small (mm) and large (1,000 km) scales. Time and space scales are interacting, which adds to the difficulty of investigating phytoplankton assemblages.

Due to all these difficulties, phytoplankton is usually not correctly sampled, either qualitatively or quantitatively, essentially because of the lack of appropriate techniques (Smayda 1998; Baretta et al. 1998).

To overcome the above listed limitations, we took advantage of the commercialization of a new instrument specially designed for in situ investigation of phytoplankton at the single cell level in an automated way (Cytosub, www.cytobuoy.com). We thus developed a new approach (Thyssen et al. 2008a) involving high frequency (up to every 15 min) phytoplankton analysis based on this instrument, which collects the shapes of pulses generated by cells when intercepted by the laser beam. We investigated the short-term variability of the phytoplankton distribution by conducting time series over two summer periods in 2005 and 2006 in the Bay of Marseilles (Thyssen et al. 2008b). To address the spatial heterogeneity of the phytoplankton distribution, we conducted the analysis of surface phytoplankton along a sailing ship track between the Azores Islands and French Brittany in April 2007 (Thyssen et al. 2009). The presented results are part of the PhD thesis of Thyssen (2008) and were also reported elsewhere (Thyssen et al. 2008a, b, 2009).

2 Materials and Methods

2.1 Sampling Strategy

For the time series, the Cytosub was fixed on a stainless steel frame and moored at the end of a quay in a sailing yacht harbor in Marseille (43°17″N, 5°22′E) at 2 m depth. Energy was supplied through a 250-m waterproof electric cable also used to continuously transfer data to a computer. Samples were collected every 30 min from 11 July to 5 September 2005.

For the spatial study, samples were automatically collected from the 14 to the 23 April 2007 during a cruise of the "Fetia Ura" (http://www.sewnergies-oceans.abcsalles.com/prive/fr/fiche.php?n=10617) sailing ship between Horta (38.6°N–28.6°W,Island of Faïal,the Azores) and Lorient (47.6°N–3.6°W, French Brittany). Seawater was pumped from the ship's central non-toxic seawater supply (situated in the center of the boat) at 1.5 m depth every 15 min for 3 min at 30 $dm^3.s^{-1}$, filling a 1 dm^3 reservoir sampled by the Cytosub 2 min after the pump stopped in order to let air bubbles disappear. The pump used a flexible impeller technology that does not squeeze the water passing through, avoiding damaging of the cells.

An autonomous sensor was coupled in both situations to the Cytosub to collect in parallel conductivity, temperature, and depth (CTD, Microcat SBE 37). An autonomous fluorometer (ECO, Wet Labs) was also coupled to the Cytosub for the time series experiment.

2.2 Flow Cytometry

The Cytosub was designed to analyze large phytoplanktonic cells (1–1000 μm and a few mm in length) and relatively large water volumes (up to 4 cm^3 per sample). It was cable (250 m) connected for energy supply and data transfer to a computer. In the flow cell, each particle was intercepted by a laser beam (Coherent solid-state Sapphire, 488 nm, 15 mW), and the generated optical signals were recorded. The light scattered at 90° (side scatter), and fluorescence signals were dispersed by a concave holographic grating and collected via a hybrid photomultiplier (HPMT). The forward scatter signal was collected via a PIN photodiode. The red (FLR), orange (FLO), and yellow (FLY) fluorescences were collected in the wavelength ranges 668–734, 601–668, and 536–601 nm, respectively. Data recording was triggered by the forward scatter signal. The shape of the signals was encoded at a frequency of 4 MHz, and data were saved in distinct 64-kb grabbers before their transfer to the computer through the connecting cable. Particles flew at a rate of 2 m.s^{-1} through the 5 μm laser beam. A priori cette définition n'est pas aussi évidente, dixit George.

2.3 Cytometric Softwares

Cytoclus software (version 2004, Cytobuoy b.v.) was used to analyze the data collected by the Cytosub. Clusters were selected by taking into account the amplitude and the shape of the different signals. In addition to five average signal heights for forward scatter (FWS), sideward scatter (SWS), and for three fluorescence signals (FLR, FLO, FLY) some simple mathematical models were assigned to each signal shape: inertia, fill factor, asymmetry, number of peaks, length, and apparent size (FWS size) (Dubelaar et al. 2003).

See Thyssen et al. 2008a, b for more details.

3 Results

3.1 Short-Term Variability

The Cytosub was programmed to run phytoplankton analyses every 30 min according to two successive settings enabling observations of the smallest cells (>1 μm) and larger cells by using a threshold to prevent saturation of the buffer memories with the smallest cell signals.
These buffer memories brought a limitation in the analyzed size spectrum (<20 μm) that is now corrected by new electronics achieving real-time transfer to a computer. Seven clusters (cell groups) were resolved on the basis of their optical properties and pulse shapes, and were labeled C1 to C7 in absence of species identification (Fig. 1). The survey lasted 2 months in summer 2005. The high frequency analysis enabled the determination of cell cycles for the clusters by combining variations of fluorescence, size, and abundance (DuRand, and Olson 1996; Vaulot and Marie 1999).

Autocorrelation calculations conducted on data (from 528 samples) collected over 12 days where meteorological conditions did not apparently disturb the clusters also yielded cell cycles of about 24 h for most of the clusters. Cell groups C2 and C5 exhibited daily abundance variations between the detection threshold and >10^3 cells.cm^{-3} (see Fig. 1b). Both groups appeared sensitive to changes in the N/P Redfield ratio. Indeed, at two periods during the survey, N/P became lower than the standard Redfield value of 16. On both occasions, we observed a peak of abundance for C5 followed by one of C2 with a 4-day interval. The other clusters responded to wind events by exhibiting peaks of abundance appearing along the same sequence. Differences in the response delay and in the interval between the abundance peak formation were linked to the intensity and duration of the related wind events.

3.2 Spatial Heterogeneity

The spatial heterogeneity of the phytoplankton distribution at a sub-meso scale sampling resolution (1–10 km) was addressed during the ACYPHAR (Atlantic flow CYtometry spatial study of PHytoplankton through Automated Recording, 14–23 April 2007) cruise between Horta and Lorient.

Six clusters were resolved by flow cytometry, and four water masses were distinguished along the ship track from the temperature-salinity relationships derived from the data collected simultaneously with an autonomous CTD (SBE 37SMP). Abundance variations were complex and illustrated a strong patchiness, as expected. A short-term variability was superimposed on a larger trend change. Different smoothing treatments of the data enabled to sort the long trend change from the short-term variability. Autocorrelation calculations were applied to the short-term variability signal to look for a possible periodicity of this short-term variability. Results showed in particular that cells of the C1 cluster exhibited a cyclic variation twice a day for their abundance, size, and red fluorescence. Significant correlations were found between cluster

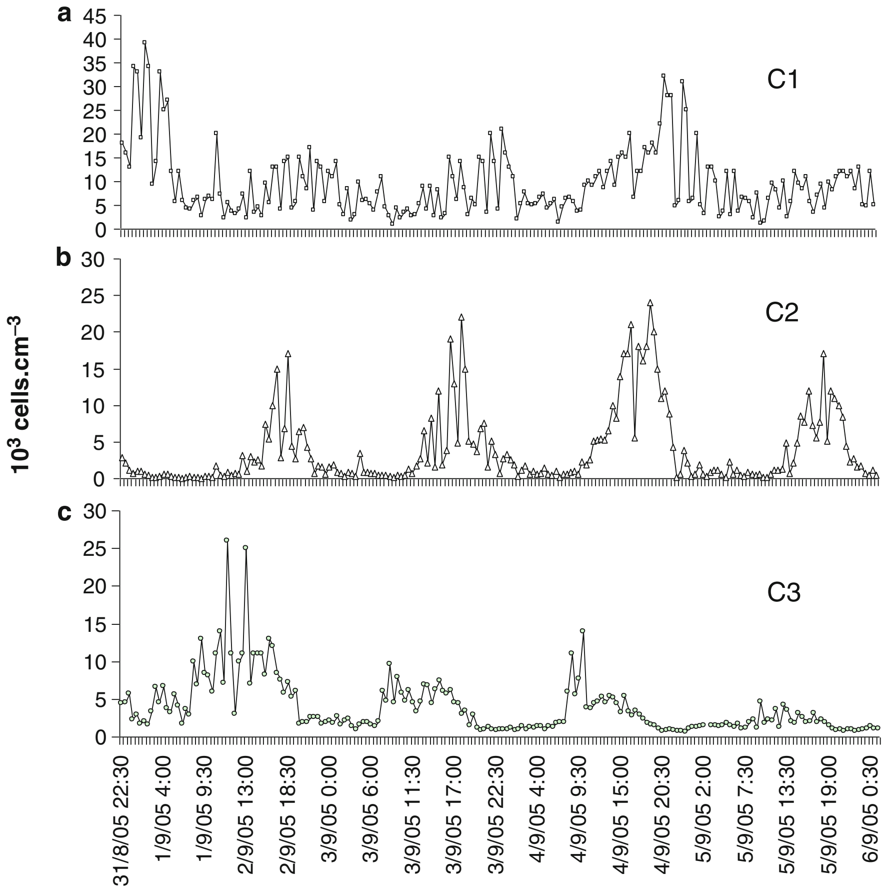

Fig. 1 Abundance dynamics of three selected clusters monitored in the Bay of Marseille during summer 2005. (**a**) Abundance dynamics of cluster C1. (**b**) Abundance dynamics of cluster C2. (**c**) Abundance dynamics of cluster C3. See Thyssen et al. (2008b) for a more detailed description. All three clusters exhibited regular daily cycles, but abundance maximal values occurred at different periods of the day, suggesting that the observed cell groups were controlled by different factors

distributions and hydrological features of the related water masses.

4 Discussion and Conclusion

The high frequency analysis of phytoplankton assemblages by using automated submersible flow cytometry opens the way to a better understanding of the phytoplankton ecosystem functioning.

As an example, the high frequency was critical to interpret the abundance variation of clusters C2 and C5. The large amplitude of the diel abundance change cannot be explained by the cell cycle itself, but it becomes consistent if assigned to diel vertical migration, which still needs to be further substantiated.

One major conclusion from the high frequency in situ time series is that the observed clusters behaved like independent entities and as such can be considered as response functional groups. Further studies are needed to explore the response of these clusters to other environmental changes and open the way to the identification of bio-indicators of specific environmental changes. Combining the high frequency in situ study of these functional groups with species identification will generate a powerful tool to better understand the functioning of phytoplankton assemblages.

In the spatial investigation of the phytoplankton distribution, the high frequency analysis was critical to achieve a spatial resolution better than 3 km and collect information relevant to the sub-meso scale. Significant relationships could thus be established between the different cluster distributions and the hydrological characteristics of the sampled water masses. It was also critical to extract the short-term variability from the data and determine the cell cycle of the different clusters (Fig. 2) by processing autocorrelation calculations as operated on the time series data. Spatial studies necessarily involve space and time variations that cannot usually be sorted out. The high frequency analysis helped in solving this difficulty.

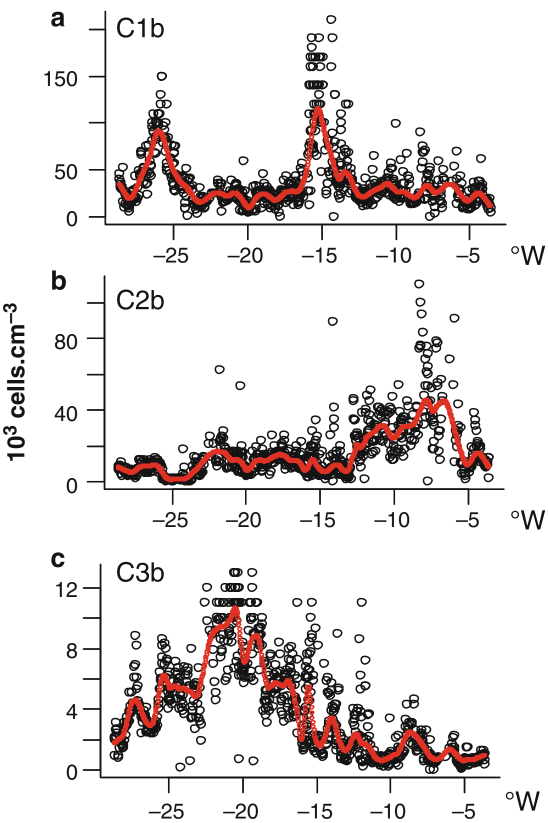

Fig. 2 Abundance dynamics of three selected clusters observed in the Northeast Atlantic Ocean in April 2007. Black dots correspond to the analyzed samples, the red line represents the smoothed data when using a LOESS method. (**a**) Abundance dynamics of cluster C1. (**b**) Abundance dynamics of cluster C2. (**c**) Abundance dynamics of cluster C3. See Thyssen et al. (2008c) for a more detailed description. All three clusters exhibited regular daily cycles, but strong abundance variations can be related to external factors, such as hydrodynamism and nutrient availability

Automated in situ phytoplankton monitoring with submersible flow cytometers appears particularly appropriate for long-term time series or impact studies for developers, coastal management, and surely aquaculture. We must point out that, to our knowledge, submersible flow cytometry is the only technique that can provide biological information in an automated way, in contrast with other automated sensors that are essentially relevant to physics or chemistry. This technology has an appealing potential for global oceanic observing systems, to which at last it could provide real biological information either by being deployed at fixed points or on board of opportunity ships. It also could prove critical with its field data to calibrate phytoplankton remote sensing.

Ongoing developments include (1) image in flow (already commercially available) to reconcile flow cytometry and species identification. A library of cell pictures and related flow cytometric signatures is under construction (cytobuoy.com) for automated identification; (2) extension of the instrument capacities to the automated analysis of heterotrophic organisms, in particular those of the prokaryotic world; and (3) the automation of the data treatment to ensure the real-time transfer of information directly usable by stakeholders as well as warnings when anomalies are detected by the instrument.

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Deep Sea Net: An Affordable and Expandable Solution for Deep Sea Sensor Networks

Pierre Valdy

Abstract Deep Sea Net is a new concept of a deep-sea sensor network built from fiberoptic microcables and battery-operated IP access nodes. Deep Sea Net fiberoptic transport segments are deployed directly from a standard ROV, avoiding the use of a costly cable layer ship. Deployment skid is abandoned on user sites ready to connect local sensors and next transport segments. Deep Sea Net is a low consumption network that can be activated by user (or sensor) demand. Lithium batteries and fine power management will give 10 years of autonomy with ¼ h operation per day. The innovative approach of Deep Sea Net will permit to build, extend, and maintain future sensor networks at very competitive prices.

1 Network Concept

Deep Sea Net is a new concept of a deep-sea sensor network to be installed beyond 1,500 m depth for servicing smart IP sensors (Fig. 1). Deep Sea Net is built from fiberoptic microcables (FOMC) and battery-operated switches (IP nodes) for long distance transport and local IP sensor interconnection. Deep Sea Net is deployed, extended, and maintained with an all-sensor suite with a standard ROV.

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2 Fiberoptic Microcable

2.1 FOMC Justification

In the deep sea, the risk of cable damage because of anchoring, fishing activity, or fish bites is very limited. When laid beyond 1,500 m depth, most of telecommunications cables have no external armor above high voltage insulation and are not buried. External protection is not critical at such depths. Telecommunications cables are laid from the surface. They need to be dense and strong to support laying forces induced by current, heave, and depth. If the cable is laid directly above the bottom, density and internal reinforcement are not critical. Telecommunications networks never sleep. Repeaters need to be powered all the time. If the network is operated only part of time, repeaters may be powered with batteries, and copper and high voltage insulation is not necessary.

In light of the above considerations, a 2-mm light cable is conceivable. A 50-km cable spool may interconnect user sites and match with repeater spanning. Such a small and light spool is compatible with standard ROV handling capacities.

2.2 FOMC Technical Issues

Two fibers are the general standard for long-distance IP transport. However, for simplifying Deep Sea Net connectors, bi-directional transmission with a single fiber is preferred. A loose metal tube structure for fiber protection seems inevitable (no water ingress, no hydrogen effect, no fiber stress). This solution will allow maintaining, in the long term, attenuation performances on

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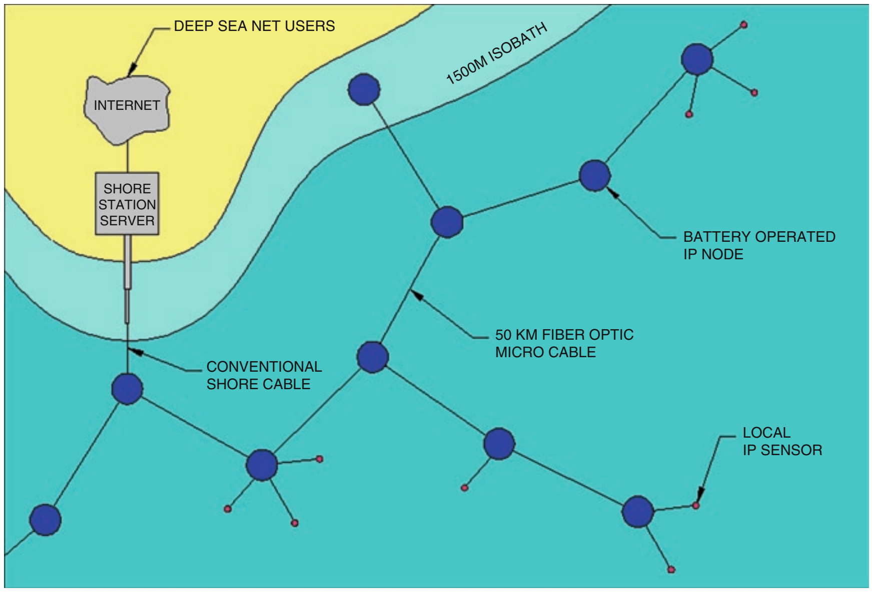
Fig. 1 Deep Sea Net concept



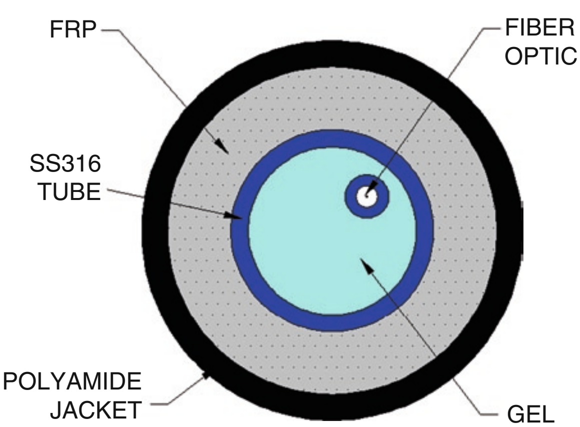


Fig. 2 Fiberoptic Microcable

long distance transport segments. For corrosion aspects, 316 stainless steel is preferred to 304, but Inconel or Hastelloy may be a better choice in the future. The choice of the final cable structure has been very difficult.

Several longitudinal reinforcements have been evaluated (second metal tube plus polyamide jacket or high performance liquid crystal polymer jacket). Finally, glass fiber in epoxy matrix (FRP) plus a tight polyamide jacket (for improving the bamboo structure cohesion and friction factor) has been selected (Fig. 2) for a light, strong, stiff, elastic, and low-torque cable.

FOMC comparison with a standard telecommunications cable: The following chart (Fig. 3) compares the general characteristics of Deep Sea Net FOMC with a low-weight telecommunications cable.

3 IP node

3.1 Electronics

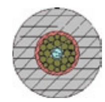
Electronic made by IFOTEC integrates elements into one single board (Fig. 4):

- 100 Mbps ADM 6999 Ethernet switch
- 3 × 100FX ports for main and secondary transport (bi-directional transmission: 1,530/1,570 nm)
- 3 × 100TX ports for local applications (with 24 V power) over Ethernet for power or bi-directional wake up (over the two Ethernet twisted pairs)
- Node wake up circuit
- Programmable timer for adjustable time-out
- Micro-controller for node and port management

3.2 Batteries

The node is powered with lithium batteries (7.5 kg/5,594 Wh). The consumption of the node is 0.5 mW in sleeping mode and 4 W in operation. This will give 10 years of autonomy with ¼ h communication per day.

Fig. 3 Comparison between standard telecommunications cable and Deep Sea Net microcable

Cable	LW Telecom	FOMC
Construction	12 FO in SST* steel armor copper tube PE* jacket	1 FO in SST* FRP* armor PA* jacket
Application	Light Weight cable for repeated telecommunications beyond 1500m	Deep Sea Net
Diameter (mm)	17	2.2
Breaking load (daN)	7000	200
Weight (kg / 50 km)	25 000	425
Volume (liters / 50 km)	14 500	242
Cost factor	1	1/6
* SST: Stainless Steel Tube FRP: Fiber Reinforced Polymer PE: Polyethylene PA: Polyamide		

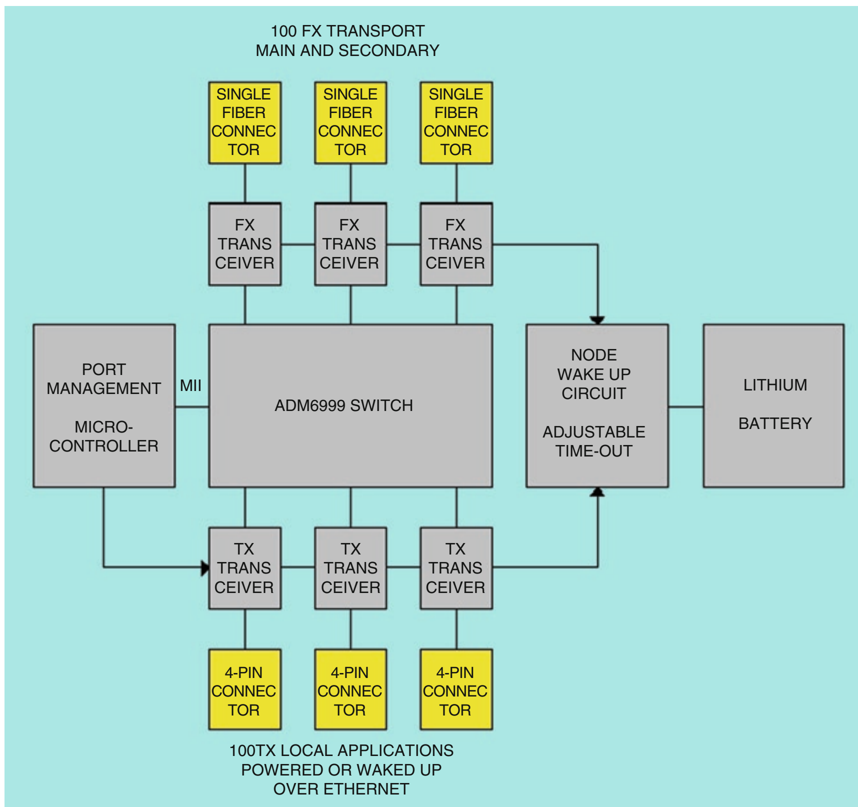


Fig. 4 IP node electronics

3.3 Node Integration

The electronic board, the six battery packs, and the six wet mateable connectors are integrated in a single flat titanium canister (Fig. 5) resisting up to 6,000 m depth pressure. The assembly is very compact, and the weight in water is compatible with ROV handling capacity (40 daN).

3.4 Wet Mateable Connectors

Electrical and optical connectors will be issued from a cooperation between Seacon Europe and Ifremer. The final objective is to develop, in the same small format, low-cost and reliable wet mateable connectors:

– Electrical connectors will integrate the Seacon Sea-Mate inserts (4 x gold plated contacts in nitrile

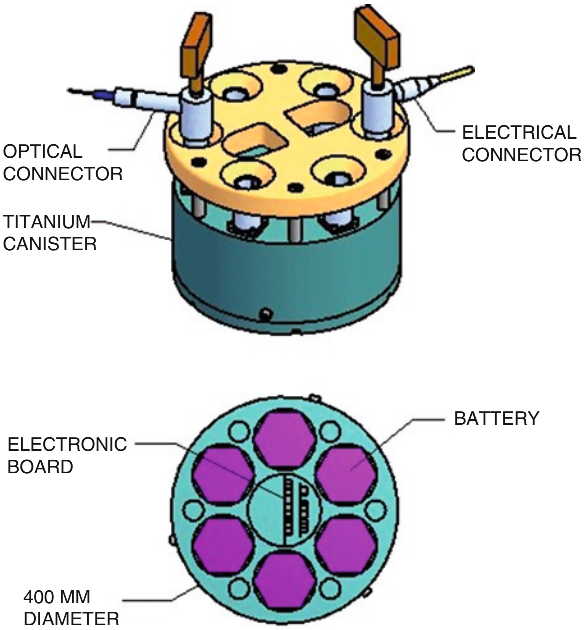


Fig. 5 IP node integration

housings). Such connectors are not insulated when not connected. Node management will allow to switch off data and power.
– Optical connectors will use expanded beam technology. Specific inserts will integrate fiber optic collimators protected behind high-pressure windows. Such connectors will have more losses than conventional ones. The Deep Sea Net optical budget is sized in correspondence.

3.5 IP Node Operation and Management

The node wake-up circuit and node timer are independent from the micro-controller. The wake-up circuit detects the arrival of the optical carrier of transport segments or the electric current of local applications. It starts the timer and powers up the switch and the attached micro-controller. When powered up, the micro-controller can respond to SNMP requests for node management.

The network supervisor may have access to each node via individual web pages with the following displays (Fig. 6):

– Connection status
– Node status (temperature/ voltage/ time-out setting: 2, 4, 8, or 16 min/ wake-up source: Fx or Tx)
– Transport port status (carrier sense)
– Port power and data status (need to be Off when no connector and On when application is powered up or waked up)
– Port wake-up event status (needs to be disabled if there are too many wake-up events)

4 Deep Sea Net Operating Modes

4.1 Sleeping Mode

All IP nodes are sleeping, ready to be waked in Standard mode, Emergency mode, or Alarm mode.

4.2 Standard Mode

Every day at a fixed time, the shore station server wakes the network during an adjustable period between 1 and 16 min. During this period, data are automatically downloaded by ftp from all applications to the shore station server to be available to the user when the network is sleeping.

4.3 Emergency Mode

The network may be waked at any moment by the user who wants to connect with its application (via http, ftp, or ssh).

4.4 Alarm Mode

The network can be waked at any moment by the application if an exceptional event is recorded. Then an e-mail is sent to the user.

Fig. 6 IP node Web page

Connection status	
Communication	1/0
Update rate	10s
Last update	0.105s
Update in progress	1/0
Last connect	11/11/06 - 09:00:38
Last disconnect	11/11/06 - 08:02:12
Update	

Node Status	
Serial number	51
Temperature	29°C
Battery voltage	2214V
Elapsed time	Im05s
Time-out setting	2/4/8/16
Wake up source (FX or TX port number)	TX2
Default configuration	On/Off

Transport port status	
FX port number	Cariner sense
1	1/10
2	1/10
3	1/10

Application port status			
TX port number	Power	Data	Wake up event
1	On/Off	On/Off	Enable / Disable
2	On/Off	On/Off	Enable / Disable
3	On/Off	On/Off	Enable / Disable

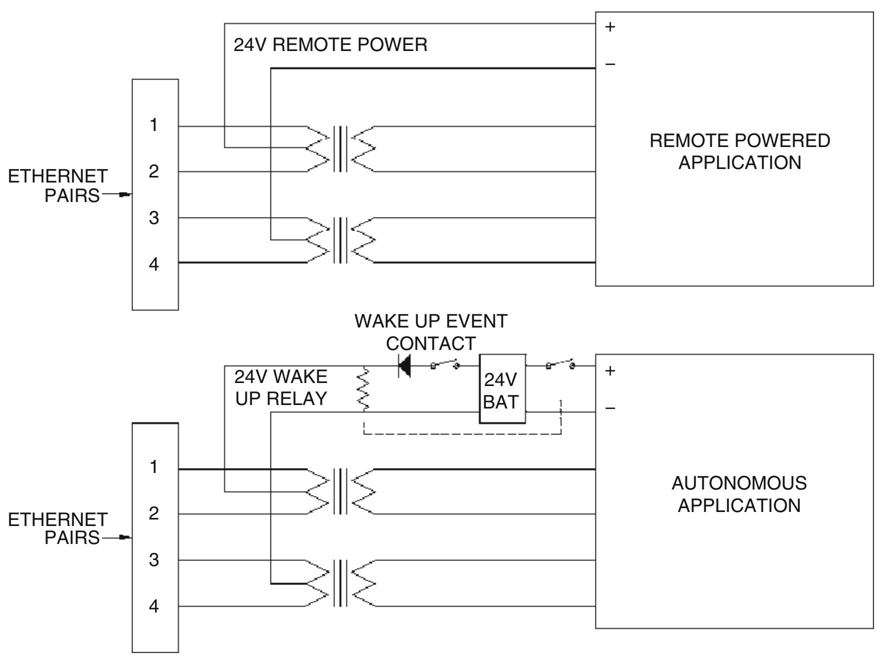


Fig. 7 Application interfaces

5 Applications and Interfaces

There are two types of applications (Fig. 7) that may be interfaced on Deep Sea Net:

5.1 *Remote Powered Applications*

These applications may be powered on demand by a 24-V phantom current through Ethernet pairs when the

IP node is awake. These applications must have low power consumption (1 W maximum). Being not autonomous, they cannot record parameters out of daily operating periods. However, it may be interesting to equip each node systematically with such applications (e.g., low power multi-parameter sensor, CTD sensor, current meter, etc.).

5.2 Autonomous Applications

Some applications need to monitor their environment regularly. They are powered by their own battery, and the same Ethernet pairs can wake them by the intermediate of a 24-V relay for data file transfer. Other applications, such as seismometers or tsunami sensors, need to send an alarm when an exceptional event is detected. In such cases, these applications can wake up the node through the same Ethernet pairs.

6 Deployment Skid

Deployment skid (Fig. 8) integrates the following components:

Rotating spool: A horizontal rotating spool is supported with roll bearings and has up to 55 km FOMC capacity. The IP node is fixed on a top flange by the intermediate of a bayonet mounting. A simple rotating spool has been preferred over a fixed spool with internal unwinding (torpedo-pack like) or external unwinding (fishing-reel like). Despite the spool's high inertia, cable loads will be limited in comparison with cable characteristics (2 daN during laying and 15 daN during maximum acceleration for 200 daN cable breaking load).

Constant tension roller: A spring-actuated roller prevents the FOMC from being overloaded during acceleration or to be slack during deceleration (0.5-m cable stroke).

Automatic brake band: At the end of the stroke, to prevent any slack, the roller actuates a brake band to brake the spool.

Trench plow: For better FOMC protection, a heavy knife is installed at the rear part of the deployment skid to bury the microcable inside the sediment (trench section: 30 mm wide/200 mm deep). A flexible and heavy

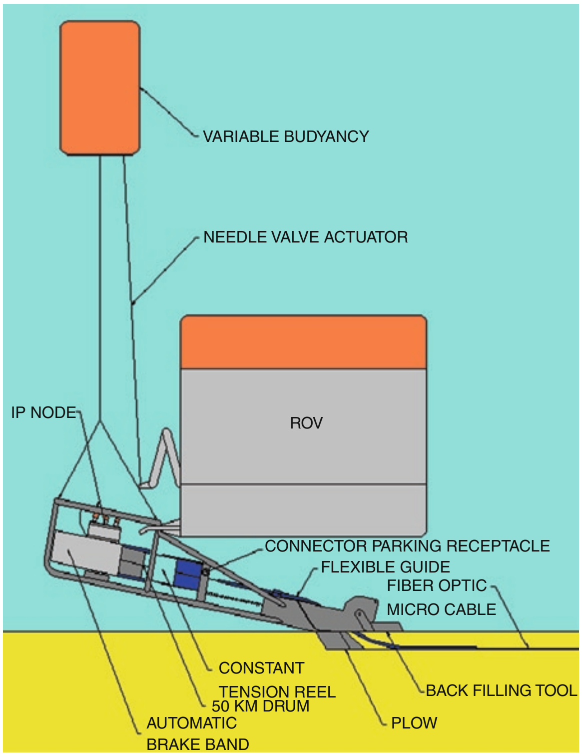


Fig. 8 Deployment skid

hose is in charge of guiding the microcable smoothly inside the trench.

Deployment: The skid is sent to the bottom under buoyancy in equilibrium between the buoyancy force and the reaction on the plow (120 kg). Due to the cable variable weight in water, the ROV has to compensate for the difference (230 kg). The ROV laying speed (1 knot) will allow installing a 50-km segment per day. The cable laying route, under full ROV control, will clear all significant obstacles on the bottom, and the skid will be driven right near to the user spot (which is in comparison difficult to achieve with conventional junction boxes laid from the surface).

7 Deep Sea Net Demonstration

A Deep Sea Net demonstrator will be tested on the Antares junction box (Fig. 9). Antares is a neutrino telescope installed in Mediterranean Sea at 2,400 m depth, offshore of Porquerolles Island. Antares' junction box is

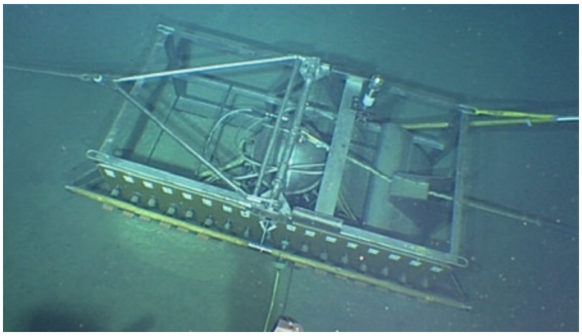


Fig. 9 Antares junction box

equipped with 16 electro-optic connectors and is connected to La Seyne sur Mer shore station with a 42-km cable. Deep Sea Net will use one of these connectors.

One ultra-low power seismometer, developed by Geosciences Azur from the Observatoire de Villefranche, will be connected to the demonstrator. The Deep Sea Net demonstrator is funded by the French National Research Agency (ANR) through the MOGLI project, which is a cooperation between Géosciences Azur and Ifremer. The final objective will be to propose an affordable solution for a seismometer network in the Ligurian Sea (offshore of the city of Nice). This site is one of the key sites proposed by the European ESONET project.

8 Conclusion

The author, as adviser of physician community for Antares marine architecture and as responsible for the Antares sea operations, knows the costs and difficulties of such installations. Cable layer ships are expensive and difficult to schedule, junction boxes are heavy and risky to deploy, and oil-filled connectors are expensive and may experience failures. When problems occur, maintenance of such a network is very difficult. Telecommunications cables, when used for shore to shore communications, are very reliable. When used for interconnecting deep-sea junction boxes, the reliability is certainly more difficult to predict.

Even with the hypothesis of 20 years' life expectancy, annual ROV campaigns are still necessary for sensor suite maintenance and implementation. Scientists who want to develop smart sensors should be interested by this alternative solution, which can be installed, maintained, and extended at affordable prices using their own naval equipment. Deep Sea Net may be operated from conventional shore cable or an offshore telecommunications buoy.

It may also be operated as an extension of an existing scientific network. Applications may be oceanographic or for homeland security. After the demonstration, the technology, which is protected by Ifremer patents, will be transferred to the industry for the benefit of deep-sea scientific applications.

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Physical, Chemical, Biological and Biogeochemical Functioning

Distribution and Long-term variation of Turbidity in Tokyo Bay

Hisayuki Arakawa, Shizuka Mizuno, Miho Narita, and Mitsuhiro Ishii

Abstract The seawater of Tokyo Bay is very highly turbid due to anthropogenic activities. We examined the distribution, properties, and long-term variations of turbidity in Tokyo Bay.

A transmissometer of three wavelengths was used in the investigation of the distribution and the properties of turbidity.

The long-term variation of turbidity was analyzed based on a Secchi disc depth data set collected from 1948 to 2006 by the Chiba Prefectural Fisheries Research Center.

1. Beam attenuation coefficient, Secchi disc depth, color of water by the Forel-Ule scale, suspended solid (SS), and chlorophyll *a* (Chl. *a*) concentration at the innermost part of Tokyo Bay (off Urayasu) in 2005–2006 indicated 7.5 m^{-1}, 1.3 m, No. 18, 9.0 mg L^{-1}, and 59.1 μg L^{-1} in summer, and 2.3 m^{-1}, 2.5 m, No. 16, 5.1 mg L^{-1}, and 27.1 μg L^{-1} in winter, respectively.
2. Contributions to the turbidity caused by dissolved organic matter, particulate organic matter, and particulate inorganic matter in summer reached values of 5%, 85%, and 9% at the innermost part of the bay (off Kawasaki), and 8%, 50%, and 42% at the mouth of the bay (off Kyonan), respectively.
3. The Secchi disc depth of Tokyo Bay decreased rapidly in the 1960s, and indicated the lowest value in 1964. Since then, the Secchi disc depths in summer have not shown marked changes. But the values in winter have increased since the end of the 1960s. The present values in winter indicated measurements close to those observed in the 1950s.
4. If as a future aim of water quality in Tokyo Bay a Secchi disc depth of >1.5 m, that is, a state under which red tide conditions are not generated, the concentrations of Chl. *a* and SS in summer must be decreased by 34% and 21.5% from the present value, respectively.

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1 Introduction

The seawater of Tokyo Bay is very highly turbid due to anthropogenic activities. It is reported that the turbidity of Tokyo Bay began to increase from the first half of the 1950s; it became the maximum at the end of the rapid economic growth of the first half of the 1970s in Japan. Then, turbidity was considered to have decreased little by little because of the implementation of various waste water regulations (Tsubota et al. 1975; Nomura 1995). However, the state of very high turbidity is continuing now. The purpose of our study is to clarify the distribution, properties, and long term variation of turbidity in Tokyo Bay.

2 Methods

Beam attenuation coefficient (λ; 488 nm m^{-1}), Secchi disc depth (m), color of water by the Forel-Ule scale, suspended solids (SS; mg L^{-1}), volatile suspended solids

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(VSS; mg L^{-1}), relative irradiance (% m^{-1}), and chlorophyll *a* concentration (μg L^{-1}) as turbidity indexes of seawater were observed every month from July 2005 to July 2006. Observation stations are shown in Fig. 1. A transmissometer of three wavelengths (Narita et al. 2006, Narita 2008) was used in the investigation of the turbidity due to dissolved organic matter, particulate organic matter, and particulate inorganic matter. The observed wavelengths were 380, 488, and 680 nm. After seawater at each station filtered by GF/F (Whatman Co., Ltd.), the turbidity due to the dissolved organic matter (C'_{DOM}) of the filtered seawater was measured by beam transmissometer. The long-term variation of turbidity was analyzed based on a Secchi disc depth data set collected from 1948 to 2006 by Chiba Prefectural Fisheries Research Center (Stations A, B, and C in Fig. 1) (Ishii et al. 2008). For data processing, 3-year running means were performed.

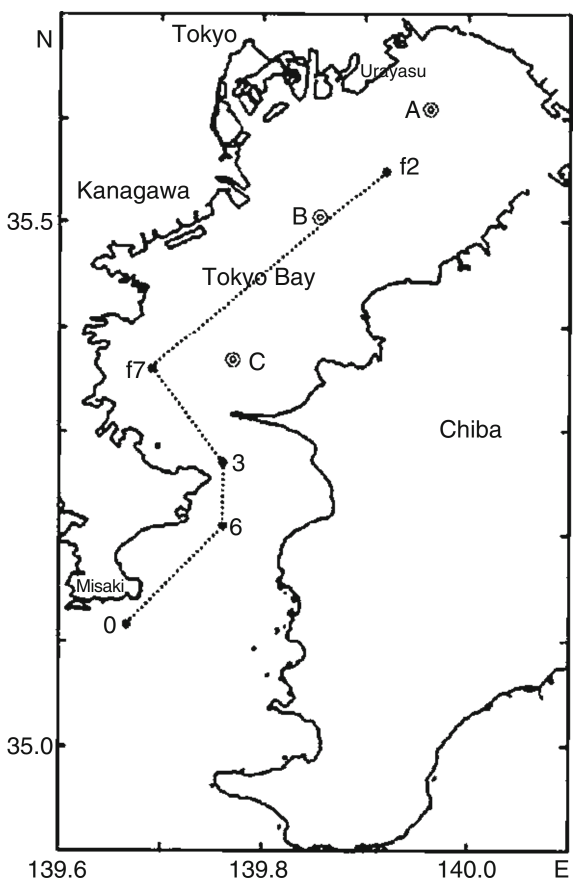

Fig. 1 Observation stations in Tokyo Bay. The field observation was conducted at Stations f2, f7, 3, 6, and O. The long-term data of Secchi disc depth at Stations A, B, and C was analyzed. A vertical section of turbidity along the line from Station f2 to Station O is shown in Fig. 2.

3 Results

The values of turbidity indexes at the innermost part of Tokyo Bay (Station A) in 2005–2006 denoted in Table 1. Beam attenuation coefficient, Secchi disc depth, color of water, suspended solids, and chlorophyll a concentration in summer (mean values from June to September) are 7.5 m^{-1}, 1.3 m, the Ule scale no. 18, 9.0 mg L^{-1}, and 59.1 μg L^{-1}, respectively, indicated very high values. These values of the seawater show a condition of red tide (Nomura 2008). However, values in winter (mean values from December to March) decreased remarkably. However, these values are higher than the value of the other eutrophic sea areas in Japan.

Vertical distributions of turbidity of dissolved organic matter (DOM), particulate organic matter (POM), and particulate inorganic matter (PIM) for July are shown in Fig. 2. The beam attenuation coefficient (C') is high for the sea surface from innermost part of Tokyo Bay to the central part of the bay, and it decreases at the mouth of the bay. Moreover, in the central part, high turbidity is seen also near the seabed. Contributions to the turbidity caused by dissolved organic matter, particulate organic matter, and particulate inorganic matter in summer were 9.6%, 70.1%, and 20.3% at the innermost part of the bay (off Urayasu; Sta. f2), and 15.6%, 45.1%, and 39.3% at the mouth of the bay (off Misaki; Sta. O), respectively. The high turbidity of the particulate organic matter near the sea surface of the innermost part of the bay is considered to be due to the high concentration of phytoplankton cells. (Narita et al. 2006).

Table 1 Values of turbidity indexes at the innermost part (Station A; off Urayasu) of Tokyo Bay in summer 2005 and winter 2006

Index	Summer	Winter
Beam attenuation coefficient (m^{-1})	7.5	2.3
Secchi disc depth (m)	1.3	2.5
Color of water (no.)	18	16
Suspended solid (mg.L^{-1})	9	5.1
Chlorophyll a (μg.L^{-1})	59.1	27.1

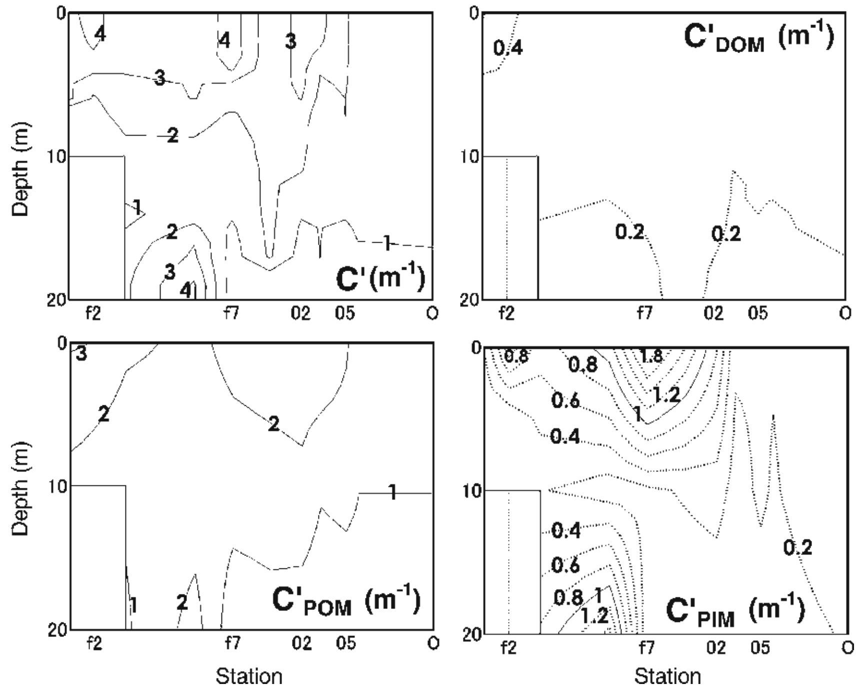

Fig. 2 Vertical distributions of turbidity of DOM (dissolved organic matter), POM (particulate organic matter), and PIM (particulate inorganic matter) in July. The index of turbidity is beam attenuation coefficient. The unit is m^{-1}

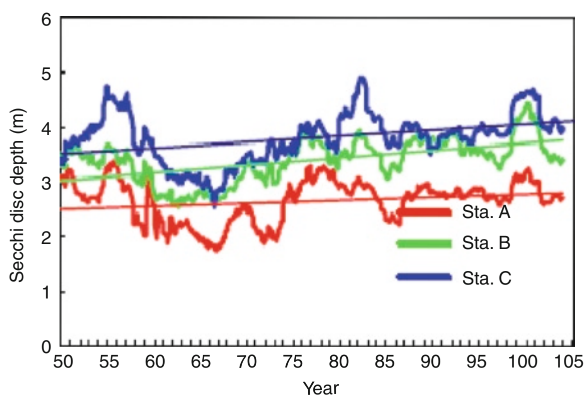

Fig. 3 Long-term trend of Secchi disc depth in Tokyo Bay. Sta. A, B, and C are inner part, middle part, mouth part of Tokyo bay

Moreover, the high turbidity seen near the surface and bottom of the central part of the bay originates not only from the particulate organic matter, but also from the particulate inorganic matter.

Long-term variation of Secchi disc depth is indicated in Fig. 3. The Secchi disc depth decreased rapidly in the 1960s, and indicated the lowest value at Station A in 1966–'67. The reduction was especially remarkable at the innermost part of the bay. Subsequently, the transparency of water has increased little by little.

The Secchi disc depth at Station A in summer, however, has not shown marked change from the 1960s to the present. However, the values for winter have increased gradually since the end of the 1960s (Fig. 4).

The past turbidity values were computed from the relationships between Secchi disc depth and the turbidity indexes (Table 2).

Secchi disc depth, color of water, beam attenuation coefficient, relative irradiance, suspended solids (SS), volatile suspended solids (VSS), and chlorophyll a concentration in 1954 were 2.6 m, no. 15 on the Ule scale, 3.0 m^{-1}, 40.3% m^{-1}, 4.3 mg L^{-1}, 2.6 mg L^{-1}, and 20.5 μg L^{-1} in summer, and 2.8 m, no. 14 on the Ule scale, 2.7 m^{-1}, 44.1% m^{-1}, 3.9 mg L^{-1}, 2.3 mg L^{-1}, and

Table 2 The past turbidity values computed from data set of Secchi disc depth

	Summer			Winter		
StaA surface	1954	1964	2004	1954	1964	2004
Secchi disc depth (m)	2.6	1.2	1.4	2.8	3.3	3.4
Color of water (No.)	15	19	18	14	13	12
Beam attenuation coef. (m^{-1})	3.0	6.9	5.7	2.7	2.3	2.2
Relative irradiance (% m^{-1})	40.3	13.5	18.8	44.1	50.0	51.8
SS (mg L^{-1})	4.3	8.9	7.5	3.9	3.4	3.3
VSS (mg L^{-1})	2.6	5.8	4.8	2.3	2.0	1.8
Chl.a (μg L^{-1})	20.5	50.3	41.3	18.1	14.8	13.7

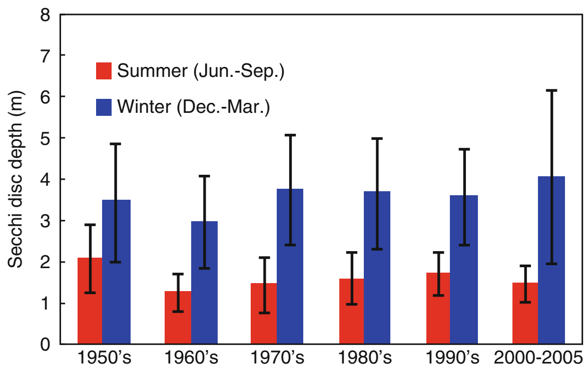

Fig. 4 Changes of Secchi disc depth in summer and winter at Station A. Vertical bars indicate standard deviation

18.1 μg L^{-1} in winter, respectively. Based on these data, we can understand that Tokyo Bay was a sea area of high turbidity even in 1954.

Compared with 50 years ago, the values of turbidity index values for this summer (2004) have increased and approximately doubled. However, in winter, they decreased by 10–20%.

4 Discussion

From the viewpoint of all turbidity indexes, Tokyo Bay in the current summer is in a state of red tide. It is reported that the number of days that the state of red tide occurs is decreasing (Nomura 1998). However, the turbidity due to particulate organic matter occurs on the surface of innermost part of the bay. In the long period of the summer, it is considered that it is in a state of red tide constantly.

The Secchi disc depth of Tokyo Bay decreased remarkably in the 1960s. This is considered to be due to the influence of the higher economic activity of Japan. Subsequently, the Secchi disc depth has increased gradually. This is considered to be an effect of the drainage regulation that started after the 1970s.

According to Ishii et al., the nutrients (NH_4–N, PO_4–P) of the sea water in Tokyo Bay are decreasing each year. Especially, they are markedly reduced from the surface waters in summer. However, the Secchi disc depth of the current summer has hardly changed from values in the 1960s. The cause of this lack of change in the Secchi disc depth needs to be clarified in the near future.

If the future aim for water quality in Tokyo Bay is a Secchi disc depth of >1.5 m, a state under which red tide conditions are not generated, the concentrations of Chl. *a* and SS in summer have to be decreased by 34% and 21.5% from the present value, respectively.

5 Conclusions

The turbidity of Tokyo Bay is very high on the surface of innermost part of the bay. About 70% of this value in summer is generated by particulate organic matter.

The Secchi disc depth of Tokyo Bay decreased rapidly from the 1950s to the 1960s; subsequently, it has increased little by little. However, the value in summer has not markedly changed compared with the lowest time (the 1960s). On the other hand, the value for winter has increased every year, and the present value is higher than 50 years ago.

Furthermore, the values of the turbidity indexes for 50 years of Tokyo Bay were estimated. The values of the turbidity indexes in summer show that Tokyo Bay has always been in a state of red tide over the last 50 years. It is necessary to examine how to reduce the turbidity in summer.

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Evaluation of Chemical Contamination in the Western Mediterranean Using Mussel Transplants

Bruno Andral, Jean-François Cadiou, François Galgani, and Corinne Tomasino

Abstract The MYTILOS project aimed to draw up a baseline report on chemical contamination of coastal seas on a Western Mediterranean scale. On the basis of the methodology developed on the French Mediterranean coast by Ifremer and the Agence de l'Eau Rhône Méditerranée & Corse (Rhône, Mediterranean & Corsica water board) since 1996 with the RINBIO monitoring program, data were collected between 2004 and 2006 along the northern and southern continental coasts and around the Balearic Islands, Sicily, Sardinia, and Corsica.

MYTILOS was supported by the INTERREG III B/MEDOC program, steered by Ifremer, and backed by Toulon Var Technologies, in cooperation with research institutions from concerned countries: ICRAM (Italy), IEO (Spain), PSTS (Sicily), IMEDEA (Balearic Islands), CSIC, Agencia Catalana de l'Aigua (Spain), INSTM (Tunisia), ISMAL (Algeria), INRH and University of Agadir (Morocco). MYTILOS formed part of the MEDICIS program and was also backed by the PNUE/PAM – MEDPOL and Rhône Méditerranée & Corse water board. Three cruises (2004, 2005, 2006), for a total of 120 days at sea, were carried out to install mussel bags for 3 months at 123 stations and to recover them.

The results obtained for 40 chemical compounds in the western Mediterranean show similarities with observations previously made on the French coast in terms of the highest measured levels and background level. The most highly impacted areas are mainly urban and industrial centers and the outlets of major rivers. A far higher dilution is seen for organic compounds than for heavy metals when going from the coast towards open sea. Metal levels measured in the open sea were often found to be similar to those in natural shellfish populations living along the coast.

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1 Introduction

Most environmental quality monitoring programs now include the use of biological indicators. This is based on the assumption that levels of trace contaminants accumulated in biological tissues represent the time and space integrated value of these contaminants in the surrounding waters. High concentrations of many contaminants in bio-indicators render the measurement of contaminant concentrations technically simpler. Variations of contaminant levels in tissues may reflect the variations in water and suspended particulate matter contaminant concentrations (Goldberg 1975; Cossa 1989; Claisse 1989), although biological variability can also affect tissue concentrations.

The MYTILOS project aimed to draw up a baseline report on chemical contamination of coastal seas on a Western Mediterranean scale (continental coasts of France, Spain and Italy, the Balearic Islands, Sicily, Sardinia, Corsica, and Maghreb). MYTILOS was supported by the INTERREG III B/MEDOC program, steered by Ifremer, and backed by Toulon Var Technologies, in cooperation with ICRAM (Italy), IEO (Spain), PSTS (Sicily), IMEDEA (Balearic Islands), CSIC, Agencia Catalana de l'Aigua (Spain), INSTM (Tunisia), ISMAL (Algeria), INRH and University of Agadir (Morocco). MYTILOS formed part of the MEDICIS program and

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was also backed by the PNUE/PAM – MEDPOL and Rhône Méditerranée & Corse water board.

MEDICIS (www.ifremer.fr/medicis) is a research program launched by IFREMER in 2003 that aims to improve knowledge on the sources, state, and fate of chemical contamination in the Western Mediterranean; and to develop tools for policy makers and maritime and coastal environment stakeholders to improve the management and prevention of chemical contamination in marine waters, ecosystems, and seafood. Through this federative approach to chemical contamination, the MEDICIS program also aims to improve the knowledge of the main processes involved in marine environment dynamics. Understanding contaminant fate requires a systematic and integrated approach to the marine environment; MEDICIS therefore includes the design and development of multiscale models relating to water mass circulation, sediment transport, exchanges at interfaces, food webs, and in situ measuring methods and instruments.

2 Materials and Methods

In the Mediterranean Sea, the species *Mytilus galloprovincialis* is widespread, but in some locations natural populations are rare or absent. The transplantation method has compensated for this scarcity and allows controlling the source, age, and stage of sexual maturity of the samples. However, implementing it on a large geographic scale introduces factors, such as variations in physiochemical characteristics and food availability in the immersion areas. Although measured concentrations in the tissue are a function of bioavailable pollutant levels in water, the bioaccumulation factor depends on mussel growth in relation to the primary food production, or trophic capacity, of the environment (Phillips 1976; Lobel and Wright 1982; Borchardt 1983). Comparison of raw data on tissue concentration between sectors of different trophic potential may be misleading. A biometric parameter representing growth must then be used to correct initial data and to produce reliable comparisons at a large spatial scale (Fischer 1984; Soto et al. 1995).

The Condition Index (CI) is an efficient indicator of physiological state and growth resulting from the environmental effect. The results acquired by the RINBIO monitoring network (Andral et al. 2001; Andral et al. 2004) since 1996 show robust condition index/contaminant concentration correlations in all geographical zones for certain contaminants. It is therefore possible to determine a correction model for these contaminant families and obtain a concentration, notwithstanding the environmental effect, which is representative of the bioavailable contaminant concentration in the environment. At a large spatial scale, this model enables the standard comparison of data notwithstanding the physiochemical and trophic heterogeneity of the target zones.

2.1 Transplantation

Because the decontamination kinetic is usually slower than that of contamination (Fowler 1982; Wang et al. 1996), the samples of *Mytilus galloprovincialis* came from a firm in Languedoc-Roussillon that harvests contamination-free mussels from the open sea. The batch was made up of adult mussels 18–24 months old, measuring about 50 mm, sorted twice according to the height of the shell through 19-mm mesh. The 3-kg samples were stored in conchylicultural pouches mounted on PVC tubing and re-immersed for 10 days so they could re-cluster prior to transplantation. Subsurface anchorages, comprising a mussel bag attached to a 30 kg weight, were maintained in open water with an 11-L float. The stations were positioned at various depths and distances from the coast. Globally speaking, bathymetry was 20–50 m in depth according to coastal configuration, and the bags were attached at depths of 8–10 m. The aim was to install each station in an equivalent continental input dilution volume to avoid being under the direct influence of one pollution source and to seek good and coherent station representativeness.

Mytilos I (March 20 mars–April 15, 2004) sampled a coastline stretching from Cartagena (Spain) to Orbelleto (Italy). Mytilos II (March 21–April 19, 2005) covered southern Italy and part of southern Spain, with sampling along the coast of the Balearic Islands, Sardinia, and Sicily. Mytilos III (May 16–June 3, 2006) covered Corsica, the southernmost coasts of Spain, and the Maghreb coasts (Morocco, Algeria, Tunisia).

2.2 Recovery

Recovery of the samples in the northern part of Italy and Spain took place from July 2 to July 21, 2004, using a 12-m fast and easy-to-handle catamaran. Taking into account the geographical scope of the two other campaigns, retrieval in the south of Italy and Spain, in the Balearic islands and Sicilia was carried out using the oceanographic vessel THETYS II from June 17 to June 26, 2005. In 2006, the oceanographic vessel L'EUROPE recovered stations located on the Maghreb coast from August 10 to August 27. These vessels provided vital logistical support (diving equipment, samples processing, and preservation), including a zodiac equipped with detection instruments (sweep sonar, vertical echo sounder) for station retrieval operations.

On board the ship, the mussels were separated and rinsed in seawater. Mortality and height of the shell were recorded. At every station, the samples were preprocessed according to standardized procedures. The mussels were opened raw, and the flesh was scraped out of the shell with a stainless steel scalpel. Shells were dried at 60°C in the oven for 48 h and then weighed. Flesh was weighed after freeze-drying. The ratio of dry flesh weight to dry shell weight (FW/SW) was used to determine a condition index (CI) for each sample.

2.3 Analysis

The following techniques were used to obtain chemical contaminant levels in samples: for lead, cadmium, and nickel: graphite oven atomic adsorption spectrometry; for mercury: atomic fluorescence. Dichlorodiphenyltrichlorethan; for DDT, DDD, DDE, hexachlorocyclohexan (αHCH, γHCH), polychlorobiphenyls (PCBs): gas-phase capillary chromatography combined with an electron-capture detector; for polycyclic aromatic hydrocarbons (PAHs): high-performance chromatography combined with mass spectrometry; non-ionic detergents such as nonylphenols [4-(para)-nonylphenol) and octylphenols (para-tert-octylphenol]; dioxins: gas chromatography combined with high-resolution mass spectrometry; brominated diphenyl ethers: gas chromatography combined with mass spectrometry – CI negative. All of the above compounds were analyzed in mussel flesh for every station, except dioxins, brominated diphenyl ethers, and non-ionic detergents, which were measured for about one in five stations.

3 Results

A total of 123 stations were retrieved out of the 149 stations deployed (82.5%).

3.1 Biometric Parameters

The analysis of shell height distribution showed that the batches were well calibrated; therefore, a slight "campaign effect" was visible in the Alboran basin where the size of the mussel shells of the samples was slightly smaller than in other areas. However, the analysis of flesh dry weight showed very marked trophic variations between study sites. Growth conditions in Alboran and the Northwestern basin were better, leading to a higher flesh dry weight. We also observed more favorable growing conditions in southern Spain, in the Ebro and Rhône input areas and, generally speaking, adjacent to major cities and ports (Palma, Barcelona, Marseille). This observation was confirmed by the distribution of the individual flesh dry weight and condition index.

3.2 Contaminants

The raw concentration results show that growth has a major impact on result distribution, in particular with regards to heavy metals. Some trace metals (Cd, Hg, Ni) showed systematically higher levels in the most highly oligotrophic zones, linked to the distribution of the mussel condition index. Concentrations of the vast majority of PCB congeners were lower than the limits of detection set at the study outset. CB 153 and 138 were the most reliable markers and were present in all samples; their distribution is similar to that of the sum of the ten congeners. Regarding PAHs, out of the 16 compounds analyzed, a large majority did not exceed the analytical limit of detection. On the scale of the three cruises, no compound showed a distribution profile identical to that of the sum of the 16 compounds. Compound distribution between stations was widely heterogeneous in comparison, for example, to that of PCBs. α HCH and γ HCH did not in any case exceed the limit of detection in coastal zones. Lastly, metabolites of DDT were the most commonly found elements. Analyses of brominated diphenyl ethers and non-ionic detergents all showed results below the analytical limit of detection.

3.3 Models

For each contaminant, adjustment parameters calculated on the basis of the raw data from this campaign are presented in Table 1.

Models were significant (p-values < 0.05) for most contaminants, with the exception of PAHs. The highest growth effect was observed for cadmium, mercury (Fig. 1), and nickel, with a variation of more than 50% in results explained by sample growth.

3.4 Adjusted Data

The value of the reference condition index is 0.11, which corresponds to the mean of the condition indices obtained from all samples. Raw data were adjusted for this value using specific parameters for every chemical compound. However, data on PAH were not processed according to this method.

At the scale of the study, the distribution of lead adjusted data was relatively homogenous, with an average value of 1.41 mg/kg and a median of 1.17 mg/kg. However, two sites were pinpointed as being particularly impacted by lead: the Portoscuso industrial site (Southwestern sub-basin), with a maximum of 8.25 mg/kg, and the zone spanning Portman to El Portus (Alboran sub-basin) from 5.3 to 6.25 mg/kg, which was home to a thriving mining industry with the dumping of $50{\cdot}10^6$ t of mining waste during the intensive extractive activities carried out during the period from 1960–1990. The maximum levels observed in the northwestern sub-basin are in the area of Barcelona (2.79 mg/kg) and in the Tyrrhenian sub-basin in Porto Ferrario at Isola d'Elba (3.05 mg/kg).

Adjusted levels of cadmium were globally homogenous throughout stations, with an average of 1.32 mg/kg and a median of 1.28 mg/kg. A few stations showed

Table 1 Adjustment parameters

	Model	R-square	*P*-value
Lead	Pb = 0.021 * CI + 0.932	6.57	0.01
Cadmium	Cd = 0.124 * CI + 0.01	84.65	0.00
Mercury	Hg = 0.008 * CI + 0.007	81.71	0.00
Nickel	Ni = 0.049 * CI + 0.41	50.25	0.00
Sum of DDTs	SDDTs = 15.398 * CI + 1.328	19.44	0.00
Sum of PCBs	SPCBs = 39.236 * CI + 5.174	14.78	0.00
Sum of 16PAHs	S16HAPs = −36.277 * CI + 43.223	1.41	0.21
Dioxins	DEF = 3.069 * CI + 0.145	47.47	0

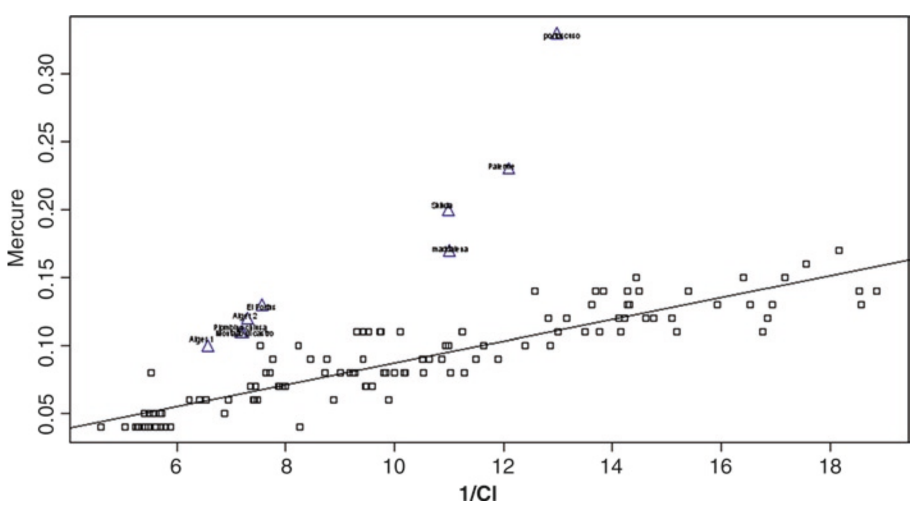

Fig. 1 Data set and model for mercury

relative peaks of around 2 mg/kg: Filicudi and Ustica stations in Tyrrhenian sub-basin (Sicily), Aguilas and Adra in Spain (Alboran sub-basin).

Several sites impacted by mercury were recorded: first and foremost the Portoscuso site in Sardinia (Southwestern sub-basin), with a maximum level of 0.31 mg/kg, witnessing significant contamination generated by a large industrial complex. To a slightly lesser degree, high levels were recorded in the SW sub-basin in Skida (0.19 mg/kg) and in the Tyrrhenian sub-basin especially in Palermo (0.22 mg/kg).

Average adjusted concentrations of nickel were around 1.1 μg/g, with a median of 0.94 μg/g. Extreme values were found in some sampling sites in the southwestern sub-basin, especially in Tunisia (Tabarka [3.18 mg/kg d.w]), Algeria (Oued Zhor [2.89 mg/kg d.w], Oran [2.47 mg/kg d.w]), Morocco (Nador [2.72 mg/kg d.w.]), and south of Spain (Fuengirola [2.44 mg/kg d.w]).

The average value of the sum of DDT compounds was 3.93 μg/kg, with a median of 3 μg/kg at the scale of the studies. Significant peaks were recorded in the Northwestern and Tyrrhenian sub-basin, especially in front of Marseille (15.47 μg/kg), Barcelona (15.17 μg/kg), and Napoli (15.34 μg/kg). In the Southwestern sub-basin, Algiers also showed a high level (10.23 μg/kg). The level recorded at the Algiers station was equivalent to the overall levels recorded at stations off the coast of the following rivers and streams: Ebro, Rhone, and, at slightly lesser levels, Tet, Aude, Herault (Northwestern sub-basin), and Tevere (Tyrrhenian sub-basin).

Regarding the sum of the ten congeners of PCBs and the CB153, the distribution shows a similar profile. The average value of the sum of PCB compounds was 14,583 μg/kg, with a median of 8,98 μg/kg at the scale of the studies. The results show the presence of sites that are impacted by PCBs, in the Northwestern sub basin (Barcelona [63.87 μg/kg], Marseille [103.52 μg/kg]), Tyrrhenian sub-basin (Napoli [91.48 μg/kg]), and Southwestern sub-basin (Algiers [51.13 μg/kg]). This characteristic presence along the coast of major urban centers is further confirmed by the values obtained in the Tyrrhenian sub-basin at La Maddalena (58.49 μg/kg), located close to a major naval base. To a lesser degree, we can also pinpoint inputs by the Ebro (20.37 μg/kg) and Rhone (37.80 μg/kg) Rivers.

Results related to the sum of the 16 analyzed molecules for PAH are expressed in raw values. Wide data heterogeneity was observed at the scale of the studies with an average value of 46.51 μg/kg and a median 44.4 μg/kg. Two peaks were identified in the Northwestern and Tyrrhenian sub-basin located at Marseille (105.5 μg/kg) and Piombino in Italy (80.8 μg/kg) near a large industrial complex.

Regarding the results for the dioxin compounds, the median level is around 0.7 ng/kg (TEQ). One peak recorded in the Marseille area reveals the existence of significant inputs of these compounds (2.66 ng/kg). Moreover, it confirms the peak measured for PCBs, which belong to the same compound family. On the project scale, we observed a similar distribution to the one of PCB congeners, with the highest values at Barcelona, La Maddalena, Napoli, and Algiers (Fig. 2).

4 Discussion

The Mytilos project confirmed the operational viability of the RINBIO methodology at the basin scale. The project's logistics, the mooring technique, and the deployment/recovery methods enabled cost minimization, plus a highly satisfactory retrieval rate taking into account the diversity of the studied coasts.

The condition index spread is indicative of the trophic heterogeneity of the Mediterranean coastlines. Overall, the waters are richer in the Northwestern sub-basin, due to the nutrients brought in by the Rhône (Minas 1989) and the Ebro Rivers, and in the Alboran sub-basin. The condition index is well related to the chemical contamination levels, especially in the case of trace metals. Regarding cadmium concentrations, the raw values measured in the Balearic Islands were two to three times higher than those measured at the mouth of the Ebro River and along the Spanish coast. For organic compounds, a relation can also be observed between the condition index and contaminant concentrations, except for PAH (polycyclic aromatic hydrocarbons).

These findings can be explained by a greater tissue growth in areas where nutrient concentrations in the waters are the highest, thus justifying the adjustment of data with reference to a standard condition index. Tissue growth is especially likely to mask or dilute the levels measured in the case of metals like cadmium, which is essentially present in the seawater in dissolved form. Its uptake and bioaccumulation via feeding (filtered particles) is negligible.

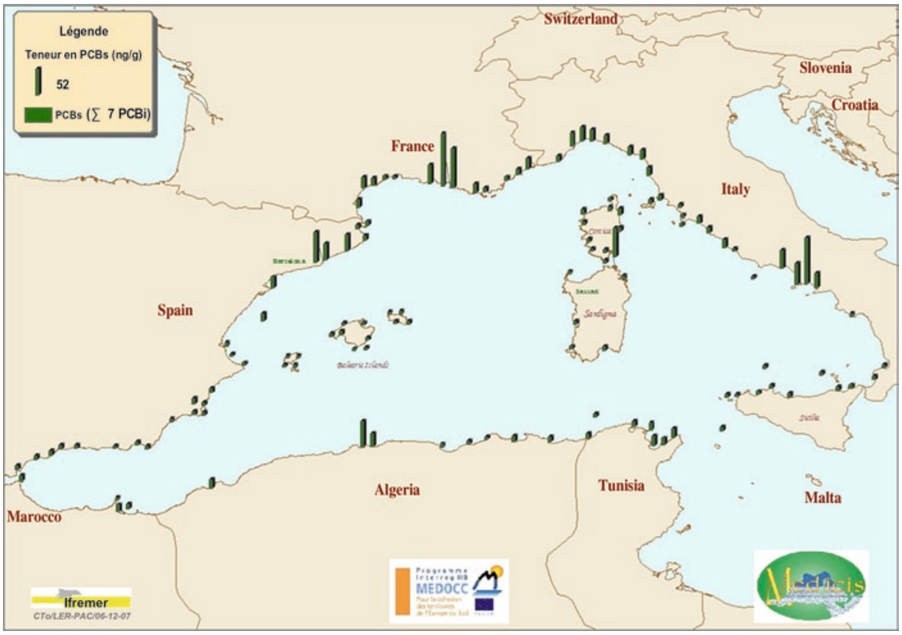

Fig. 2 PCBs level in mussels (? seven PCB indicators, ng/g)

In a more general way, the significant connection between the condition index and levels of contaminants in mussel tissue makes it possible to adjust the measurements and make comparisons between them, and thus to identify contaminated sectors within the network. The most highly impacted zones were mainly situated adjacent to urban and industrial centers and the outlets of major rivers.

Acknowledgements Thanks to experience sharing, all project partners were able to judge the easy implementation of this methodology and familiarize themselves with its main concepts through an active participation in all operations.

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First Biological Data on the Marine Snails *Osilinus turbinatus (Gastropoda, Trochidae)* of Eastern Coasts of Algeria

Sabrine Boucetta, Farid Derbal, Zitouni Boutiba, and M. Hichem Kara

Abstract The main objective of this study is to develop our knowledge of the ecology and the biology of the malacological fauna from interdital zone of the eastern coast of Algeria, with a particular interest for a marine snail, *Osilinus turbinatus*. Two coastal sites were chosen for the study: one is subjected to domestic pollution (Rizi-Amor beach, bay of Annaba), and the other one remote from anthropic activities (Oued Ghanem beach, Bay of Chetaibi). The harvest of the top shell was carried out during an annual cycle between December 2007 and December 2008 at a rate of a taking away per season. The number taken away was limited to avoid the disturbance of the population in place and thus to preserve the resource. Samplings were done in the morning between 10:00 and 12:00 a.m. to avoid the displacement of the top shell toward the rock faults according to the light and food.

Different environmental indexes (specific diversity, density, biomass) were determined to better understand the structure and variations of the malacologic population, notably in unsettled medium. The growth and sex ratio are given. The discussion is general and comments on the different biological and ecological aspects. We surveyed three species among the Trochidae family: *Osilinus turbinatus*, *O. articulatus*, and *Gibbula divaricata*. The density and the biomass of *O. turbinatus* was very appreciable in the two sites. Data on the morphology and relative growth of *O. turbinatus* are presented.

1 Introduction

With the patellid limpet, the marine snails are considered the most primitive of the Prosobranchia Gastropoda (Gaillard 1987). These marine mollusks colonize the rocky substrates of the intertidal zone. With 56 species found in the Mediterranean Sea (Conti and Ceccettib 2003), their taxonomy has always remained a subject of controversy (Hickman 1998; Donald *et al.* 2005). Not very known on the Algerian coasts, the marine snails are represented primarily by the genus *Osilinus* with a dominance of the species *O. turbinatus* and *O. articulatus*. We introduce here the first descriptions given of the ecobiology of *O. turbinatus* of the east coast of Algeria.

2 Material and Methods

The marine snails *O. turbinatus* were collected in February 2008. Two coastal sites were harvested: one is subjected to domestic pollution (Rizi-Amor beach, Bay of Annaba, is located between the Cap de Garde in the west, Ras El Hamra: 7°16' E and 36°68' N, and Cap Rosa in the east, 8°15' E and 36°38' N), and the other one is located far from anthropic activities at Oued Ghanem beach, Bay of Chetaibi. Samplings were done in the morning between 10:00 and 12:00 a.m. (Fig. 1). We introduce some data concerning the ecology and biology: associated invertebrate fauna, density of the marine snails,

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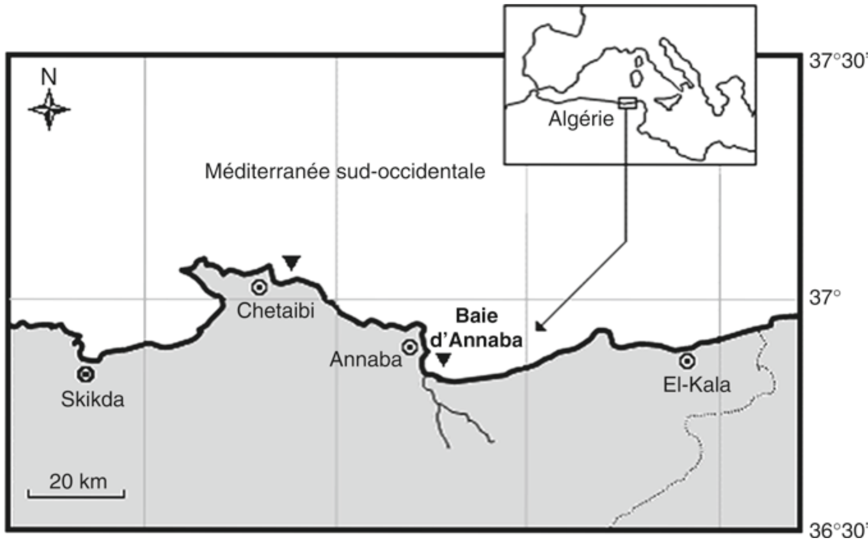

Fig. 1 Geographical limits of sampling sites (▼) of *Osilinus turbinatus* off the eastern coast of Algeria.

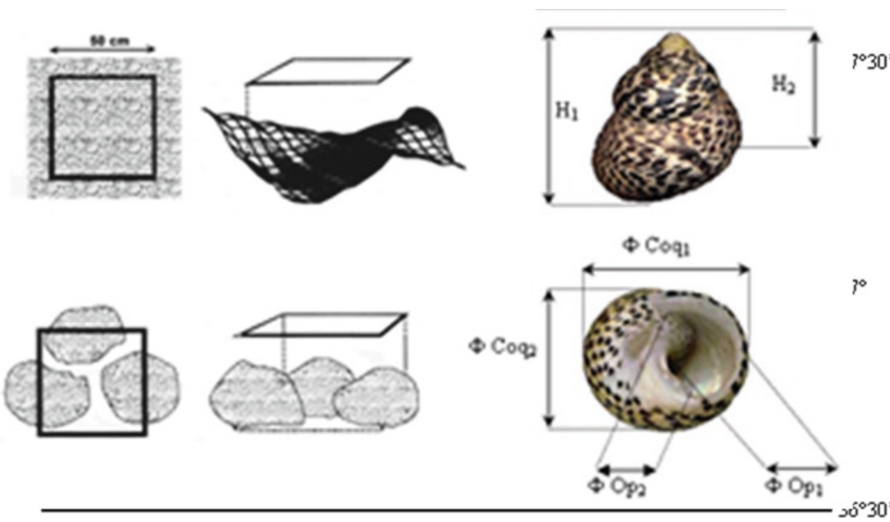

Fig. 2 From left to right: method of harvesting the top shells using one quadrate, measurements performed on marine snails Osilinus turbinatus collected along the eastern coast of Algeria (H_1 and H_2: height maximum and minimum; Φ Op_1 and Φ Op_2: peristomal diameters; Φ Coq_1 and Φ Coq_2: diameters of the shell).

relative growth, length-weight relationship, and sex ratio (Bode *et al*., 1986) of *O. turbinatus*.

The harvest of the top shell on the rocks was carried out in the intertidal zone, between 0 and 50 cm depth, using one quadrate of 0.5 m^2 area (Fig. 2). In each quadrate, we took everything inside, including the associated invertebrate fauna. The operation was carried out linearly for approximately 300 m. The samples, maintained alive in plastic cans, were then forwarded to the laboratory for the sorting and identification of the various collected species).

3 Results

The invertebrate fauna that cohabits with marine snails from the intertidal zone of the eastern coast of Algeria are represented by the mussels *Perna perna* and *P. picta*, the shells *Littorina* sp, the polychaeta *Nereis pelagica*, the patellid limpet *Patella caerulea, P. intermedia*, *P*. spp, and the polyplacophora *Acanthochitona crinitus*.

In the two sites, the marine snails are represented by two species: *Osilinus turbinatus* (6 ind · m^{-2}) and *O. articulatus* (1.5 ind · m^{-2}). The study of the biometrical

Table 1 Comparison of the regression equations and coefficient of allometry of the metric characters measured according to the height of the shell (H_1) in the *Osilinus turbinatus* population of Annaba Bay (An) and Chetaibi Bay (Ch). *: $P \leq 0.05$; ***: $P \leq 0.01$

Function	Zone	r	Regression equation	Coefficient of allometry	Allometry	Limit values (mm)	
						Max.	Min.
$H_2 = f(H_1)$	An	0.951	$H_2 = 0.993\ H_1 - 0.249$	$H_2 = 0.563\ (H_1)^{0.993}$	Isometry	18.67	4.23
	Ch	0.942	$H_2 = 1.268 H_1 - 0.413$	$H_2 = 0.386(H_1)^{1.268}$		26.43	2.19
$\Phi\ Op_1 = f(H_1)$	An	0.925	$\Phi\ Op_1 = 1.093\ H_1 - 0.171$	$\Phi\ Op_1 = 0.674\ (H_1)^{1.093}$		10.66	2.98
	Ch	0.823	$\Phi\ Op_1 = 1.003\ H_1 + 0.002$	$\Phi\ Op_1 = 1(H_1)^{1.003}$		13.89	0.14
$\Phi\ Op_2 = f(H_1)$	An	0.957	$\Phi\ Op_2 = 1.086\ H_1 - 0.272$	$\Phi\ Op_2 = 0.534\ (H_1)^{1.086}$	Majorant (***)	14.03	4.08
	Ch	0.878	$\Phi\ Op_2 = 1.348\ H_1 - 0.441$	$\Phi\ Op_2 = 0.632(H_1)^{1.348}$		18.18	3.01
$\Phi\ Coq_1 = f(H_1)$	An	0.830	$\Phi\ Coq_1 = 1.060\ H_1 - 0.464$	$\Phi\ Coq_1 = 0.343\ (H_1)^{1.060}$	Isometry	22.38	6.61
	Ch	0.840	$\Phi\ Coq_1 = 1.187\ H_1 - 0.517$	$\Phi\ Coq_1 = 0.304(H_1)^{1.187}$		38.62	2.92
$\Phi\ Coq_2 = f(H_1)$	An	0.960	$\Phi\ Coq_2 = 1.174\ H_1 - 0.542$	$\Phi\ Coq_2 = 0.284\ (H_1)^{1.174}$		20.17	5.2
	Ch	0.848	$\Phi\ Coq_2 = 1.251\ H_1 - 0.529$	$\Phi\ Coq_2 = 0.295(H_1)^{1.251}$	Majorant (*)	36.39	4.86

relationship of *O. turbinatus* showed that the maximal height of the shell grows differently compared to the other measured parameters (peristomal diameter Φ Op1 and Φ Op_2). In both sexes and both sites, all the studied features grow less quickly than the maximal height of the shell. The statistical analysis showed an absence of sexual dimorphism at *O. turbinatus* (Table 1) of Oued El Ghanem beach. On the contrary, at the site Rizi Amor, we found three parameters providing evidence of a sexual dimorphism: the minimal high (H_2), the peristomal diameter (Ø Op_1), and (Ø OP_2).

The morphological study of the population of *O. turbinatus* showed a highly significant correlation (Annaba Bay: $0.83 \leq r \leq 0.96$; $P \leq 0.001$, Chetaibi Bay: 0.82) between the various measured parameters and the height of the shell (H_1). The growth is generally isometric except for the Φ Op_2 and Φ Coq_2 parameters, which grow more quickly than the height of the shell (H_1) of the Chetaibi Bay population. Allometry between the height of the shell and its weight is of the isometric type, thus translating into a ponderal growth at the same rate/rhythm as the linear growth. The regression equations of the length-weight relationship are: $Pt = 2.415\ H_1 - 2.113$ (Chetaibi Bay) and $Pt = 2.153\ H_1 - 1.718$ (Annaba Bay).

The population was structured in the following way: 176 males (43.56%), 175 females (43.31%), and 53 immature (13.11%) in the station at Oued Ghanem beach. The sex ratio total of *O. turbinatus* is equal to 58.27, a value in favor of the males ($\chi^2 = 45.872$; $P > 0.05$).

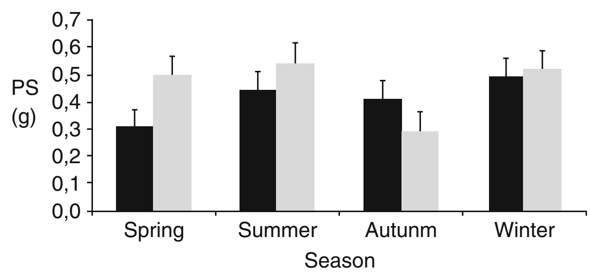

Figure 3 Seasonal comparison of the biomass of *O. turbinatus* between the stations of Rizi Amor and Oued Ghanem

The analysis of variance showed very highly significant differences for seasonal biomass ($P < 0.000$; Fobs = 12.91) at Rizi Amor ($P < 0.000$; Fobs = 11.39) at the Oued Ghanem beach station (Fig. 3).

4 Conclusions

This study is a contribution to the knowledge about Trochidae of the eastern coast of Algeria. Various ecological and biological aspects were considered for the use of these gastropods in the biosurveillance of the littoral ecosystems.

The survey of Trochidae showed the presence of three species (*O. turbinatus, O. articulatus, and G. divaricata*). The temporal follow-up of the natural populations of *O. turbinatus* showed the highest density at Oued Ghanem beach compared to the second polluted sectors.

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Combining Monitoring Networks, Hydrodynamic Modelling and Satellite Data to a Better Understanding of the Trophic Functioning of Coastal Waters in Normandy

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Abstract The Normandy Hydrological Coastal Network (Réseau Hydrologique du Littoral Normand or R.H.L.N.) is a monitoring network running since 2001 in order to reach two main goals:

1. To monitor the quality and assess the eutrophication status of coastal water bodies.
2. To contribute to the implementation of the Water Framework Directive (W.F.D.) surveillance program in the Seine Normandy hydrographic district.

Hydrobiological data acquired from 2001 to 2007 on 18 coastal and 4 transitional water bodies showed a clear contrast between trophic systems in the western and eastern side of the Cotentin peninsula, and the Bay of Seine.

West and North Cotentin are oligotrophic systems characterised by low terrigenous inputs, and therefore show "classical" biogeochemical cycles, with low nutrient concentrations, and low chlorophyll *a* levels (<10 μg · L^{-1}). East Cotentin shows an enrichment gradient southwards, due to the influence of the baie des Veys waterbasin bringing relatively rich nutrient inputs to the coast, which support a high phytoplankton production (20–25 μg chl. *a* · L^{-1}). But despite of moderate signs of dystrophy, the biogeochemical cycles remained "classical", with a complete nutrient depletion of the water column during the phytoplankton bloom.

The eastern Bay of Seine, which is under the influence of four waterbasins including the Seine (70,000 km^2), shows totally different patterns, characterised by high nutrients winter stocks and continuous terrigenous inputs reloading the ecosystem with nutrients. These inputs thus prevent from nitrate depletion at any time, allowing a succession of phytoplankton blooms to occur throughout the productive period, with average chlorophyll levels of 20–40 μg · L^{-1}, and maxima reaching 70 μg · L^{-1}. Modifications were also observed among phytoplankton communities. However, despite of these eutrophication signs, the eastern Bay of Seine waters did not show oxygen depletion probably thanks to a high hydrodynamism.

Combined with these in situ data, satellite ocean-colour data (through a specific algorithm developed for turbid coastal zones (F. Gohin et al. 2002)) revealed a one-month delay in the start of the productive period between the western Cotentin and the central Bay of Seine, and allowed the assessment of the spatial heterogeneity of chlorophyll distribution in each water body. Hydrodynamics modelling with six regional bidimensional models allowed optimising the in situ monitory strategy though the careful evaluation of the relevance of sampling stations to represent the trophic status of each water body.

1 Introduction

In the framework of the Nitrate directive and of the OSPAR convention (OSlo PARis convention for the protection of the North East Atlantic) common procedure

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for eutrophication assessment, a regional survey of nutrients loads and phytoplankton blooms in 2001 led to setting up a regional monitoring network. Its strategy aimed at understanding the trophic functioning of Normandy coastal waters and assessing their eutrophication level. From 2002 to 2006, this monitoring network was progressively intensified in terms of number of sampling stations and of monitoring frequency. This built up an important set of data, thus giving a good knowledge of coastal waters functioning.

This development was also undertaken as a specific task in the implementation of the W.F.D. (Water Framework Directive-2000/60/CE). This directive is the core of a new water policy in Europe, based on an integrated water management at the water basin scale, including continental (rivers, lakes), transitional (estuaries, lagoons) and coastal surface waters, as well as groundwaters. Its central objective is to reach a "good status" of all European waters within 2015. Towards this ambitious goal, the W.F.D. sets major requirements such as to:

1. Characterize the water basin (delimitation of water bodies, evaluation of their chemical and ecological status, evaluation of human pressures, economical analysis)
2. Elaborate a monitoring program
3. Elaborate a management plan and a program of measures, defining the specific objectives for each water body and the measures to be taken to reach them.

W.F.D. puts special emphasis on biology, by taking into account several biological quality elements: phytoplankton, benthic macroinvertebrates, macroalgae and angiosperms, fishes. Thus, the requirements of this directive regarding the elaboration of classification tools and monitoring programs for coastal and transitional water bodies are central objectives in the R.H.L.N. ("Réseau Hydrobiologique du Littoral Normand") project, for the phytoplankton biological quality element.

In this context, the R.H.L.N. monitoring strategy was finalised for a routine network with two major goals:

i. To answer to the monitoring requirements of the W.F.D.
ii. To satisfy regional needs regarding the trophic status and functioning of coastal waters, and the development of phytoplankton including toxic species; these needs are tightly connected with human activities dependant upon seawater quality, such as seashell farming and harvesting, which are of major economical importance in the region as Normandy is the first region in France for production of mussels and the second for production of oysters.

2 In Situ Monitoring Network

The Hydrobiological monitoring network in Normandy coastal waters ("R.H.L.N.") was developed in 2001 on the basis of the former national phytoplankton network (REPHY) operated by Ifremer since 1984, which had five sampling stations along Normandy (Le Goff and Riou 2005).

The number of sampling stations was thus increased progressively, from 10 in 2002 up to 33 at the maximum effort level in 2006. In addition, the sampling frequency was doubled to one sampling every two weeks, during the growing season (March–October) after an initial sampling in February (assessment of nutrients concentrations at the end of winter, i.e. winter stock). This increased monitoring frequency brought a better understanding of the high temporal variability in the most productive areas of the coastline. Physical and chemical parameters monitored are temperature, salinity, turbidity, dissolved O_2, nutrients concentrations [NO_3^-, NO_2^-, NH_4^+, Si $(OH)_4^-$, PO_4^{3-}].

Concerning biological parameters, chlorophyll *a* and pheophytin concentrations are measured in subsurface, and phytoplanktonic flora are identified and counted at the species level.

Finally, with increasing knowledge along years, the sampling strategy was revised, ending up with a routine version of the monitoring network (cf. Fig. 1):

i. Some stations showing the same environmental patterns and thus bringing redundant information, were excluded.
ii. Sampling frequencies were adjusted to the national monitoring recommendations developed for W.F.D. (surveillance control and operational control) and finalised in 2007 through the Circulaire 2007/20 from the Environment French Ministry; i.e. 1 per month every month between March and October (growing period) for chlorophyll; 1 per month all year long for phytoplanktonic flora.
iii. In addition to W.F.D. monitoring, complementary stations and/or increased sampling frequency (2 per month) during the growing period were implemented for regional purposes (waterbodies

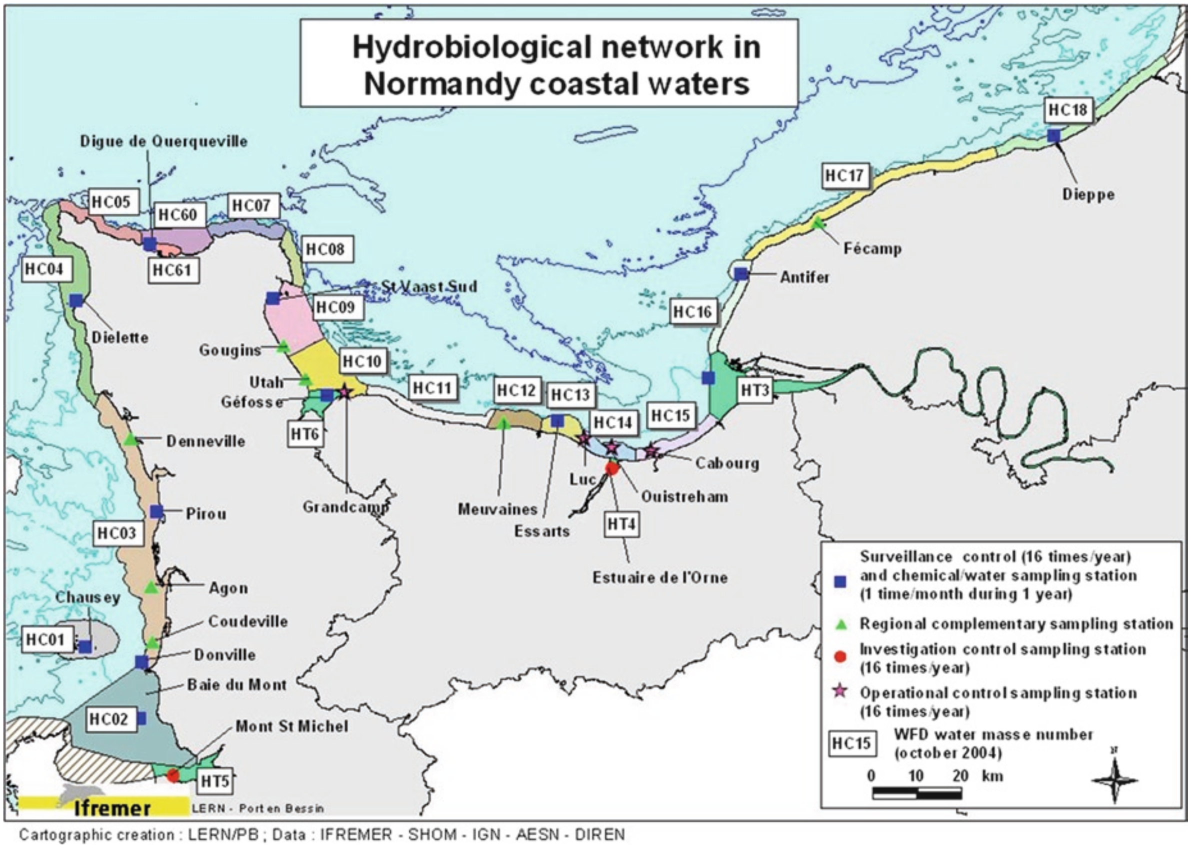

Fig. 1 Maps of R.H.L.N. ("Réseau Hydrobiologique du Littoral Normand" – Hydrobiological monitoring network in Normandy coastal waters) sampling stations

supporting activities sensitive to water quality, or showing a high temporal variability).

iv. Location of sampling stations was eventually adjusted, after a study of the relevance thanks to hydrodynamical models (cf. Section 3 below) and to satellite images, in order to ensure their representativeness of the status of the water body.

3 Modelling

Two types of numerical models, hydrodynamical and ecological, were developed (cf Fig. 2):

1. A complete set of bidimensional hydrodynamical models was elaborated, including a global model (400 m mesh size) covering the whole Normandy coastal zone, and a series of local models (75 m mesh size) to simulate local conditions with a better spatial resolution. Both types integrate physical driving forces, among which tidal and wind-generated currents play an important role on coastal waters dynamics and circulation (Riou et al. 2007). These models are able to simulate the fate of

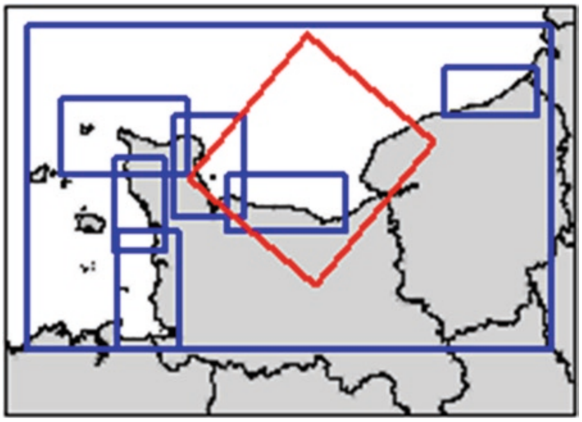

Fig. 2 Localisation and extension of hydrodynamical models (*in blue*) and ecological model (*in red*) along Normandy coastline

freshwater and pollutants discharges into coastal waterbodies.

2. An ecological model was developed on the Bay of Seine, coupling a three-dimensional hydrodynamical model including suspended sediment transport and a biological model simulating nutrient (N, P, Si) cycles, their consumption by phytoplankton (with discrimination of diatoms and flagellates), and their respective contribution to phytoplankton biomass regulation (Cugier 1999). A further development of the model occurred more recently with a modelling chain connecting models on the Seine river basin, the Seine estuary and the bay (Cugier et al. 2005). Field and satellite data are of particular interest for calibration and validation of such models.

A specific use of hydrodynamical models within the R.H.L.N. project was to simulate currents circulation in each water body, in order to assess the relevance of R.H.L.N. sampling station(s) as representative for the waterbody's status. Fig. 3 shows examples of the circulation pattern (trajectories along 7 days of tidal cycles) of particles of water dropped at the sampling stations in "HC03 – Ouest Cotentin" waterbody.

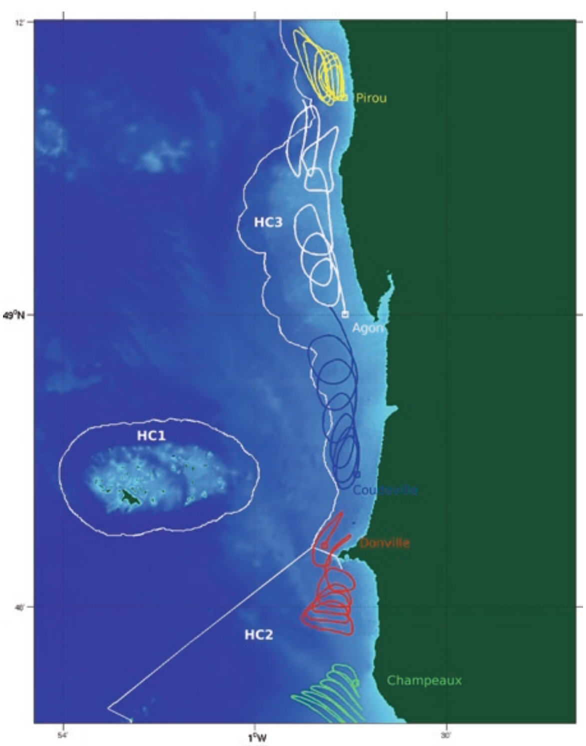

Fig. 3 Maps of simulated particles trajectories (2D hydrodynamical models) along 7 days of tidal cycles after their launch at the sampling stations (*red dot*) in water body number [$HC0_3$]

The hydrobiological model of the Bay of Seine was used to evaluate the fate of nutrient inputs from the Seine and from coastal rivers, during their consumption by phytoplankton. Water and nutrient discharges from the Seine are well estimated thanks to heavy field monitoring along the estuary. Though they largely overwhelm the discharges from coastal rivers, an estimation of these smaller inputs and of their impact on phytoplankton growth, especially in summer, was nevertheless performed, in order to be able to discriminate their respective contribution to phytoplankton production along the growing season (Fig. 4).

4 Satellite Data

Ocean-colour data from satellite were used to evaluate chlorophyll concentration in surface waters (Fig. 5). Data from Sea-viewing Wide Field-of-view Sensor (SeaWiFS) and more recently from MOderate Resolution Imaging Spectroradiometer MODIS (NASA) and from Medium Resolution Imaging Spectrometer Instrument MERIS (ESA) were processed with a specific treatment algorithm (Gohin et al. 2003, 2005), as the original data treatment algorithm developed by NASA was adapted to clear oceanic waters, and had thus to be adjusted for turbid and chlorophyll-rich coastal waters, which are characteristic for Normandy coastlines. In situ measurements are also used to calibrate satellite maps. The combined use of in situ and satellite data, and ecological models, are described in (Gohin et al. 2008).

5 Results

Some results of this integrated monitoring strategy coupling in situ measurements, satellite data and numerical modelling are presented here. R.H.L.N. in situ monitoring brought a good knowledge of nutrients and chlorophyll evolution along years; this allowed an interannual and spatial (between stations and between waterbodies) comparative analysis. First of all, this monitoring provided with a precise characterization of hydrobiological

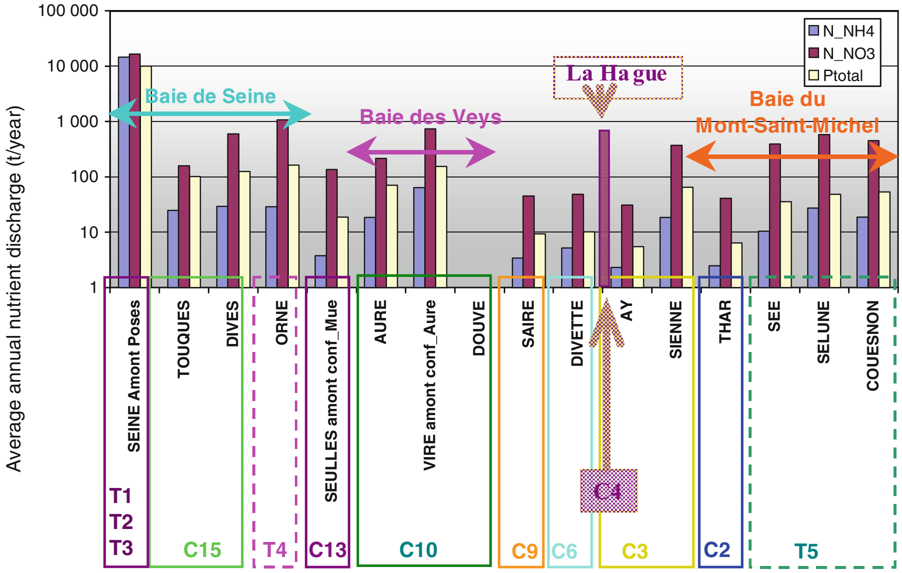

Fig. 4 Average yearly nutrients discharges from Seine and Lower-Normandy coastal rivers (in t/year; discharges estimated over the period 1990–1999 from data collected by the Hydrometric and Quality Monitoring networks on rivers) and identification (coding) of the water body

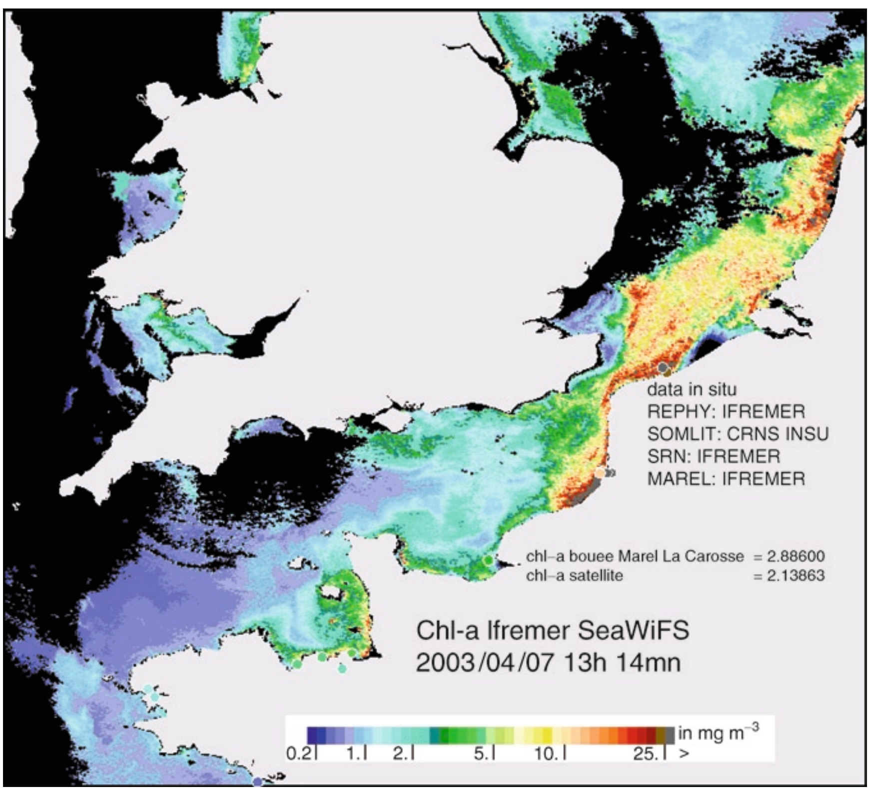

Fig. 5 Chlorophyll concentration from satellite

cycles in coastal waters. It clearly showed contrasted situations, with an enrichment gradient from the Cotentin western coast to the Eastern Bay of Seine. Figure 6 illustrates examples of this gradient for three stations, showing the temporal evolution of nitrate and chlorophyll concentrations during 3 years (2001 to 2003) with contrasted climatic conditions. In Western Cotentin, the phytoplankton spring bloom, with a chlorophyll peak reaching 5 µg · L^{-1}, leads to a complete depletion of nitrate in the water column, which in return limits phytoplankton growth. In Bay of Veys, similar nitrate depletion is observed, but the chlorophyll peak concentration is higher, reaching 25 µg · L^{-1}. The eastern part of the Bay of Seine (Cabourg station) shows a very different pattern, with a high production level (chl. maximum concentration reaching up to 70µg · L^{-1}) and no complete nitrate depletion (except very briefly in dry summers like 2003), but a continuous enrichment of the water body by river inputs, which supports a succession of increasing chlorophyll peaks.

Within the characteristic pattern observed for each zone, both from in situ monitoring and satellite images, an important interannual variability is also observed, for instance regarding the start period of the spring bloom (1 month earlier in the dry and hot year 2003), and the maximum concentration reached.

The ecological model simulates the fate of nutrients and the development of phytoplankton blooms, and appeared to be coherent with in situ observations. It was also able to reproduce satellite observations both in term of temporal cycles and in term of spatial location and extension of phytoplankton blooms (Gohin et al. 2008). Figure 7 illustrates an example of simulated maximum dinoflagellates concentrations simulated for a dry year (1993) and a wet year (1995). It shows a more intense development in dry years, with a maximum production along the south-eastern part of the Bay of Seine. On the contrary, wet years are less productive regarding dinoflagellates, and the maximum of biomass is located on the north-eastern limit of the bay, in front of Antifer cape.

The model is also able (i) to identify which nutrient is limiting phytoplankton growth at any time and (ii) to discriminate the respective influence of the nutrient discharges from Seine and coastal rivers, and thus can reveal the contribution of summer inputs to dinoflagellates blooms.

Finally, a major prospective use of the model was to simulate the effect of different levels of nutrients discharges limitations, either for nitrate or for phosphorus or both nutrients. They revealed, for instance, that 50% limitation in nitrate discharge reduces by 50% dinoflagellates

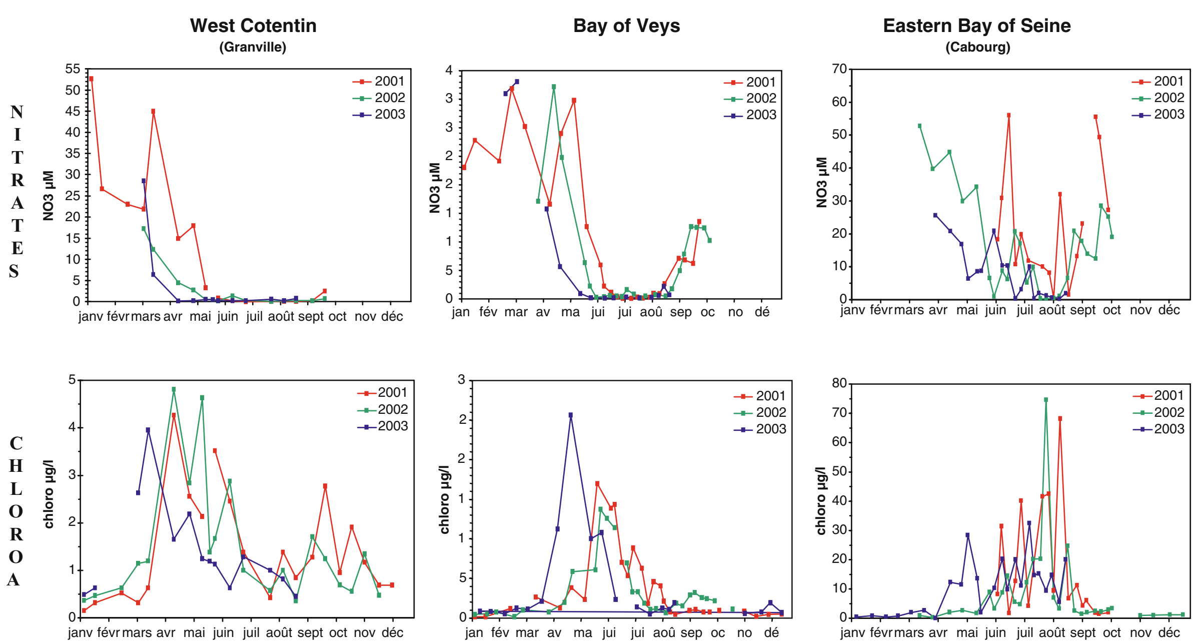

Fig. 6 Compared temporal evolution of nitrate and chlorophyll concentrations between years 2001 and 2003 in three contrasted waterbodies along the trophic gradient

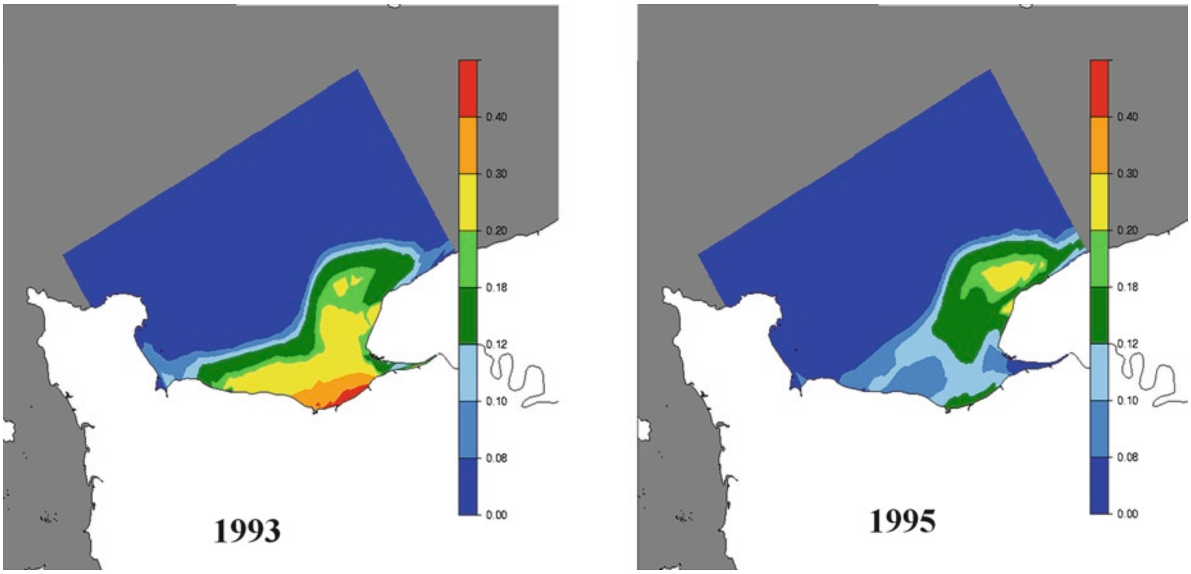

Fig. 7 Simulated yearly maxima of dinoflagellate biomass (expressed in μmol L^{-1} N) for a dry year (*left*) and a wet year (*right*) (Cugier 1999)

production without impacting significantly diatoms production (Cugier 1999). These simulations are of major interest in term of decision-making tools, as they help define the level of effort required regarding the limitation of nutrient discharges from human activities, for instance for the elaboration of programs of measures at the water-basin level. To further develop this modelling approach, a combined use of models developed on the Seine water basin, its estuary and the bay of Seine was performed by Cugier et al. (2005).

To conclude, a last type of results from the R.H.L.N. is the classification of the ecological status of coastal waters, for the "phytoplankton" biological quality element of the W.F.D. and its related criteria (chlorophyll concentration, frequency of blooms). Table 1 illustrates, for each coastal waterbody in Normandy, the yearly classification and the integrated classification over a 6-years period, using the 90th percentile metrics and the classification grid adopted for "Atlantic waters" within the European intercalibration exercise.

6 Conclusion

The combined approach developed in Normandy by the RHLN network, based upon in situ monitoring, satellite images and modelling, brought an important set of data allowing a precise comprehension of nutrient cycles and phytoplankton dynamics in coastal waters. They provided with a good evaluation of trophic functioning and a solid assessment of eutrophication level. Moreover, thanks to the use of satellite data, a large scale evaluation was accessible. This has major interests, among which (i) to assess spatial variability and heterogeneity of phytoplankton dynamics, (ii) to assess spatial heterogeneity within waterbodies and between waterbodies, more completely than in situ monitoring, and (iii) to allow a characterisation of phytoplankton production at large scale, offshore of the limits of coastal waters, which makes this technique a powerful tool for the implementation of the monitoring requirements of the new European directive for a marine strategy (2008/56/CE).

Numerical modelling was used to assess the hydrodynamic functioning of waterbodies, and helped evaluate the pertinence of the location chosen for in situ monitoring stations. Ecological modelling, simulating nutrients fate and phytoplankton development, was able (i) to reproduce observed spatial and temporal variations, (ii) to evaluate yearly production for diatoms and dinoflagellates groups, and (iii) to assess the effect of different levels of nutrients discharges limitations on phytoplankton production; these latter results are of specific interest for decision-makers for the definition of measures to be taken on the waterbasin,

Table 1 Example of ecological classification of coastal waterbodies based on the chlorophyll *a* 90th percentile (in $\mu g \cdot L^{-1}$)

		Chlorophyll-a $\mu g \cdot L^{-1}$: 90th percentile (March to October)								
		By sampling station							by waterbody	
Waterbody	Station	2001	2002	2003	2004	2005	2006	Global 2001–2006	Global 2001–2006	
HC01	Chausey	4,63	2,77	2,89	1,62	1,70	2,34	2,28	2,28	0–5 $\mu g \cdot L^{-1}$
HT5	St Michel						10,90	10,90	10,90	5–10 $\mu g \cdot L^{-1}$
	Mt St Michel 2007									10–20 $\mu g \cdot L^{-1}$
HC02	Cancale Est						2,36	2,36	3,13	20–40 $\mu g \cdot L^{-1}$
	Champeraux						3,13	3,13		>40 $\mu g \cdot L^{-1}$
	Hacqueville				2,97	1,48	3,38	2,87		
HC03	Agon				1,71	1,03	4,32	1,82	2,80	
	Coudeville						2,77	2,77		
	Denneville						1,19	1,19		
	Donville	3,00	2,85	1,95	2,34	1,82	2,26	2,66		
	Pirou	2,31	2,41	3,70	3,03	1,88	4,10	3,46		
HC04	Bameville				3,16	2.01	3.19	2.66	2.82	
	Dielette									
	Jobourg				5,24	1,47	1,49	2,62		
HC05	Gruehy				4,49	1,97		3,43	3,43	
HC60	Cherbourg				4,60	1,81	1,88	3,86	3,86	
HC61	Digue Querqueville					2,03	1,79	2,14	2,14	
HC07	Cap Lévi				3,44	2,30	0,97	3,06	3,06	
HC08	Réville				5,54	7,97	5,50	6,79	6,79	
HC09	Les Gougins						7,01	7,01	7,32	
	La Hougue			4,14	5,40	4,73	5,09	7,23		
HT6	Géfosse		16,63	10,26	6,89	9,79	7,42	10,54	10,54	
HC10	Grandeamp	10,37	8,04	10,41	6,89	7,18	6,71	9,01	8,83	
	St Germain de Varre	10,13	8,80	3,01	3,28	5,56		5,35		
	Utah Beach						7,76	7,76		
HC11	Porten Bessin				9,34	6,44	9,74	8,63	8,63	
HC12	Meuvaines				4,89	3,87	2,69	4,74	4,74	
HC13	Les Essarts				2,97	2,69	1,75	2,82	2,82	
HC14	Luc sur mer	4.72	9.85	12.61	8.39	4.79	8.99	8.50	16.01	
	Ouistreham	27,98	17,32	18,47	13,39	9,89	11,64	19,05		
HT4	Estuaire de l'Orne									
HC15	Cabourg	41,57	5,44	18,84	7,77	8,97	17,35	13,76	13,76	
HT3	Carosse		6,97	14,16	20,16			18,63	18,63	
HC16	Antifer		21,69	9,61	7,90	21,27	16,96	12,72	12,72	
HC17	Fécamp						2,99	2,99	3,90	
	St Aubin (76)				2,25	7,32		5,97		
HC18	Dieppe				2,53	4,76		2,97	2,97	

in the framework of management plans and program of measures that need to be elaborated within 2009, as required by the W.F.D.

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Impact of Hydrocarbons on Marine Microbial Communities

Christine Cagnon, Magalie Stauffert, Lionel Huang, Cristiana Cravo-Laureau, Marisol Goñi Urriza, Sylvain Bordenave, Sandrine Païssé, Pierre Caumette, and Robert Duran

Abstract The coastal areas are periodically exposed to pollution by crude oil, in a chronic or accidental way. This leads to a rupture of the equilibrium of the bacterial communities in the coastal ecosystems. In other respects, the important metabolic diversity present in these ecosystems suggests an important enzymatic potential for hydrocarbon degradation. We intend to evaluate the impact of oil pollution on coastal bacterial communities and the biodegradation potential.

Analyses of bacterial communities by molecular approaches allowed the characterization of the structure and the dynamic of these communities in chronic polluted sites (mats of Berre Lagoon, Provence, France) or in punctually contaminated laboratory microcosms (mats of Salins-de-Giraud, Camargue, France; sediments of the Benoît aber, Bretagne, France). Genomic and transcriptomic T-RFLP (Restriction Fragment Length Polymorphism) analysis based on 16S rRNA encoding genes showed changes in bacterial diversity and community structures revealing the adaptation of microorganisms to oil pollution.

Identification of naturally occurring differences between unpolluted and polluted microbial communities' transcriptomes have also been demonstrated by the differential display method. Several functional bacterial mRNA were characterized as differentially expressed according to experimental conditions. The role of these genes in the response of hydrocarbon must be elucidated.

We also develop an approach to characterize adaptive genes associated to mobile elements.

1 Introduction

The coastal ecosystems are particularly exposed to pollution by hydrocarbons, whether this pollution is accidental or chronic. Once in the environment, these hydrocarbons can have an impact on the bacterial communities either by a direct toxic effect or by inducing changes of the environmental parameters. The biodegradation of hydrocarbons by natural microbial populations represents one of the primary mechanisms by which petroleum and other hydrocarbon pollutants are eliminated from the environment. We investigate the impact of hydrocarbon contamination on microorganisms and microbial communities. We follow the framework presented in Fig. 1. First, dynamic approaches following the modifications of bacterial community structures are developed. Secondly, the bacterial response to the presence of pollutant is investigated. This corresponds to an explicative approach analyzing both the diversity of metabolic genes during the contamination period and the identification of genes expressed under oil contamination.

Different marine microbial communities were studied: coastal sediments or microbial mats developed at the water-sediment interface in coastal zones. In order to compare these environments in non-contaminated and contaminated conditions, samples were maintained in microcosms or mesocosms at the laboratory (Fig. 2).

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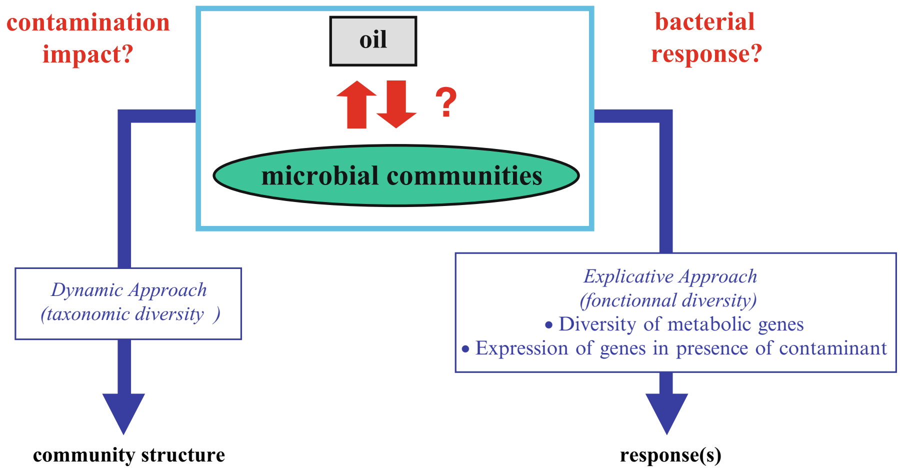

Fig. 1 Framework for studying the impact of hydrocarbon contamination on microbial communities

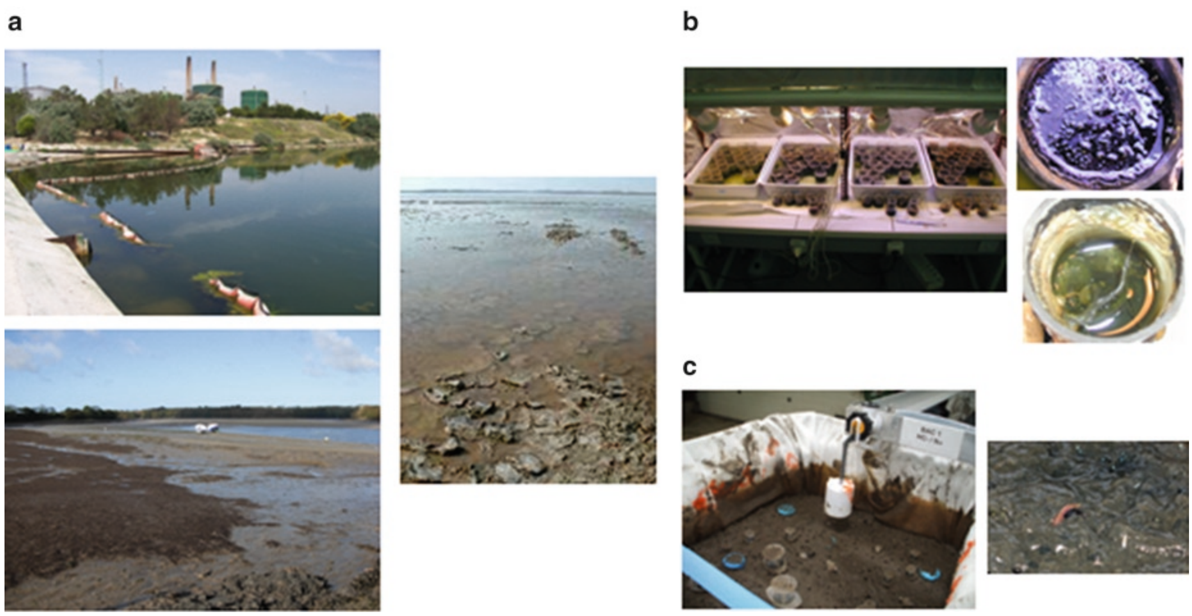

Fig. 2 Different marine microbial communities studied. (**a**) Microbial mats and coastal sediments in natural environments; (**b**) microbial mats in microcosms; (**c**) coastal sediments in mesocosms

2 Oil contamination impact on microbial community structure

2.1 *In Situ Studies*

One *in situ* study was carried out on a chronically oil-polluted retention basin sediment located in the Berre Lagoon (Etang de Berre, France). Hydrocarbon content analysis clearly revealed a gradient of hydrocarbon contamination in both the water and the sediment following the basin periphery from the pollution input to the lagoon water. A lack of alkanes at the input station suggested an important alkane biodegradation in the sediments at this station (Païssé et al. 2008). Microbial biodiversity analyses, performed by terminal-restriction fragment length polymorphism (T-RFLP) based on the polymorphism of ribosomal

16S genes (Païssé et al. 2010) showed a strong correlation of the bacterial community structure with the hydrocarbon content (Païssé et al. 2008).

The phylogenetic analysis highlighted a particular community of the input station with a predominance of deltaproteobacteria and especially bacteria mainly involved in the sulfur cycle. Thus, the bacterial community of the sediments of this station had a particular structure associated with hydrocarbon concentration, which indicates the adaptation of this community related to an important biodegradation of hydrocarbons (Païssé et al. 2008).

2.2 Microcosm Studies with Adapted Sediments

In order to evaluate the degradative capacities of the adapted bacterial community of the input station, this community was analyzed in controlled conditions at the laboratory. Petroleum was added and its composition compared to that of abiotic control. Fifty to sixty percent of alkanes disappeared after 14 days. Concerning the major polycyclic aromatic hydrocarbons (PAHs), naphthalenes and phenanthrenes and their methylated forms, 60–80% are biodegraded in 14 days. The minor aromatic compounds, with four and five aromatic cycles, started to be degraded at day 14. These results showed the great capacity of this community to degrade hydrocarbons quickly (Païssé et al. in press).

Analysis of the active bacterial community by T-RFLP based on the ribosomal gene expression products (16S rRNA transcripts) showed a separation of non-contaminated control and contaminated sample as soon as 3 days of incubation. Then the active bacterial community was stabilized at day 12, as the community of the control continued to diverge. The active community structure was therefore strongly dependent on oil contamination (Païssé et al. in press).

2.3 Microcosm Studies with Non-adapted Sediments

Photosynthetic microbial mats developed in a pristine hypersaline environment from Camargue salterns (Salins-de-Giraud, France) were used. They were maintained in microcosms under laboratory conditions for 1 year. Half of them were contaminated by Erika fuel. A rapid biodegradation of PAHs was observed.

The modification of the community structure was followed by T-RFLP analyses based on polymorphism of ribosomal 16S genes. The evolution of microbial communities during the incubation period was observed in both control and contaminated mats. However, changes were more important under fuel contamination (Fig. 3), especially after 2 and 3 months of incubation. These results clearly illustrated the impact of oil contamination on bacterial community structure (Bordenave et al. 2007). However, after 5 months and until the end of the experience, the contaminated mats were quite similar to the non-contaminated ones, indicating that the microbial mat ecosystem has high resilience capacities (Bordenave et al. 2007).

In addition, the responses of the metabolically active bacterial community were evaluated by T-RFLP based on 16S rRNA transcripts. The structure of the active bacterial community was modified as soon as the oil was added to the microcosms. This result suggested a fast adaptation of the microbial mat to oil contamination (Bordenave et al. 2007).

All these results confirm the impact of oil contamination on bacterial community structure.

3 Bacterial Responses to Oil Contamination Functional Diversity

The other part of our investigation consists of estimating the metabolic response to the presence of pollutant, especially its biodegradation potential. For this purpose, we investigated the diversity of two families of genes coding for enzymes involved in hydrocarbon biodegradation. The genes for naphtalene dioxygenase and benzyl succinate synthase were chosen as model genes for aerobic and anaerobic biodegradation, respectively.

These genes were expressed after 24 h of contamination, and T-RFLP analyses showed that their biodiversity was not affected during the entire incubation period or according to experimental conditions. Thus, no apparent functional response could be observed

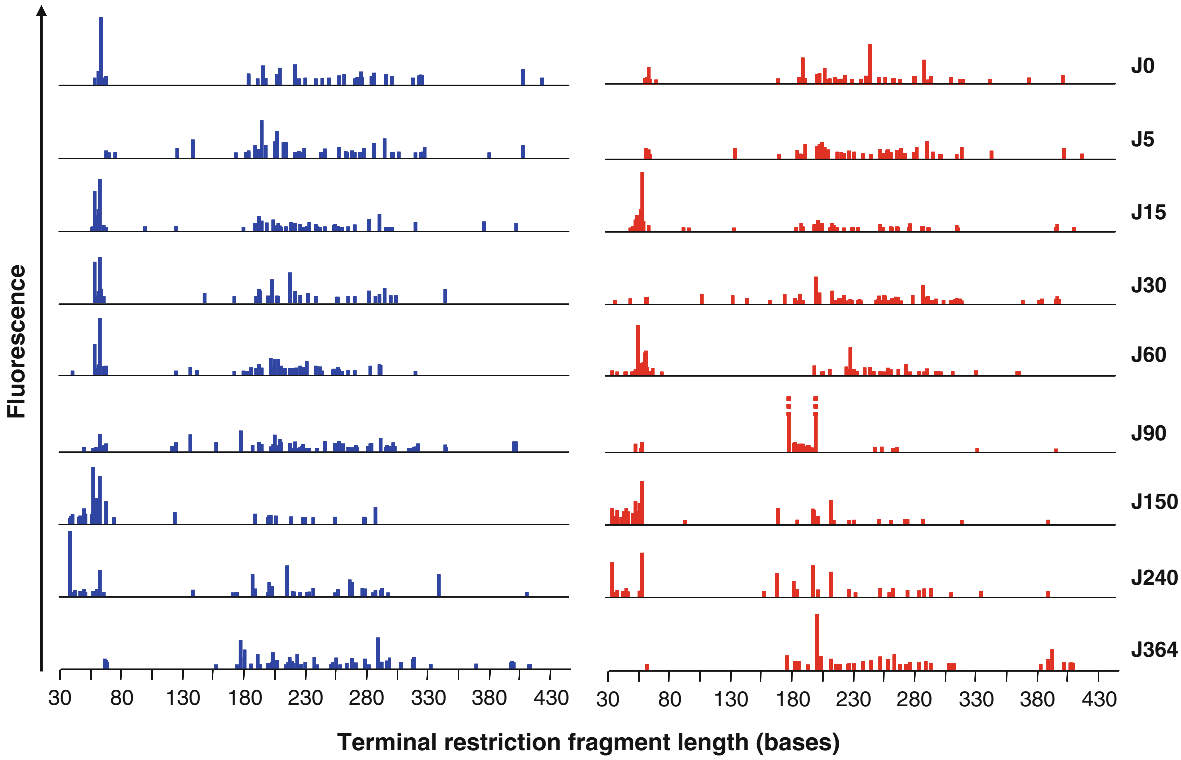

Fig. 3 T-RFLP (Restriction Fragment Length Polymorphism) profiles based on 16S rDNA of non-contaminated (*left, in blue*) or oil contaminated (*right, in red*) non-adaptive microbial mat microcosms

with these two biodegradative gene families. These results led us to investigate for unknown genes that are differentially expressed under fuel contamination.

3.1 Expression of New Genes in Response to Hydrocarbon Pollution

A first approach was to use the differential display method (Bordenave et al. 2010) comparing RNA from two different conditions as non-contaminated and contaminated conditions. Genes were detected as differentially expressed. Most of them did not show homology with sequences of known genes or presented homology with genes encoding protein of unknown function.

Some of them showed homology with genes coding for proteins potentially involved in response to the presence of pollutant-like transporters or membrane proteins. In all cases, their real role must be further characterized. Indeed, the fact that many conserved hypothetical proteins were differentially displayed could be very interesting since this kind of method may suggest a function of some proteins of unknown function found in many different bacteria.

We also developed a second approach for the identification of genes expressed under oil contamination consisting of target mobile genetic elements. Indeed, the adaptation of the microorganisms is mainly explained by horizontal transfer processes. Mobile genetic elements are involved in these processes, and genes giving a selective advantage are usually found in such elements. The mobile genetic elements are thus an ideal target for the search for new genes involved in the response to pollution.

We are especially interested on integrons. They are genetic structures able to acquire or disseminate genes and also to express them. The only stable gene (*intI*) is coding for an integrase, responsible for the integration of new genes. Then, we are following the expression of this gene in order to evaluate the implication of such an element in the adaptation to pollution.

The last gene integrated is located just after the *intI* gene and is the most expressed. So we have developed a method to clone only the genes at this particular position in integrons from the metagenome because they are the best target to find new adaptive genes in integrons (Huang et al. 2009). We are now exploring the genes from coastal sediment studied in the DHYVA project (Stauffert et al. 2010).

4 Conclusion

In conclusion, all our results show the hydrocarbon impact on active community structure and the microbial adaptation to such pollution. This impact may be due to hydrocarbon toxicity or to its potential use as an organic substrate for bacteria, or just to the environment physical–chemical condition changes.

For bacterial response, no biodiversity modification has been detected for the biodegradative model genes investigated, but their early expression could be detected. Differential analysis approaches allowed the identification of several differentially expressed genes according to the experimental conditions. An accurate characterization of these differentially expressed genes must be performed in order to determine their role in hydrocarbon contamination response. Quantitative analyses of the expression of these genes all along the incubation period are also explored in order to characterize their regulation mechanisms.

Finally, we are developing new molecular tools to understand more precisely the adaptation of microbial community to hydrocarbon contamination.

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Chemical Defense of Marine Organisms Against Biofouling Explored with a Bacterial Adhesion Bioassay

Mercedes Camps, Linda Dombrowsky, Yannick Viano, Yves Blache, and Jean-François Briand

Abstract Marine biofouling is a complex process of colonization of submerged surfaces, whatever their natural or artificial nature. After a biochemical conditioning step, pioneer bacteria begin to settle on the surface to constitute a biofilm.

Progressively, microalgae, especially diatoms, and fungi contribute to the biofilm expansion and increase the complexity in terms of structure as well as functions. The next important step will be characterized by the settlement of macroalgae and macroinvertebrates (e.g., mussels and barnacles).

Nevertheless, some marine organisms are known as almost free of fouling. The main organisms of concern are macroalgae and invertebrates such as ascidians, gorgonians, corals, or sponges. One of the possible mechanisms responsible for this ability is the chemical defence via the production of secondary metabolites to limit the fouling process.

In order to identify natural purified metabolites from algae possibly involved in such processes, and then, potentially used as a biocide in antifouling paint, bioassays were achieved. These bioassays are based on the initial step of biofouling. Some pioneer bacteria were used as target organisms. The inhibition of in vitro monospecific bacterial biofilm formation was evaluated in microplates through fluorescence detection.

Several molecules including controls were tested simultaneously, which allowed us to verify the reliability of the assay.

In addition, a small quantity of molecules is required and the detection is faster with a multiwell plate reader.

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1 Introduction

Biofouling is a clearly defined colonization process of hard substrata mainly observed in aquatic ecosystems. Hard substrata may be artificial or natural. In the latter case, if the substratum is living, the phenomenon is known as "epibiosis." Despite the high variability of environmental conditions and the wide diversity of organisms involved, a similar overall sequence is observed: after an initial step during which the submerged surface undergoes biochemical conditioning, pioneer bacteria will attach irreversibly to the surface and progressively form a multiple species biofilm also involving microalgae (especially diatoms) and fungi (see synthesis in Wahl 1989; Fusetani 2004). This initial part of the fouling process is known as "microfouling". The subsequent attachment and growth of macroalgal spores or various macroinvertebrates larvae constitutes the "macrofouling."

Defense mechanisms, together with tolerance and avoidance processes, were developed by some macroorganisms to limit epibiosis, which can lead to damage of the surface, shading (in the case of phototrophs), or competition for the nutrients, and in the most severe cases, the death of the fouled organisms. Mechanical, physical, and chemical defenses correspond to a widespread adaptation in marine environments. The latter is based on the production of secondary metabolites toxic to the foulers.

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Antifouling is generally defined as a way to prevent foulers. In its broadest sense, it includes both the defensive bioprocesses to limit epibiosis and human technology applied to artificial submerged facilities. Particularly the colonization of vessel hulls leads to major economic losses due to both increased fuel consumption and the higher frequency of dry-docking operations required. Recent developments are due to the progressive banning of "historically" efficient, but environmentally harmful biocide-based paint products (Tributyl-tin, Irgarol, etc.). The new challenge is to find effective but environmentally friendly antifouling biocides. This objective has led researchers to look at the natural defense mechanisms of macroorganisms (e.g., DeNys and Steinberg 2002). The identification of effective biocides requires tools useful to screen the numerous candidates. This is why antifouling bioassays have been developed.

Overall marine algae represent a major source of bioactive secondary metabolites (Blunt et al. 2008), including antifouling activities (Bhadury and Wright 2004). Among them, brown algae have long been studied in the laboratory (e.g. Culioli et al. 2008). More than 80 percent of the purified molecules in the family come from *Dictyota* spp. (Phaeophyta, Dictyotales), and half of the brown algae secondary metabolites isolated are cyclic diterpenes isolated from *Dictyota* spp. As these species exhibited generally a low level of fouling and were low grazed, they appeared to be a potentially interesting model for a chemical defence mechanisms study. In addition, some antifouling bioassays were already achieved on diterpenes isolated from *Dictyota* spp. (Schmitt et al. 1998; Barbosa et al. 2007).

So this paper describes the antifouling activity of purified natural products from *Dictyota* sp., sampled in a lagoon near Toulon. The activity was evaluated through a bacterial adhesion assay, and the results discussed in the light of existing literature.

2 Materials and Methods

2.1 Sampling, Purification and Identification of Natural Products

The marine brown algae *Dictyota* sp. was collected in July 2007 in Le Brusc Lagoon (Mediterranean coast near Toulon) during a proliferation. The shade-dried material (57 g dry wt.) was extracted with mixtures of $CHCl_3$/MeOH (1:1 v/v; 3 × 800 mL) at room temperature. The concentrated extracts were combined to yield, after solvent removal, 5.9 g of a dark brown lipophilic extract. This extract was subjected to column chromatography on silica gel (Si60, 40–63 μm, Merck) with a solvent gradient from *n*-hexane to EtOAc and then from EtOAc to MeOH. The fraction eluted with 40% EtOAc in *n*-hexane was further purified by repeated semi-preparative high-performance liquid chromatography (Merck Purospher Star RP-18e 5 μm; 10 × 250 mm; 3 mL/min) using H_2O/MeCN (20:80) to afford a new compound, dictyol E (62 mg). From the fraction eluted with EtOAC/*n*-hexane (50:50), two known metabolites, hydroxycrenulide (25 mg) and dictyol C (18 mg), were also purified by reverse-phase HPLC with H_2O/MeCN (10:90) as eluent. Finally, the new metabolite DL9.2 (28 mg) and fucoxanthin were purified in the same manner from the fractions eluted respectively with EtOAC/*n*-hexane (60:40) and EtOAC/*n*-hexane (70:30). These compounds have been fully characterized on the basis of their spectroscopic data: 1D NMR(^{1}H, ^{13}C and DEPT), 2D NMR (COSY ^{1}H-^{1}H, NOESY ^{1}H-^{1}H, HSQC and HMBC), MS, IR, UV, and α_D.

Eleganolone and eleganediol were obtained from the brown alga *Bifurcaria bifurcata* (Fucales, Sargassaceae) harvested from Quiberon (Brittany, French Atlantic coast) using a previously described protocol (Culioli et al. 1999).

TBTO and capsaicin were purchased from Sigma-Aldrich.

2.2 Bacterial Adhesion Assay

Pseudoalteromonas sp. D41 was grown on VNSS (Vaatanen Nine Salt solution) at 25°C and sampled in the stationary phase. After centrifugation, cells were suspended in sterile artificial seawater (ASW) to reach an optical density of 0.6 at 600 nm. Then 200 μL of ASW was inoculated on the border-row wells of the microplate (sterile black polystyrene NUNC, Bioblock, Illkirch, France) and 100 μL of the bacterial suspension on other wells using an eight-channel pipette; 100 μL of dilution of standard biocides (TriButyl Tin Oxyd, TBTO & Capsaicin) and purified molecules were added to the latter wells. All the concentrations were tested in triplicates. Then 100μL of ASW was added to six wells to constitute the bacterial adhesion control. After 15 h, the nonadhered bacteria were eliminated by three successive hand washings (36 g L^{-1} sterile NaCl solution). The adhered bacteria were fixed for 1 h 30 min at 4°C with 36 g L^{-1} sterile NaCl solution containing 2% formaldehyde, then stained (20 min) by adding 200 μL of 4μg mL^{-1} DAPI (Sigma-Aldrich, St Quentin, France). The excess stain was removed by three hand washings (36 g L^{-1} NaCl solution). The DAPI was then solubilized in a 200 μL 95% ethanol (15 min under shaking). Fluorescence was measured (λ_{exc} = 380 nm, λ_{em} = 495 nm) using an Infinit 200 micro-plate fluorescence reader (TECAN, Lyon, France).

2.3 *Modelization and Statistical Analyses*

The dose-response curve fitting and the determination of the EC_{50} and hillslope for each molecule were achieved using GraphPad Prism software.

2.4 Results and Discussion

2.5 *Which Molecules Were Identified from Dictyota sp.?*

Ten molecules were purified and identified from the crude extract of *Dictyota* sp. Nine were cyclic diterpenes and the last one a classic pigment in the brown algae, the carotenoid Fucoxanthin. Among the cyclic diterpenes, four were tested: dictyol C and E, hydroxycrenulide (extented sesquiterpenes) and a new dolabellane (DL 9.2) (Fig. 1).

2.6 *What Are the Bioassay Characteristics?*

As reported in several publications, microbial communities result from the first step of the biofouling process and are often involved involved in the regulation

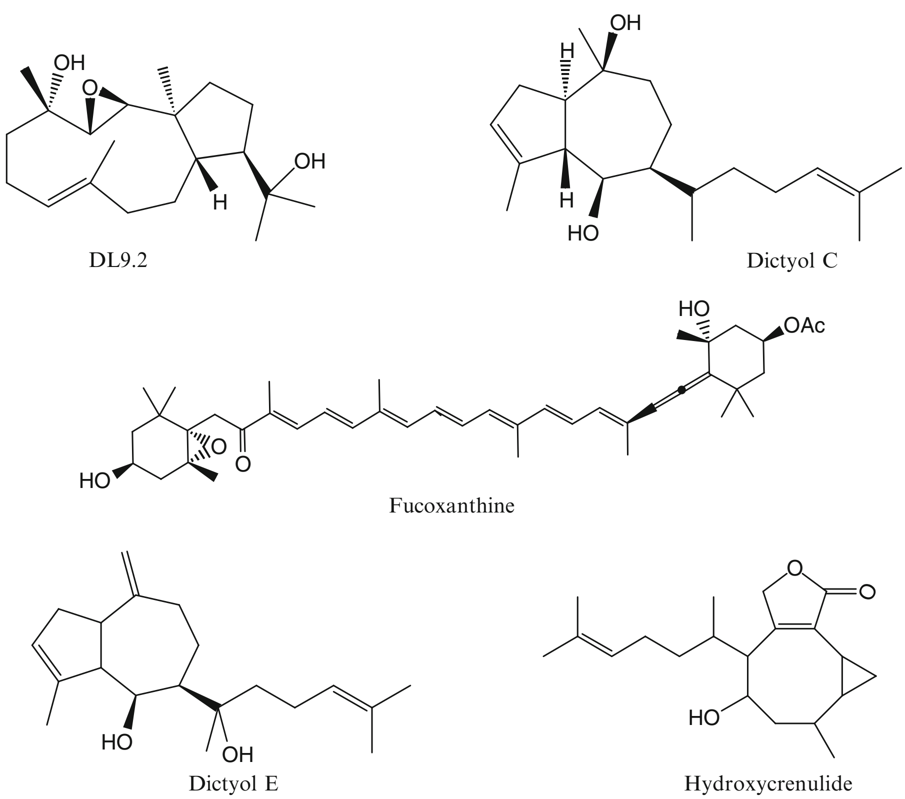

Fig. 1 Structure of the tested diterpenes

process of the macrofouling. Consequently, our argument is that inhibition or elimination of initial biofilms would probably, at least in some case, limit the settlement of macrofoulers. This is why we chose marine pioneer bacteria, isolated from the Bay of Brest (Brittany) by Ifremer (Leroy et al. 2007). We also isolated others pioneer bacteria from the bay of Toulon, and work is in progress to compare the sensitivity of strains from different origins. We chose to evaluate the adhesion process, which appeared to be more relevant than a classical antibacterial test.

To improve the reliability of the bioassay, microplates were used. Additional advantages are that a small quantity of molecules is required and detection is faster with a multiwell plate reader.

2.7 What About the Activity of Natural Products?

The activity of the two standard biocides (TBTO and capsaicin), the three molecules (two cyclic diterpenes and a carotenoid) isolated from *Dictyota* sp., and two linear diterpenes from *Bifurcaria bifurcata* was determined after the modelization of a sigmoid dose-response curve. Two parameters can be determined, both the EC_{50} and the hill slope. The linear diterpenes (eleganolone and elegandiol, Culioli et al. 1999) were added for comparison.

The reproducibility of the bioassay was established thanks to the determination of six EC_{50} and hill slopes for the TBTO, which exhibited, in addition, the lowest EC_{50} and the lower hill slope among the tested molecules. On the contrary, capsaicin had a low activity (Fig. 2 and Table 1).

Dictyol E and hydroxycrenulide had low activity (EC_{50} > 250 μM, data not shown). The new compound 9.2 and the fucoxanthin exhibited a similar moderate activity (EC_{50} nine times higher than TBTO but two times lower than capsaicin), as the linear diterpenes. Dictyol C was the most powerful molecule with an EC_{50} of 32 μM but a shallow hill slope in the case of capsaicin (Fig. 3 and Table 1).

2.8. What Can We Infer from These Activities?

One of the purified molecules, Dictyol C, showed a significant antifouling activity against pioneer bacteria. This secondary metabolite, which is present in

Table 1 The activity of the tested molecules

Biocides	*n*	EC_{50} (μM)	S.D.	Hill Slope	R^2
TBTO	6	9.2	5.9	−5.6	0.930
Capsaicin	1	149	–	−0.87	0.846
DL 9.2	1	78	–	−3.95	0.700
Dictyol C	1	32	–	−0.84	0.964
Fucoxanthin	1	74	–	−1.89	0.923
Eleganolone	1	75	–	−3.94	0.985
Eleganediol	1	61	–	−2.63	0.997

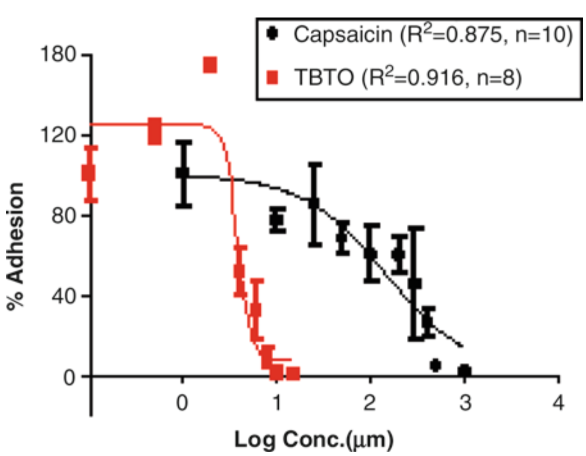

Fig. 2 Sigmoid dose-response curves of standard biocides

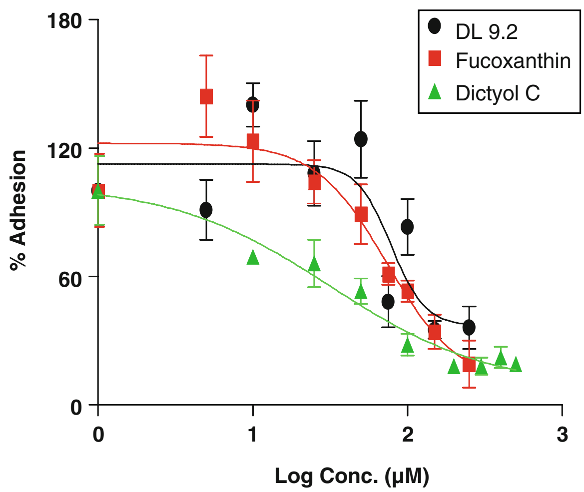

Fig. 3 Sigmoid dose-response curves of purified natural molecules (DL9.2, fucoxanthin and dictyol C) from *Dictyota* sp.

relatively high proportion in the seaweed *Dictyota* sp. studied, could be implicated in the chemical defence mechanisms of the species. It would be interesting to determine its seasonal variation and to try to connect it with the fouling sequence of the species. Geographical variations could also be investigated in order to precisely determine its specificity in terms of target organisms. TBTO was confirmed as a one of the most powerful biocides. But its toxicity against a wide range of organisms, including non-targeted ones, has led to its banning. On the contrary, capsaicin, which was reported as a potential antifoulant (Xu et al. 2005), displays a rather low potential according to our study.

Cyclic diterpenes were already studied for their antifouling properties. Antisettlement assays with larvae of barnacles (i.e., *Balanus amphitrite*) were conducted on two families of cyclic diterpenes, Renillafoulins (from *Renilla reniformis*, Octocorallia) (Keifer et al. 1986, Clare et al. 1999) and Kalihinenes (from *Acanthella cavernosa*, Porifera). Some of the latter exhibited lower EC_{50} than $CuSO_4$ (Okino et al. 1995, 1996; Hirota et al. 1996). As already said, cyclic diterpenes represent a high proportion of secondary metabolites in *Dictyota* spp. Dolabellanes (the same family as our new compound 9.2) from *Dictyota pfaffii* inhibited the settlement of *Perna perna* ("mussel test"). In addition, these laboratory results were coherent with field studies (*cf*., phytagel, where crude organic extracts of marine organisms are incorporated into hard, stable gels that serve as substrata, Barbosa et al. 2007). Dictyol E and B acetate, pachydictyol and dictyotal (extented sesquiterpenes from *Dictyota* spp.) were also reported as settlement inhibitors of three marine invertebrates (including *Bugula neritina*, Schmitt et al. (1998).

Our results showed a higher activity for the dictyol C compared to dictyol E. All these elements provide evidence that such cyclic diterpenes could be involved in the prevention of fouling in *Dictyota* spp.

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Temporal Evolution of Metals in the Two Most Industrialized and Densely Populated Gulfs of Greece, via Metal Accumulation by *Mytilus galloprovincialis*

Vassiliki-Angel Catsiki

Abstract The status and trend of chemical contamination of the coastal marine environment through the monitoring of sessile marine organisms is widely established (i.e., the mollusc bivalve *Mytilus galloprovincialis* often used in the "mussel-watch" programs). This paper aims to describe temporal trends of metals by the means of metal accumulation in *Mytilus galloprovincialis* at the most industrialized and densely populated gulfs in Greece.

Municipal, industrial wastes, and naval activities are the three main pollution sources responsible for the decline of the Saronikos (Central Aegean Sea) and Thermaikos (North Aegean Sea) Gulfs' water quality. In addition, three rivers flowing into Thermaikos contribute to the surcharge of its waters. Note that the coasts of the Thermaikos gulf and its adjacent area host the most extended and productive mussel aquacultures in Greece.

Mussels were collected from selected stations frequently:—four times per year from Saronikos Gulf (1986–2006) and one to two times per year from Thermaikos Gulf (1993–2007). They were monitored for the metals Cu, Cr, Ni, Zn, Fe, and Mn, and occasionally for Cd and Pb. Data were treated statistically in order to calculate trends.

Metal concentrations varied considerably during the studied period in both gulfs. Evolution patterns were not common for all metals.

In Saronikos Gulf, metal values in mussels reached their high levels during the periods 1988–1990 and 1994–1998. The operation (since 1996) of the Athens waste treatment plant together with a certain deindustrialization of the area is the probable cause of a decreasing trend in Ni, Cd, and Cu; Cr and Mn presented no trend, while Zn was increasing.

In the case of Thermaikos Gulf, metal values in mussels reached their high levels during the 2004–2007 period, while lower levels were noted during 1996–1998. An increasing trend was recorded for Cu, Cr, Ni, Zn, Fe, and Mn that for some metals were of 150% of magnitude, a fact that needs a special attention.

1 Introduction

The worldwide increasing trend of marine pollution requires control strategies and routine monitoring of contaminants in the marine environment, frequently performed through the monitoring of sessile marine organisms and especially mussels (i.e. "mussel-watch" programs – Langston and Spence 1995). Mussels are recognized as pollution bioindicator organisms because they accumulate pollutants in their tissues at elevated levels in relation to pollutant biological availability in the marine environment (Phillips 1976). The present paper aims to describe temporal trends of metals by the means of metal accumulation in mussel *Mytilus galloprovincialis* in the most industrialized and densely populated gulfs in Greece: Saronikos (Central Aegean Sea) and Thermaikos (North Aegean Sea – Fig. 1).

The two gulfs suffer from an important water quality decline caused by anthropogenic activities. In Saronikos Gulf, three main pollution sources are implicated: municipal and industrial wastes as well as naval activities. Note that the biggest port of Piraeus

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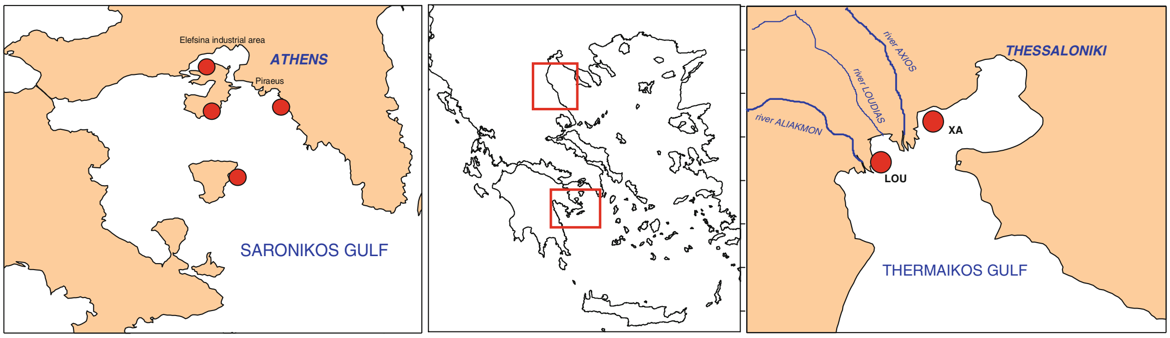

Fig. 1 Sampling stations in Saronikos and Thermaikos Gulfs

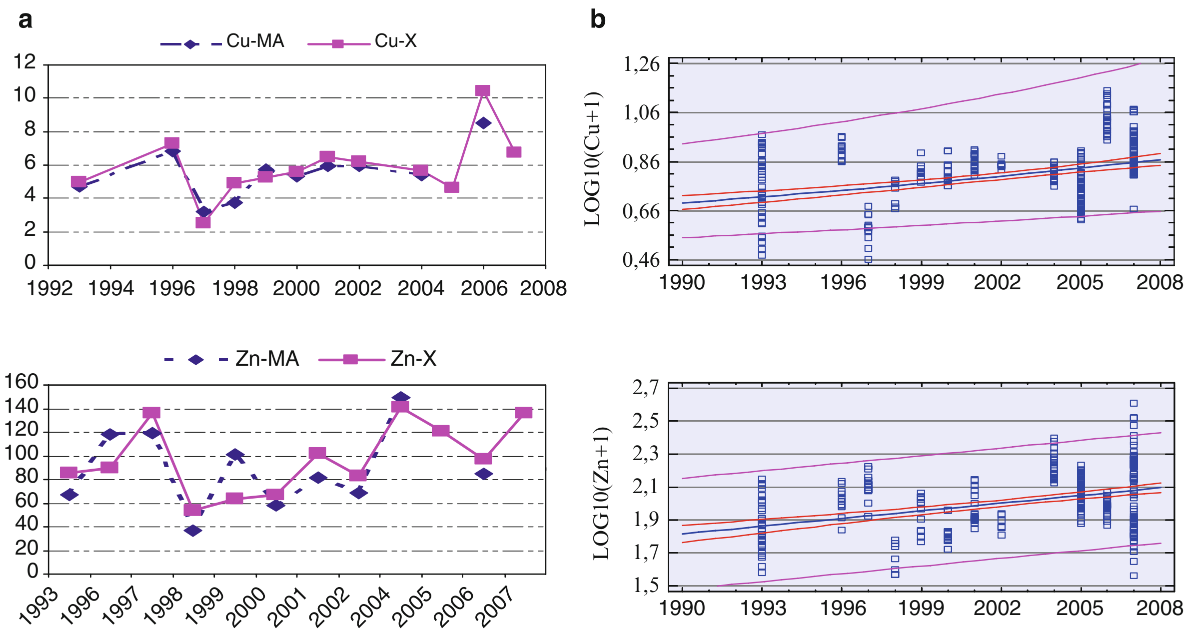

Fig. 3 (**a**) Evolution of Cu and Zn levels in mussels from Thermaikos Gulf, (**b**) Cu and Zn temporal trends

and the largest industrial zone of Elefsina, hosting almost 50% of the industrial activity, operate in the area. The short renewal time of the surface waters (reaching 2–3 months) limits the extent of pollution (Frilingos and Barbetseas 1990). In addition to the impact of municipal, industrial wastes and naval activities, Thermaikos gulf is influenced by river runoff (Axios, Loudias and Aliakmon rivers) and agricultural activities. Moreover extended mussel aquacultures (70% of the whole production of Greece) have been settled in the gulf.

2 Materials and Methods

Mytilus galloprovincialis samples were collected from selected stations (four times per year from Saronikos Gulf from 1986–2006 and one to two times per year for Thermaikos Gulf from 1993–2007). Four to six replicate samples during each sampling occasion were monitored for Cu, Cr, Ni, Zn, Fe, and Mn, and occasionally for Cd and Pb. The accuracy of the chemical analysis was tested with the simultaneous analysis of reference materials (DORM2). Data (on a dry weight

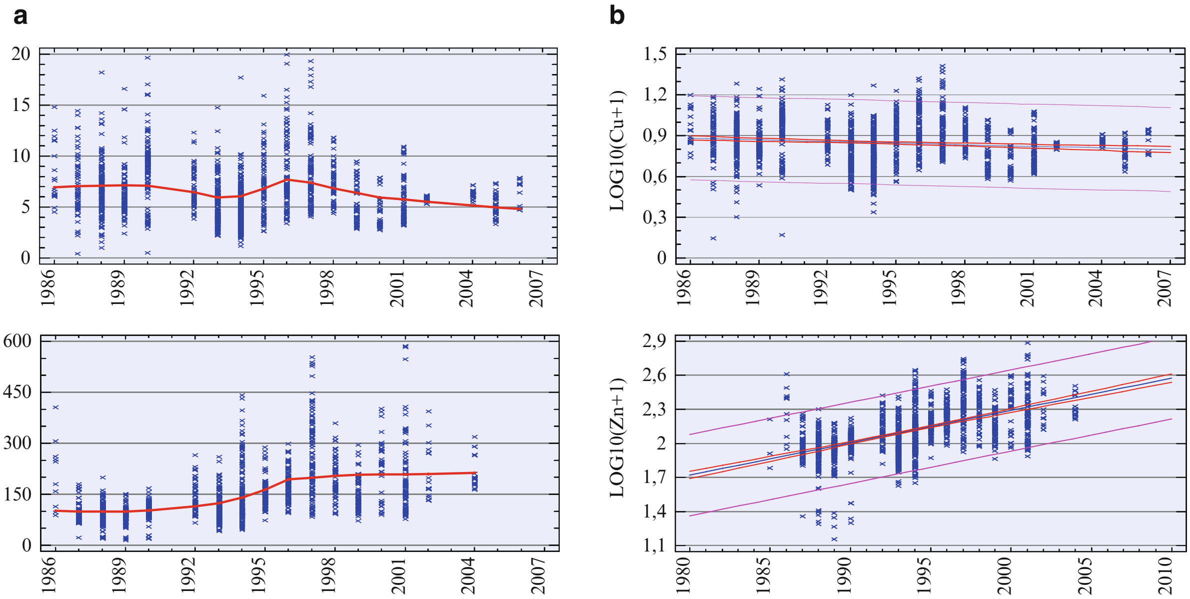

Fig. 2 (**a**) Evolution of Cu and Zn levels in mussels from Saronikos Gulf, (**b**) Cu and Zn temporal trends

basis) were treated statistically after logarithmic transformation in order to calculate trends using the STATGRAPHICS software package.

3 Results and Discussions

Metal concentrations varied considerably during the studied period in both gulfs. Evolution patterns were not common for all metals. In Saronikos Gulf, metal values in mussels reached their higher levels during the periods 1988–1990 and 1994–1998 (Fig. 2). The decreasing trend in Ni, Cd, and Cu are attributed to the operation since 1996 of the Athens waste treatment plant together with a certain deindustrialization of the area. However, Zn increased, probably due to the augmentation of the sewage quantity released from Saronikos (WWTP of Psytalia personal communication 2007) since this metal is abundant in the sewage. Cr and Mn presented no trend.

In the case of Thermaikos Gulf, metal values in mussels reached their high levels during the 2004–2007 period, while lower levels were noted during 1998 (Fig. 3). An increasing trend was recorded for Cu, Cr, Ni, Zn, Fe, and Mn, for some metals reaching a magnitude of 150%, which should be seriously taken into account by decision makers. Although waste treatment operates for the Thermaikos Gulf, it does not seem to reduce metal pollution in the Gulf effectively.

The above-mentioned mussel-watch programs effectively detected trends in metal concentrations and provided information to stakeholders in order to evaluate their control measures against pollution.

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Clipperton, a Meromictic Lagoon

Loïc Charpy, M. Rodier, and G. Sarazin

Abstract In 2005, we participated in the Jean-Louis Etienne expedition to investigate the lagoon of Clipperton Island (10°17′N 109°13′W) and the surrounding oceanic waters. Since 3,000 years ago, the date of the first registered closure, the lagoon has suffered a succession of reopenings. Its actual situation, a meromictic lake, seems to have been the same for 150 years. Brackish waters of the upper layer (0–10 m) are oxygenated, while salted waters of the deep layer (>20 m) are anoxic. The transition layer between brackish and salted waters is characterized by a temperature inversion of 1.6°C. Mean DIN (dissolved inorganic nitrogen) (0.39 ± 0.08 µM) and SRP (soluble reactive phosphate) (0.13 ± 0.01 µM) concentrations in the upper layer of Clipperton Lagoon were close to the values reported for other French Polynesian atoll lagoons. Taking into account the methodological problems of previous measurements, the physical characteristics of the lagoon do not seem to have changed significantly since the last expeditions (1980).

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1 Introduction

Clipperton Island (10°17′N 109°13′W) is the only coral atoll in the eastern Pacific. This ring-shaped atoll, completely enclosing a lagoon of 4 km^2, is one of the most remarkable models of biogeochemical processes known. This atoll is also a nesting site for thousands of seabirds, resulting in a net input of nutrients into the atoll from their droppings. Clipperton, now uninhabited, has been the subject of relatively little scientific investigation, especially biogeochemical studies. The main part of the lagoon is 2–5 m deep (Jost 2005) and is colonized by dense beds of macrophytes (Couté and Garrouste 2009). The lagoon also has three deep basins with stagnant waters at the bottom (Niaussat 1986). In these basins, Carsin et al. (1985) and Niaussat (1986) described a strong halocline at a depth of 10 m with brackish waters at the surface. The upper layer is oxygenated, whereas the deep saltwater layer is anoxic and contains high concentrations of hydrogen sulfide (H_2S).

In 2005 we took part in the Jean-Louis Étienne expedition to study the lagoon of Clipperton Island. The objective of this intensive sampling program was to extend knowledge of the physical and chemical structure of the lagoon, with a special focus on the deep basins.

Based on these new data, we also assess the evolution of the lagoon's status since the last scientific expeditions.

2 Materials and Methods

We sampled the whole lagoon of Clipperton from 8 February to 4 March 2005 (Fig. 1). Water samples were collected with a Niskin bottle (5 L) at 0, 5, 10, and 20 m in deep basins and at 0 m at the shallower stations.

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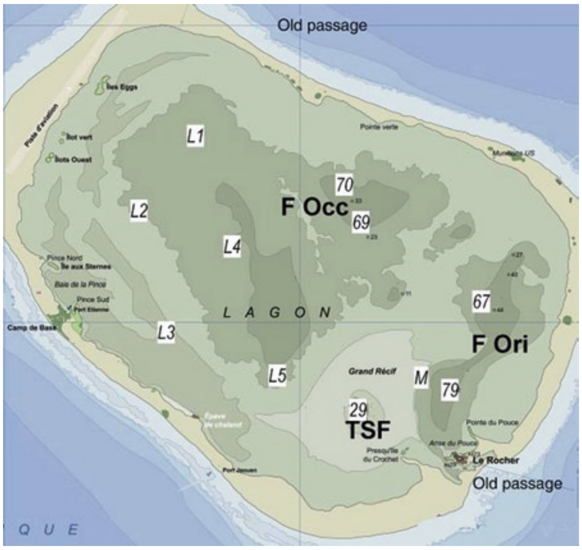

Fig. 1 Sampling stations

Bathymetric records were taken by manual soundings and depth finder. Salinity, temperature, pH, and dissolved oxygen were measured using a YSI 600 probe. The pH probe was calibrated using NBS (NIST) buffers.

Samples for soluble hydrogen sulfide (Total $\sum H_2S = [H_2S] + [HS^-]$) were stored in ampules and analyzed by colorimetry using a Merck Spectroquant™ kit in the "Laboratoire de Géochimie des eaux" of Paris 7 university.

Ammonium (NH_4^+) and soluble reactive phosphate (SRP) were analyzed immediately after sampling. Nitrate (NO_3^-) was analyzed after the expedition on $HgCl_2$-preserved samples (Kattner 1999). Ammonium concentrations were determined with a Turner Design TD-700, using the fluorometric and *o*-phthaldialdehyde method described in Holmes et al. (1999). Nitrate concentrations were determined by colorimetry using a Technicon® Auto-analyzer II and standard techniques (Strickland and Parsons 1972). In the anoxic layer, results are given only as dissolved inorganic nitrogen (DIN = $NH_4^+ + NO_3^-$) because the classical sampling method used for all of the samples may have created artifacts due to the contact with the oxygen from air. Soluble reactive phosphate concentrations were measured manually with a Cecil® CE 1011 spectrophotometer (cell length 10 cm), using the molybdenum blue reaction (Murphy and Riley 1962).

3 Results

During the 2005 expedition, the bathymetry of the lagoon was partially recorded. The western deep basin (F Occ) was 33 m deep, and the eastern basin (F Ori) was deeper (44 m), close to the previous published values (Niaussat 1986). However, in the deepest basin, called "Trou sans fond" (TSF, perimeter 439 m), we never found a depth greater than 37 m, which is much less than the depth given on the old maps, namely 94 m.

Turbidity, temperature, salinity, pH, and dissolved oxygen concentrations were measured at 21 lagoonal stations. Examples of recorded profiles in the deep basins are shown in Fig. 2. The upper layer of the lagoon (0–10 m) was turbid with a Secchi disc value between 2 and 3 m and desalted with an average salinity value of 5.70 ± 0.01. This upper layer was well oxygenated, with maximum values of oxygen saturation (160%) at the surface at the close of day. The pH ranged from 7.13 to 9.95 with an average of 8.3 ± 0.3.

In the deep basins, the water column was stratified. A two-layer system has already been described by Niaussat (1986), but we found an additional very turbid intermediate layer at depths between 11 and 15 m. This turbidity was due to the accumulation of material in the pycnocline produced by a strong density gradient (Fig. 2), especially in the TSF. The temperature profiles in this intermediate layer were characterized by temperature anomalies in most cases with some differences from one basin to another. In the F Ori basin, the temperature increased from 26.6°C to 27.5°C at 15 m and then decreased further down. In the TSF, temperature increased from 25.7°C to 27.3°C at 15 m and then remained constant down to the bottom. In the F Occ this temperature anomaly was observed in only 50% of the profiles. Below the intermediate layer, the deep layer (20 m to the bottom) had an average salinity of 34.73 ± 0.03 and an average pH of 6.52 ± 0.27. This deep layer was an anoxic and H_2S-rich zone (3,785 µM H_2S in F Ori).

Nutrient data collected in the lagoon during the 2005 expedition are set out in Table 1. In the upper layers soluble reactive phosphate (SRP) concentrations ranged from 0.1 to 0.3 µM, with an average of 0.13 ± 0.01 µM. The mean concentration of dissolved inorganic nitrogen (DIN) was 0.39 ± 0.08 µM (0.24 ± 0.04 NH_4^+,

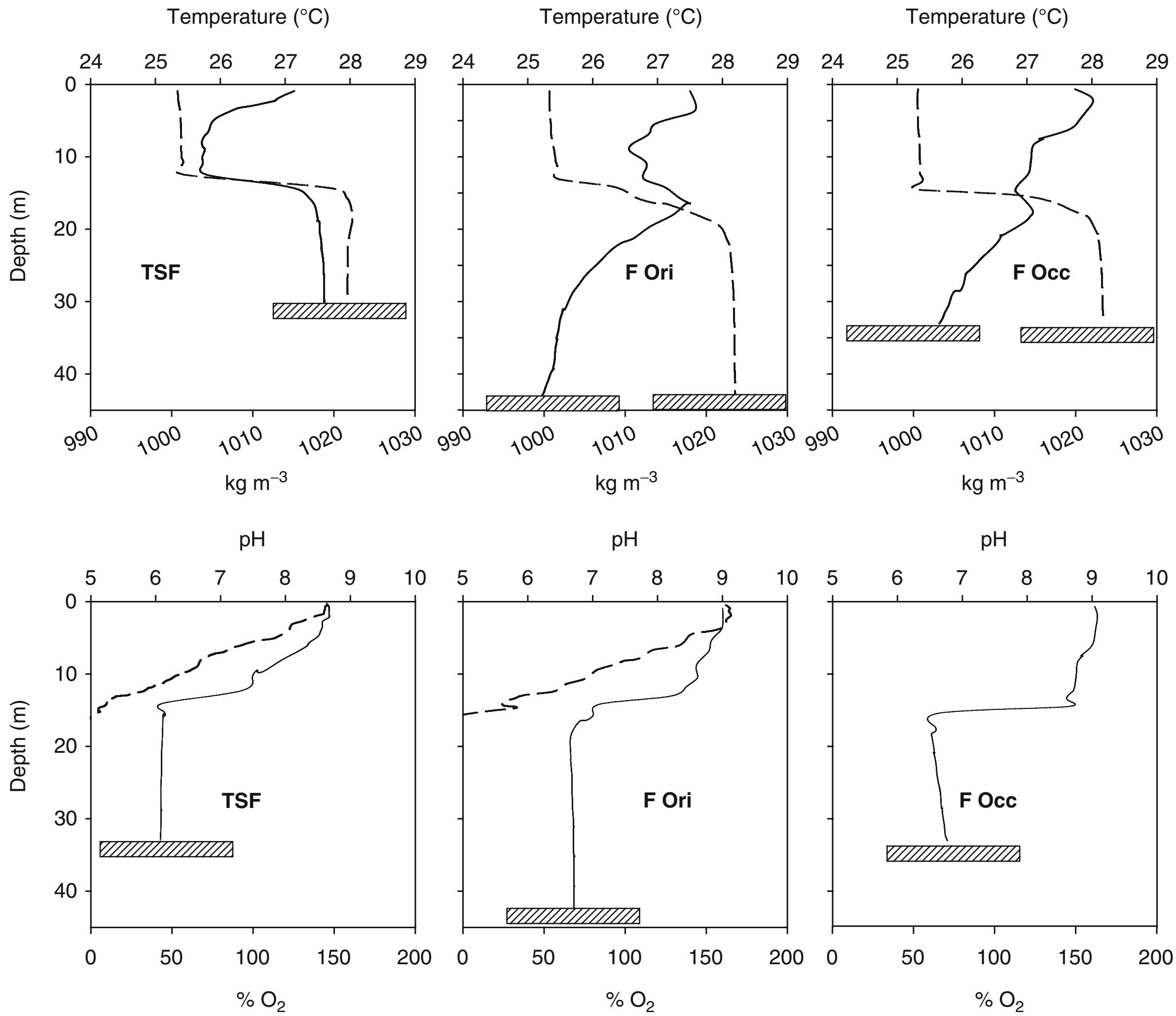

Fig. 2 Temperature (solid line), density (dotted line), pH (solid line), and % oxygen saturation (dotted line) profiles in the three lagoon basins

0.12 ± 0.05 NO_3^-, and 0.03 ± 0.01 NO_2^-) and the DIN/SRP ratio 3.69 ± 0.86. In the deeper, anoxic layer, the concentration of DIN increased strongly: DIN = 44.9 ± 1.1 μM. We could not measure the SRP concentration because of interference due to H_2S in the colorimetric method (Grasshoff et al. 1983).

4 Discussion

Knowing the date of the closure of the lagoon is important to gain an understanding of the modifications of the lagoon structure and functioning. The date of the last closure is still uncertain, but can be estimated from sailors' reports and maps. The first map, drawn by Sir Edward Belcher in 1839, showed two passages: one at the NE and the other at the SE end of the island. These passages were no longer shown on the maps drawn by Sir Hennig Master in 1849 and Le Coat de Kerveguen in 1858. However, in 1858, the lagoonal waters were still salty. Three years later, in 1861, the American officer Griswold reported that the lagoon water was fresh and drinkable. This observation was not invalidated by any of the scientific expeditions that explored this part of Pacific from 1898 onwards. Hence, the last closure of the marine lagoon must have occurred between 1839 and 1849, and its transformation into a lake completed by 1861. The separation between lagoon and ocean close to the SE old passage, which appears on the map drawn by Sir Edward Belcher in 1839, is very narrow, allowing some entry of seawater, as observed during the 2005 expedition.

In the three basins, we found a two-layered system, clearly separated by a strong density gradient implying no mixing between the brackish surface layer and the deeper anoxic waters (Fig. 2). Thus, the present observations confirm the meromictic character of the deep basins due to the last closure of the lagoon in around 1849. The use of a multiparameter probe during our expedition evidenced a third intermediate layer between brackish and saline waters that was not reported in previous expeditions, probably because adequate equipment was lacking. This layer within the density gradient presents a temperature anomaly with a temperature inversion between 1°C and 1.6°C at TSF and F Ori basins, extending down to the bottom in the TSF basin (Fig. 2). The anomaly of temperature around 17 m is inside the strong pycnocline. For that reason, waters at that depth are not affected by surface fluxes, and temperatures may remain quite constant throughout the year while the surface layer cools in winter. This may explain the temperature anomalies in the intermediate layer. In addition, a reduction process in this layer may lead to energy being released.

Nutrient surface concentrations in the lagoon were relatively low (DIN = 0.39 ± 0.08 µM and SRP = 0.13 ± 0.01 µM). These low values are unexpected in an ecosystem inhabited by very large colonies of seabirds and so enriched by bird droppings. Nevertheless, we may assume that the low DIN and SRP concentrations observed in the upper layer are the consequence of a strong nutrient uptake by primary producers of the lagoon as attested by the elevated total gross primary production reported by Murphy and Kremer (1983).

To assess evolution since previous scientific expeditions between 1938 and 1980, we compared our results with earlier data. The salinity range measured during our expedition in 2005 from the beginning of January to the end of March (5.3–6.3) was higher than the salinity ranges reported in previous expeditions (Table 2). The increase in salinity during the last decades could merely be due to an increase of entry of seawater in the SE part of the lagoon. The pH measured in deep basins in 2005 differed from previous values (Table 2). However, the

Table 1 Nutrient concentrations in Clipperton lagoon

Site	Depth (m)	SRP (µM)	NH_4^+	NO_3^-	NO_2^-	DIN
TSF	0.5	0.17	0.03	0.04	0.03	0.10
	5	0.19	0.62	0.05	0.02	0.69
	10	0.08	0.38	0.06	0.02	0.46
	20					32.43
F Ori	0.5	0.03	0.30	0.06	0.01	0.37
	5	0.08	0.30	0.07	0.04	0.41
	10	0.12	0.40	1.09	0.23	1.72
	20					88.40
	0.5	0.08	0.45	0.05	0.02	0.52
	5	0.12	0.41	0.05	0.02	0.48
	10	0.14	0.40	0.09	0.02	0.52
	20					28.58
F Occ	0.5	0.12	0.02	0.07	0.01	0.10
	5	0.14	0.03	0.06	0.01	0.10
	10	0.19	0.03	0.07	0.01	0.11
	20					
	0.5	0.14	0.16			
	5	0.18	0.18	0.04	0.01	0.23
	10	0.28	0.18	0.06	0.02	0.26
	20					30.13
L1	0.5	0.16	0.23	0.08	0.01	0.32
L2	0.5	0.14	0.20	0.06	0.01	0.27
L3	0.5	0.09	0.10	0.06	0.02	0.18
L4	0.5	0.11	0.14	0.08	0.01	0.23
L5	0.5	0.06	0.24	0.07	0.03	0.33

Table 2 Salinity and pH in the two layers

	0–10 m		>20 m		References
Date	Salinity	pH	Salinity	pH	
1967	3.7–4.0	ND	ND	ND	Niaussat 1986
1968	3.6–5.3	7	33.7–34.5	10	Ehrhardt 1976
1976	4.1–4.6	7.3–8.8	32.9–35.2	6.9–7.0	Niaussat 1986
1980	4.2–4.4	8.7–9.2	31.7–34.0	4	Carsin et al. 1985
2005	5.3–6.3	7.1–9.2	33–35.1	6.1–6.7	This study

1968: pH paper indicator

1976, 1980: Delayed analysis

2005: In situ probe pH_{NBS}

use of pH paper indicators in 1968 and the lag time between sampling and pH measurements in 1976 and 1980 lend the published data a degree of uncertainty. In this case, differences may be ascribed to the methods, which have been improved recently. Finally, the value of 3,785 μM H_2S measured in 2005 is of the same order of magnitude as the value of 3,120 μM given by Carsin et al. (1985) for the TSF deep layer.

There are very few data on nutrient concentrations in Clipperton. Murphy and Kremer (1983) reported higher DIN concentrations in the upper layer of the lagoon: 2.1 μM (1.5 NH_4^+, 0.2 NO_3^-, and 0.4 NO_2^-) and no detectable SRP. As already mentioned for the physical parameters, this discrepancy between past and present data may be ascribed to the improvement of or differences in the methods. Thus, Murphy and Kremer's high NH_4^+ values could be due to the lag between sampling and analysis, and their undetectable SRP to the lower sensitivity of their analytical method.

The meromictic character of the lagoon seems the same since previous expeditions even if the upper layer salinity increased, probably because of an increase in seawater entrance.

No comparable meromictic and lagoonal system is described in the literature. There are very few completely closed atolls. The only one that has been relatively well studied is Taiaro (Charpy and Blanchot 1998; Leis et al. 1998; Dufour and Berland 1999; Dufour et al. 1999, 2001; Andréfouët et al. 2001). However, the Taiaro lagoon has a high salinity (>42) from the surface to the bottom and is well oxygenated. Niau Lagoon is probably more similar to Clipperton, with an anoxic and H_2S-rich layer. However, only one unpublished preliminary study has been carried out.

The most important consequence of the closure of Clipperton lagoon since 1849 was the rapid desalination of the upper layer of the lagoon, leading to a meromictic system. This process was due to a positive precipitation-evaporation balance. Indeed, in the area of Clipperton, precipitation exceeds evaporation (Da Silva et al. 1994). The rapid salinity decrease has progressively caused the death of marine organisms and a shift from marine to brackish or freshwater communities. Thus, the shallow part of the lagoon was covered by a freshwater sea grass Potamogetonaceae species, *Potamogeton pectinatus* (Couté and Garrouste 2009).

The second consequence of the lagoon transformation to a meromictic ecosystem was the creation of an H_2S-rich saline anoxic layer below 20 m and the death of living oxygenic organisms at depth.

Fewer than 150 years were enough to transform the marine lagoon of Clipperton into a meromictic ecosystem and dramatically modify the lagoon community and functioning. This transformation is the consequence of closure and a positive precipitation-evaporation balance.

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Experimental Characterization of the Oceanic Water Exchanges in a Macro-tidal Lagoon

Cristèle Chevalier, Jean Luc Devenon, and Gilles Rougier

Abstract In the macro-tidal lagoons of the Indian Ocean, coral reefs can be momentarily submerged by water at high tide and partially emerged at low tide. This process contributes to lagoon and open sea exchanges, although the reefs are often considered as impervious and the water fluxes assumed to occur only through the passes. To gain insight into spatial variability of fluxes at a reasonable cost that moorings alone are not able to provide, we developed an original experimental approach combining small ship side-mounted ADCP (Acoustic Doppler Current Profiler) measurement following transects through passes and near reefs with more classical high-resolution ADCP moorings.

This new strategy of measurement was exemplified at the occasion of an experimental campaign on the Mayotte Lagoon and used afterward in Tulear Lagoon. The results of this experiment are presented. Particularly, first, it is shown how the mounted ADCP data are collected.

A specific tidal analysis methodology is then proposed to obtain the spatial variability of the tidal component of the current through the passes and above the reef. Then, all these analyses allow us to estimate the tidal-induced fluxes through the passes and above the reefs and to evaluate their respective part in water lagoon renewal.

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1 Introduction

Tropical lagoons are impacted by climatic changes, but also by direct anthropogenic activities. Water quality is directly related to the water renewal time, which is regulated by passes and by the coral reef barrier. Possible reef modifications (destruction induced by anthropic or weather-intense phenomena caused by climatic change, sudden cessation of the general conditions suitable for their improvement, water level increase) could have major consequences on the functioning of coral reef lagoons. To be able to forecast the reef tolerance versus modifications of the lagoon functioning conditions, analysis of the hydrodynamics regulated by these reef barriers and the possible evolution of their regulating effects is necessary.

In some meso or macro tidal lagoons of the Indian Ocean, the water that is over-running above the reef modulates strongly fluxes through passes during the tidal period. The Mayotte Lagoon and the Tulear Lagoon, located in the Mozambique Channel, are examples of these meso-tidal reef lagoons. The pronounced tidal character of these lagoons generates peculiar hydrodynamics.

In this study, we focused on the cross-reef tidal fluxes. Our aim was to determine the importance of the hydrodynamic control of the reef on the open sea/lagoon water exchanges as well as to compare fluxes occurring above the coral reef and through passes.

In order to gain insight into the spatial and temporal variability of these fluxes at a reasonable cost that several moorings would not enable to provide, we developed a specific experimental approach. This approach combined measurements obtained with a small ship side-mounted ADCP along transects with more classical high-resolution ADCP moorings. This experimental

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strategy will be presented first. Then, the specific methodology of the analysis will be described. In the third part, results will be presented. The tidal-induced fluxes through passes and above reefs, as well as their respective parts in water lagoon renewal will be particularly discussed.

2 Field Experiment

2.1 Study Site

The field observation was performed in the Tulear Lagoon and in the northeast lagoon of Mayotte. These two lagoons that are located in the Mozambique Channel and experience quite similar external tides dominated by semi-diurnal waves.

Even if their orientation is opposite, their morphology is similar. Their water body is about 20 km long and 2–10 km wide, opened at two passes of about 2–4 km. The north pass of the Mayotte Lagoon and the south pass of the Tulear Lagoon are the narrowest (2 km) and the deepest (50 m at the Mayotte lagoon; 20 m at the Tulear lagoon). These lagoons are partially closed to the Mozambique Channel by the coral reef. The Mayotte Reef is a well-developed fringing reef, whereas the Tulear one is mainly a sand- and mud-covered, dead reef. Even if the geological origins of these reefs, their external aspects, and their widths are different, their influences in the flow control seem to be the same. At high water, the reef is submerged (2–3 m at the Mayotte lagoon), whereas it is weakly immerged (50 cm at the Mayotte lagoon) or emerged (at the Tulear lagoon) at low tide.

However, the Mayotte Lagoon is deeper (about 20 m) than the Tulear one (about 10 m), and the nature of the entrance water varies according to the lagoon. At the Tulear Lagoon, the in-flow is usually constituted by water issued from the Mozambique Channel, but sometimes, due to external conditions such as wind, a small quantity of fresh water arising from the Fiherena and Onilahy Rivers might get inside. At the Mayotte Lagoon, the entrance water can come either from the Mozambique Channel (water getting in through the north pass and above the reef) or from the south lagoon (water getting in through the south pass).

2.2 Sensor Deployment

Our observations were performed from November 9, to 24, 2006, at the Mayotte Lagoon and from September 9 to October 12, 2007, at the Tulear Lagoon. To obtain both spatial and temporal variability of hydrodynamics, we deployed moorings on which ADCPs were installed, and we achieved ADCP measurements along the transect. Then, a field survey along the pass and, if possible, along the reef (at the Mayotte Lagoon) was conducted.

2.2.1 Moorings Deployment

At the Mayotte Lagoon, the north pass is divided into two channels (Fig. 1). One ADCP was been moored at each of them; the others were moored in the south pass and along the reef. At the Tulear Lagoon, the south pass is divided into two channels as well. One ADCP was moored in each of them and in the north pass; another was moored not exactly on the coral reef for safety reasons, but a little bit farther from it. Finally, in each lagoon and during each campaign, four ADCPs were deployed at the locations indicated in Fig. 1. Accounting for the shallow water depth and for accuracy considerations, ten 1-m bins with a dwell time 1 min were chosen for the vertical ADCP velocity profiles.

In order to facilitate the ADCP deployment network, and also to be able to get measurements in a region with very shallow water, ADCPs were set up in plastic containers, weighted with concrete or lead, and placed on the seabed. This kind of mooring can be deployed with two divers and a small boat. Even with this light mooring techniques and with a current velocity sometimes reaching up to 2 m/s, these moorings remained unperturbed and did not move during the experiments.

2.2.2 Description of Transect

Because of the shallowness, the boat used during these field campaigns had a quite small draught. Inspired by the usual method to measure currents in rivers, the ADCP was set up on the boat with tubulaa r stainless steel frame along the inflatable fender. With a low wavy sea and with a boat velocity of about 3 knots,

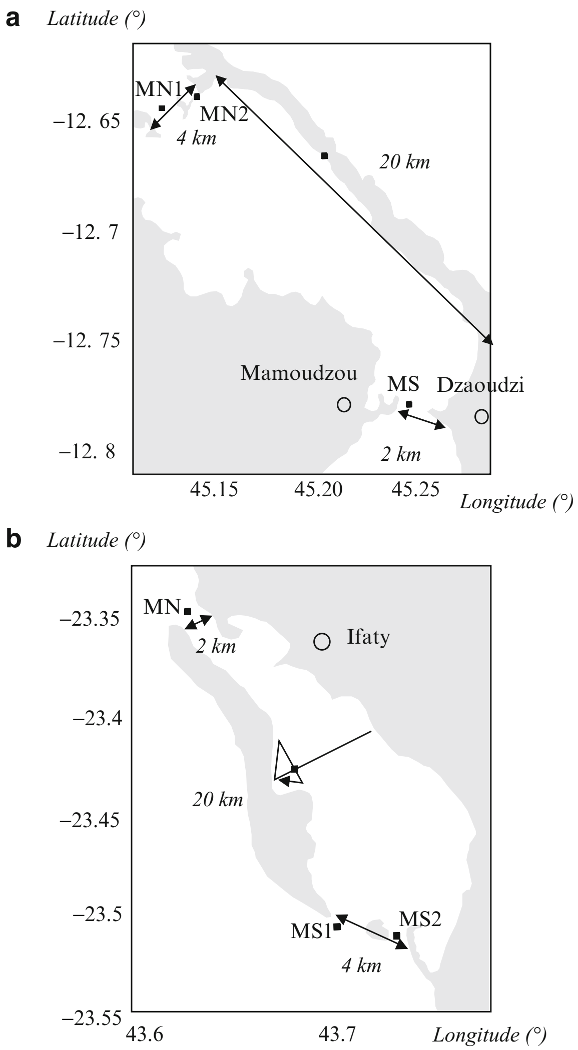

Fig. 1 Study site: Mayotte Lagoon (**a**) and Tulear Lagoon (**b**), and sensor deployment: (↔:Transect; ● Mooring)

velocity measurements were found by comparison in agreement to within 5 cm/s with those obtained with the ADCP moorings.

To associate these two kinds of velocity data, in order to quantify incoming and outgoing fluxes, transects were made crossing over moorings, either in passes or along reef barriers as presented in Fig. 1.

2.3 Data Description

Tides were found to be mainly semidiurnal for both sites with a spring and neap tide modulation (tidal waves M2, S2, and K1). The campaign duration was chosen to cover these two situations. During these experiments, the tidal amplitude in Mayotte, (respectively Tulear) was about 2 m (respectively 1.5 m) during the spring tide and 0.5 m (respectively 0.25 m) during the neap tide. Velocity time series in the middle of passes were obtained in the two lagoons.

For logistical raisons, the Mayotte campaign (10 days) could not last as long as the Tulear one (50 days). During the Mayotte campaign, the wind was weak and the sea was quite calm, while during the Tulear campaign, the wind was a little stronger with more waves. Nevertheless, transects could be performed despite not acquiring some data because of technical problems with one of our devices.

The measured velocities were almost the same from the bottom to the surface with only a weak phase lag. A few examples of velocity time series are presented Fig. 2. The main direction of the velocities was perpendicular to the pass and to the reef and alongshore orientated in the middle of the lagoon. However, at the north mooring and at the south mooring of the Mayotte Lagoon, we could observe a variability in the velocity direction between the ebb and the flow. Indeed, the velocity followed the coral reef respectively toward the southeast and the southwest at low tide, whereas, the influence of the last was weak (the velocity direction was directed towards the north) when the tide was high. This phenomenon was less pronounced for the Tulear Lagoon data.

The classical spectral analysis of the velocity confirms the influence of the mean semi-diurnal and diurnal tidal waves M2, S2, and K1, but also reveals the presence of other harmonic components, such as quarter diurnal waves (M4 for the Tulear Lagoon and MS4 for the Mayotte Lagoon). These quarter diurnal waves are probably the result of fluxes above the coral reef depending on the coral reef semi-diurnal immersion. Classically, the velocity reverses with the tide, but sometimes (during neap tide) the velocity is not seen to present an inversion, notably at the north mooring of the Tulear Lagoon and at the south mooring of the Mayotte Lagoon. This lack of inversion indicates the great influence of the non-tidal velocity component at these locations (maybe a South-North alongshore current along the Madagascar coast and an eddy in front of Dzaoudzi Island).

Finally, the side-mounted ADCP revealed the variability of the velocity caused by the reef concerning

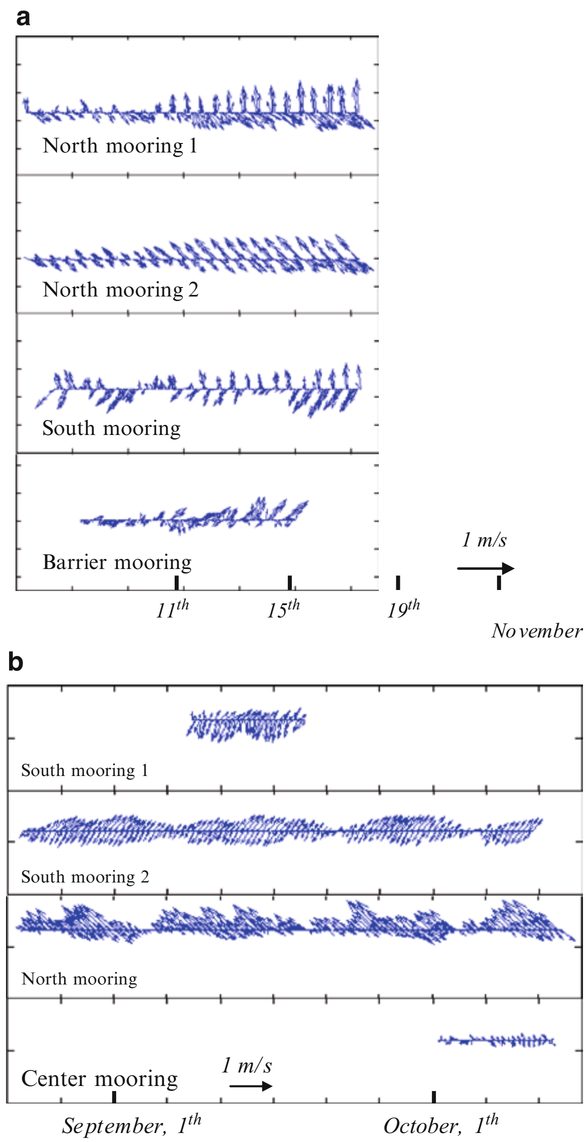

Fig. 2 Example of velocity stick plots obtained from ADCP moorings at the Mayotte Lagoon (**a**) and the Tulear Lagoon (**b**)

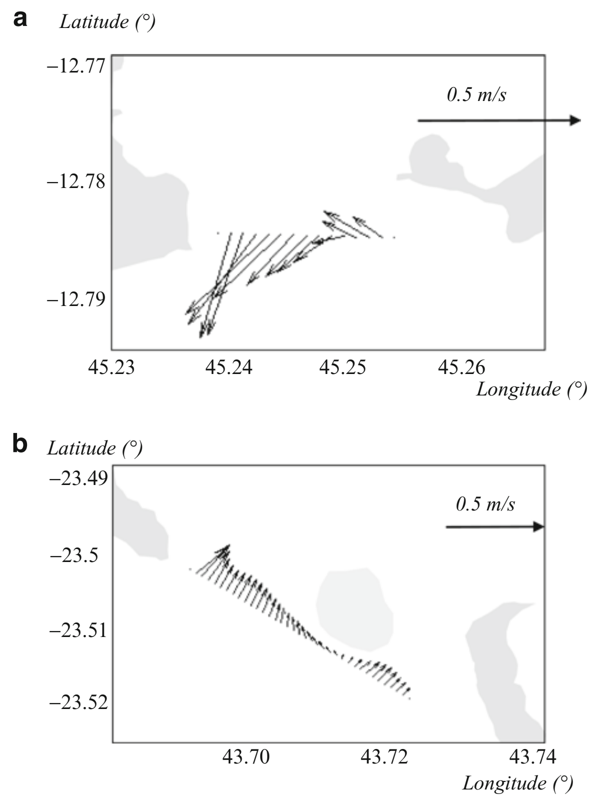

Fig. 3 Example of velocity measured along the south pass of Mayotte Lagoon, 14 November between 9 and 9.30 a.m., during high tide (**a**), and along the south pass of Tulear Lagoon, 6 October, between 8 and 9 a.m., 2 h after the low tide (**b**). The coral potatoes pictured in the middle of Tulear picture are partially submerged

the topography of the bottom (Fig. 3) and also the tidal evolution of the tidal signal.

2.4 Data Processing

In order to obtain accurate information about the spatial variations, the velocities had to be estimated at the same time for each transect. Indeed, the side-mounted ADCP mixes the spatial and temporal variability of the velocity because of the transect duration while the tidal signal varies. For this purpose, the temporal series of the mooring data can be used to obtain temporal information at each point all along the transect. This is made possible by using both the information of moored ADCP and the transect data.

As identical transect work was repeated during the campaign, many instantaneous data could be gathered at the same location. Then, at each transect location, the spectral amplitudes of the above identified tidal components, as well as the unresolved low frequency signals (period more than 1 day), were extrapolated from values available at the mooring location. These were developed according to a low degree polynomial functions of the distance to the mooring. The polynomial coefficients were obtained by a least square technique owing to the velocity measurements available along the transect. This enables obtaining the main signal all along the transect without necessarily fulfilling the Shannon criteria everywhere with regard to the higher tidal frequency encountered. Figure 4 illustrates

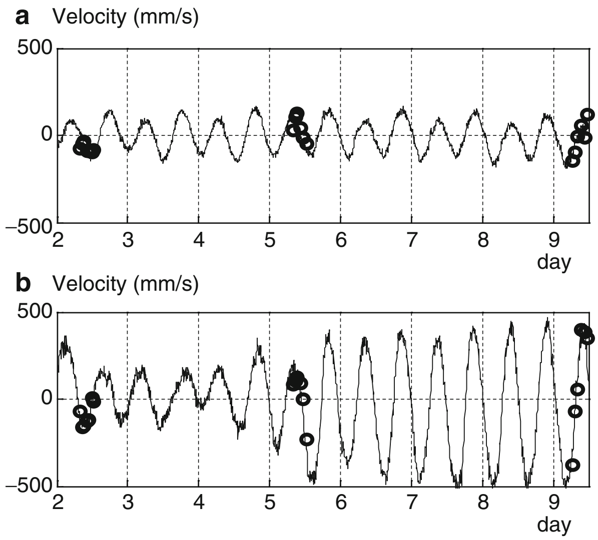

Fig. 4 Measured velocity (*black points*) and calculated velocity (*black line*) in the middle of the south pass of Tulear Lagoon. (**a**) Represents the east component and (**b**) the north component

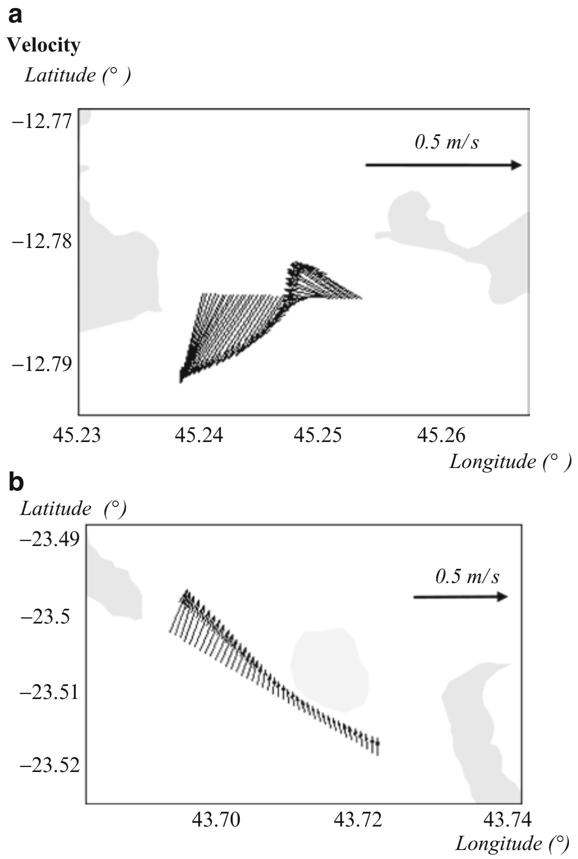

Fig. 5 Estimated velocity in the south pass of Mayotte, 14 November, at 9 a.m. (**a**), and along the south pass of Tulear Lagoon, 6 October, 8 a.m., 2 h after the low tide (**b**)

the so-estimated velocity with the in-situ data in a location in the Tulear Lagoon.

2.5 Velocity

The resulting velocities are in accordance with measurements. The adopted model for spatial and temporal variation is not bad since correlation coefficients are better than 0.9 and the residual errors (between 3 and 4 cm/s) are under the measurement error values. Figure 5 illustrates the velocity at the Mayotte Lagoon and at the Tulear Lagoon. The results provide the main spatial features of the velocity in passes. In the Tulear Lagoon passes, the velocity is almost homogenous and follows the bathymetry. On the contrary, at the Mayotte Lagoon, the processing revealed an eddy in front of the south pass. This corroborates the numerical study of Gourbesville and Thomassin (2000), and this could affect the estimate of bulk fluxes with regard to the one that would result from only one mooring observation. Consequently, spatial and temporal information about velocity enables us to improve flux estimation.

2.6 Fluxes

During the Mayotte campaign, a mean flux of about 3,000 m^3/s flowed in through the north pass, whereas a flux of 1,500 m^3/s went out of the lagoon through the south pass (Figs. 5 and 6). Neglecting other minor effects on density variation, the water volume conservation law implies that a flux of about of 1,500 m^3/s flows out of the lagoon above the coral reef. If we consider the velocity constant all along the coral reef, and set the value obtained at the reef mooring at each point of the entire reef, we obtain a rough approximation of the flux above the reef of around to 2,000 m^3/s, which is coherent with the above estimation.

At the Tulear Lagoon, a flux of 1,000 m^3/s for the north pass and 2,000 m^3/s for the south pass went out (Fig. 5). Thus, we can assume that a flux of about 3,000 m^3/s flows into the lagoon above the reef. In addition to other possible effects, this high level flux can be explained by the strong waves, which break on the coral reef.

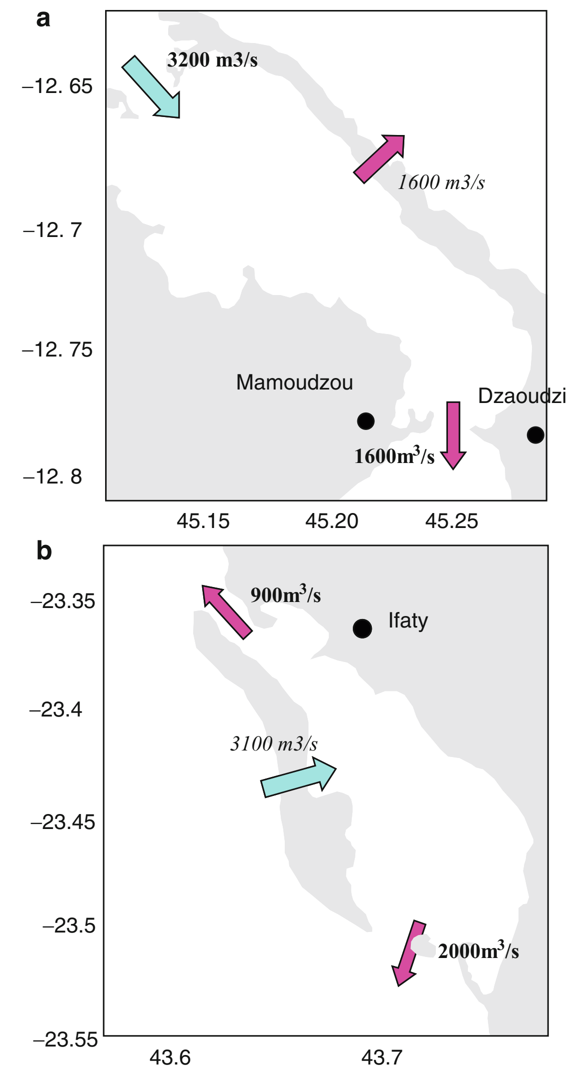

Fig. 6 Exchange flux in the Mayotte (**a**) and Tulear (**b**) Lagoons

These estimations point out the importance of the flux above the reef and enable us to calculate bulk water renewal times, which were respectively 7 days for the Mayotte Lagoon and 4 days for the Tulear Lagoon during our campaigns.

3 Conclusion

This study in two meso-tidal lagoons of the Mozambique channel reveals the great influence of the coral reef on the circulation. Probably due to the tidal wave interaction with the reef, a quarter diurnal tidal wave is created. Different orientations of the velocity could be noticed at the flood and at the ebb for the Mayotte Lagoon. This could be due to the variation of the vertical section offered to the water passage above the coral reef induced by the water level tidal variation. Finally, this processing shows that the flux above the reef reaches the same magnitude as the one through passes.

A new strategy of data acquisition and adapted data processing techniques to obtain spatial and temporal variations of current at a reasonably experimental cost has been developed. Finally, the velocity description at the boundary of lagoons will allow us to improve the numerical circulation model, which will be necessary to evaluate the residence time of water masses much more accurately.

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Modification of the Berre Lagoon Pelagic Ecosystem Since the 1980s

Floriane Delpy, Delphine Thibault-Botha, and François Carlotti

Abstract The Berre lagoon is a semi-enclosed basin Northeast of Marseilles (South of France) that has been largely impacted by human activities for several decades. The implantation in the northern part of the Saint Chamas power plant (deviation of the Durance River) in 1966 led to inputs from large freshwater bodies of water within this system.

The lagoon communicates with the Mediterranean Sea through the canal of Caronte. Up to 2005, the inputs of freshwater were largely uncontrolled, leading to strong seasonal variations (2 to 4.10^9 $m^{-3}.y^{-1}$) with minima observed in summer – early fall and maxima from fall through late spring. This led to important horizontal and vertical spatial as well as temporal variations in salinity, showing strong stratification taking place seasonally, particularly in the southern part where marine water forms the bottom layer.

Since September 2005, inputs of freshwater have been modified following the EU's compulsory requirement to reduce salinity variations. This has resulted in strong modifications of the ecosystem, such as disappearance of abundant species (i.e., rotifers), introduction of several marine species/genuses (i.e., chaetognaths, ctenophores), and alteration of trophic level interactions.

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1 Introduction

The Berre Lagoon (South of France, northwest of Marseilles) is a brackish water basin covering an area of 155 km^2. It communicates in the southwestern sector with the Mediterranean Sea through an artificial canal, the canal of Caronte, and receives four small natural rivers (the Cadière, the Arc, the Durançole, and the Touloubre), which represent only ~11% of the total freshwater influx. The EDF-derived canal (St Chamas power station) is the main contributor of freshwater into this coastal lagoon. The last intensive studies of this environment were done ~20 years ago (Kim 1982, 1985; Arfi 1991; Gaudy et al. 1995). The zooplankton community was then dominated by two species: the rotifer *Brachionus plicatilis* and the invasive copepod *Acartia tonsa*. The latter is an American species introduced through ballast waters from cargo ships.

This ecosystem is strongly influenced by the release of freshwater through the EDF-Saint Chamas power station. The amount of freshwater and silt used to show very large variations, which did not allow the ecosystem to reach a steady state. Since 2006, the freshwater release has been modified in order to reduce perturbations; the volume introduced into the lagoon will be regulated on a weekly basis and will not exceed 1.2 10^6 $m^3.year^{-1}$. Since then, a survey of the seasonal variations of salinity, temperature, populations of mussels, macroalgae, and benthic invertebrates has been planned. In addition, a study of the population of plankton is actually being conducted for a 1-year period. This aim of the project is to characterize the actual lagoon functioning and to estimate the evolution of the zooplankton community.

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2 Materials and Methods

The study is based on three permanent stations located in the North (SA1), in the center (SA2), and in the South (SA3). Physical (temperature and salinity) characteristics are recorded continuously. Phytoplankton and microzooplankton were sampled at the surface and at 1 m from the bottom. Vertical net tows were done at each station from the bottom to the surface for total abundance and species composition and for biomass.

3 Results and Discussion

Temperature and salinity data (surface and bottom) showed respectively weak or strong differences compared to 1985. Seasonal temperature variations are clear both on the surface and bottom layer, with summer temperatures around 24–25°C (6–8°C higher than in 1985), and low winter temperatures around 6–7°C. While in 1985 salinity never reached 20 in the surface layer and was at times as low as 5, 2007–2008, continued recording shows salinity over 20 most of the year (Fig. 1). Bottom salinity shows the same trend except for the most marine station. A phytoplankton community sample (June 2, 2008) shows a majority of marine species, in contrast to the sample from 1985. Already Arfi (1991) noted a decrease in the contribution of freshwater species. Freshwater organisms, like the Chlorophycea, which were important in the Northern station, have not been observed so far.

No clear spatial variations were observed, apart from a larger concentration of dinoflagellates at SA3, which could be linked to the changes in salinity, which allowed the establishment of a larger marine component in the lagoon. A large contribution of toxic or potentially toxic species, although none reached critical concentration for food consumption of shellfish, is observed. The presence of toxic species might be of concern for the growing mussel farming activities.

Overall phytoplankton and microzooplankton concentrations appeared in the same range as in previous studies. Microzooplankton were largely dominated by flagellates.

Since the 1980s, the Berre Lagoon has been characterized by a large abundance of a very few number of species. This semi-closed ecosystem was largely dominated by *Acartia tonsa*, a small copepod introduced through ballast waters from the Chesapeake Bay, by the rotifer *Brachionus plicatilis* and meroplankton larvae. In our study (Table 1), *A. tonsa* remained the most

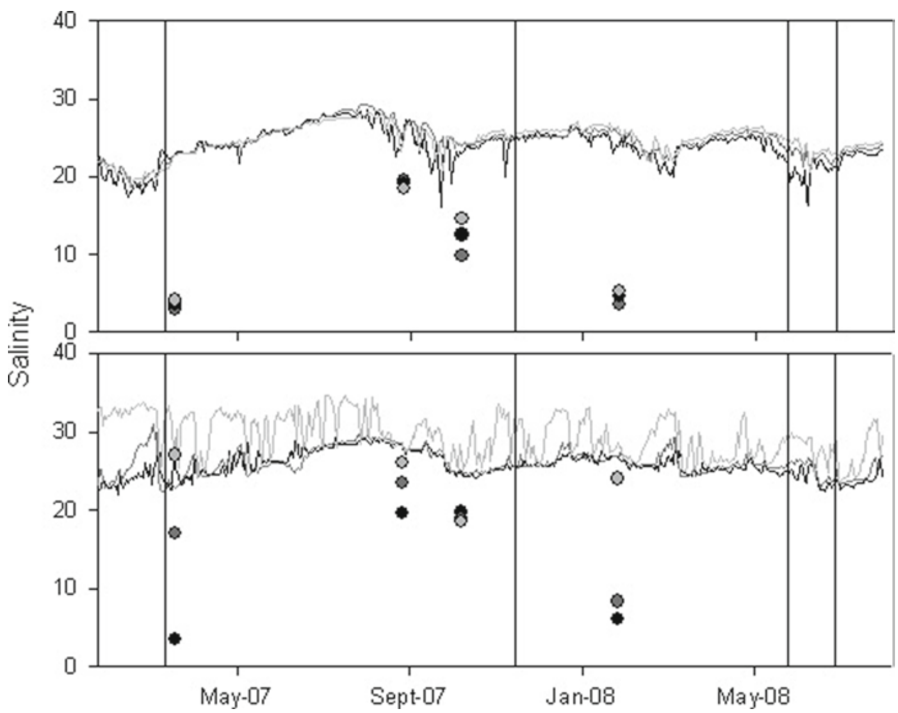

Fig. 1 Salinity daily mean at the surface(*top*) and at the bottom (*bottom*) from April 2007 to May 2008 for stations SA1 (black), SA2 (dark grey), and SA3 (light grey). Gaudy et al. (1995) data are presented (dots). Vertical lines indicate sampling dates for our study

Table 1 Zooplankton abundance (individuals. m^{-3}) per station and sampling dates

	10-May-07			*29-Nov-07*		*5-May-08*			*2-June-08*		
Abundance (individuals m^{-3})	*SA1*	*SA2*	*SA3*	*SA2*	*SA3*	*SA1*	*SA2*	*SA3*	*SA1*	*SA2*	*SA3*
Copepods	745	965	3,003	602	865	1,260	663	302	3,392	1,808	2,181
Acartia tonsa	744	965	3,003	576	847	22	91	46	2,556	983	1,683
Eggs	0	0	0	0	0	1,210	467	136	38	4	1
Nauplii	0	0	0	9	0	22	90	117	783	808	432
Gelatinous zooplankton	3	3	1	249	113	1	1	3	0	0	0
Other zooplankton	834	287	858	589	77	7,728	8,599	4,549	38,072	56,856	37,100
Total abundance	1,582	1,255	3,862	1,440	1,055	8,989	9,263	4,854	41,464	58,664	39,281

abundant species, but no rotifers have been observed. The difference in total abundance observed between 2007 and 2008 samples is mainly linked to the use of two different nets (80 and 200 μm), the difference being linked only to a better sampling of some meroplankton larvae. *A. tonsa*, the main copepod species, showed clear seasonal variations as shown by the percentage of the different development stages. The copepod community was dominated by eggs in May when the population showed stronger reproductive activities.

The population of *A. tonsa*, although well established, has shown a large decrease since 1985 (Gaudy et al. 1995): i.e., at SA3 30,850 individuals.m^{-3} reported in June 1985 and only 2,503 individuals.m^{-3} in 2008.

The other obvious change in the Berre System is the disappearance of the rotifer *B. plicatilis* (which reached over 20,000 individuals.m^{-3} [Gaudy et al. 1995]). It has been shown (Cervetto et al. 1995, 1999) that *A. tonsa* and *B. plicatilis* can tolerate large ranges of temperature (15–22°C) and salinity (1–72); therefore, the changes in the flow of freshwater cannot explain these differences. However, several marine species were more abundant (i.e., tintinnids) or appeared, such as predatory gelatinous zooplankton: i.e., jellyfish, chaetognaths, and ctenophores, which represent some real threats for the local species and economic activities. The appearance in fall 2006 of large numbers of the predator ctenophore *Mnemiopsis leidyi* caused great concern among the fishermen and aquaculture community. Blooms of the jellyfish *Aurelia aurita* are also reported very often in the Canal of Caronte. Still fairly large numbers (considering the type of net used) of fish roe and larvae are present.

The lagoon is therefore increasingly showing the characteristics of a marine lagoon (higher salinity) with new species coming from the Gulf of Fos as well as new introduced species. The presence of strong predators (i.e., gelatinous zooplankton) might in turn have a large impact on the structure of the ecosystem.

Once a marine lagoon before 1863, and with the deepening of the Canal of Caronte, then a brackish lagoon with localized low salinities with the opening of the power station in 1966, now the lagoon is looking for new economic development of aquaculture and sustainable fishery.

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Length-Weight Relationships and Reproduction of Three Coastal Sparidae (*Diplodus cervinus cervinus, Boops boops, and Spondyliosoma cantharus*) of the Eastern Coast of Algeria

Farid Derbal, Sarah Madache, Naima Boughamou, and Mohamed Hichem Kara

Abstract This study is part of the development of the eastern coast of Algeria. Its main objective is to increase the knowledge of the biology of the fish assemblages living in the shallow sea bodies with a major interest in species that inhabit remarkable ecosystems, such as the *Posidonia oceanica* seagrass beds. We introduce a synthesis of data on the length-weight relationships and sexual cycle of three coastal sparidae of economic interest, *Diplodus cervinus cervinus, Boops boops*, and *Spondyliosoma cantharus*.

1 Introduction

In the coastal region of eastern Algeria, artisanal fishermen are catching various species of Sparidae, which are of high commercial interest (200–700 Da/kg). This family is represented by ten genuses and 20 species frequently inhabiting various biotopes of the tray and the continental slopes (Derbal and Kara, 2001). The gathering of information concerning the biology of this family is necessary for the management of the exploited stock. This study presents some biological aspects of three coastal Sparidae, the zebra sea bream *Diplodus cervinus cervinus*, the bogue *Boops boops*, and the black sea bream *Spondyliosoma cantharus*.

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2 Materials and Methods

Fishes (203 *D. cervinus cervinus*, 684 *B. boops*, and 501 *S. cantharus*) were sampled along the eastern coast of Algeria between 2001 and 2005 (Fig. 1) using gillnets and crossbows. The relationship between the overall length (Lt) and the eviscerated weight (We) were established monthly and carried out using the program Fishparm 3.0 (Saïla et al. 1988). The various aspects of reproduction studied here are spawning period, length at first sexual maturity, structure of the population, and fecundity (Fig. 2).

3 Results

Within the total population studied, the length-weight relationship is majorant for *S. cantharus* and minorant for the other two species. Spawning occurs in spring for *S. cantharus*, at the beginning of the summer for *B. boops*, and during all the summer period for *D. cervinus cervinus*. In the latter, the values of the relative fecundity and diameter of ovocytes are comparatively low (Fr = 199 eggs, Do = 624 µm). However, this species has higher prolific capacity (Fa_{max} = 463,000 eggs). For the three species, the sex ratio is in favor of females (χ^2 Dcc = 2.22; χ^2 Bb = 6.82; χ^2 Sc = 243.4; $p < 0.05$). Lastly, the smallest size of first sexual maturity is recorded for the bogue (13.9 cm), and the largest one is recorded for the zebra sea bream (25 cm) (Table 1).

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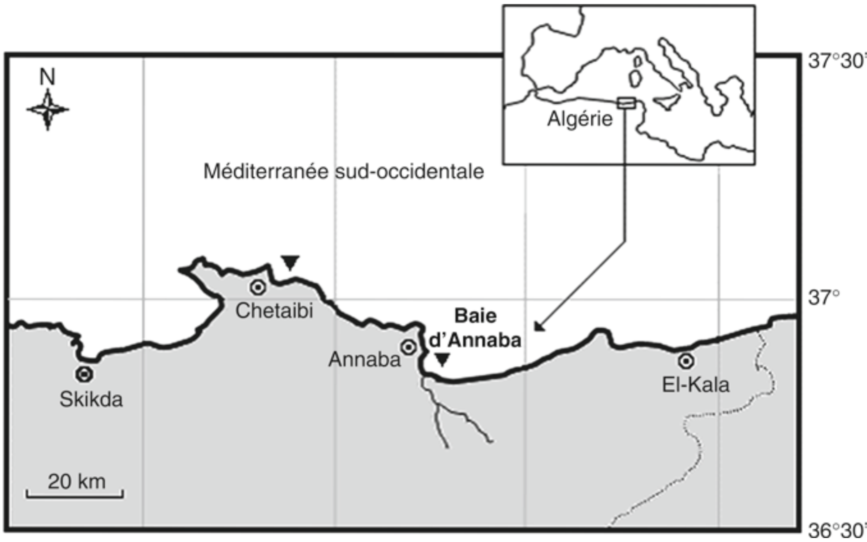

Fig. 1 Geographical limits of sampling sites (▼) of *Diplodus cervinus cervinus*, *Boops boops*, and *Spondyliosoma cantharus* of the eastern coast of Algeria

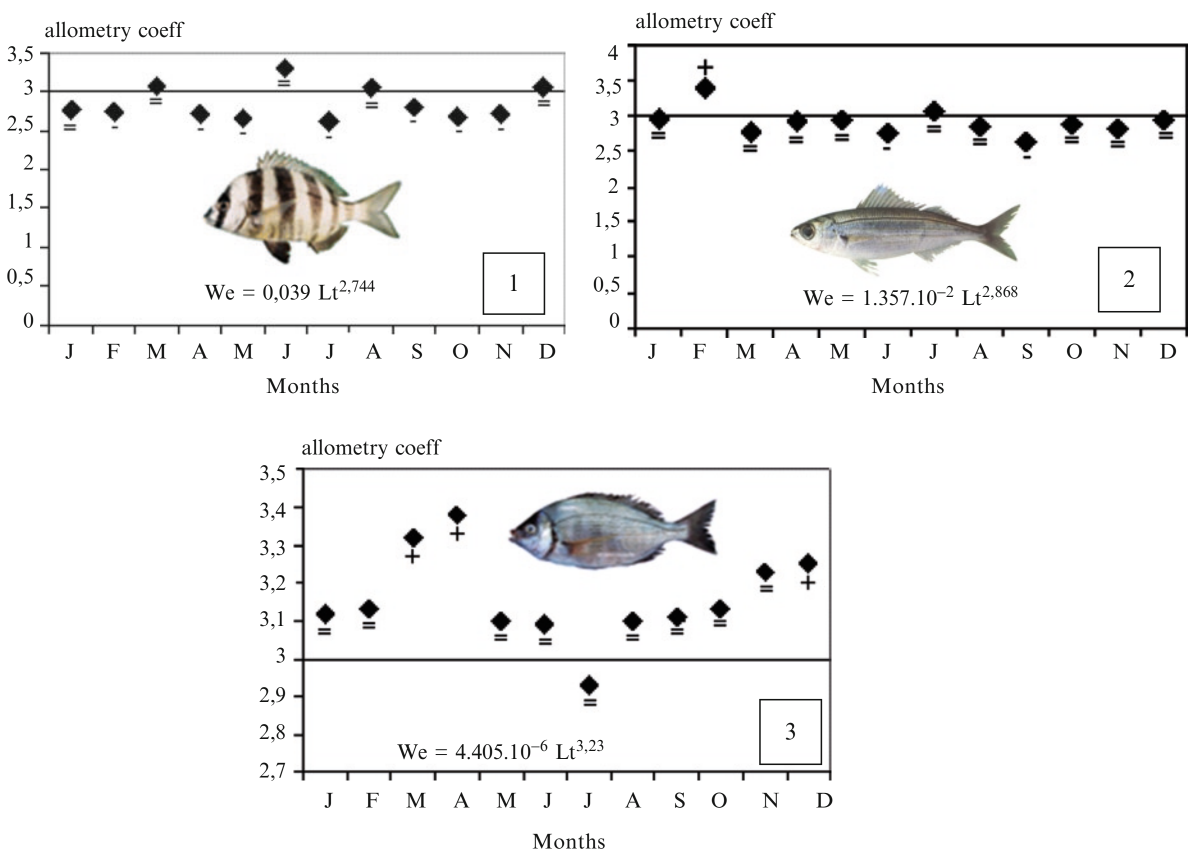

Fig. 2 Monthly variations in the allometry coefficient of the length-weight relationships for (1) *Diplodus cervinus cervinus*, (2) *Boops boops*, and (3) *Spondyliosoma cantharus* of the eastern coast of Algeria. =, isometry; +, majorant allometry; -, minorant allometry; We, weight; Lt, total length

Table 1 Spawning period, fecundity, structure of the population, and length at first sexual maturity of *Diplodus cervinus cervinus*, *Boops boops*, and *Spondyliosoma cantharus* from the eastern coast of Algeria

Species	Spawning period	Relative (Fr) and absolute (Fa) fecundity and diameter of eggs (Do)	Structure of the population (%)	Length at first sexual maturity (cm)
Diplodus cervinus cervinus	June–September	Fa = 130,000 – 463,000 eggs Fr = 199 eggs Do = 624 µm	F = 50.7 M = 20.8 J = 26.4 F + M = 2.1	25
Boops boops	May–June	Fa = 110,800 – 350,280 eggs Fr = 2,388 eggs Do = 950 µm	F = 54.5 M = 44.6 F + M = 0.9	13.9
Spondyliosoma cantharus	April–May	Fa = 100,320 – 222,040 eggs Fr = 478 eggs Do = 626 µm	F = 76.5 M = 12 M + F = 11.5	19.3

F: female, M: male, J: juveniles, F+M: hermaphrodit

4 Conclusion

The numerical importance of the females of the three species of Sparidae can be considered to be a sign of the dynamic imbalance of the exploited populations. *D. cervinus cervinus* is a species that deserves very special attention because it is becoming rare in numerous regions of Mediterranean Sea.

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Investigating and Assessing of the Quality of Seawater in the Marseille Coastal Zone: An Approach Using Lipid Class Biomarkers

Madeleine Goutx, Marie Duflos, Catherine Guigue, Jonathan Lucien, and Marc Tedetti

High dynamic anthropogenic activities in urban coastal areas are harming the aquatic environment, which may result in partially high contamination levels near urban wastewater discharges and corresponding coastal areas. However, the occurrence, fate, and distribution of numerous anthropogenic contaminants and xenobiotics are still poorly understood. In this chapter, we present strategies and methods focused on lipid biomarkers developed for characterizing and assessing the anthropogenic impact on Marseille's coastal environment.

In marine seawater, lipids mainly originate from natural sources. However, they are also concentrated in domestic and industrial waste waters. Thus, natural lipids may act as a sink for hydrophobic pollutants. Comprehensive studies of the distribution and dynamics of dissolved lipids have focused on the transport of lipid DOM by rivers, estuaries, and urban discharge at land/ocean interfaces. Only a few studies have reported on the distribution of dissolved lipid classes in the open sea (see Goutx et al. 2009 and references therein). Studies reporting on interactions between natural lipid variations and hydrophobic pollutant concentrations in seawater are scarce. Analytical limitations due to low lipid class concentrations in the open ocean are a major obstacle to the study of dissolved lipids in marine systems. The traditional methods of thin-layer or column chromatography require a significant amount of material, which can be problematic in field studies. The TLC/FID technique involving an "Iatroscan" apparatus allows detection of low concentrations of lipids (including total hydrocarbons) without prior splitting of the extract, which opens very interesting prospects by removing a major obstacle to the study of neutral and polar lipid classes in the marine environment. This analytical technique involves a qualitative separation of the lipid extract by thin-layer chromatography on Chromarods coupled to a quantification of separated compounds by flame ionization detection. We contributed significantly to the improvement of the Iatroscan lipid analytical protocol (Delmas et al. 1984; Goutx 1988; Gérin and Goutx 1993; Parrish et al. 1996; Striby et al. 1999). As this methodology requires a small extract for lipid class separation compared to current methodologies, it requires less seawater and therefore seemed to be a priori a technique of choice enabling the exhaustive sampling required for describing short temporal or spatial scale variability of chemical organic species in marine ecosystems submitted to human pressure (Fig. 1).

The bacterial response to lipid discharge in seawater may be assessed through the measurement of bacterial hydrolysis rates impacting on Iatroscan-separated acyl-lipids, using a specific and sensitive assay recently set up in our laboratory and based on the use of a tritium-labeled natural substrate (Bourguet et al. 2003). In addition, tests were conducted to estimate the bacterial fraction of the microbial community responsible for the measured activities. With the substrate ELF-palmitate, whose product of hydrolysis is insoluble (Nedoma et al. 2007), targeted cells (7%) were observed in pure cultures of lipase" + "bacteria" (Duflos et al. 2009). Iatroscan lipid analysis, lipase assay using radio labeled triglycerides as substrate,

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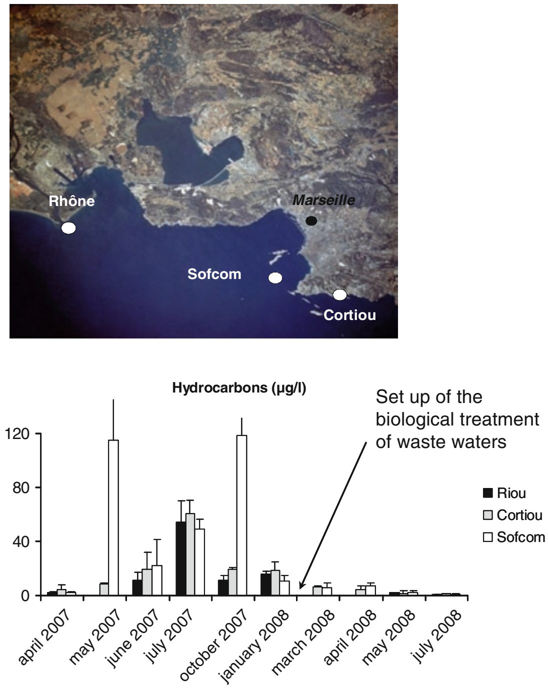

Fig. 1 Marseille Bay (*upper* panel): Time series of Iatroscan-measured total hydrocarbon concentrations (*lower* panel)

and lipase + cell labeling using ELF fluorochroms may be useful tools for a better understanding of the marine lipid cycling and identification of pathways through aquatic food webs.

However, while these methods are adapted to process study in the marine environment, high frequency samplings of chemical and biological parameters appear one of the major requirements for assessing the quality of seawater. Development of new sensors (Tedetti et al. 2009) and the AUVs carrying them is an essential challenge for future oceanographic studies and monitoring of the marine environment.

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Size Distributions of Low Molecular Weight Dicarboxylic Acids, Ketocarboxylic Acids and α-Dicarbonyls in the Marine Aerosols Collected over Okinawa Island, the Western North Pacific

Manuel Lazaar and Kimitaka Kawamura

Abstract Marine aerosols were collected at Cape Hedo, Okinawa, Japan (128.25°E, 26.87°N), using an Andersen middle volume impactor (M.V.I.) sampler at a flow rate of 100 L min^{-3} and pre-combusted quartz filter (80 mm in diameter).

The aerosols were studied for low molecular weight straight chain (C_2–C_{12}) and branched chain (C_4–C_6) dicarboxylic acids and related compounds using a capillary gas chromatography after dibutyl ester derivatization. A MVI sampler segregated aerosol particles at nine stages (<0.43, 0.43–0.65, 0.65–1.1, 1.1–2.1, 2.1–3.3, 3.3–4.7, 4.7–7, 7–11.3, and >11.3 µm in diameter). Homologous α,ω−dicarboxylic acids (C_2–C_{12}) and aromatic (phthalic) diacids were detected in the aerosols together with unsaturated dicarboxylic acids, ω-oxoacids (C_2–C_9), and α-dicarbonyls (glyoxal and methylglyoxal).

In all the size ranges, oxalic acid (C_2) was found as the most abundant dicarboxylic acid, followed by malonic (C_3) and succinic (C_4) acids. Glyoxylic (ωC_2) acid, 2-oxoethanoic acid, was the most abundant ketoacid followed by ω-oxobutanoic acid (ωC_4).

We found that all the water-soluble organic compounds are concentrated with peaks at fine particle sizes between 0.65 and 2.1µm, except for pyruvic acid, which showed two peaks in both fine and coarse sizes.

Oxalic acid (C_2, 4.4–70.6 ng m^{-3} with average of 23.9 ng m^{-3} and median of 15.5 ng m^{-3}) comprised 54–80% (average 67%) of the total diacid concentrations.

The maximal concentrations of small diacids at particle sizes of 0.65–2.1 µm suggest that they are produced by gas-to-particle conversion via photochemical oxidation of precursor organics and subsequent condensation on the pre-existing fine particles during long-range atmospheric transport.

They may also be produced by heterogeneous reactions in the atmospheric particles, including dusts and cloud droplets.

1 Introduction

In the western North Pacific, atmospheric transport of Asian dusts and pollutants is enhanced in spring by a strong westerly wind. Asian outflows control the air quality of the marine atmosphere, but also influence the marine environments by supplying the dusts and pollutants via dry and wet deposition.

Okinawa Island is located on the western rim of the North Pacific (ca. 600 km to Asian continent). We collected a series of size-segregated marine aerosols at Cape Hedo, northern edge of Okinawa, Japan (128.25°E, 26.87°N), and analyzed the aerosol samples for molecular compositions of water-soluble organic aerosols using a capillary GC and GC/MS. Here, we report the size distributions of carboxylic acids and dicarbonyls and discuss their characteristics in relation to the source regions and atmospheric photochemical processes.

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2 Samples and Methods

Size-segregated aerosol samples were collected using an eight-stage Andersen-type middle volume impactor (M.V.I.) sampler at a flow rate of 100 L min^{-3} and pre-combusted quartz filter (80 mm in diameter). Size cuts of the particles (back-up filter and eight-stages) were <0.43, 0.43–0.65, 0.65–1.1, 1.1–2.1, 2.1–3.3, 3.3–4.7, 4.7–7, 7–11.3, and >11.3 µm in diameter. The aerosols were extracted with organic-free pure water. The extracts were dried and then reacted with 14% BF_3/n-butanol to derive –COOH and –CHO groups to butyl esters and dibutoxy acetals, respectively. The ester and acetal fractions were determined using a capillary gas chromatography and GC/MS.

3 Results and Discussion

Homologous α,ω–dicarboxylic acids (C_2–C_{12}) and aromatic (phthalic) diacids were detected in the samples together with unsaturated dicarboxylic acids, ω-oxoacids (C_2–C_9), and α-dicarbonyls (glyoxal and methylglyoxal). In all the size ranges, oxalic acid (C_2) was found as the most abundant dicarboxylic acid, followed by malonic (C_3) and succinic (C_4) acids (Fig. 1). Glyoxylic (ωC_2) acid, 2-oxoethanoic acid, was the most abundant ketoacid followed by ω-oxobutanoic acid (ωC_4).

We found that all the water-soluble organic compounds are concentrated with peaks at fine particle sizes between 0.65 and 2.1µm (see Fig. 2 for oxalic acid), except for pyruvic acid, which showed two peaks in both fine and coarse sizes.

Oxalic acid (C_2, 4.4–70.6 ng m^{-3} with average of 23.9 ng m^{-3} and median of 15.5 ng m^{-3}) comprised 54–80% (average 67%) of the total diacid concentrations. The maximal concentrations of small diacids at particle sizes of 0.65–2.1 µm suggest that they are produced by gas-to-particle conversion via photochemical oxidation of precursor organics and subsequent condensation on the pre-existing fine particles during long-range atmospheric transport. They may also be produced by heterogeneous reactions in the atmospheric particles, including dusts and cloud droplets.

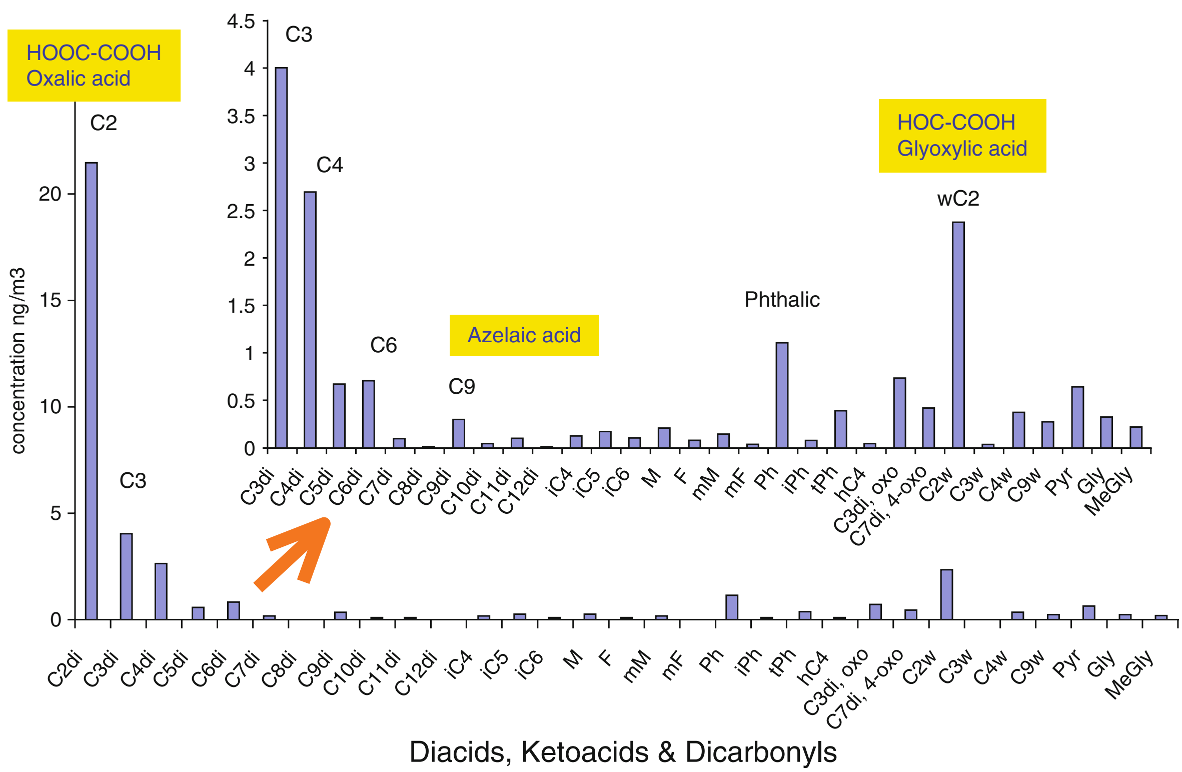

Fig. 1 Averaged molecular distributions of low molecular weight dicarboxylic acids, ketocarboxylic acids, and a-dicarbonyls in the marine aerosol particles collected from Cape Hedo, Okinawa

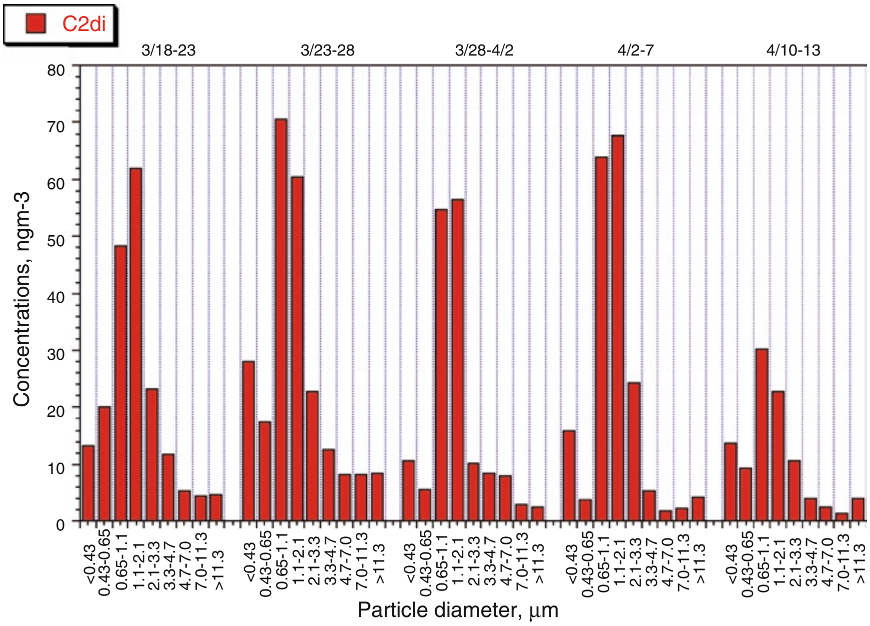

Fig. 2 Size distributions of oxalic acid in the marine aerosols collected from Cape Hedo, Okinawa, from March 18 to April 13, 2008

Dynamics of Two Greenhouse Gases, Methane and Nitrous Oxide, Along the Rhone River Plume (Gulf of Lions, Northwestern Mediterranean Sea)

Danielle Marty, Valérie Michotey, and Patricia Bonin

Abstract The concentrations of two greenhouse gases, nitrous oxide (N_2O) and methane (CH_4), and the bacterial processes involved in their production (nitrification and denitrification for N_2O, and methanogenesis for CH_4) were determined in a water column, along the Rhône River plume, in the Gulf of Lions, northwestern Mediterranean Sea. High concentrations of dissolved CH_4 and N_2O were recorded in the surface waters, up to 1,300 nM for CH_4 and 40 nM for N_2O, versus, 2–3 nM, and 5 nM in the open ocean surface waters. Along the Rhône River plume, the highest concentrations of CH_4 were detected near the river mouth, whereas N_2O remained present throughout the plume, up to the marine station. Bacterial production rates of N_2O and CH_4 indicated that the Rhône River plume constituted an area favorable for the development of nitrifiers, denitrifiers, and methanogens, promoting their activities in suspended particles.

Nonetheless, no direct relationship could be found between the concentration and the production of the biogases, suggesting that the dissolved gases were not necessarily produced in situ, in surface seawater, but could be produced in deep water or bottom sediment, and then transported to the surface waters.

The presence of extremely high concentrations of CH_4 and N_2O, in superficial seawater implies they can escape directly to the atmosphere; consequently, these near-shore waters enriched in greenhouse gases may play an important role in the atmospheric increase of both CH_4 and N_2O concentration, and thus in the global climate change.

1 Introduction

Among greenhouse gases, CO_2, at around 380 ppm in the atmosphere, is one of the major greenhouse gas, followed by CH_4, under 2 ppm, and N_2O, less than 1 ppm (IPCC 2007). Despite its low atmospheric mixing ratio (1.774 ppm) and short atmospheric residence time (about 10 years), CH_4 is considered as the most potent greenhouse gas after CO_2 due to its higher effectiveness (20–30 times) in absorbing infrared radiation, and its involvement in tropospheric and stratospheric chemistry (Wuebbles and Hayhoe 2002). N_2O, with a steady-state level at 319 ppb, is a climatically important trace gas because of its long life (180 years), its radiative impact about 200 times that of CO_2 on the molecular basis, and its role as a catalyst in reactions leading to the destruction of the stratospheric ozone layer (Nevison et al. 1999).

CH_4 and N_2O are two greenhouse gases showing several common characteristics. They are primarily produced by biogenic processes, which occur in natural and disturbed ecosystems. CH_4 and N_2O are trace constituents of seawater (2–3, and 5 nM, respectively); in contrast, supersaturations of these dissolved gases are observed in the shallow coastal zones (Capone 1991; Kiene 1991), which could contribute, respectively, to 75% and 60% of the global oceanic emissions of CH_4 and N_2O to the atmosphere (Ferrón et al. 2007).

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The aims of this study, which were carried out within the framework of the interdisciplinary European project METROMED (Dynamics of the MattEr TRansfer and BiOgeochemical Cycles: Their Modeling in Costal Systems of the MEDiterranean Sea), were to investigate the spatial and temporal variations of CH_4 and N_2O concentrations and intensity of bacterial processes of CH_4 production (methanogenesis) and N_2O production (denitrification and nitrification) in the water column along the Rhône River plume, in the Gulf of Lions, northwestern Mediterranean Sea.

2 Materials and Methods

In the northwestern Mediterranean Sea, the Gulf of Lions is an area of 1.7×10^4 km^2 that extends from the Provence region to the Pyrenees mountains. The Rhône River is the major source of suspended particulate matter in the Gulf of Lions, providing more than 80% of its total terrigenous inputs (Courp and Monaco 1990). Suspended and settling particles are distributed in nepheloid structures, distinguishable by their higher density of particles compared to the surrounding water. They form a well-defined multilayer system, including a thin, superficial nepheloid layer (SNL), spreading from the river mouth, and a benthic nepheloid layer (BNL), which extends over the greater part of the continental shelf. Such coastal plumes play an important role in the fluxes of materials between the estuaries and oceanic margins, and they are characterized by a distinct salinity structure and high biological production relative to ambient shelf water.

Water samples were taken along a transect, composed of nine stations, extending from the mouth of the Rhone River toward the open ocean, with three main stations: proximal station A (43 m depth) near the river mouth, distal station B (75 m depth) in the middle of the plume, and marine station C (95 m depth), out of the plume (Fig. 1). Along the nine stations of the transect, water samples were collected in SNL and BNL to determine salinity and temperature together with CH_4 and N_2O concentrations. At the three main stations, water samples were collected along vertical profiles, every 10 or 20 m for stations A or B and C, respectively. CH_4 and N_2O were measured in all the water samples, and the processes involved in their production were measured in SNL and BNL.

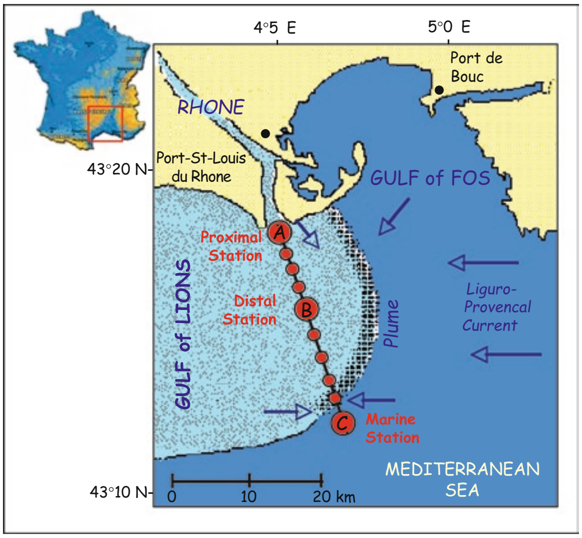

Fig. 1 Study area – transect along the Rhône River plume

The data obtained from the seven sampling campaigns were clustered as season set data.

3 Results and Discussion

3.1 Nitrous Oxide

Concentration and Production of N_2O (Figs. 2–5)
In all the area influenced by the Rhône River, the N_2O concentration ranged from 10 to 50 nM (Fig. 2). The highest values were found in the plume and the benthic nepheloid layer. In marine station C, N_2O concentrations were close to equilibrium with the atmosphere (about 5 nM).

The data showed supersaturation of about two to ten times throughout the salinity gradient; no conservative dilution pattern was observed, indicating the presence of bacterial production in the studied area.

Nitrous oxide concentrations were determined in the different stations throughout the year. There were significant differences between stations located in the surface nepheloid layer (StA-S, StB-S StC-S, Fig. 3a), with higher concentrations at station A ($p = 0.006$), but no significant differences between the stations in the bottom nepheloid layer (StA-B, StB-B, StC-B, Fig. 3a). The effect of the sampling time was not significant ($p = 0.524$, Fig. 3b). In contrast, except at the surface of the marine station, N_2O production rates were about

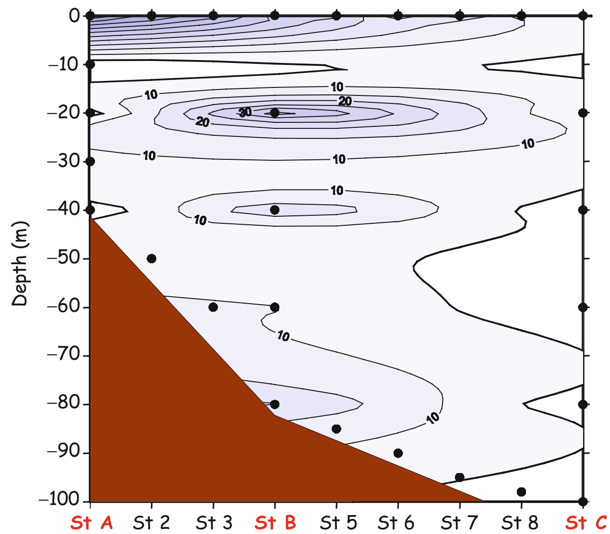

Fig. 2 N_2O distribution along the Rhône River plume transect: Station A to Station C

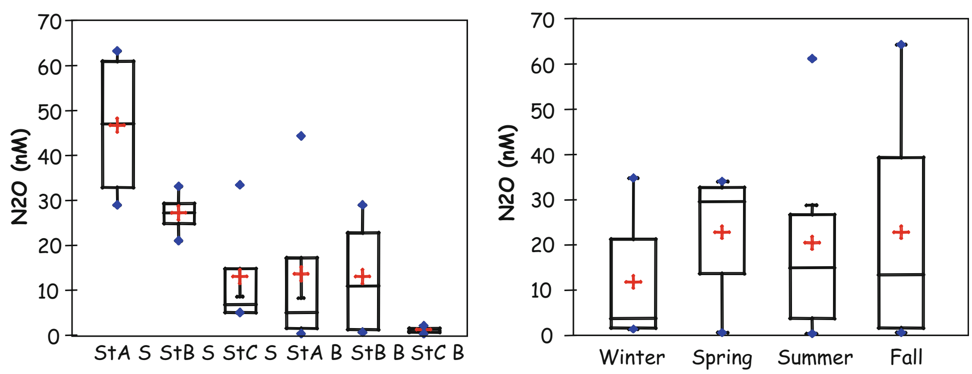

Fig. 3 Effect of sampling site and time on nitrous oxide distribution at stations A, B, and C (S = surface water and B = bottom water); (**a**): Sampling station effect; (**b**): sampling season effect

the same in all stations (Fig. 4a, $p = 0.760$), showing, however, a strong seasonal effect (Fig. 4b, $p = 0.004$).

Maximal N_2O production rates were observed during summer and spring, whereas the lowest values occurred during winter and fall months (Fig. 4b).

The production of N_2O may come from incomplete denitrification or nitrification processes. By using acetylene as inhibitor, we were able to characterize the contribution of each process in the production of N_2O (Bonin et al. 2002). Without the addition of acetylene, total N_2O production coming from the both processes can be measured, whereas in the presence of 10 Pa C_2H_2, nitrification is inhibited and N_2O becomes only from partial denitrification.

In almost all stations, whatever the sampling time, N_2O production could be measured showing the wide occurrence of N_2O production in estuaries (Fig. 5). At station A, near the Rhone River mouth, denitrification and nitrification were responsible for the N_2O production; denitrification, occurring probably in anoxic microniches within particulate material, was the main process. In other stations, in the surface layer, most of the N_2O was produced by nitrification, with the percentage of N_2O originating from nitrification increasing from the river mouth to the sea. In contrast, denitrification was the main process leading to N_2O accumulation in the bottom layers (Fig. 5).

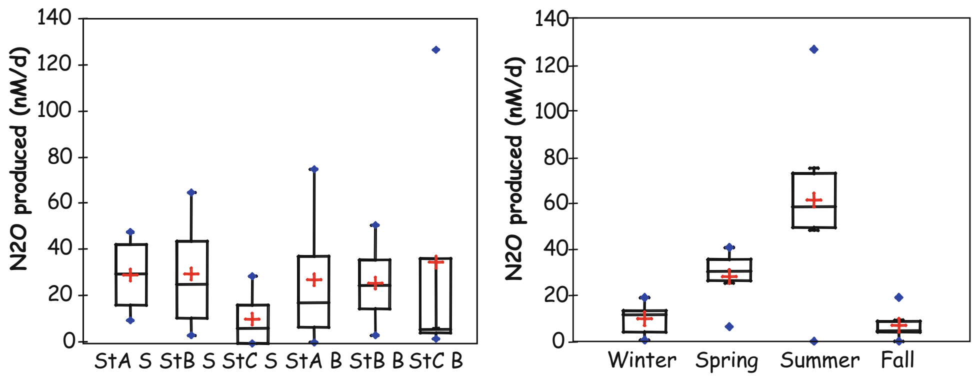

Fig. 4 Effect of sampling site and time on bacterial nitrous oxide production at stations A, B, and C (S= Surface water and B = bottom water); (**a**): sampling station effect; (**b**): sampling season effect

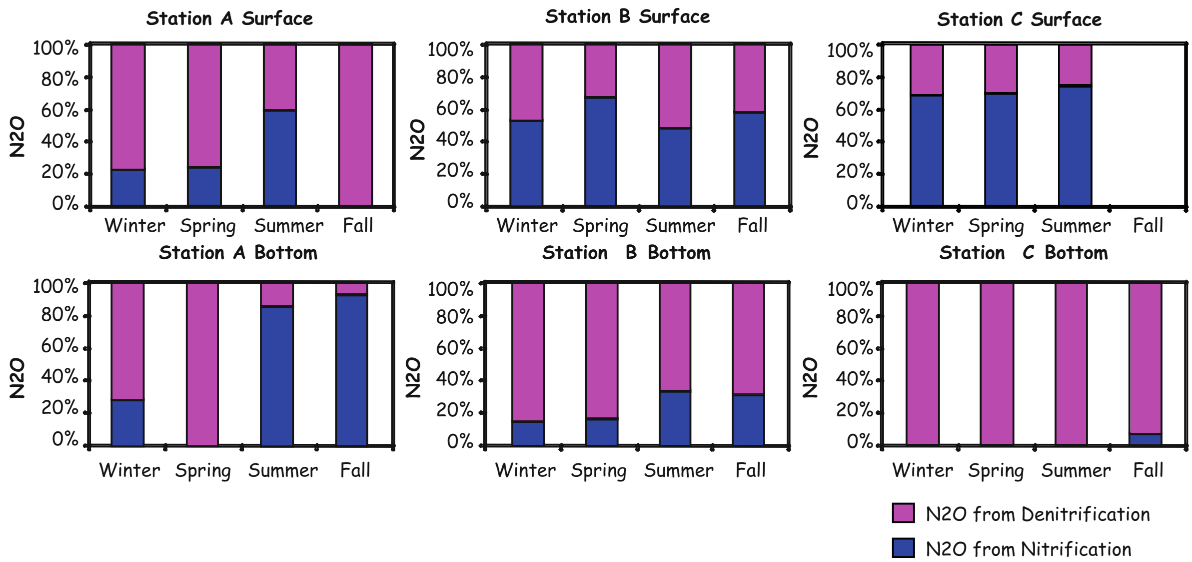

Fig. 5 Bacterial processes responsible for N_2O production at stations A, B, and C in surface and bottom nepheloid layers

3.2 Methane

Concentration and Production of CH_4 (Figs. 6–9)

Important concentrations of dissolved CH_4 were observed in surface water, with more than 1 µM CH_4 at station A (versus 2–5 nM for open ocean) (Fig. 6). The CH_4 concentration decreased along the transect from the river mouth to the sea, with an average of 1,200 nM at station A, 470 nM at station B, and 250 nM at station C.

CH_4 concentration decreased also with depth, with a slight decrease at station A, about 900 nM CH_4 at 40 m depth, and a drastic decrease at stations B (80 m depth) and C (100 m depth), until around 50 nM CH_4 (Fig. 6).

The decrease in dissolved CH_4 with depth suggests that the high CH_4 concentrations detected in the surface layer originated from in situ production in surface nepheloid layer.

Dissolved CH_4 was mainly present at station A, in surface and bottom water (Fig. 7a, $p = 0.002$). Whereas CH_4 remained present in surface water until the marine station C; in bottom water, dissolved CH_4 was essentially observed at station A, with very low concentrations in stations B and C.

The seasonal distribution showed a slight seasonal effect (Fig. 7b, $p = 0.033$), except in winter, when both the low temperature and low flux of material particulate did not favor bacterial methane production.

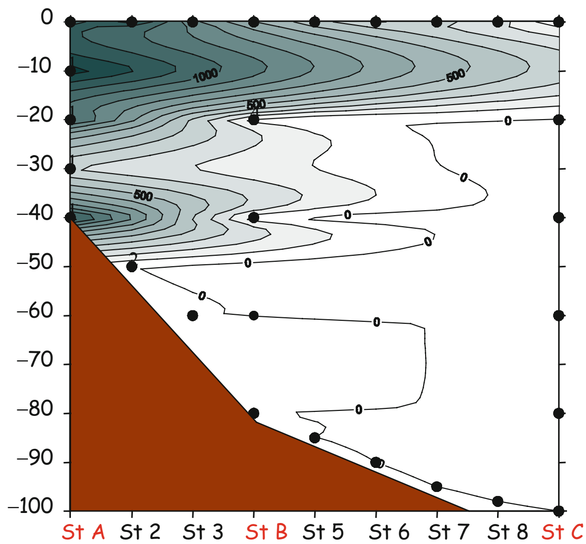

Fig. 6 CH_4 distribution along the Rhône River plume transect: Station A to Station C

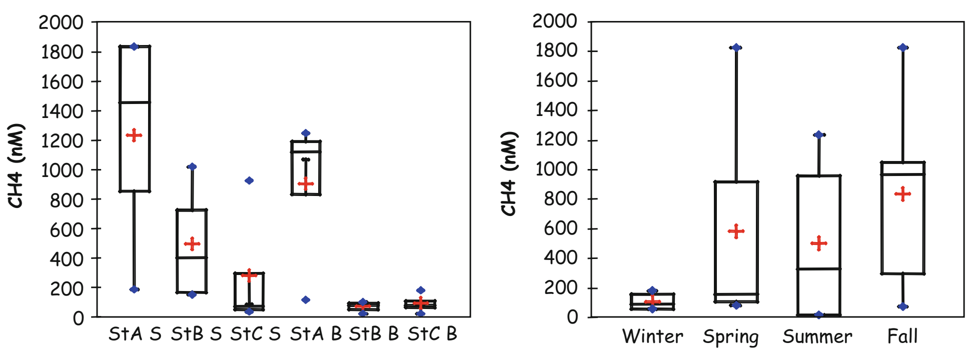

Fig. 7 Effect of sampling site and time on methane distribution at stations A, B, and C (S = surface water and B = bottom water); (**a**): sampling station effect; (**b**): sampling season effect

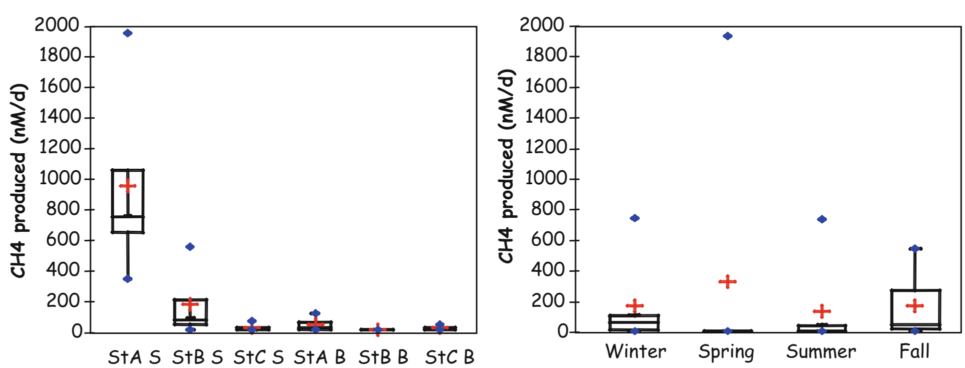

Fig. 8 Effect of sampling site and time on archaebacterial methane production at stations A, B, and C (S = surface water and B = bottom water); (**a**): sampling station effect; (**b**): sampling season effect

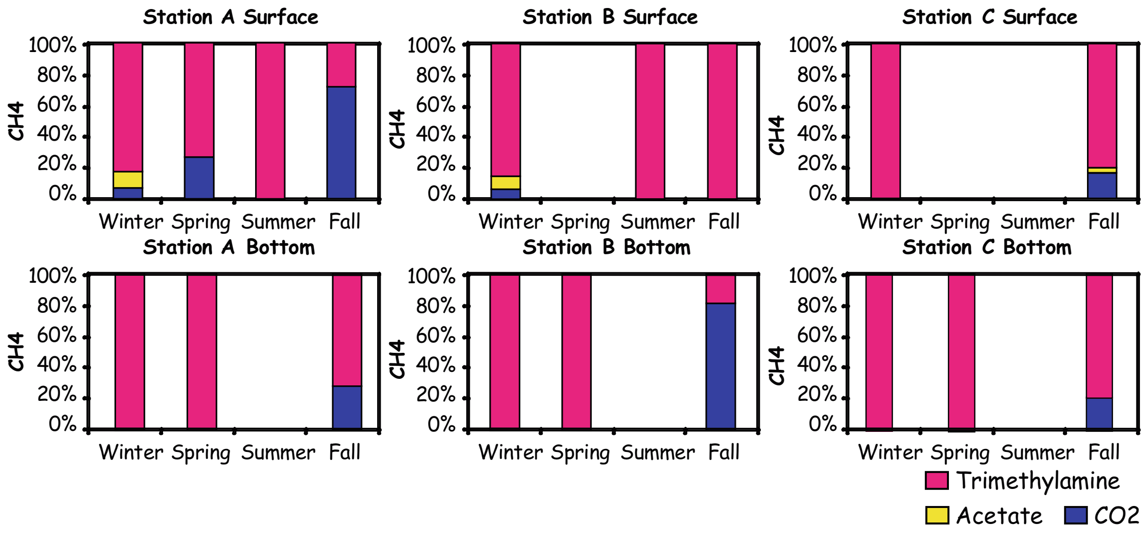

Fig. 9 Archaebacterial processes responsible for CH_4 production at stations A, B, and C, in surface and bottom nepehloid layers

Unexpectedly, a methanogenesis process was essentially present in the surface nepheloid layer, with higher values at station A (Fig. 8a, $p = 0.004$). The decrease in CH_4 production with depth suggests that CH_4 was produced in situ in the surface nepheloid layer, probably in anoxic microniches within particulate material favoring bacterial processes.

Lower methanogenic activity was observed in spring and summer, when Rhône river influx contained lower material particulates (Fig. 8b, $p = 0.719$).

Among the three metabolic pathways involved in methane production processes, carbonate respiration ($H_2 + CO_2$) is a ubiquitous pathway used by 80% of methanogenic archaebacteria, the aceticlastic reaction (acetate) is essentially found in freshwater biotopes, and the methylotrophic pathway (methylamines and methanol) in biotopes with salinity gradients, such as estuarine and intertidal environments. Methylamines are non-competitive substrates, whereas H_2–CO_2 and acetate are preferentially removed by sulfate-reducers, thermodynamically more efficient than methanogens, in using these substrates.

Along the Rhône River plume, in the surface as well as in the benthic nepheloid layers, the greatest part (76.4%) of CH_4 was produced by metabolically versatile methanogens using the methylotrophic pathway; a smaller part (22.4%) was produced by carbonate respiration, and a minor part (1.3%) by aceticlastic reaction (Fig. 9), this last pathway being essentially present in surface desalted water, unfavorable to sulfate reducers.

4 Conclusion

High concentrations of dissolved CH_4 and N_2O were recorded in the surface waters up to 1,300 nM for CH_4 and 40 nM for N_2O, versus, 2–3 nM, and 5 nM in the open ocean. Along the Rhône River plume, the highest concentrations of CH_4 were detected in the surface layer of the desalted station, near the river mouth, whereas N_2O remained present throughout the transect, up to the marine station. Bacterial production rates of N_2O and CH_4 indicated that the Rhône River plume constituted an area favorable for the development of nitrifiers, denitrifiers, and methanogens, promoting their activities in suspended particles (Marty et al. 2001). Nonetheless, no direct relationship could be found between the concentration and the production of the biogases, suggesting that the dissolved gases were not necessarily produced in situ in surface seawater. Important gas levels could be either produced in deep water or bottom sediment, and transported to the surface waters.

The presence of extremely high concentrations of CH_4 and N_2O in superficial seawater implies they can easily escape to the atmosphere; consequently, these near-shore waters enriched in greenhouse gases may play an important role in the increase of the atmospheric concentration of both CH_4 and N_2O, and thus in the global climate change.

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Aerobic Metabolism of Vitamin E by Marine Bacteria: Interaction with Free Radical Oxidation (Autoxidation) Processes

Mina Nassiry, Sophie Guasco, Abdelkrim Mouzdahir, Patricia Bonin, and Jean-François Rontani

Abstract The degradation of vitamin E by aerobic bacterial communities, isolated from marine sediment and microbial mat samples, was investigated. PCR-DGGE profiles and cloning/sequencing experiments revealed that biodegradation of vitamin E in sediments is mainly carried out by strains belonging to the genera *Idiomarina* and *Bacillus* for which the DGGE pattern matched with the pattern obtained from the second sediment subculture.

Biodegradation processes appeared to involve an initial ω-oxidation of the isoprenoid side chain and subsequent β-oxidation sequences affording 2,5,7,8-tetramethyl–2(2'-carboxyethyl)-6-hydroxy-chroman (α-CEHC). This compound was not accumulated at the end of the growth, showing that the bacterial degradation of vitamin E is not limited to its isoprenoid side chain.

In cultures still containing residual sediment, the presence of metabolites with a shortened side chain and an opened chroman ring (e.g., α-tocopheronolactone and α-tocopherylhydroquinonolactone) attested to the simultaneous involvement of biodegradation and autoxidation processes.

The induction of autoxidation during these incubations was attributed to some of the sediment components, which could act as catalysts of free radical reactions.

In oxic environments, the combination of free radical oxidation and aerobic biodegradation processes should result in a very fast degradation of vitamin E. Different pathways are proposed to explain the formation of the different compounds resulting from these interactions.

1 Introduction

Aerobic bacterial communities able to degrade vitamin E efficiently were enriched from marine sediment and microbial mat samples. The degradation of vitamin E by these bacteria appeared to be mainly carried out by strains belonging to the genera *Halomonas*, *Idiomarina*, and *bacillus*. It involves an initial ω-oxidation of the isoprenoid side chain and subsequent β-oxidation sequences resulting in the formation of α-CEHC.

This compound was not accumulated at the end of the growth, showing that the bacterial degradation of vitamin E is not limited to its isoprenoid side chain (Fig. 1).

In cultures still containing residual sediment, the presence of metabolites with a shortened side chain and an opened chroman ring (e.g., α-tocopheronolactone and α-tocopherylhydroquinonolactone) attested to the simultaneous involvement of biodegradation and autoxidation processes. The induction of autoxidation during these incubations was attributed to some of the sediment components, which could act as catalysts of free radical reactions. In oxic environments, the combination of free radical oxidation and aerobic biodegradation processes (Fig. 2) should result in a very fast degradation of vitamin E. Since the aerobic bacterial metabolism of the isoprenoid side chain of this

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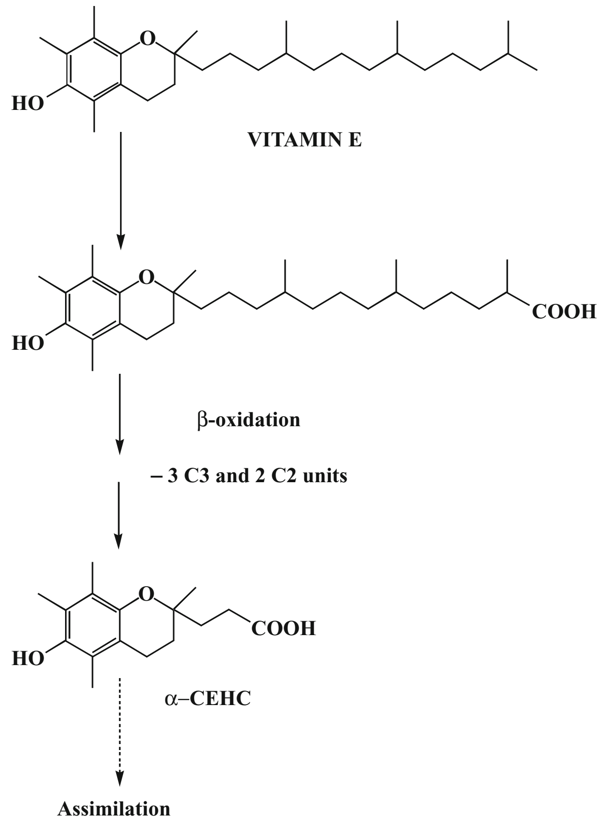
Fig. 1 Metabolism of vitamin E by aerobic marine bacteria



compound takes place without significant production of metabolic intermediates, the degradation of vitamin E by aerobic marine bacteria will not constitute a likely source of isoprenoid compounds in the marine environment.

In order to simulate diagenetic degradation processes, Goossens et al. (1984) previously studied the thermal degradation of vitamin E (under nitrogen) and observed the formation of prist-1-ene. These authors concluded that the degradation of tocopherols could thus constitute a significant source of pristane in ancient sediments and crude oils.

Although it is generally thought that in sediments a part of tocopherols could escape biodegradation once incorporated into macromolecular structures (Goossens et al. 1984), the very high efficiency of autoxidation (Rontani et al. 2007) and aerobic biodegradation processes (Rontani et al. 2008) towards vitamin E suggests that only a weak part of tocopherols could become sequestered into the sedimentary record. These compounds could thus be a likely potential source of prist-1-ene (and thus of pristane) only in environments with high primary production or where sediments are buried under strongly reducing conditions.

There is a real need for compounds sufficiently stable and specific to act as markers of oxic conditions of sedimentation. Some of the metabolites of vitamin E identified (resulting from the combination of autoxidation and biodegradation processes) could constitute very specific tracers of oxic conditions. However, as we could see above, aerobic biodegradation of vitamin E oxidation products is not limited to their isoprenoid side chain, and the opened chroman ring is also quickly consumed by bacteria. Consequently, a good preservation of these compounds in sediments seems very unlikely.

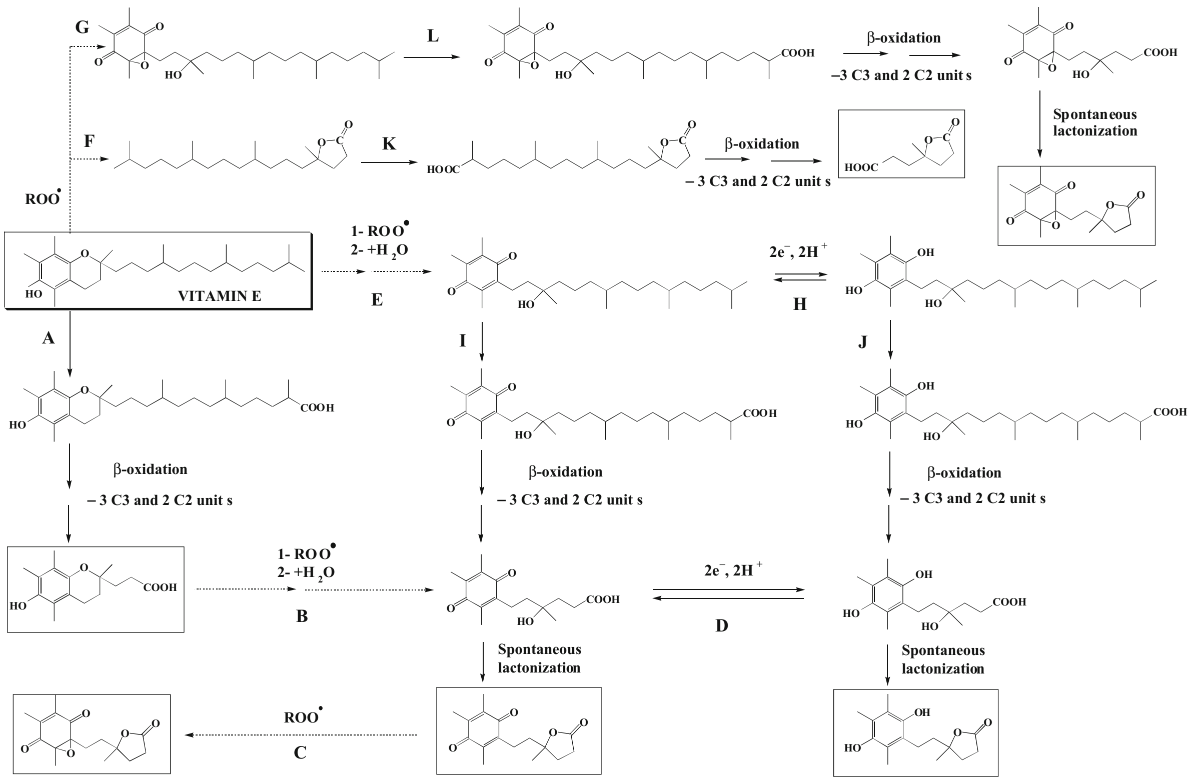


Fig. 2 Interactions between aerobic bacterial degradation and autoxidation of vitamin E

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The MERMeX Program for the Mediterranean Sea

Richerd Sempéré, Xavier Durrieu de Madron, and Cécile Guieu

Abstract The French community working in marine biogeochemistry and biological ecosystems is currently structured to initiate the MERMeX project (Marine Ecosystems Response in the Mediterranean Experiment; http://MERMeX.com.univ-mrs.fr/). This project is part of the large program MISTRALS devoted to the understanding of the Mediterranean environment. MISTRALS is led by the "Institut National des Sciences de l'Univers" (CNRS-INSU), and it includes several projects, such as HYMeX, related to the study of the hydrological cycle, and CHARMeX, related to the study of the atmospheric chemistry in the Mediterranean basin. MERMeX aims at deepening the current understanding of the Mediterranean marine ecosystems to better anticipate their upcoming evolution in the context of global and anthropogenic changes. It focuses on the response of ecosystems to modifications of physico-chemical forcing at various scales, both in time and space, linked to changing environmental conditions and increasing human pressure. Here, we present general features of the Mediterranean Sea ecosystem functioning as well as the key questions that need to be addressed concerning the Mediterranean Sea to predict the response of the Ecosystem to global change in the twenty-first century as well as ideas for implementation. Most of the information given here will be extended and fully described in the review paper published by the MERMeX group (2011).

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1 Discussion

Physically, the Mediterranean is a semi-enclosed basin surrounded by continents and is characterized by very short ventilation and residence times (~70 years) when compared to the other oceans (200–1,000 years). A real thermo-haline circulation takes place in the Mediterranean basin and is driven by Atlantic spreading in the basin as well as surface water convection in the Gulf of Lions, the Adriatic Sea. and the Levantine basin. The main external sources of water in the Mediterranean basin are Atlantic water spreading through the Gibraltar Strait followed by the contribution of rivers and the Black Sea. Freshwater inputs play a significant role in the Mediterranean Sea since they significantly enhance the primary productivity at local scales and play a major role in the balance of water inputs through the Strait of Gibraltar. Since the damming of the Nile, the northern rivers (mainly the Rhöne, Pô, and Ebre Rivers) are the major source of freshwater and terrigenous particles to the Mediterranean basin (Sempéré et al. 2000; Ludwig et al. 2009 and references therein). Because of its weak cloud coverage, the Mediterranean Sea is also subjected to stronger solar radiation (Cristofanelli and Bonasoni 2009) in comparison with oceanic areas of similar latitude, which leads to a small limitation of photosynthesis by algae and plants by light, a high level of

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photoreception, potentially damaging phytoplankton, and a strong photochemical reactions in the surface layer. Biogeochemically, the Mediterranean has long been known as an impoverished area with relatively low nutrient concentrations and is characterized by a general west- to-east gradient of increasing oligotrophy. The elemental stoichiometry in all compartments (i.e., particulate and dissolved inorganic and organic) reveals an excess of carbon, a deficiency in phosphorus relative to nitrogen, and a sporadic silicate deficiency (Béthoux et al. 2002). Macro nutrient concentrations mainly depend on the exchanges through the Straits of Gibraltar and Bosphorus, atmospheric depositions, and river discharges. Continental inputs are characterized by a strong variability, dominated by extreme events (i.e., large river floods and Saharan dust deposits) due to the climatic specificities of this region.

The Mediterranean has a rich biodiversity and an elevated proportion of endemic species. Despite its small size (0.82% of the world‘s ocean surface) and relative oligotrophy, from 4% to 18% of the world‘s marine diversity, depending on the phylum considered, is concentrated the Mediterranean (WWF/IUCN 2004). Another important issue is the relative isolation of deep-sea communities, not only with respect to those of the Atlantic, but also between those in the Eastern and Western Mediterranean, separated by the Sicilian Channel. Demographically, the Mediterranean is subject to strong human pressures that threaten marine ecosystems. The countries bordering the Mediterranean currently have a combined population of about 450 millions people, and its 26,000 km of coast supports a population estimated at 132 millions inhabitants, as well as intensive farming and industrial activities. The population greatly increases during the summer tourist season, with the Mediterranean being the world‘s leading tourist destination with about 200 millions people arriving per year. Land-based (farming, domestic, commercial, and industrial) activities generate large volumes of waste water, which supplies nutrients, organic matter, toxicants, and pathogens that impact coastal marine environments. Growth of populations and change of traditional and recreational fishing practices along the coasts have led to the expansion of sea fishing and an increase in landings. All these characteristics make the Mediterranean a single ocean region experiencing many climatic and anthropogenic forces (Fig. 1). This region is expected to be an area particularly sensitive to current and future trends of these forces.

According to the last IPCC report (IPCC 2007), the atmospheric temperature has increased by 1–2°C in the last decade in the Mediterranean basin, which is four times the increase reported for the earth, and atmospheric surface temperatures are projected to increase by 0.6–4°C. This warming will probably increase the upper layer stratification and decrease the deep ocean ventilation as well as reduce new nutrient input into the upper layer. This could result in an increasing trend of small cells of dominating phytoplankton communities that in turn would modify the entire structure and efficiency of the food web. Moreover, the amount of light biologically available per day would increase with the stronger stratification,

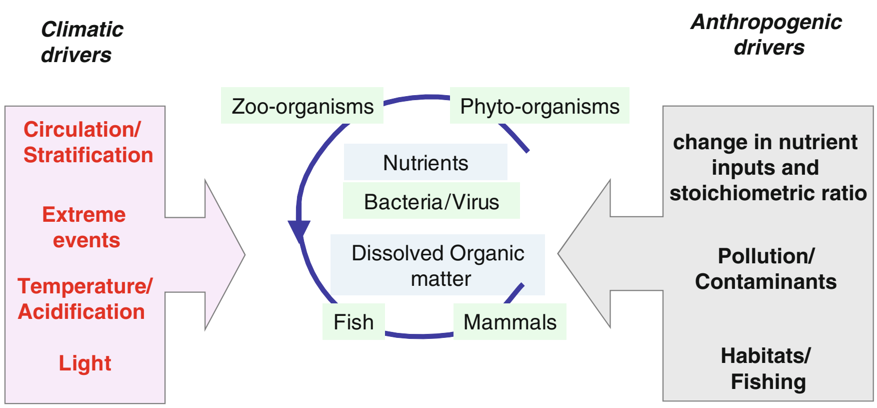

Fig. 1 Main climatic and anthropogenic factors that may impact Mediterranean marine ecosystems in the twenty-first century (according to different scenarios) for eastern and western basins. These factors will be specifically studied during the MERMeX Program

which would lead to a strong algal species selection in the upper layer. Lowering the photosynthetic production would lead to a decrease of the associated flux of organic detritus into the ocean interior, which could lead to a change in their biogeochemical cycle. A warming effect will further lead to a decrease in pH (acidification), modification of the Revelle factor, and changes in the CO_2 air-sea fluxes, leading to a change in the carbonate system and subsequently altering the input of anthropogenic CO_2 by the ocean. The rising population density (resident and non-resident) is expected to have repercussions on the nutrient and contaminant inputs to the Mediterranean (Fig. 2).

In the MERMeX project (http://MERMeX.com.univ-mrs.fr/), we propose a comprehensive, integrated approach considering the continuum between the coastal zone and the open sea and its interfaces, including ocean-continent, ocean-atmosphere, and water-sediment to describe and model the current state of the Mediterranean ecosystems and the complex interactions existing between the environmental and human factors precisely. We present the French initiative MERMeX for a large biogeochemical program in the Mediterranean and call for international collaboration. The French MERMeX project will be implemented from ca. 2012 to 2022 in the framework of 'MISTRALS' program (Fig. 2). The program involves several partner agencies with specialized expertise and technical resources. In addition to INSU, these agencies include Meteo France, CNES and IFREMER, and French Universities. One of the objectives of the French MERMeX project is to promote international efforts to develop an operational strategy that provides integrated end-to-end capabilities at the regional, national, and local levels within a multi-hazard framework. The objectives focus on the understanding of the ecosystem response to likely changes in physical, chemical, and socio economic forces induced by global change and induced by growing anthropogenic pressure at the regional scale. We wish MERMeX to be based on an original approach that will consider the whole continuum of continent-coastal zone-open sea in relation to the atmospheric dimensions in order to describe the present state of the Mediterranean Sea and consider its evolution towards a disturbed state because of the alterations caused by different forces. Understanding the changes going on in the Mediterranean Sea require effective long-term observation systems. One of the

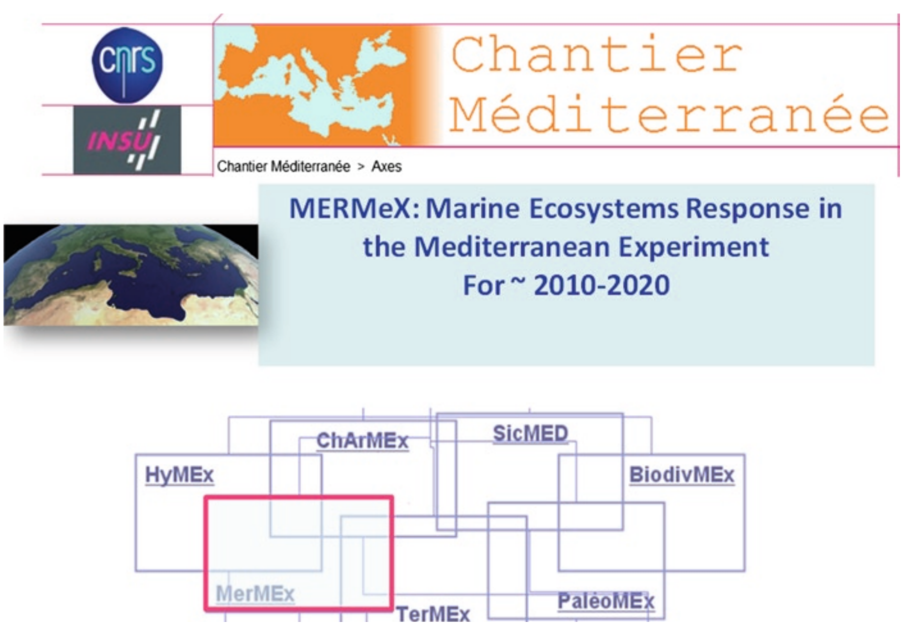

Fig. 2 Framework of the French program Chantier Méditerranée so-called 'MISTRALS' (http://www.dt.insu.cnrs.fr/c-med/c-med.php). The MERMeX program has strong links with other Mediterranean-related programs such as CHARMeX (for atmospheric chemistry; https://CHARMeX.lsce.ipsl.fr/), HYMeX (for Hydrometeorology: http://www.HYMeX.org/) and Biodivmex (for Biodiversity)

major aims is to be able to propose scenarios of changes and foresee the variations that ecosystems will experience in the future.

The efforts of the group are based on the five following work packages:

WP1 Impact of hydrodynamic changes on Mediterranean biogeochemical budgets (PIs: P.Conan, F.D Ortenzio, C.Estournel)

WP2 Ecological processes: biogeochemistry and food web interactions (PIs: F. Carlotti, F. Van Wambeke, C. Melon)

WP3 Land-sea interactions including intense events (PIs: C. Radakovitch, C. Rabouille)

WP4 Natural and anthropogenic atmosphere-sea interactions (PIs: F. Gazeaux, K. Desboeufs, M. Mallet)

WP5 Ecosystem base management feed-back to services linked to ecosystem functioning (PIs: P. Koubbi, S. Gasparini)

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Hydrocarbon Degradation in Coastal Muddy Areas and Anoxic Ecosystems (DHYVA Project): Role of Bacterial Mechanisms and Bioturbation Effects on the Biodisponibility of Organic Pollutants

Magalie Stauffert, Lionel Huang, Isabelle Vitte, Ronan Jézéquel, Cristiana Cravo-Laureau, Georges Stora, Christophe Pécheyran, Christine Cagnon, Frank Gilbert, Marisol Goñi Urriza, François-Xavier Merlin, David Amouroux, Philippe Cuny, and Robert Duran

Abstract Muddy areas and more or less coastal anoxic zones play a key role in maintaining the integrity of estuarine and coastal ecosystems. By their localization, these areas are frequently exposed to pollutant damage, such as oil spills or accidental events, and accumulate various pollutants released by continental hydrosystems.

Due to their composition (fine silt sediments), actual remediation techniques are not adapted. These artificial compounds accumulate in different sites, constituting "pollutant reservoirs" that are threats for the ecosystem as well as for human health.

Microorganisms play a key role in hydrocarbon degradation. However, most of our knowledge has been obtained by studies on isolated bacterial strains. In addition, bacterial cooperation within different microbial groups has been demonstrated for the degradation of complex hydrocarbon mixtures. Silt sediments are located at the oxic/anoxic interface where various bacterial metabolisms coexist and follow one another through the function of the tides.

Moreover, burrowing organisms can influence the bacterial metabolisms by making the oxygen penetration easier. It thus seems judicious to tackle the problem of hydrocarbon degradation in a global way, trying to understand how bacterial metabolisms interact in the degradation of the pollutants and to estimate the bioturbation effects of these activities.

This is the main objective of the project DHYVA (Dégradation des HYdrocarbures dans les VAsières), with a special focus on anaerobic degradation mechanisms, about which we have limited knowledge.

(*) CEntre de Documentation, de Recherche et d'Expérimentation sur les pollutions accidentelles des eaux

1 Introduction

In natural environments, the fate of hydrocarbons depends on abiotic processes (such as photo-oxidation and chemical oxidation) and biotic processes, such as microbiological activity. It is well known that microorganisms play a major role in their degradation (Van Hamme et al. 2003). Burrowing macrofauna play a role in both hydrocarbon transfers at the sediment–water interface and subsequent hydrocarbon biodegradation in the sedimentary column (Gilbert et al. 1996). These organisms can affect, directly or indirectly, the microbial metabolisms by modifying the oxic/anoxic boundaries.

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For these reasons, it is interesting to define the importance of the effects of burrowing organisms and the bacterial community on hydrocarbon degradation.

2 Materials and Methods

An experimental device was set up in order to create the cycles of tides at the "CEDRE" laboratories (Brest, France). These mesocosms (Fig. 1) were established with silt sediment (Aber-Benoît, Brest, France). Four conditions (in triplicate) were applied: a negative control (only sediments), sediments with oil pollution (BAL 110), sediments with *Nereis diversicolor* (burrowing organisms), and sediments with oil pollution and bioturbation. Samplings were carried out over 9 months (15 days, 1, 3, 6 and 9 months) for chemical and microbiological analysis. Luminophores (provided by the University of Kiel, Germany) were used in this experiment. They are fluorescent inert particles used as conservative tracers to quantify the burrowing process induced by the fauna. Luminophores were deposited on the upper part of each experimental mesocosm.

Hydrocarbon pollution and burrowing impacts on bacterial community structures were investigated by T-RFLP (Terminal Restriction Length Polymorphism). This technique allows obtaining a molecular fingerprint of the total and active bacterial community (DNA and RNA analysis). Stratification in the sediments and temporal evolution of bacterial communities in each condition were studied.

A cultural approach allowed the isolation and the characterization of bacterial consortia and bacterial strains able to degrade hydrocarbons in anoxic and oxic conditions. In order to discover the genes involved in the bacterial community response to the pollution, we investigated the integrons, mobile genetic elements involved in adaptation mechanisms.

First, integrase diversity was determined and their transcription level estimated in order to evaluate their implication in the adaptation to petroleum compounds.

Second, thanks to clone library analyses of their first gene cassettes, we expect to find new adaptive genes.

3 Results and Discussion

The macrobenthic compositions within mesocosms were compared using the quantitative Sanders coefficient based on the species dominances and the qualitative Sorensen coefficient calculated with the presence/absence of the species (Pischedda et al. 2008).

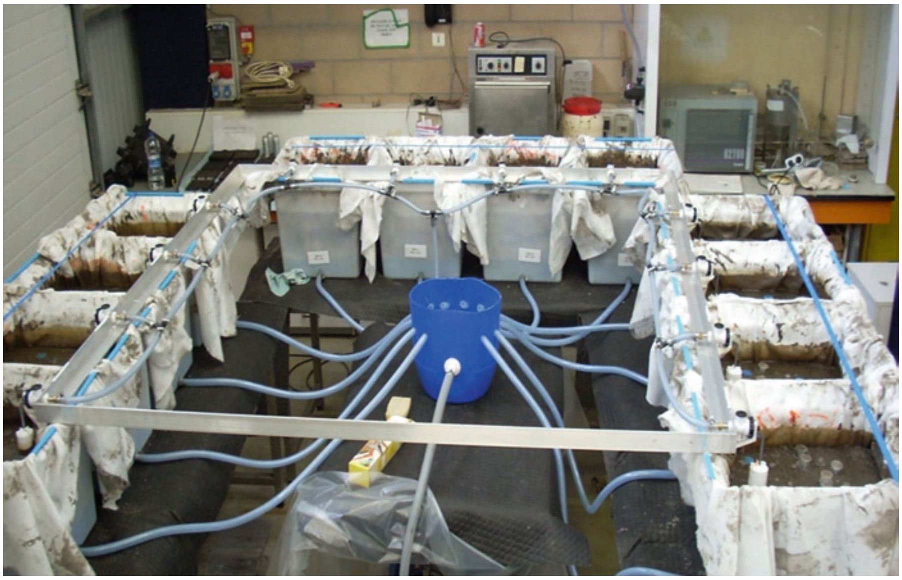

Fig. 1 Microcosm experimental set up. Mud sediments from Aber Benoit were maintained in microscosms at the "CEDRE" laboratories (Brest, France); 12 microcosms (30 L of mud each) were subjected to four conditions: three controls (only sediments), three polluted with oil (BAL 110), three with bioturbation where *Nereis diversicolor* was added, and three subjected to both pollution and bioturbation. In the center, the collector on the lifting table collects tide waters. New sea water was added to each tide cycle

Crude oil is a complex organic matrix, containing a large variety of molecules ranging from light alkanes to heavy polyaromatics. The oil degradation and the influence of oxy/anoxy cycles on the fate of hydrocarbon compounds in muddy sediments were evaluated by following the evolution of oil composition throughout the kinetics of degradation by gas chromatography coupled with a mass spectrometer (GC-MS) and by high-performance liquid chromatography (HPLC). The degradation of hydrocarbon compounds was quantified in order to measure the importance of the biodegradation phenomena.

Crude oils contain some metallic elements, mainly nickel and vanadium, which form complexes with heavy molecules such as porphyrins. The metals found in oil show a concentration of up to several hundred ppm. By analyzing the metal content of the petroleum after every step of biodegradation, we would like to determine the speciation of the metallic elements and to which molecules they are associated.

4 Conclusions

This project answers to a strong demand for the development of biological strategies for cleaning coastal ecosystems, particularly those exposed to pollutants.

The expected results will provide new insights on biological mechanisms involved in determining the fate of hydrocarbons in the environment at the oxy/anoxy interface.

In the long term, they should provide useful knowledge for a better management of the microbial resources in situ and to the establishment of new molecular tools for the evaluation of the impact of hydrocarbons on ecosystems.

These tools should provide simple and specific molecular indicators leading to the appreciation of the impact of oil pollutions on the biological functioning of the ecosystems.

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Impact of Red Mud Deposits in the Canyon of Cassidaigne on the Macrobenthos of the Mediterranean Continental Slope

Georges Stora, André Arnoux, Eric Duport, Christian Re, Magali Gérino, Gaston Desrosiers, and Frank Gilbert

Abstract Since 1967, red mud produced by an aluminum plant by the process of aluminum extraction from bauxite has been discharged into the canyon of Cassidaigne (North Mediterranean Sea) at 320 m depth. To study the effect of these dumpings on the macrobenthic fauna of the continental slope, sediments were sampled in September 1991, 1997, and 2002 from stations located on both sides of the canyon, at between 250 and 2,200 m depth. For each station, a faunal analysis was coupled with analyses of sediment granulometry and heavy metal content.

The species collected belong to deep mud assemblages characteristic of the bathyal zone of the Mediterranean Sea. We observed a progressive decrease with depth in species richness and average population density. Such a decrease is usual on the continental slope and cannot be attributed to a particular anthropogenic disturbance of the ecosystem.

Hierarchical analyses do not differentiate communities under the dependence of the deposits from communities not impacted by red mud. Correlations between environmental variables and communities fail to demonstrate a particular incidence of titanium, the optimal marker for red mud. The distribution of the benthic populations is directly dependent on changes in bathymetry and associated parameters, such as sedimentation rate and food availability.

There is a significant, long-term variability in the qualitative and quantitative composition of the communities of the continental slope, which are directly under the influence of the Mediterranean Northern Current and the global evolution of climatic conditions.

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1 Introduction

In the past century, the lack of global policies for reducing waste production has resulted in a growing use of the deep sea as a repository for waste, the latter ranging from harmless to highly dangerous to humans. Industrial wastes discharged into the ocean accumulate on the sea bed with a possible detrimental effect on the deep-sea benthos (Gage and Tyler 1991). Since 1967, red mud produced by an aluminum plant as a result of extracting bauxite has been discharged into the canyon of Cassidaigne (North Mediterranean Sea) at 320 m depth. To study the effect of these dumpings on the macrobenthic fauna of the continental slope, sediments were sampled in September 1991, 1997, and 2002 from stations located on both sides of the canyon, at between 250 and 2,200 m depth (Fig. 1).

2 Material and Methods

A USNEL box corer with a surface area of 0.25 m^2 and a height of 0.80 m was used. Two cores were taken at each station, and two sub-samples of 0.1 m^2 were taken

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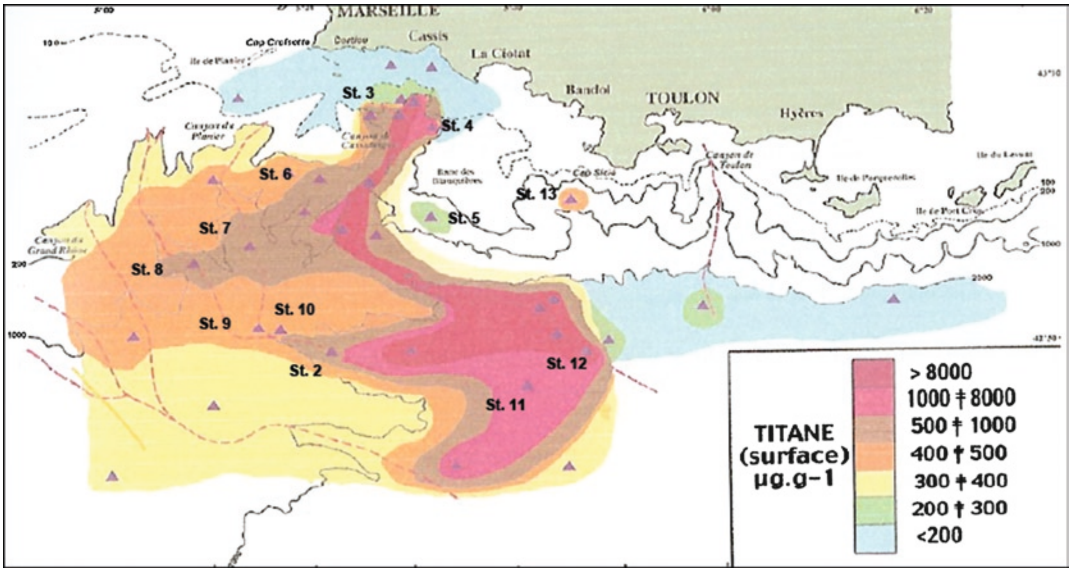


Fig. 1 Location of the stations and dispersion of the waste as characterized by titanium concentrations (specific marker for red mud) at the water-sediment interface and in the sedimentary column

from each core. Samples were sieved through a 250-µm mesh and sorted. In most cases, organisms were identified to the species level.

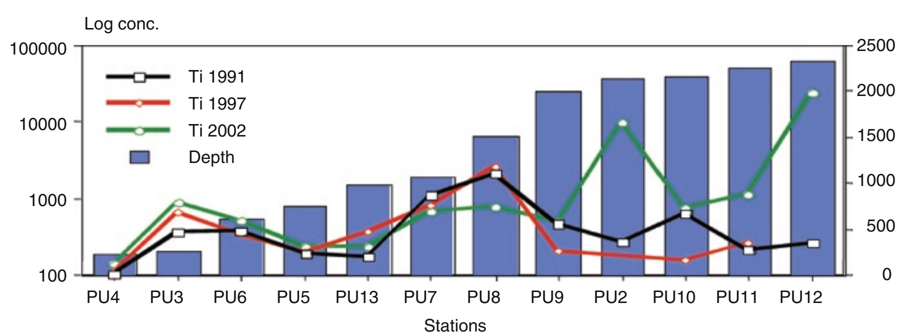
Geochemical analyses (grain size analysis, Cu, Pb, Cr, Ni, Ti, Fe, Al, and Va) were carried out on an average sediment sample obtained by mixing and homogenizing two subsamples of 0.79 dm^2 taken from different layers (surface, 0–3, 3–6, and 6–9 cm).

Data were processed using the classical and synthetic methods described by Clarke and Warwick 2001.

3 Results

See Figs. 2 through 5.

Fig. 2 Titanium concentrations in the surface sediment of the 12 sampling sites



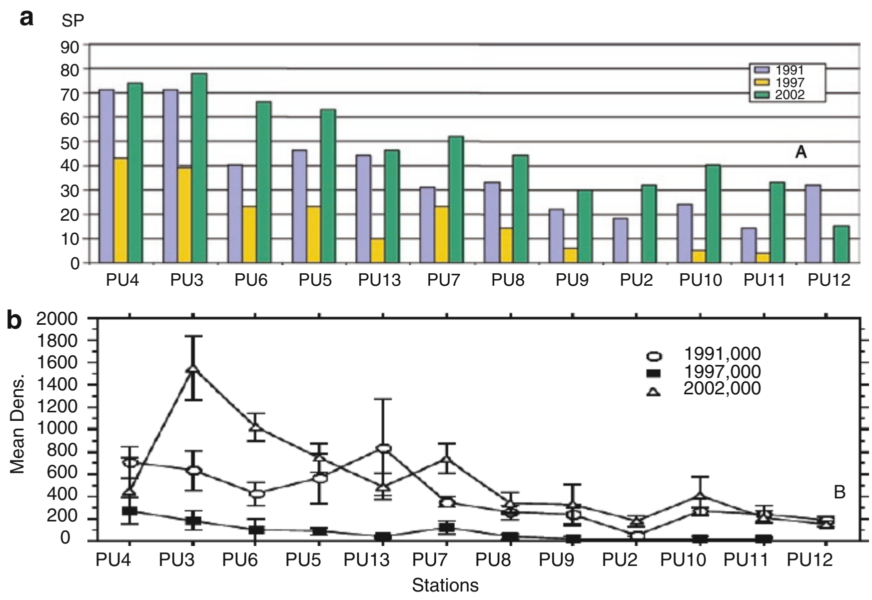


Fig. 3 Species richness (**a**) and density per m² (**b**) of macrobenthic communities at each sampling site

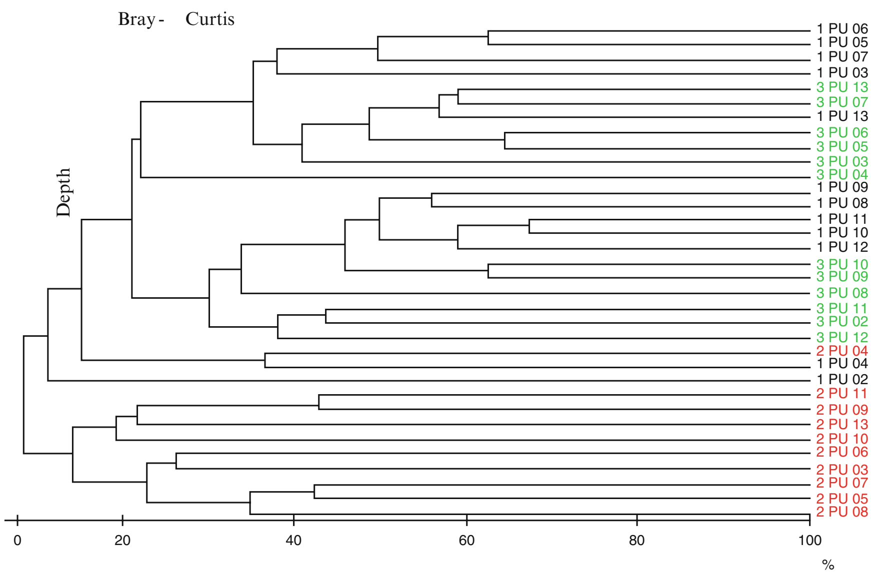


Fig. 4 Dendrogram of the 12 stations at different sampling years (*black* 1991, *red* 1997, *green* 2002) using group average clustering from Bray-Curtis similarities

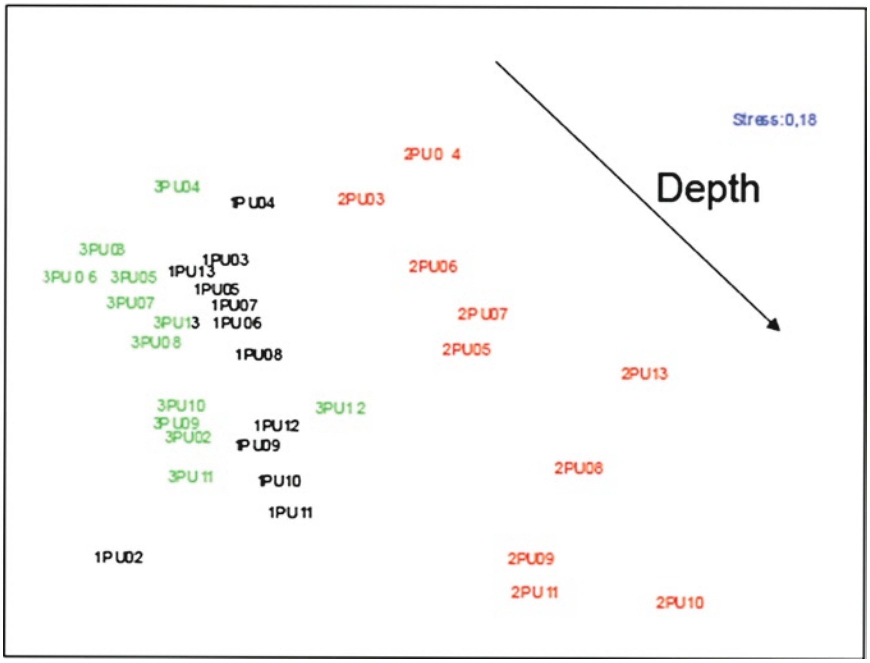


Fig. 5 MDS ordination of the 12 sampling sites during the 3 sampling years (*black* 1991, *red* 1997, *green* 2002) based on Bray-Curtis similarities

4 Discussion and Conclusions

The species collected belong to deep mud assemblages characteristic of the bathyal zone of the Mediterranean Sea. We observe a progressive decrease in species richness and average population density with depth. Such a decrease is usual on the continental slope and cannot be attributed to a particular anthropogenic disturbance of the ecosystem.

Hierarchical analyses do not differentiate communities under the dependence of the deposits from communities unimpacted by red mud. Correlations between environmental variables and communities fail to demonstrate a particular incidence of titanium, the optimal marker for red muds. The distribution of the benthic populations is directly dependent on changes in bathymetry and associated parameters such as sedimentation rate and food availability.

There is a significant, long-term variability in the qualitative and quantitative composition of the communities of the continental slope, which are directly under the influence of the Mediterranean Northern Current, and the global evolution of climatic conditions.

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Coastal Seawater Pollutants in the Coral Reef Lagoon of a Small Tropical Island in Development: The Mayotte Example (N Mozambique Channel, SW Indian Ocean)

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Abstract Mayotte Island, a small volcanic island, is surrounded by the largest coral reef lagoon of the Indian Ocean (almost 1,500 km^2), with all coral reefs types represented: ribbon barrier reefs, lagoonal and fringing reefs, and also coastal mangroves.

Since 1975, Mayotte has shown an incredible development of its population (today about 180,000) and of its economy, with increases in consumer goods, fresh water use, construction of concrete houses and buildings, roads, truck and car parks, harbor and airport activity, gas and oil consumption, etc. At the same time, land vegetation coverage has decreased (there are still burnings of areas for agriculture and charcoal making) as land erosion has increased in some areas.

As a consequence of this development of towns, villages, roads, harbor and airport traffic, which is mainly along the coastal areas, today an increase in the turbidity and the pollutants in the surface waters of the coast has been observed. Pollution was analyzed by monitoring the midlittoral oyster (*Saccostrea cucullata*): some trace metals, as Zn, Cd, Hg, and Cr; some polyaromatic hydrocarbons, such as naphthalene, pyrene, and fluoranthene; and some polychlorobiphenyls (PCB), even some that are particularly toxic, were found.

Associated with this degradation of the coastal water quality and human impacts in general, in the decades since 1997, a decrease in the vitality of the fringing reef communities that surround the islands (predominant coral coverage ratio <5%) has been observed. Therefore, the human impacts are stronger than the natural stresses, such as the ENSO and/or coral bleaching events, and the crown-of-thorns starfish *Acanthaster* infestations. Muddy flats now overlay inner areas of the fringing reef flats. Linked with this, coastal fish populations have decreased.

1 Introduction

Amounts of various pollutants, such as trace metals, polyaromatic hydrocarbons (PAHs), and polychlorobiphenyles (PCBs), in the coastal waters are difficult to determine in a common and systematic monitoring of the coastal water quality. Quantitative bioindicators are consequently used. In the cold and temperate seas, mussels, filter feeders that are very common along the coastlines, are currently used for determining pollution levels (as part of the world "mussel watch").

Mayotte (Comoro Archipelago) is a small, eroded volcanic island (land: 376 km^2, for >1,500 km^2 of coral reef lagoon) in the northern part of the Mozambique Channel; the tidal range reaches up to 4 m (Thomassin et al. 1989). With a wet tropical climate, as in other Indopacific regions, the mussels living in the midlittoral rocky fringe are the very small *Brachydontes variabilis* (1 cm long); this species cannot be used as a bioindicator of the surface seawaters because of its

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size. This is not the case for the larger mussel *Modiolus* (*M. auriculatus* Kraus), but they live in the infralittoral muddy-sandy bottoms and seagrass beds, and are used for subsurface seawater monitoring (for example, in Reunion, J.-P. Quod, personal communication; and Tahiti, Bourdelin 1997).

Consequently, in Mayotte Lagoon, since 1997, to monitor the pollution of surface coastal seawaters, we have sampled the oyster, *Saccostrea cucullata* (Born), commonly used for this purpose in other countries (under the name *Crassostrea commercialis* in Australia or *S. cucullata tuberculosis* in New Caledonia); this oyster belt is located just below that of *B. variabilis* (Fig. 1).

Mayotte is a new overseas French department that has shown important economic development since 1975–1977. Consequently, the population has exploded from approximately 40,000 habitants in 1974 to more than160,000 in 2007. Most of this population lives along the lagoonal coasts. Linked to the wet tropical climate and the huge recent human impacts, such as buildings and road works, increase of cars, gas and oil consumption (for electric plants), deforestation (burnings of lands for agriculture and woods for charcoal, etc.), the terrestrial erosion of the lateritic soils (rich in Fe oxides and Al silicates), have drastically increased, with an important recent infilling of the bays and coastal bottoms inducing a lowering of the water transparency. The "green" landscapes have turned "red" in many places on the islands in only 30 years. Today, only 46% of the freshwater of the rivers is in good condition, as it has been polluted by washings, dumping, etc.

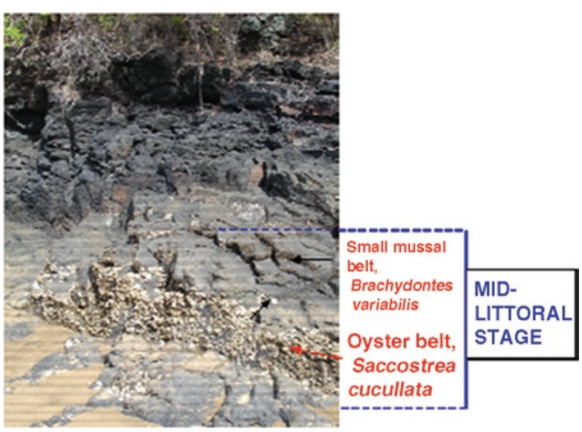

Fig. 1 Small mussel and oyster belts on a rocky shore in Mayotte in the midlittoral stage

Consequently, all pollutants, including trace metals, PAHs, and PCBs, reach the coastal lagoonal waters, brought by air and winds, rainfalls and river flooding, domestic and industrial untreated sewages, underground seepages from dumping grounds, etc.

Previous pollutant surveys in Mayotte (Arnoux 1998; Arnoux and Thomassin 2002) were restricted to the eastern coasts: the more populated areas of Mamoudzou town and its neighbourhoods, Pamandzi Island with the airport and recently the country's gas tanks, and the Longoni harbor vicinities. In the scope of the IFRECOR Program ("Initiative Française pour l'Etude des REcifs CORalliens"), the 2007 monitoring included all shorelines of the Main Island and of the Pamandzi Island washed by the lagoon (Fig. 2).

2 Materials and Sampling Time

Oysters were collected by one of us (J.W.) on rocky capes all around the Main Island, Pamandzi Island, and a few rocky lagoonal islands (as check samples) at the end of the austral spring season (October 2007), when the oyster gonads are not yet well developed (most liposoluble organic pollutants are supposed to be eliminated according to the low fat content), their development and spawning occurring during the wet and hot season (December to April–May). Oyster flesh (100 individulas) was kept frozen (−18°C). The LHMA scientists performed the pollutant analyses.

3 Trace Metals

The potential sources of pollution in the lagoonal waters are: (1) the freshwater inputs after rainfalls from streams and rivers (some polluted), village drainages and sewages, road wash-off, etc.; (2) metallic materials throw directly into the water, even with oils, old iron frameworks and tanks, aluminum cans, lead batteries, rubbish, etc., plus the zinc-galvanized corrugated iron plates used for house roofs and fences.

Consequently, the main two sources of metal are: (1) the geochemical nature of the soils coming from the "mother volcanic rocks" after alteration, for which a "ground-loading level" is pointed out, and (2) anthro-

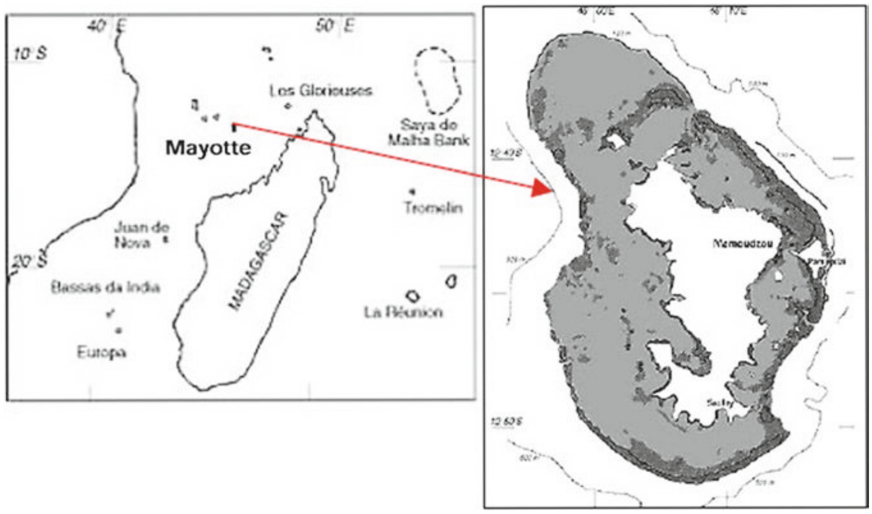

Fig. 2 Location of the Mayotte Islands in the SW Indian Ocean. General view of the insular platform belt with ribbon barrier reefs (up to 200 km long) with inside lagoonal and fringing reefs (195 km long for 210 km of coastlines) and coastal mangroves (735 ha)

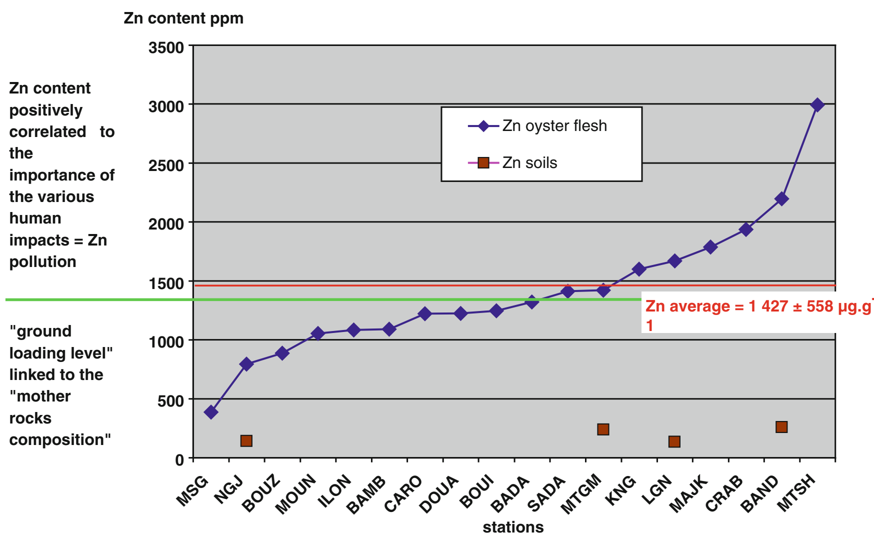

Fig. 3 Mayotte Islands: zinc content in *S. cucullata* oyster flesh: classification of the stations according to increasing values; distinction between values linked to the "ground-loading level" and anthropogenic pollution

pogenic contaminations, estimated as the quantity of metal measured in the oyster flesh reaching up to the values of this "ground-loading level" (Fig. 3).

S. cucullata was shown to be a very efficient bioconcentrator for several metals (metallic elements) in comparison to metal contents:

In the muddy lagoonal sediments:
For Hg: 4–7 times more
For Cd: 0 up to 115 times more
For Cu: 1.5–73 times more
For Zn: up to 85 times more
In the land soils:
For As: average 15,000 times more
For Cd: 3–5 times more
For Zn: 6–17 times more
For Cu: 4–10 times more

Peculiar anthropogenic impacts are demonstrated, mainly for:

Zinc: highest values recorded along the more populated coasts in the northeast

Nickel;

Cadmium: highest values recorded along the powerless populated coasts;

Lead and copper: concentrated in the northern region of the Main Island and in relation to the composition of the volcanic "mother rocks" and/or to the recent increasing human impacts (extensive growth of some villages)

Mercury and even chromium.

4 Polyaromatic Hydrocarbons (PAH)

Those compounds, commonly found in the oyster flesh, are: pyrene (>10 μg · kg^{-1} dw), naphtlene (>15 μg · kg^{-1} dw), acenaphtylene, fluoranthene, and benzo(b)fluoranthene, the two last being associated with polluted freshwaters from urban areas where streams and untreated sewage run easily down into the lagoon.

The Σ_5PAHs content (≈<Σ_{16}PAHs) varies between >1 up to 83 μg · kg^{-1} dw, average = 39 ± 23 μg · kg^{-1} dw.

According to the fluoranthene/pyrene ratio, it is possible to differentiate the two origins for these PAHs: in stations where the pyrene content is high (14–17 μg · kg^{-1} dw), the origins of these PAHs is petrogenic (as near the rocky capes), while, where the fluoranthene content is maximal and the pyrene one absent or low, the origin is pyrolitic (in relation to land fires and charcoal burnings). However, pyrene shows a light correlation with the total PAH content and the Σ_7PCB content, which points to a link between these pollutants (presence of pyrene in the electric transformer oils) (Fig.4).

5 Polychlorobiphenyl (PCB) Hydrocarbons

Six levels of the total PCB content [= (Σ C_{118} + C_{138} + C_{153} + C_{180}) × 100/41] are identified (cf. Wafo et al. 2005, for the method). The highest value reaches up to 50 μg · kg^{-1} dw. It is considered as the reference for "non-polluted waters" in the USA, with a temperate climate, but it is not valuable in wet tropical climate conditions. At Mayotte, this reference value is lower, near 30 μg · kg^{-1} dw. Around Mayotte, three shorelines show the highest PCB content (Fig. 5b): eastwards where the urban population and small industries are concentrated (W Pamandzi, Mamoudzou-Koungou conurbation, and Dembeni); northwards with the large villages showing a recent population explosion (Mtsamboro-Msahara conurbation), but also westwards where important scattered villages are located (Mtsangamouki, Acoua, Boueni, and Kani Kely).

The PCB "toxicity index" (Fig. 5c) [ti = Σ (C_{118} + C_{123} + C_{126} + C_{167} + C_{169})] confirms that the northern area of the Main Island is the more impacted.

The PCB "degradation index" (di = C_{153}/C_{138}) confirms this finding, with the highest values along the northern shorelines. Two hypotheses can be considered to explain such a situation: the congeners C_{153} and C_{138} are less degraded, or an "auto up-keeping" occurs in some lagoonal areas from the PCB sources induced by the hydrodynamic streams in the lagoon.

From 1997–2007, we observed a general increase of the PCB contents in the oysters in the eastern areas of the lagoon when, as is the case in a few stations, these bivalves have not already died, killed by pollutants coming from semi-industrial areas.

6 Conclusions

Around the Mayotte Islands, trace metal contents in midlittoral oyster flesh vary tremendously from one site to another, according to the geochemical composition of the surrounding mother rocks and their alterites, with metals being washed away to the lagoonal seawaters by rainfall, streams, and rivers, in dissolved or particulate forms; the various human inputs, some concentrated in towns, villages, or rubbish area and carried by aerosols and/or dissolved substances in freshwater and then reaching seawaters. Concerning the geochemical nature of the mother rocks and their alterites, there is a "ground level" for each metal. When a metal exceeds its ground level in the oyster flesh, it is possible to identify the anthropogenic impacts, such as, for example, Ni, Zn, Cd, Hg, and even Cr. Some metals are correlated with others, such as Cr with Ni, Cu with Pb, and As with Zn, while Hg does not show a clear metal association. Zn is clearly correlated to the anthropic parameters, confirming other results obtained from Mayotte soils analyses (Bozza et al. 2008).

The more common PAHs uncounted in midlittoral oyster flesh are, in decreasing order of frequency, and that quantitatively: naphtlene, pyrene, acenaphtylene, fluoranthene, and benzo(b) fluoranthene. Sources for total PAHs, as well as for naphtlene and pyrene, which are predominant in the first ones, seem extremely dispersed all around the islands and are not concentrated only in the urban centers. However, the highest contents of fluoranthene and benzo(b)fluoranthene seem associated to freshwater inputs rich in PAHs and issued from the crowded urban centers where polluted waters, sewage, and streams easily reach the lagoon.

Even if the USA's "non-polluted water" total PCB reference level has just been reached in Mayotte, it appears that oysters are relatively contaminated by total PCB, mainly near large urban zones: the eastern and northern coasts of the Main Island and western coast of Pamandzi Island. Concerning the evolution of the situation

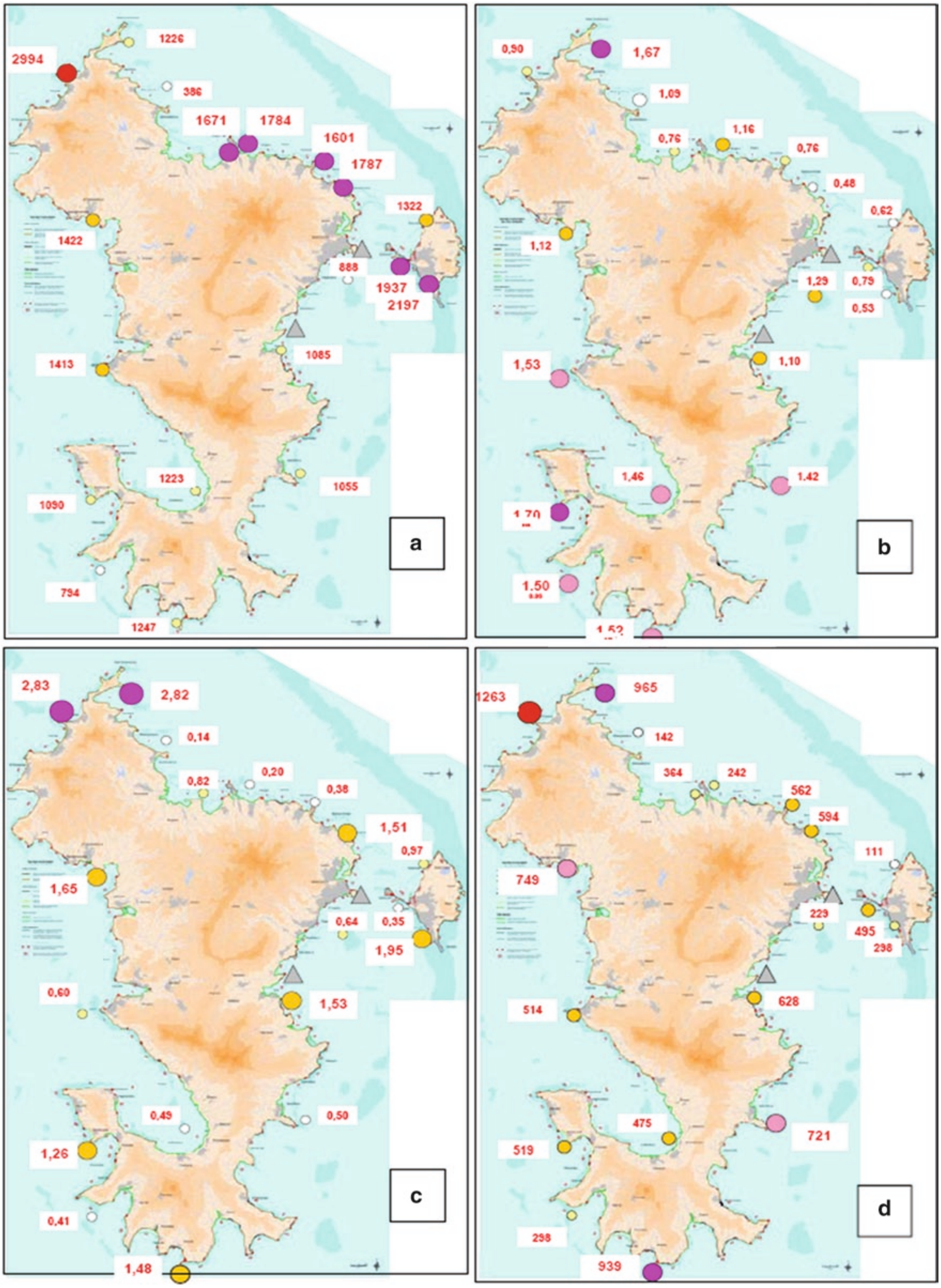

Fig. 4 Distribution of some metal contents in the oyster flesh around Mayotte: (**a**) zinc; (**b**) cadmium; (**c**) lead; (**d**) copper

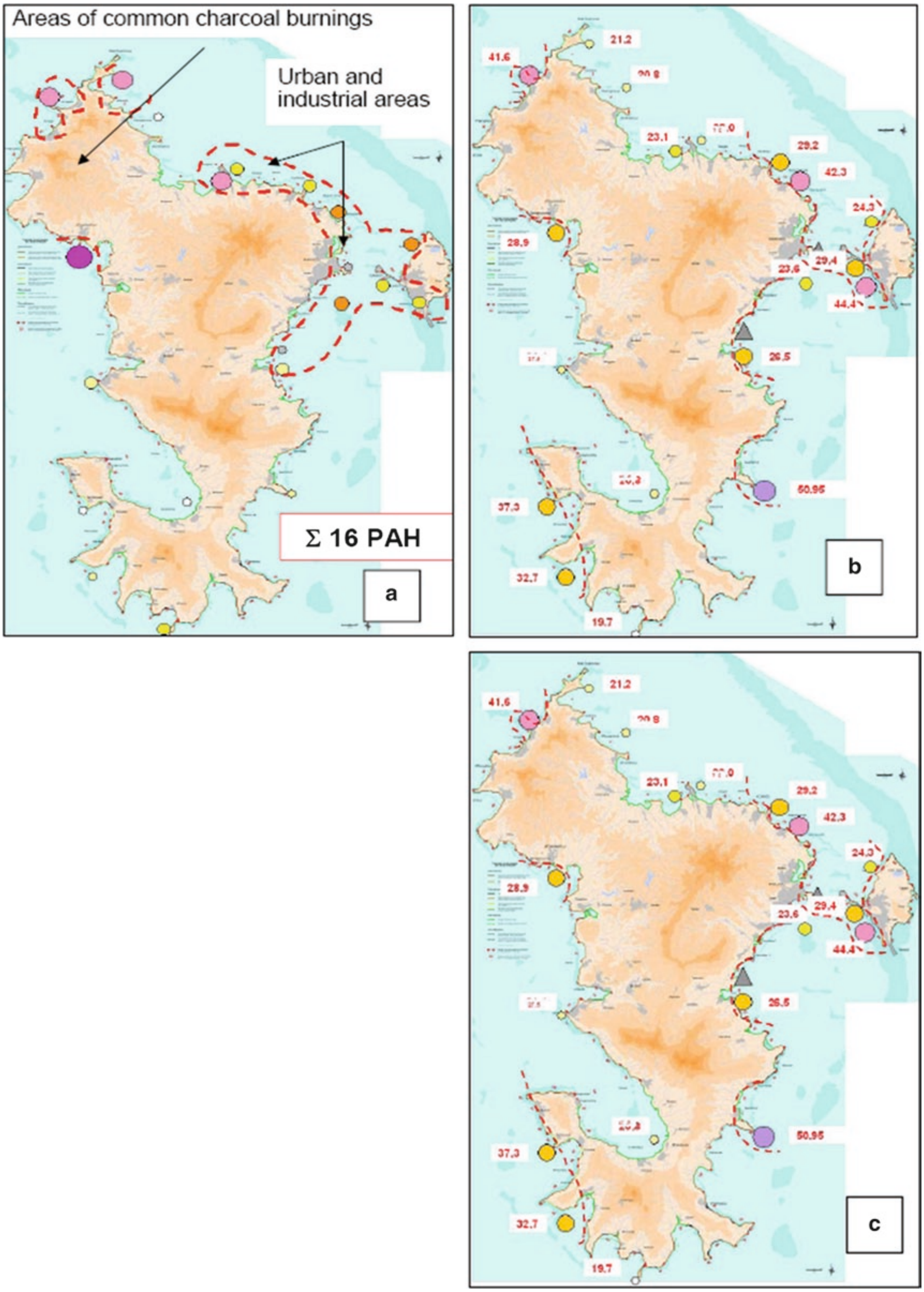

Fig. 5 Mayotte: distribution: (**a**) Σ_{16} PAH content; (**b**) total PCB content; (**c**) PCB toxicity index in the oyster flesh

between the years 1977 and 2007, an important degradation in the quality of the coastal waters has occurred in the eastern lagoon, around the Mamoudzou–Dzaoudzi strait, and down the Mamoudzou–Majikavo conurbation on the Main Island and the Dzaoudzi–Labattoir–Pamandzi conurbation on the Pamandzi Island. This is linked to an explosion of the urban population and the anthropogenic impacts induced. The same phenomenon has probably occurred along the southern coast of Mamoudzou–Passamaïnti and is increasing. The same has occurred for the Main Island's northern shorelines with the impacts from the large, developing villages.

Consequently, with this degradation of the coastal surface seawaters of the lagoon, a degradation of the coral coverage and vitality along the fringing reef fronts (Wickel and Thomassin 2005, 2006) has been observed. Correspondingly, coral and coastal fish communities are in decline.

A new balance between human development in the Mayotte Islands and the evolution of the natural marine ecosystems is currently being established.

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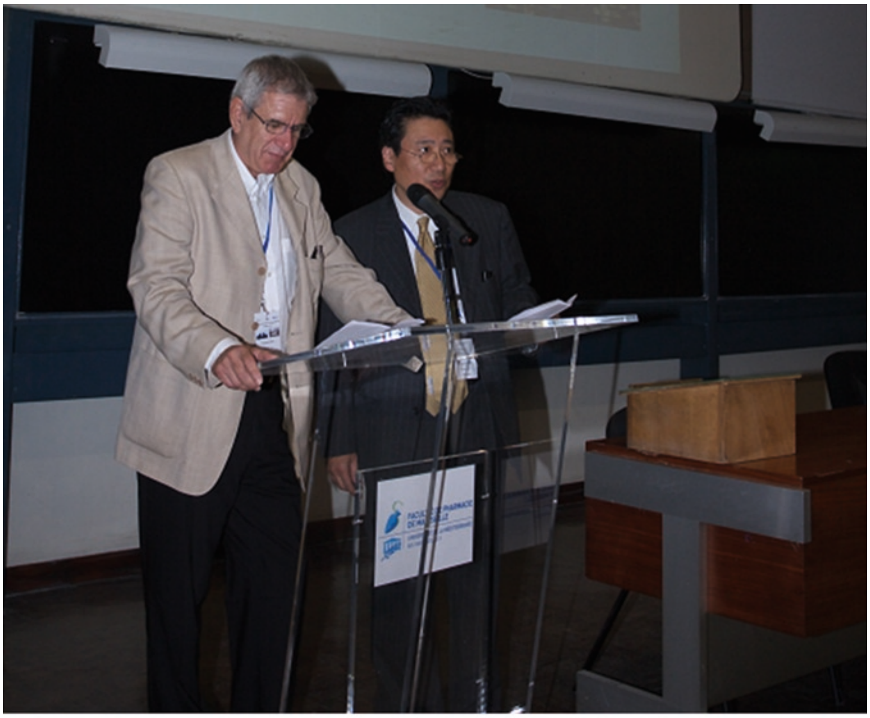

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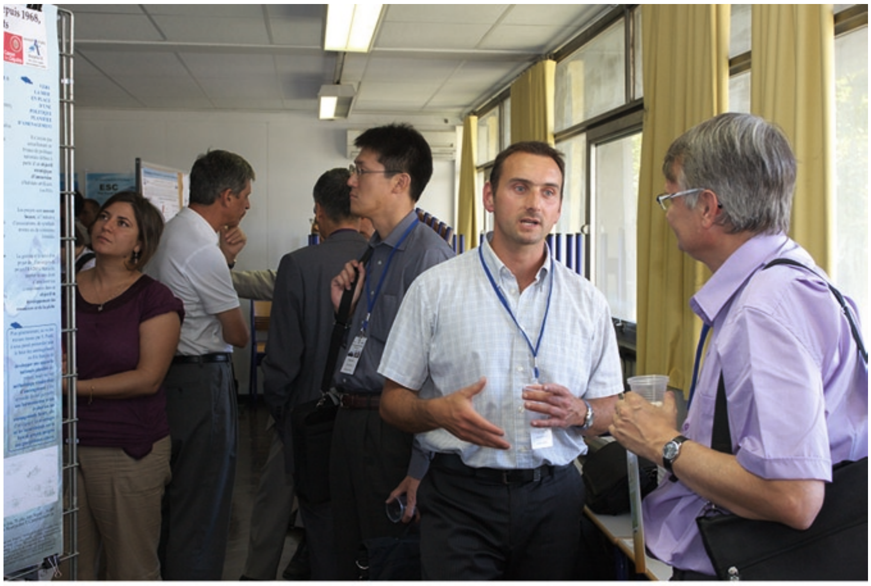
Dr. Gérard VERON (IFREMER), Prof. TANIGUCHI Akira (Tokyo University Agriculture, Okhotsk), Prof. Teruhisa KOMATSU (University of Tokyo), Dr. Eric DELORT (In Vivo / IXSEA), Prof. Ivan DEKEYSER (Director of Centre d'Océanologie of Marseille), Mr Pierre GAIGNEUX (Marseille Administrative Court), and several other participants

Prof. IMAWAKI Shiro, President of SFJO; Prof. Hiroki YAGI, (vice-President of SFJO, Otaru University of Commerce), Dr. Didier SAUZADE (IFREMER), Dr. FURUKAWA Keita (National Institute for Land and Infrastructure Management), Dr. Michel DENIS (Centre d'Océanologie de Marseille), Dr. IIBUCHI Toshio (Marine Ecology Resarch Institute) and several other participants

A general view of participants: Prof. NAKAGAWA Heisuke (Hiroshima Universitu), Dr. Yves HENOCQUE (IFREMER), Dr. Christian (Ministry of Industry), Dr. IIBUCHI Toshio (Marine Ecology Research Institute), Prof. IMAWAKI Shiro (JAMSTEC), Dr. Michel DENIS (Centre d'Océanologie de Marseille), Dr. Didier SAUZADE (IFREMER), Prof. HAMANO Akira (National Fisheries University)



A poster session

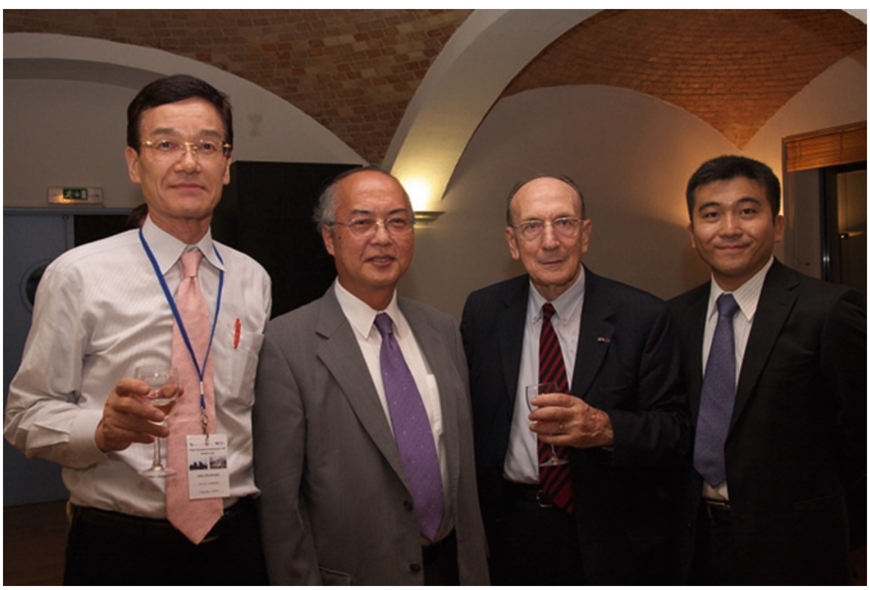
Poster session: Dr. Didier SAUZADE (IFREMER), Prof. EBATA Keigo, (University or Kagoshima), Prof. Denis BAILLY (University of Brest)

Meeting between young researchers, with Dr. Lounis MEBAREK and Mathias GIRAULT

During a coffe break: *On the left*: Prof. TAKAI Rikuo (President of Tokyo University of Marine Science and Technology), Prof. TANIGUCHI Akira (Tokyo University Agriculture Okhotsk), Prof. Bernard QUEGUINER (Centre d'Occéanologie de Marseille)

Signature of the twinning agreement between Tokyo University of Marine Science and Technology and Université de la Méditerranée. *From left to right*: Prof. TAKAI Rikuo (President of Tokyo University of Marine Science and Technology), Prof. Yvon BERLA ND (President of Université de la Méditerranée), Mr BAMBA Masahiro (General Consul of Japan in Marseille)

Congratulations and exchange of gifts after the signature of the twinning agreement between Tokyo University of Marine Science and Technology and Université de la Méditerranée. Prof. TAKAI Rikuo (President of Tokyo University of Marine Science and Technology) and Prof. Yvon BERLAND (President of Université de la Méditerranée)



Unformal meeting after the signature ceremony. From the left to right: Prof. IMAWAKI Shiro (JAMSTEC, President of Société franco-japonaise d'Océanographie of Japan), Mr BAMBA Masahiro (General Consul of Japan in Marseille), Prof. Hubert-Jean CECCALDI (President of Société franco-japonaise d'Océanographie of France) and Mr IWATA Yoshikazu (Vice-Consul for cultural Affairs)

Meeting for the synthesis of meetings in Maison de la Culture du Japon (Nihon Bunka Kaikan) in Paris. Prof. KOIKE Yasuyuki and Prof. ARAKAWA Hisayuki (Tokyo University of Marine Science and Technology), Prof. KOMATSU Teruhisa and Prof. TAKAGI Ken (University of Tokyo and Representative of Techno-Ocean Network), Prof. NAKAGAWA Heisuke (University of Higashi-Hiroshima), Mr Christian VICENTY (Ministry of Economy), Dr. IIBUCHI Toshio (Marine Ecology Research Institute), Prof. YAGI Hiroki, Vice-President of SFJO (Otaru University of Commerce, Otaru), Dr Richard SEMPERE (Director of research laboratory in Centre d'Océanologie of Marseille), Dr. FURUKAWA Keita (National Institute for Land and Infrastructure Management), Dr. Patrick BARAONA (President of Marine Cluster of Mediterranean), Dr. Guy HERROUIN (IFREMER and Marine Cluster of Mediterranean), Dr. Yves HENOCQUE (IFREMER, Bureau of Strategy) and Emeritus Prof. Hubert-Jean CECCALDI (Academy of Marseille, President of Société franco-japonaise d'Océanographie of France)

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