DEEP
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Desbruyères et al.

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Deep-sea Genetic Resources

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by Ifremer / Deep and Biomar departments

Ifremer

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DEEP-SEA GENETIC RESOURCES

by
Département « Etudes des écosystèmes profonds »" (DEEP)[1]
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Two third of our Planet are covered by oceans more than 3,000m deep (≈ 307 millions km²) and the mean depth of the sea is approximately 3,800m. The volume of the deep ocean (aphotic) is about 1,280 Millions of km^3 while the volume of terrestrial ecosystems is only 125 Millions of km^3. Therefore, the deep-sea ecosystem is by far the largest complex biome on the Earth. However, it suffers from a general disinterest of the public and decision makers due to its remoteness and inaccessibility. Edward Forbes "azoic theory" is still deeply rooted in most people brains and during public conferences, a large part of the audience being still amazed to hear that macroscopic life does exist in the deepest part of the sea floor (11,000m depth).

Characteristic features of deep seabed ecosystems.

Until the seventies, the deep seafloor was considered as relatively desert, due to the fact that in absence of light, all the deep-sea food-web is relying on organic matter synthesized at the sea surface by photosynthesis, and deposited as vertical fluxes of particles andthat decay by microbial heterotrophic processes during their descent. The benthic biomass in the deep ocean is thus generally low (of the order of $1g/m^2$ wet weight) and the biological turn-over is slow. Conversely, the specific diversity of abyssal sediments is high, whereas deep-sea is considered as one of the main reservoirs of biodiversity in the world. In a much debated paper, Grassle and Maciolek (1992) estimated by calculation based on $21m^2$ samples, that the deep sea would have 10^7 macrofaunal species living on and inside the sediments. More conservative estimations have been published since this paper, but every deep-sea cruise is still bringing its batch of amazing taxonomic novelties. Even when their

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biomass and turn-over look negligible, organisms living on the deep-sea floor contribute to carbon cycling by the terminal mineralisation of organic matter. Conversely, a large part of the human chemical pollution affects the deep-sea benthos, directly through the oceanic food web. Several *in situ* experiments have demonstrated that slow-evolving deep-sea communities could become very dynamic when disturbance is occurring; their high proportion of rare species allows them to respond to a large set of unpredictable events (e.g. diverse inputs of organic matter or chemical energy). If our knowledge of deep-sea animals is still in its infancy and subject to re evaluation, the situation is by far worst when considering the reduced knowledge of the microbial realm (from microbe to virus).

Our view of the deep seafloor has changed with the exploration using modern technologies which demonstrated that the deep seafloor is scattered with heterogeneous habitats where dynamic biologic processes occur.

- Situated at the edge of continental margins, canyons are hotspot ecosystems characterised by a high biodiversity. These features act as pathways for organic matter transportation from the land to the deep-sea. Canyons contain a diversity of habitats, such as hard-rocks wall, and mobile sediments with probable high degrees of endemism.
- Seamounts are undersea mountains with steep slopes, the presence of hard and soft substrates, large depth ranges from abyssal to sub-littoral and geographic isolation. It is estimated that 100,000 seamounts over 1000m in height exist in the world ocean. Very few (350) have been sampled and the question of endemism in such isolated structures is still pending. A group of French and Spanish scientists working on the endemism of the seamount fauna in Southwest Pacific demonstrated that rather than functioning as areas of endemism seamounts may be highly productive and support numerous species in small areas; therefore the previously hypothesized endemism of seamounts is likely due a bias resulting from under-sampling of a mosaic of habitats.
- Anoxic areas are situated in areas where mid-water oxygen minima intercept the continental margin or other geological features, resulting in sediments with very low oxygen concentrations (Oxygen Minimum Zones or OMZs). Seafloor OMZs typically occur between 200m and 1000m. Despite very low oxygen concentrations, microbial and metazoan life thrive in these ecosystems.
- Hydrothermal vents, discovered in 1977 on the Galapagos Rift, have been found in all oceans, in back-arc basins and associated with arc-volcanoes. In these areas, cold seawater penetrates through cracks in the oceanic crust or in mantellic rocks. The fluid gets heated and is depleted of oxygen and magnesium while being loaded with other metals and gazes. The superheated fluid (350°-400°C) rises back to the seafloor where it mixes with ambient cold seawater. These active vents are associated with dense metazoan population representing amazing biomass (i.e. kgs /m^2) and high biological production made by few taxa ($\approx$ 600 metazoan species presently described over the world,

90% endemic). Those species rely on bacterial chemoautotrophic production made by dense and diverse microbial communities colonizing a gradient of habitats varying from high temperature fluids to cold ambient sea and substrate. and edifices are also present in The same environments, rich in polysulfide inactive deposits harbours dense psychrophilic microbial communities. Those environments will be in the upcoming years likely subject to mineral exploitation. Only 93 hydrothermal fields have been presently explored and described so far, each of few hundred square meters along the 60,000 km long hydrothermal ridges of which only a small fraction have been explored. In other words, this represents a very limited portion of the total potential habitat..... The hydrothermal vent sites are subject to aperiodic extinctions and re-colonization of new active areas. This instability of venting is linked to the spreading rate of the ridges, high in ultra-fast ridges (Southern EPR[2]), low in slow ridges (MAR and SWIR).

- Cold seeps were discovered in 1983 by 3270m in the Florida escarpment. They are characterized by seepages of cold fluid with high concentration of methane. They are found on active and passive continental margins. The methane may have biogenic or thermogenic origins. Cold seeps are also rich in hydrogen sulphide produced by the bacterial reduction of sulphate coupled with the oxidation of methane by microbial consortia. On passive margins, these cold-seeps are frequently associated with areas of oil reservoirs (e.g. Gulf of Guinea, Gulf of Mexico). The cold seep fauna is taxonomically close to that of hydrothermal vents and is composed of a high proportion of endemic species harbouring bacterial endosymbionts. Since their discovery, aproximately 230 animal species have been described from cold-seeps. The stability of cold-seep habitat is supposed to be higher whereas the population dynamics of the major species appear slower to those observed in hydrothermal vents.
- The massive input of organic matter (e.g. whale carcasses, wood, kelp remains) at the deep water/sediment interface is first used by scavengers or wood-borers.Opportunistic species follow enriching the local sediments. The accumulation of their faecal pellets and the local decay of organic matter by microbial decomposition produce hydrogen sulphide which is secondarily used by chemoautotrophic microbes. The metazoan communities that develop, share some cognate taxa with vent and seep communities.

State-of-the-art technology for deep-sea biological studies in France.

State-of-the-art technology is of primary importance to study deep-sea ecosystems (location, mapping, exploration and sampling). Important technological considerations have to be taken in account when discussing the census of marine

[2] EPR : East Pacific Rise; MAR: Mid-Atlantic Ridge; SWIR: South-West Indian Ridge.

deep seabed resources, due to the difficulties in accessing environments that are extreme in terms of pressure and sometimes temperatures. Since the late '70s the technological progresses (multibeam echo sounders, submersibles, in situ sensors ...) and the cooperation between disciplines, allowed an access to small fragmented geological features as hydrothermal vents, cold seeps, canyons, sponge reefs, deep sea corals situated on continental margins, seamounts, oceanic and volcanic ridges and back-arc basins. In these fragmented areas, where physical and chemical gradients are sharp (at centimetre scale), sampling from the surface using grabs, corers, dredges and trawls are not only non scale-adapted but also destructive for fragile environments. These fragmented habitats represent a small proportion of the deep seabed surface which is dominated by sedimented areas fuelled by organic input from surface layers, but are the most productive habitats on the deep-floor. Despite the present interest of the scientific community on these fragmented habitats, the importance of psychrophile genetic resources within the soft-sediment deposit-based ecosystem still has to be evaluated.

Ifremer is a public independent institution undertaking research and development activities in the field of marine sciences. It ranks among the small number of research institutions in the world continuously involved in deep-sea investigations during the last decades. This includes the development of research vessels, man-operated submersibles, ROV and specialized sampling equipment for deep-sea research, the deployment of these equipments mainly for scientific cruises conducted by its own research teams or academic teams, but also sometimes for commercial purpose.

Ifremer owns and operate several vehicles to undertake deep-seabed research. Since the 70's, the French scientific community has access to deep-submergence manned submersible (Cyana 3000m and since 1984 Nautile[3] 6000m with presently account for more than 1500 dives) and developed a dedicated instrumentation for sampling, measuring and experimenting on the deep-sea floor. Since 1998, the ROV "Victor 6000"[4] (to date more than 4000 hours on the bottom for scientific research) is used by scientists for a large part of the cruises studying deep-sea ecosystems. Its modular equipment allows mapping, imaging and experimental work at a centimetre scale. Moreover, maintaining sampled organisms intact and alive, as well as culturing them, requires sophisticated and expensive technologies. Controlling *in situ* techniques (e.g. ALCHIMIST and CHEMINI based on *FIA*) for physical and chemical characterization of habitats, but also for *ex situ* experimentation under pressure (several under-pressure aquaria developed by laboratories of "*Université Pierre et Marie Curie*" and "*Université de Bretagne Occidentale*" are used aboard the scientific fleet) are one of the prerequisites for maintaining and cultivating organisms under pressure aboard research vessels and in the laboratory. The autonomous underwater vessel (AUV)[5] Aster^{X} is an autonomous shallow and deep-sea vehicle cruising at a depth of 3000m and allows mapping and detecting active areas of the seabed (multibeam and fishery echo sounders, ADCP ...).

3 http://www.ifremer.fr/flotte/systemes_sm/engins/nautile/nautile.htm

4 http://www.ifremer.fr/flotte/systemes_sm/engins/victor.htm

5 http://www.ifremer.fr/flotte/systemes_sm/engins/asterx.htm

On going deep-sea biology programmes in France.

Currently, Ifremer's main programmes of basic research related to exploration and exploitation of the biodiversity of the ocean floor include a programme (Geode) focused on fragmented chemosynthesis based ecosystems situated on continental margins and ocean ridges (cold-seeps and hydrothermal vents) and a second programme aiming to study human impact on deep-sea ecosystems (nodule mining impact, ultra deep oil). Part of these programmes are co-sponsored by European Commission (e.g. HERMES focused on biodiversity hotspots along European continental margins, EXOCET/D on development of the technology for the study of biodiversity in fragmented deep-sea environments, MARBEF and MARINE GENOMICS, two E.U. "networks of excellence" on marine biodiversity and genomics).

Marine Genetic resources in the deep-sea.

Deep sea ecosystems play an important role in carbon cycling, mineralization, stabilization of the greenhouse gazes (e.g. methane). Deep-sea genetic resources are also part of the legacy for the Humankind and for the Planet for its role in functioning and evolution of the biosphere. Beyond the numerous economical considerations that confer to deep-sea genetic resources a high economical potential, both due to the variety of molecules discovered and to be discovered, and to the wide range of potentially interesting genes due to molecular adaptation to extreme conditions, non economical considerations are not least. Due to the wide variety of extreme environments, deep-sea encloses treasures of molecular adaptation to extreme conditions, all of which may help understanding the selective processes allowing evolution and persistence of life forms in extreme conditions. The heterotrophic deep-sea environment, poor in oxygen, rich in methane, sulphur or carbon dioxide, is on Earth the most reminiscent of the so called "primeval soup" where life appeared spontaneously on earth and from which all living creatures are derived (Martin and Russell, 2002). The bacteria and *Archae* living specifically in those extreme environments may be (?) the closest descendant of those first life forms (Morell, 1997). The description of biodiversity and genetic resources from deep sea may therefore be the source of a fundamental knowledge to retrace the history of life on our planet, with the first apparition of living forms relaying on nucleic acids for their life, reproduction, persistence and evolution. Despite its remote location, deep sea environment is not protected from human global impact and massive extinction. It appears indeed clearly that the marine environment ranging from coastal and surface waters to deep-sea areas, is not constituted by isolated entities, but encloses strongly interconnected and interdependent ecosystems. Knowledge and preservation of genetic resources in Deep Ocean is therefore of critical interest for economic potential, as well as to preserve our patrimony in terms of biodiversity and understanding of life history on Earth.

If concerns grew over the impact of scientific research on fragile ecosystems, several initiatives were adopted by scientific international bodies to minimize the impact of scientific cruises on these fragmented ecosystems characterized by high endemism. Ifremer is contributing to international cooperative programmes as "CENSUS OF MARINE LIFE" aiming to take a census of marine species (projects ChEss, CoMarge, Mar-Eco and CeDAMar) and to InterRidge an initiative for international cooperation in Oceanic Ridge studies. In both international programmes, debates have been organized on impact of scientific research on fragile ecosystems and ended in recommendations ("codes of conduct") to minimize the disturbance on marine organisms and habitats during scientific cruises. Threats to these biodiversity hotspots will probably increase in the upcoming decades concomitantly to the likely "industrialisation" of the deep ocean (deep-sea fisheries, oil, mineral mining) of which impact would overtake by far that of academic research. Bioprospecting marine genetic resources in the deep seafloor is at the present time mostly focussed on microorganisms living in extreme habitats (hydrothermal vents, cold-seeps) and seldom if ever focussed on metazoans. Thus the impact of bioprospecting in the deep-sea that needs small volume of samples is of the same order that impact of basic research in microbiology and must follow the same precaution exposed in different "codes of conducts".

Marine microbiology and extreme deep-sea environments: Basic research and bioprospecting of marine resources.

Marine microbiology is a field of research characterised by its rapid expansion mainly due to (i) the two successive scientific revolutions based on molecular biology and genomics sensu lato and to the (ii) discovery of the fundamental contribution of the microbial compartment to the understanding of life and biogeochemical cycles in the oceans. Two examples illustrate this phenomenon: new picoplankton groups like *Prochlorococcus*, *Synechococcus* were recently found to play a key role in ocean biomass turnover; the recent discovery of the microbial consortium responsible for the anaerobic oxidation of methane (AOM) shed partially the light on the degradation of methane seeping from the sediments along the continental margin and contributes to the methane cycle and the regulation of greenhouse effect. Despite massive efforts and the use of powerful methods based on metagenomics (cf. Sargasso Sea metagenome by C. Venter et al. 2004), inventory of microbial diversity in marine environments is still far from complete and will require at the very least several decades. Viruses are the most common biological entities in the oceans by an order of magnitude in addition to micro and picoeukaryotes, Bacteria and Archaea. Despite the fact that most of the viruses are still unknown, being sometimes documented only by environmental clone sequences, their key roles in geochemical cycles, microbial population dynamics and lateral gene transfer are gradually emerging as a new paradigm in ocean sciences.

With the advent of the metagenomic methods allowing a direct access to genes contained in DNA extracted from crude samples, gene libraries represent a more and more important type of genetic resource, emerging in parallel to strain collections. As for isolated microorganisms, the jurisdictional status of crude samples, and their associated genetic resources, collected beyond national jurisdiction is still unclear. However, considerations developed for isolated strains still apply to crude samples: when contracting with a private investor or a private society, the best guaranty to protect the investor's rights is still to provide samples collected in your own country jurisdictional area. By default, samples collected beyond national jurisdiction can, under current regulations, insure a rather good but not absolute control of the associated genetic resources.

In the future, microbial genetic resources will encompass microbial collections, gene libraries and crude samples.

Although the future is unpredictable, it is likely that biotechnology primarily marine biotechnology will play a much more significant role in the 21st century than it did during the last century. Marine biotechnology is a multidisciplinary research field, which includes marine biology and chemistry, molecular biology, organic and natural product chemistry, biochemical engineering, physics, pharmacology, and medicine. One of the most exciting aspects of marine biotechnology is bioprospecting: the search for new natural compounds that can be used as novel pharmaceuticals, healthcare products, agrochemicals and novel bioremediation agents for environmental protection. So far, more than 15 000 molecules have been isolated and described from different marine sources including invertebrates, algae and marine microbes. The number of novel compounds continues to grow as new discoveries are made in diverse marine ecosystems.

Among the various groups of marine macro-organisms which have been the most widely employed producers of bioactive molecules, are the sessile (sponges, tunicates and bryozoans) or slow moving (snails) life forms. In many cases, microorganisms present in the tissues of these higher organisms are suspected to be involved on the biosynthesis of active biomolecules (or to be the actual source of bioactive molecules). For this reason, the search for novel sources of new bioactive molecules is particularly challenging, as the interrelationship between micro-organisms and their host macro-organisms can be very complex.

Microorganisms continue to provide an exciting source of novel metabolites. Interest in this particular field is due in part to the ability of many organisms to produce these natural products through fermentation, thus minimizing supply problems. Compared to terrestrial sources, the search for new metabolites from marine micro-organisms is only beginning and it can be expected that in the near future marine organisms will satisfy the demand for new metabolites from terrestrial biological sources. Undoubtedly micro-organisms, whose huge genetic and biochemical diversity is only beginning to be explored, look likely to become a rich source of novel chemical entities for new drugs.

Among the bioactive natural products reported to-date, a significant number have progressed to clinical trials, mainly for cancer treatment. However, other life threatening diseases such as infections, inflammatory processes, heart disease, circulatory system disorders, hyperlipidemia, Alzheimer diseases, etc, will also benefit from the quest for new drugs from marine ecosystems.

Aside from the so called "secondary metabolites", other biomolecules -such as biopolymers- could be of great utility for the biotechnology industry. Prokaryotic biopolymers offer a number of novel material properties and are finding increasing uses in the biotechnology and biopharmaceutical industries. The physical, rheological and biological properties of bacterial exopolysaccharides and bacterial pigments, for example, can be exploited for a number of products and bioactive molecules ranging from emulsifiers to adhesives. Another example of marine bacterial biopolymers with commercial promise, are the poly hydroxyalkanoates (PHAs), a class of biodegradable plastics as potential substitutes for petroleum-derived polymers.

Relatively little is known about deep ocean ecosystems, but many scientists now believe that the deep sea harbours the most diverse ecosystems on earth along with an exceedingly rich biodiversity. This biodiversity holds tremendous potential for human benefit. Increasing scientific and biotechnological interest is now being focused on the potential of organisms living in extreme environments including the deep sea, deep sediments, cold seeps and deep sea hydrothermal ecosystems. These organisms have developed unique survival strategies to survive extreme conditions such as darkness, cold or hot temperatures, chemically toxic and highly pressurized environments. It is now widely accepted that extremophilic microorganisms will provide a valuable resource not only for exploitation in novel biotechnological processes but also as models for investigating how biomolecules are stabilized when subjected to extreme conditions.

For almost two decades Ifremer has been investigating the deep ocean from cold seeps to deep sea hydrothermal vents. The collection of microorganisms recovered from the different oceanographic cruises has been screened for enzymes (primarily thermostable enzymes), novel biopolymers including exopolysaccharides and poly hydroxyalkanoates and, more recently, and secondary metabolites. Indeed, deep-sea hydrothermal vents now offer a new source of fascinating microorganisms well adapted to these extreme environments. Over the past 20 years, an increasing number of new genera and species of both hyperthermophilic and mesophilic bacteria have been isolated from these deep-sea ecosystems. This new bacterial diversity includes strains able to produce novel molecules such as enzymes, polymers and other bioactive molecules. The actual collection of microorganisms owned by Ifremer and originating from deep sea ecosystems, increases with the different sampling cruises performed by the French institute. To date up to 1300 isolates have been recovered from diverse origin. Those presenting a biotechnological interest have been protected by deposit in international collections.

Novel exopolysaccharides with innovative rheological, physical and biological properties have been isolated from deep-sea bacteria along with novel biodegradable poly hydroxyalkanoates and new chemical structures have been characterized. Such biopolymers are produced under laboratory conditions far different from those conditions encountered in their natural habitats. The search for novel enzymes by the industry relied initially on the screening of collections of microbial strains for the appropriate biocatalysts. Among the collections of microorganisms of interest, the marine extremophiles were particularly attractive since the diversity of physico-chemical conditions (pressure, temperatures, pH, salt contents, etc) and metabolic properties encountered is huge. Besides already commercialised products like DNA polymerases, many others (proteases, lipase/esterases, alcohol dehydrogenases, phytases, etc.) have been characterized. Whenever the industrial process requires improvement of the natural enzyme, the availability of many strains belonging to the same species or genera allowed the use of directed evolution to comply with the industrial specifications. The trend is to consider a whole industrial process and to optimise it in combining chemical reactions and enzyme driven reactions. To this end, marine extremophiles still hold a great potential.

Property rights and deep marine resources: Problems and suggestions.

The deep ocean is a potentially huge source of important compounds, a source that science and scientists have just begun to explore. Less than 10 bioactive molecules from sponges, deep coral and microbes have been investigated thus far, and many are under preclinical development or clinical trials. The study of deep sea biodiversity is in its infancy, and it can be expected that the discovery of new habitats and their associated life forms will yield new natural products and drugs.

Given that affordable technology for the exploration on the abyss is currently being developed allowing the discovery of new ecosystems, given that the biochemical diversity in the ocean is higher than on land, given the tremendous progress in molecular biology and chemistry, more deep sea innovative natural products and drugs are expected in the very near future.

Compared number of bioactive compounds with pharmaceutical potential (due to the much higher incidence of significant cytotoxic activity) extracted from marine versus non marine species (Fig. 1).

M.H.G. Munro et al. / Journal of Biotechnology 70 (1999) 15–25

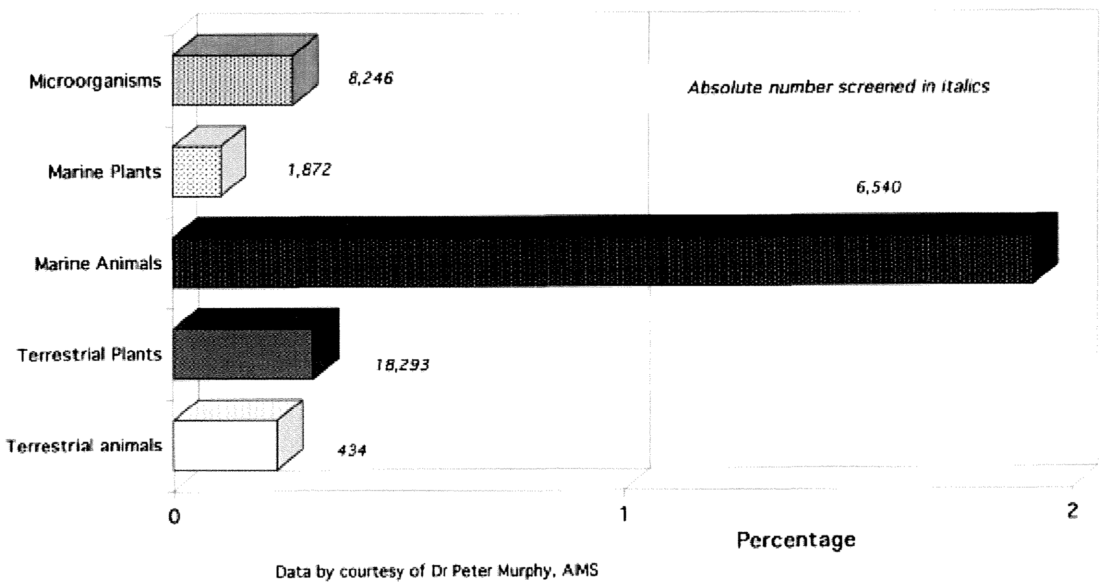


Fig. 1. Distribution of samples with significant cytotoxicity in the NCI's preclinical screen.

In the course of drug and new metabolites discovery, marine biotechnology is going to play a pivotal role in this new millennium. The exploration of untouched ecosystems will provide tremendous opportunities for the discovery of new bioactive molecules for human benefit. But this exploration must be protected from destructive practices. And because much of the deep sea lies beyond the zones of national jurisdiction, it is up to the international community to manage and protect this great resource for all of humankind.

The genetic resources of the deep-sea extreme environments, mainly hydrothermal vents, were clearly identified since the eighties by several institutions, among which Ifremer, as a strategic resource for biotechnology. Considering the limits of the national jurisdictions currently regulated by the UNCLOS (United Nations Convention on The Law of the Sea), and the practical experience gained within the frame of projects partly designed to collect samples in different national contexts, it has rapidly been obvious that, despite some drawbacks, the most secure way to enter bioprospecting of genetic resources was either to stay within the limits of its own national jurisdiction or to go beyond national jurisdictions in international water bodies.

Based on the samples of fluids, rocks, sediments and animals collected in deep-sea extreme environments (hydrothermal vents, cold seeps, subsoils, and deep-sea trenches), collections of strains have been established by various institutions having access to sampling technology. Among thousands of strains, a relatively low number of bacteria and archaea have been described, published and deposited in international reference microbial collections (Pasteur's CIP, DSMZ, ATCC, JCM...). However, most of the isolated strains are still under control of each institution involved in isolation steps and subjected to tight limitations in their diffusion to third parties, often based on industrial contracts. These collections of unpublished strains constituted the core of the genetic resource for biotechnology. Their legal property status might not be clearly defined on the basis of international jurisdiction but a full property is clearly claimed by the institutions responsible for their development and their curation.

The use of these microbial genetic resources as starting point for the development of commercial products or processes requires huge investments and specific conditions. First, the status of deep-sea bed genetic resources beyond national jurisdiction cannot indefinitely be unclear, from open access to common heritage of humankind. Second, the intellectual property rights (IPR) related to marine genetic resources should be properly addressed. Concerning these two points, Ifremer as the French public research institute in charge of managing the national oceanographic equipments, considers that the open access status is still adequate. However, the impact of sampling, either for research or for bioprospecting should be kept as negligible as possible in these extreme environments, especially when recovery timescale is long. To deal optimally with these genetic resources, it is suggested to exclude from IPR all crude samples, gene libraries and microbial collections, all of these containing, under different forms, natural genetic information. This is particularly important since enforcing IPR on genetic resources could prevent their future use in completely unexpected fields potentially full of promise and impair the development of innovative products. However, IPR should apply to products (i.e. recombinant proteins, compatible solutes, etc.) or processes obtained through various techniques using the genetic resources. These two principles should insure that in areas beyond national jurisdiction no one could establish the ownership on the genetic resources. On the other hand, building a collection of microorganisms or gene libraries from deep-sea crude samples requires huge investments and the ownership of the resulting collections and gene libraries have to be clearly recognised.

Annexe:

FRENCH DEEP-SEA BIOLOGICAL CRUISES

The exploration of deep seabed areas started at the end of the nineteenth century with the British research oceanographic vessel Challenger (1872-1876). Many deep seabed expeditions have also been led by France since the beginning of the 19's century. Ifremer holds a database containing information on the biological cruises. The Biocean database (www.ifremer.fr/biocean) was designed to collate the extremely large volume of data collected from different deep-sea ecosystem studies conducted by Ifremer's department of 'Etudes des Ecosystems Profonds' (Deep-Sea Ecosystem Studies). Metadata available on the internet have been used for this report. Ifremer research objectives are mainly located in the Atlantic Ocean and in the Mediterranean Sea, though some cruises occur in the Pacific Ocean and some used to occur in the Indian Ocean, according to the different programs. Table I provides an overview of the biological expeditions led by Ifremer in the deep sea.

DEEP-SEA ECOSYSTEMS	Nb cruises
Sedimentary Ecosystems	61
Hydrothermal vents	17
Canyons and continental margins	12
Canyons and Cold Seeps	3
Cold seeps	2
Nodules ecosystem	2
Deep coral reefs	1

Table I: Overview of deep-sea ecosystems explored by Ifremer french biological team since 1967 and available in Biocean database.

Sedimentary ecosystems

Expeditions devoted to the study of these ecosystems aim at assessing and explaining the diversity, distribution and abundance of marine life (Fig.2). They are focused on deep sea areas largely unexplored in order to expand human knowledge of extreme ocean ecosystems and to improve their conservation.

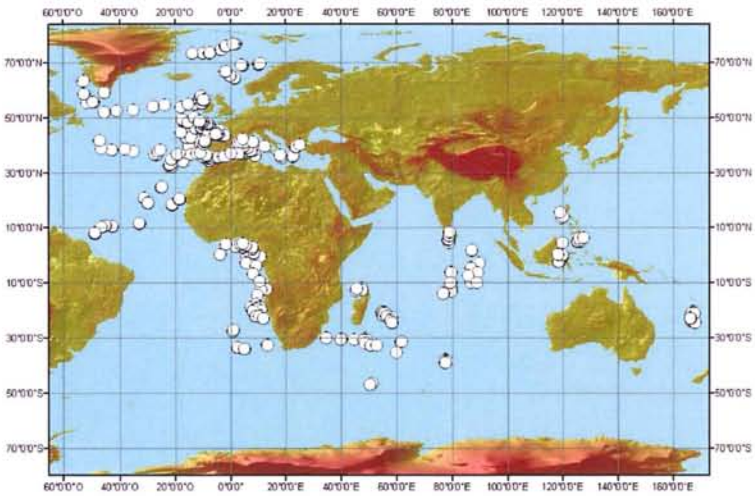


Fig. 2: Distribution of the data on deep-sea fauna from French cruises, stored in the BIOCEAN data-base (soft-sediment habitats)

Older cruises, conducted during the earliest days of deep-sea oceanography, were essentially exploratory expeditions along the continental slopes (Fabri et al, 2006). Since the 1980s an important change in the approach of deep-sea biological research has taken place. Descriptive oceanography has given way to more comprehensive studies. Sampling strategies have become more focused on limited areas in the deep sea and aim at understanding the functioning of these marine ecosystems.
Sedimentary ecosystem studies are now focused on canyons, coral reefs and nodule fields.

Canyons

Since 2000 two canyons have been extensively studied (Fig. 3):

- Zaire Canyon on which 10 cruises occurred among them one with the Victor ROV,
- Var Canyon on the Mediterranean continental margin; up to now 5 cruises have been organised but three are coming soon in the framework of the European 'Hermes' program.

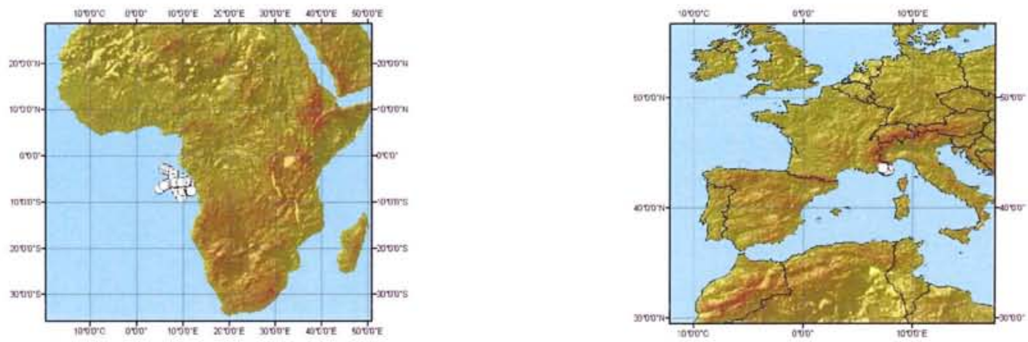


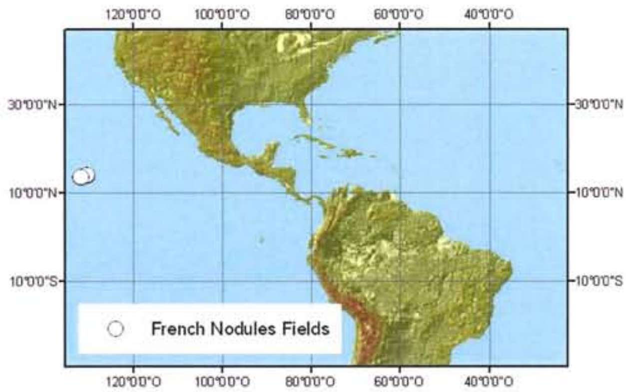
Fig. 3: Maps of French operations in the Zaïre and Var canyons.

Cold water coral reefs

With regards to the establishment of MPAs (Marine Protected Areas), consistent with international law and based on scientific information, cold water corals are being more and more studied along the continental margins. Ifremer has led one cruise in 2001 on the Ireland coast, and participated in two cruises in the north-east Atlantic Ocean and two in the Mediterranean Sea.

Nodule ecosystems

In 2004, Ifremer conducted a scientific cruise on the French mining claims in the Clarion Clipperton Fracture Zone in the North East Pacific Ocean in order to have the baseline for an impact study of potential nodule exploitation (Fig. 4). This first biological cruise followed 49 cruises led by geologists on this area. Although samples are still under process (specific identifications), first results have led to the conclusion that habitat diversity between nodules fields would sustain a higher biodiversity than surrounding abyssal plain.

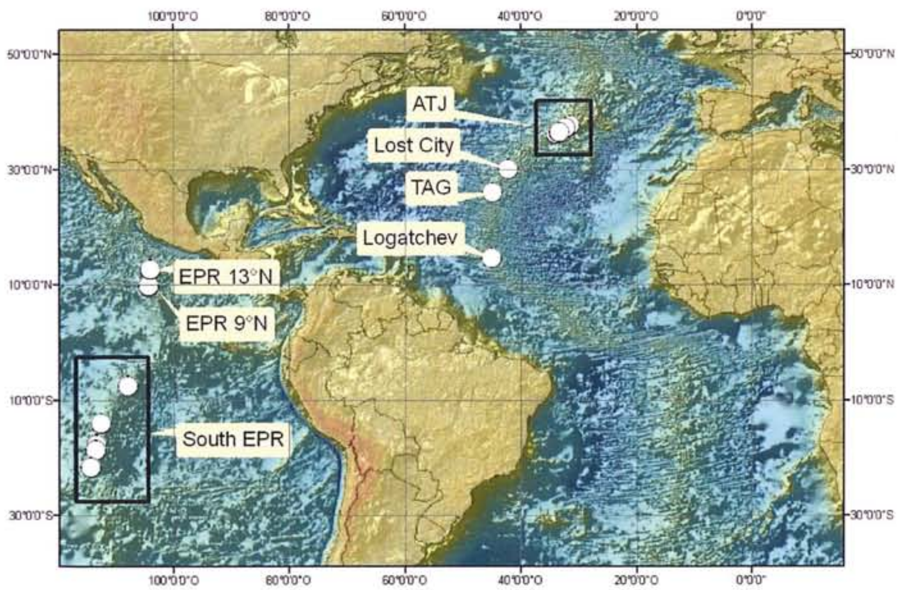


<u>Fig. 4</u>: Location of the French nodule mining claims in the Clarion-Clipperton Fracture Zone in the North-East Pacific Ocean.

Chemosynthetic ecosystems

<u>Hydrothermal vents</u>

The first French biological expedition BIOCYATHERM (1982) explored hydrothermal vents located on the fast spreading East Pacific Rise near 13°N. In 24 years, EPR/13°N has been visited by 8 French biological oceanographic cruises (BIOCYATHERM 1982, BIOCYARISE 1984, HYDRONAUT 1987, HERO 1991, HOT 1996, HOPE 1999, AMISTAD 1999, PHARE 2002, BIOSPEEDO 2004) and new sites have also been explored at 9°N and along the south part of the ridge as shown on figure 5.



<u>Fig. 5</u>: French cruises on EPR and MAR hydrothermal vents.

In 12 years, 7 biological cruises operating a submersible have been led by the French biological community (DIVA2 1994, MICROSMOKE 1995, MARVEL 1997, PICO 1998, ATOS 2001, EXOMAR 2005, MOMARETO 2006) on the Mid-Atlantic Ridge at the Azores Triple Junction (ATJ). During MICROSMOKE and EXOMAR expeditions devoted to microbiology, three sites have been sampled further south on the Mid-Atlantic Ridge: Lost City, TAG and Logatchev (fig. 4). The focus of ocean science is much more diversified than at the time of conventional oceanographic expeditions. Modern oceanography is becoming more interdisciplinary, e.g. the upcoming expedition SERPENTINE (March 2007) organised by Ifremer to determine the geochemical, biological and microbiological variability of hydrothermal processes on new sites recently discovered South to Logatchev vent field.

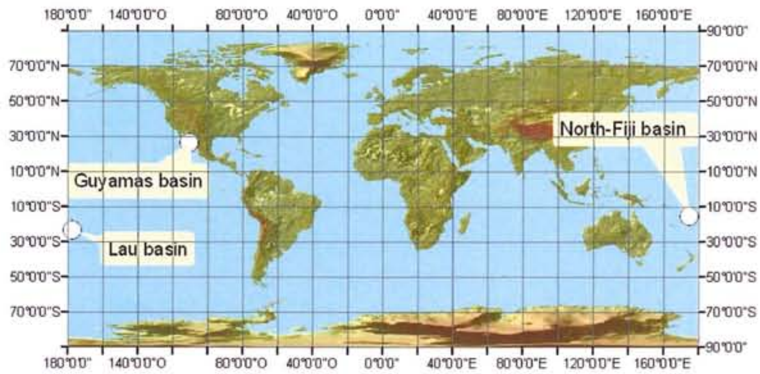


Fig. 6 : Location of Guaymas basin in the Gulf of California and of North Fiji and Lau Back-Arc Basins.

ATJ has become the subject of systematic observations under the international program called MOMAR ("Monitoring the Mid-Atlantic Ridge"),. This monitoring program results in an increase of the frequency of expeditions, some of them with a submersible and others to regularly recover moorings with small oceanographic vessels.
In addition to these cruises organised on French research vessels, French scientists have regularly been invited on US cruises, and vice versa.
French scientists have been also involved in cooperative projects with the Japan Agency for Marine-Earth Science and Technology (JAMSTEC) and organised two expeditions on back-arc basin hydrothermal vents in 1989, STARMER 2 in the North-Fiji basin and BIOLAU in the Lau basin (Fig. 6). Ifremer also cooperated with UNAM (Universidad National Autonoma de Mexico) (for the study of vent biology and microbiology in the Guaymas Basin, for the GUYANAUT expedition in 1991. The Guaymas basin is located in the Gulf of California. It is characterized by the co-occurrence, in a soft-sediment area, of hydrothermal venting and cold seepage activity. A new expedition (BIG) would take place in the same areas in the upcoming years

Cold Seeps
Since 1985, cold-seeps areas were visited by French biologists on geological expeditions on the subduction trenches off Japan, on the Peruvians margin and on the Barbados accretionary prism. Since 1998, a series of French oceanographic cruises have been organised either in the Mediterranean Sea on the Nil fan (MEDINAUT 1998, NAUTINIL 2003, to come Medeco 2007) or on the active margins of Angola and Gabon on Regab Site (BIOZAIRE1 2001, BIOZAIRE2 2001, BIOZAIRE3

2003) as shown on Figure 7. Another area of localised fluid escape features has been visited off Norway (pockmarks on the Storrega slope and mud volcanoes on Hakon Mosby) by French biologists in the frame of the European project HERMES in cooperation with the German teams (MPI Bremen).

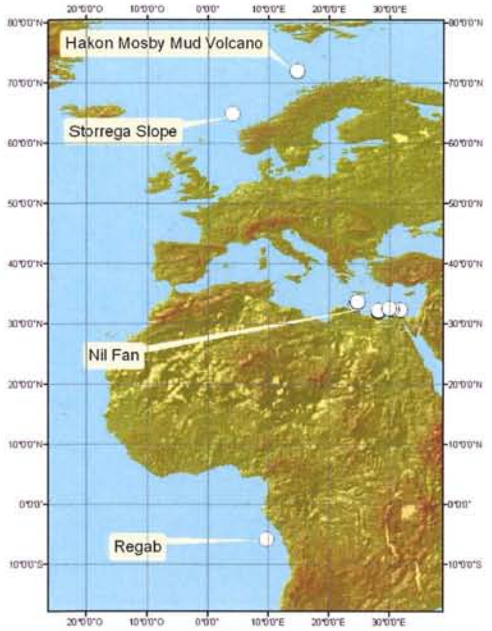


Figure 7: Location map of cold-seeps ecosystems visited by Ifremer biological teams in the Mediterranean Sea, on the active margins of Angola and Gabon and off Norway.

Reference:
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