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Exploring the life in the ocean: how do we know what is there?

Our lives are really rather two-dimensional. Our terrestrial environment is a very thin layer on the Earth's surface. Realistically it extends upwards only to the height of a large tree (about 50m), because nothing lives permanently above this level (roof gardens on skyscrapers don't count!). In contrast the ocean extends to 11km in depth, has an *average* depth of some 3.8km, and covers >70% of the Earth's surface. It is a vast three-dimensional environment without any parallel on land. We tend to ignore this environment because we are neither part of it nor can we "see" it, and its very scale makes it hard to comprehend.

Nevertheless the oceans and seafloor provide a vast reservoir of non-living and living resources, whose value and variety is still being recognised and whose exploitation and ownership presents problems for both technology and international law. There are organisms living out their lives at every point between the ocean surface and the bottom of the deepest trenches. These organisms and their interactions with each other and with their environment contribute hugely to the sustainability of "our" terrestrial life, particularly through their effects on nutrient recycling and climate. If we are to benefit from the ocean, attempt to "manage" it in any effective way and predict the consequences of any changes to it, we must understand how the chemistry, physics and biology interact and how its inhabitants contribute to, and perhaps even determine its condition.

Our challenge is to explore this habitat, to discover its communities and to understand how they are so successful, bearing in mind that the habitat is both hugely demanding and that it comprises more than 99.5% of the living space on our planet [1].

How do we know what organisms are there?

How can we find out what animals live in the deep sea, how many of them there are and how they interact with each other? Most of what we know about the terrestrial ecosystem derives from our ability to see it. We can often count individuals, watch their behaviour, and observe their development. We can follow their movements and migrations, learn how their communities are structured, and note how they interact with other species. We can even manipulate their environments and see how they respond.

In the deep ocean all this is largely impracticable. Most of the ocean is dark and we leave our visual sense be-

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hind when we penetrate much below the surface. A terrestrial biologist with a pair of binoculars has a useful visual range of several kilometres. A submersible observer, even with the most powerful lights, can see no more than ten or twenty metres. If seawater was optically like air it would be so much easier to see what is there. In clear air a jumbo jet passenger can distinctly see details of the land and the sea surface from the usual cruising height of about 10000-12000m. This distance is similar to the maximum depth of the ocean, yet a passenger looking over the side of a ship sees almost nothing of what lies below. Sampling the populations with nets from a surface ship presents formidable problems, and is sometimes compared with trying to sample the terrestrial fauna at night from a balloon or helicopter, hundreds or even thousands of metres above. Carl Petersen, the great Danish fisheries biologist, wrote in a landmark paper on sampling the seafloor *"A dredging ship may be compared with an airship towing a dredge over Copenhagen, catching a policeman in one street and a perambulator in another; and from these it draws conclusions as to the whole population of the town"* [2].

Figure 1. *HMS Challenger* (shown here in the logo for the Challenger Society for Marine Science) was a wooden-hulled naval corvette, adapted for science by the removal of all but two of her guns. She was 61m in length, had a displacement of 2300 tons and carried about 220 officers and crew and six civilian scientists. A modern research ship of similar size might carry a total complement of about 40, half of whom would be scientists.

Early inklings

Two hundred years ago it was generally assumed that the deep sea was devoid of life. After all, how could anything survive in the cold, the dark and under such high pressure? Gradually this myth was dispelled by the efforts of the mid-19th century ocean pioneers, who fished with nets as deep as they could go and always found thriving populations of animals new to science. Between 1872 and 1876 the great round-the-world expedition of *H.M.S. Challenger* studied everything from protists to fishes [3,4] and its nets collected more than 4000 new species from depths of up to 4 km (Figure 1). Building on this and subsequent sampling, a new 10-year initiative, the Census of Marine Life, is aimed at achieving a more comprehensive survey of the life in the ocean [5].

Figure 2. An opening/closing Rectangular Midwater Trawl system (RMT1+8). The lower net has an 8m^2 mouth area and mesh of 4.5mm, the upper one a mouth area of 1m^2 and a mesh of 0.33mm. This combination allows a wider size range to be sampled than would be possible with a single net. The nets open and close simultaneously on receipt of an appropriate acoustic signal from the ship. (Photo: Southampton Oceanography Centre)

Nets have been the main tool for sampling the oceans ever since those early days but what they catch still remains largely a matter of luck, although the advent of new technology has helped to reduce the uncertainty. The catching method may still be similar but the precision with which the net is fished (and therefore the information associated with the sample) is now infinitely greater [6].

Today's methods

Nets that open and close on command were first devised in the late 19th century. Nowadays nets in midwater or on the seafloor are operated by mechanical triggers or acoustic, optical or electrical signals sent from the ship [6] (Figure 2). Meters monitor the flow through the net and the angle at which it is fishing; other sensors on the net can measure the depth, temperature, salinity, light scattering, chlorophyll, daylight, bioluminescence or any other parameter of interest that can be converted into an electronic signal. If acoustic devices are mounted on the seafloor as well as the net, its three-dimensional path relative to the bottom can be determined very precisely. Very detailed knowledge can be gained of exactly where the animals taken by the net were living and what their environment was like.

Unfortunately most of the animals will be dead, killed by the trauma of capture, decompression, and exposure to the relatively high temperature of the surface

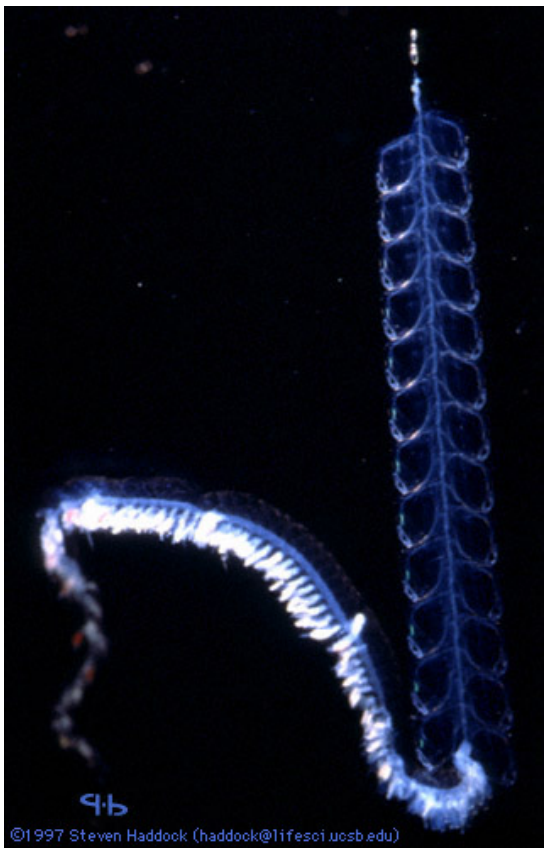

Figure 3. Gelatinous animals are very abundant in the upper ocean but are very fragile and difficult to sample properly. This siphonophore (*Bargmannia*), photographed in situ from a submersible, rapidly disintegrates when caught in a net. (Photo: Steve Haddock, MBARI)

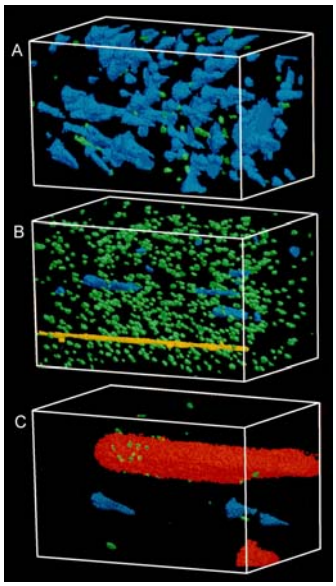

Figure 4. A 3-dimensional reconstruction of the flashes recorded as bioluminescent animals hit a mesh screen mounted on the Johnson-Sealink manned submersible.. In (A) at 16m depth, small clouds of luminescence (col-oured blue) indicate the numbers of small copepod crustaceans and short flashes (green) indicate single-celled dinoflagellates. In (B) at 61m, the dinoflagellates dominate and a long glow (yellow streak) is produced by a euphausiid shrimp (krill) stuck on the screen. In (C) at 249m, luminous organisms are much less abun-dant and the signals are dominated by the large lumi-nous clouds (coloured red) produced by a comb-jelly (*Euplokamis*). The dimensions of the white frames are 0.35 x 0.35 x 4.0m. (From ref. 7, courtesy Dr E.A.Widder, HBOI)

waters through which the net is recovered. As a result the ecology of the deep sea was for many years based largely on the anatomy of dead specimens. Today's deep-sea biologists have new tools that are gradually adding ecological insights to the anatomists' earlier interpretations of those corpses in the nets. We still have to be aware that net samples are always biased and the catch may not be a truly representative sample. Nets can only be towed slowly, so any active animal can probably avoid them (very active predators such as squid occasionally go deliberately into a net to feed on what has already been caught, and then may not escape before it closes behind them). Delicate animals, especially gelatinous ones (Figure 3), are easily damaged or destroyed by the abrasion of the meshes and nets are justifiably criticised for catching only the slow, the stupid, the greedy and the indestructible. The sizes of organisms in the oceans span at least eight orders of magnitude, from viruses to whales. Nets can handle five of those size orders with varying degrees of success (from 20μm phytoplankton cells to 2m tuna) but other methods are needed at the extremes [1]. We cannot filter a whale, nor can we harpoon a virus.

The abundance of the animals captured in a net is calculated by dividing the number of captured animals by the volume of water it has filtered. This just gives an average value of individuals per cubic metre and cannot tell us whether they were taken in a single group or were evenly dispersed along the path of the net. One ingenious method of establishing the detailed distribution of some bioluminescent organisms involves stretching a nylon mesh across a 1m diameter circular frame, mounting it on the front of a submersible and focusing an image-intensified video-camera on it. When the submersible is driven slowly through the dark waters bioluminescent organisms flash or glow as they are hit by the mesh and, with experience, their different signals can reveal the kind of organism involved [7]. Analysis of the record of flashes along the track of the vehicle gives a clear picture of the organisms' 3-dimensional distribution (Figure 4).

"Remote" methods

In seawater sound travels much further than light. Echo-sounders rely on the returning echoes to give information about the seafloor at all depths. Animals in midwater also reflect sound waves and are visible as deep scattering layers. By transmitting cones of sound into the water, using a range of different frequencies, it is possible not only to identify particular kinds of animals by analysing the returning echoes, but also to look at their three-dimensional distributions in the ocean (commercial fishermen have this technique down to a fine art – but only for fish-sized animals). Smaller animals reflect higher frequency sound waves (i.e. shorter wavelengths) but higher frequencies have less range,

so echo detection with a hull-mounted system is impracticable for smaller animals at depths of much below 200m or so (e.g. mesozooplankton 0.2-20mm in length). Instead the sound transmitter needs to be positioned closer to the depth of the target animals. This is usually achieved by placing the acoustic system in a streamlined vehicle and towing it at the required depth behind a research vessel, or by mounting it on a mooring at an appropriate depth. The stream of data it generates can then be used to analyse the distribution of animals in the water flowing past the acoustic transmitter.

Light *can* be used to sample the organisms. Particles in the water flowing through a light beam generate specific shapes and shadows and can be continuously counted. Such an Optical Particle Counter (OPC) can be combined with a video-camera to identify the organisms with considerable accuracy. New laser illumination systems are capable of generating holographic images of organisms that are only a few microns across as the plankton flows past an optical sensor. All these remote techniques allow the oceanographer to sit in the comfort of the ship's lab (or even the lab ashore) and see the images relayed from the depths and displayed on a screen. But there is more to be gained from this than just numbers of organisms: these methods now give us information on the spatial distribution of *individuals*, rather than just populations, and provide biological data on the same time and space scales as physico-chemical data. We can now see clearly how, at a very small scale, the physics determines many biological distributions. Conversely, on similar scales, the activities of the organisms affect the ocean's chemistry.

So far these remote methods have been used mostly from hull-mounted installations or in towed vehicles. Their potential has been hugely increased by the technological advances of recent years [3] and underwater sensors of all kinds can now be mounted on manned submersibles [8], remotely operated vehicles (ROVs) and Autonomous Underwater Vehicles (AUVs). Tethered or free-floating platforms in midwater, robot profilers that are programmed to ride up and down a vertical wire on a mooring, "landers" that settle on the bottom and can later be recalled, all these provide new opportunities for long-term automated measurements and observations of the biology of the deep ocean.

They are already exposing the naïvety of our earlier views. Acoustic and optical sensors on ROVs and manned submersibles (Figure 5) are showing that zooplankton are not nearly as uniformly distributed as we once assumed from our net samples. Often they are densely aggregated into layers just a few cm thick, usually associated with physical discontinuities [7, 20]. The ecological consequences of these aggregations are far-reaching for they provide quite different opportunities for interactions between the organisms.

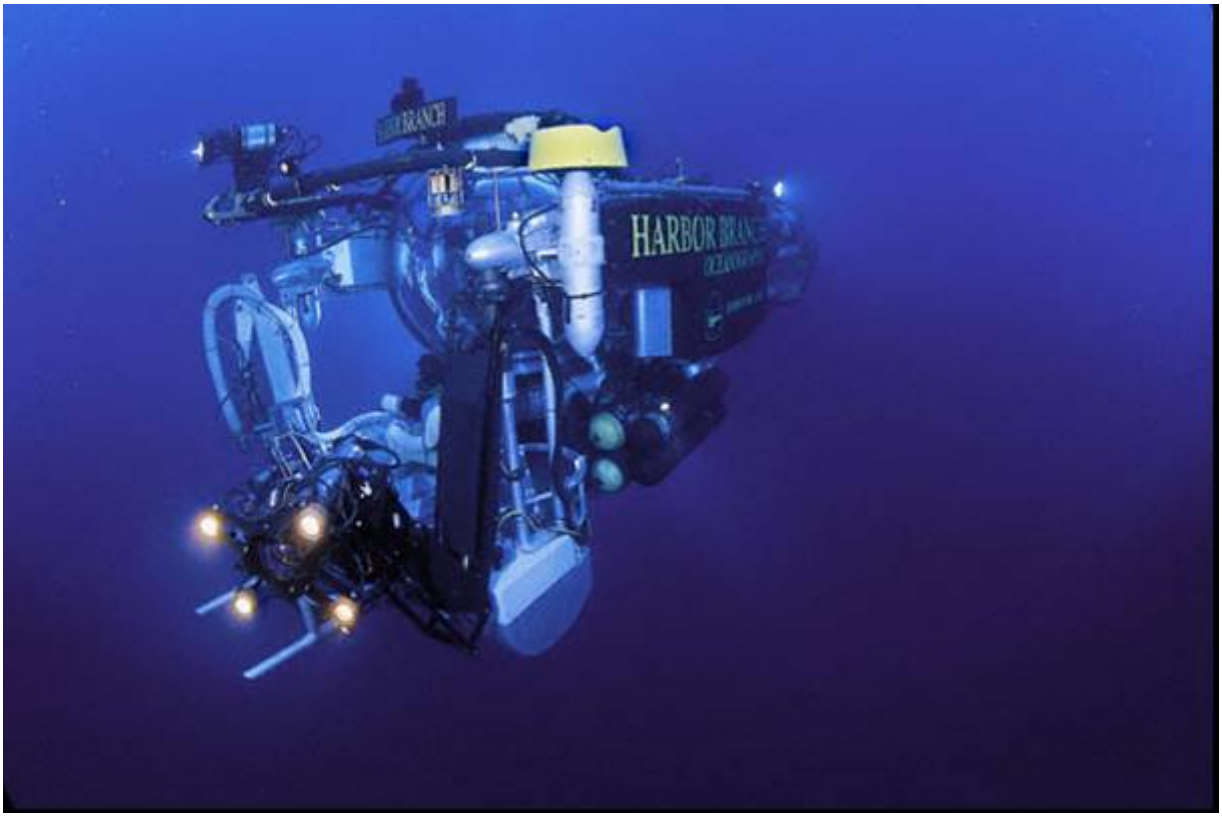

Figure 5. A side view of the Johnson-Sealink manned submersible. It has a depth limit of 1000m and the front acrylic sphere (approx 2m in diameter and 12.5cm thick), holds a pilot and observer and provides all-round visibility. A high resolution TV camera with four lights is mounted at lower right and the flexible hose is part of a sampling system that can suck specimens from the water (or the seafloor) and deliver them to storage containers for return to the surface (photo: HBOI)

We never expected research nets to be very good at catching active fishes but comparisons between acoustics and net data can be humbling. In the Gulf of Oman in 1997 there was a very strong night-time scattering layer at about 42m depth, which moved rapidly downwards whenever the (illuminated) research vessel stopped on station (Figure 6). Previous work had suggested that this was caused by a 3-4cm long lantern-

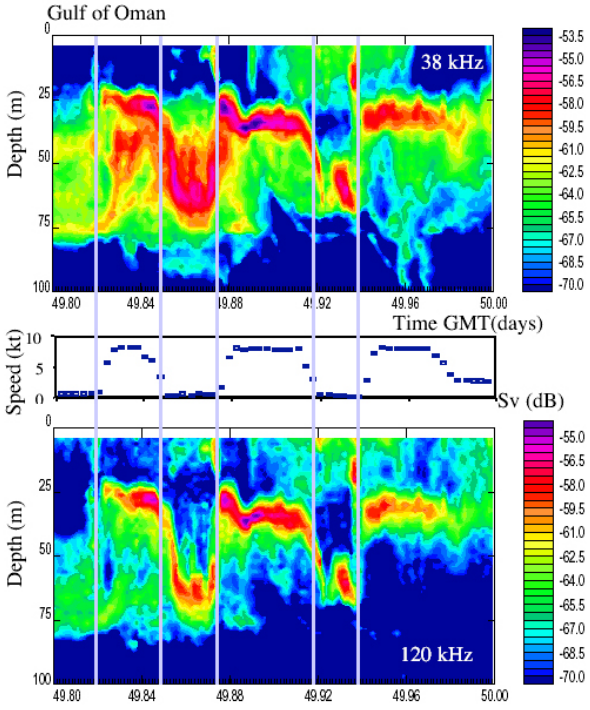

Figure 6. An avoidance reaction to ship's lights observed between approx 2300h and 0400h during the night of 18-19 February 1997 (time is measured in Julian days, from 49.80-50.00) in the Gulf of Oman. The upper panel shows the 38kHz echogram, the lower panel the 120kHz echogram and the middle panel the ship's speed. As the ship slows to stop on station (marked by the vertical lines) the midwater scattering layer descends from about 25m to 75m. The scattering layer is largely composed of the lanternfish *Benthosema pterotum* (below). (From ref. 8, courtesy of Prof. G.Griffiths, SOC)

Figure 7. The 3-4cm long lanternfish *Benthosema pterotum* is adept at avoiding midwater trawls such as the RMT1+ 8 shown in figure 2. Acoustic assessments of abundance (figure 6) are much more accurate than nets. (Photo: Peter Herring/ imagequest-marine.com)

fish, *Benthosema pterotum* (Figure 7). The acoustic data indicated that there were about 4 fish per m^3. A midwater trawl with a mouth area of $8m^2$ fishing at the same depth as the scattering layer at a speed of 1m s^{-1} caught just 1 specimen per $1000m^3$ of water filtered! For these animals the net appears to have an efficiency of less than 0.1% [9]. Nets are also poor samplers of gelatinous animals. This was emphasised by recent comparisons between an optical plankton counter, a zooplankton imaging system, and nets, operating simultaneously in the Gulf of Mexico. The nets underestimated fragile zooplankton by 300-1200% relative to the imaging data, and the OPC probably undercounted particles by 30% [10]. Other comparative studies of nets and OPCs show rather better congruence and it clearly depends on both the kinds of organism and on their population densities.

One way of knowing how organisms are really distributed in 3 dimensions would be to follow the advice of one past oceanographer, who suggested using an endothermic nuclear device to freeze a km^3 of ocean, which could then be slowly thawed and the co-ordinates of every organism in it recorded! Soil biologists do this sometimes – but on a much smaller scale. Even if this were possible it would only represent one instant of time and would say nothing about the dynamics of that community, information which our remote sampling devices are beginning to provide.

Going into the environment

Best of all for an enthusiastic biologist is to go and look, however limited the view. Almost 70 years ago William Beebe risked his life to be lowered to depths of 800m in a steel sphere, the bathysphere, suspended on a wire from a surface vessel. His reports of what he saw through the porthole were inspirational and triggered the manned exploration of the depths now undertaken in submersibles such as *Alvin* and the *Johnson-Sealink* [11]. Just as Everest was the goal for terrestrial moun-

Figure 8. Specimens of the giant squid *Architeuthis dux* are hardly ever taken in nets but, like this one in Aberdeen, are sometimes washed ashore after death. Accurate assessment of their abundance is consequently almost impossible. (Photo: Martin Collins)

taineers, so reaching the deepest point in the oceans was the goal for the 20th century ocean pioneers. Jacques Piccard and Don Walsh succeeded in 1960. Diving in the U.S. Navy's bathyscaphe *Trieste* they reached the bottom in the Marianas Trench at a depth of almost 11km [12]. No-one has ever done so again.

However well a biologist may think he knows the fauna from looking at hundreds of net catches, the experience of seeing the same animals in their own environment is breathtaking. When seen alive, the undistinguished lumps of jelly found in a net become gossamer-delicate animals of exquisite beauty yet fearsome predatory capability. Fishes illustrated horizontally in textbooks may in reality always hang vertically in the water. Unimaginable swimming styles and behavioural idiosyncrasies suddenly expose the fallibility of previous interpretations based on dead specimens.

Nevertheless the observations from manned submersibles, and the images obtained by unmanned ROVs and AUVs, have their own limitations, quite apart from their cost. Observers use their eyes and vehicles use their cameras: both require artificial illumination in the black depths of the ocean. The ranges of camera lights are limited to a few metres; beyond this the darkness swallows up the inhabitants. The eyes of many animals are attuned to the low intensities of their own bioluminescence and are easily damaged by our supernova-like floodlights. Some animals are even attracted to our lights like moths to a flame. The "behaviour" we can suddenly see may not be entirely normal, so attempts are now being made to observe the fauna with red lights, which the animals cannot see.

Submersibles and unmanned vehicles are inevitably noisy and intrusive, just as are nets and towed vehicles, so the active avoiders of our nets may be equally proficient at avoiding our underwater vehicles (and our lights). Indeed we should routinely expect active animals to avoid our samplers. Despite all our technological sophistication we could easily remain completely ignorant of a large, active, deep-sea animal that chose to avoid our puny samplers.

The giant squid is really known only because dead specimens float to the surface and then occasionally wash ashore (Figure 8), and because its beaks are found in the stomachs of sperm whales (much better samplers than our nets!). It is salutary to remember that in the last few years we have discovered or recognised new species of baleen and beaked whales (which routinely come to the surface) [13] as well as the huge "megamouth" shark (which does not)[14], while an unknown species of squid at least 7m long has recently been seen and recorded on video from our submersibles [15]. These are very large animals, so how many unknown smaller species must there be and, more importantly, what are their roles? It is salutary to remember that the single most abundant photosynthetic or-

ganism in the ocean (*Prochlorococcus*) was only discovered in the late 1980s! New methods of looking at marine microbes, using molecular tools, are disclosing a staggering variety of species and relationships that would be quite impossible to recognise with more traditional tools (and marine viruses are probably even 10 times more abundant……) [16,17]. Molecular methods are also refining the taxonomy of many groups of oceanic animals and, if automated, could be mounted on subsurface vehicles to monitor species' distributions.

For many years the deep-sea floor was regarded as a relatively uniform and undisturbed environment with low numbers of species ("biodiversity") when compared with terrestrial habitats. In his 1971 book "Life in the sea" Gunnar Thorson estimated that out of a million known species only 160,000 were marine, and 98% of these lived on the seafloor. In 1992 a detailed study of the macrofauna (animals retained on a 0.3mm sieve) in box-core samples taken at depths of 1500-2500m off New Jersey found 798 species in the 91000 specimens -- and 58% of them were new species [18]. Extrapolating from these data the authors put forward a conservative estimate of 10 million macrofaunal species worldwide! The estimate generated a lot of controversy and depends on the accuracy of the extrapolation process, but it is clear that the biodiversity of the seafloor can be very high. Midwater biodiversity is much lower, however, largely because the habitat is more uniform.

Our underwater vehicles allow us to focus our attention on individuals of the larger animals and on some of their activities in the water. With the new technology we can recover medium-sized individuals alive and undamaged for experimental studies in the ship or shore laboratory. This is done either by drawing the animal into a sealable container with a suction sampler or, even better, manoeuvring a cylindrical open container round the animal and then closing both ends. Capturing an animal intact in this way with a submersible or ROV requires great pilot skill but the more active swimmers still escape. Combined with the *in situ* observations it gives us an unrivalled opportunity to discover the extraordinary capabilities of these animals.

With a little help from the animals

Underwater vehicles, towed bodies, landers, platforms and nets all have great limitations in that none of their counters, cameras and captures occur in undisturbed (*i.e.* normal) conditions. Even this limitation can now be overcome by enrolling the animals in the sampling/observing process. Tags were originally placed on key (commercial) species of fishes in the hope that later recapture would provide data about their movements and migrations. Tags are now smaller and can incorporate all manner of sensors. They can be temporarily glued or otherwise attached to larger jellies, crustaceans, fishes, turtles, diving birds, seals, whales and

dolphins to give us a picture of their movements, their diving habits, their heart rates and many other parameters, all of which can be downloaded automatically to a satellite for transmission ashore. Recapture is no longer necessary. The larger the animal the greater its potential for instrumentation. The miniaturisation of digital still and video-cameras now allows them to be temporarily attached to whales and seals, sharks, turtles and penguins and they are providing the first animal's-eye views of feeding and social behaviour [19]. As miniaturisation continues so more and more potential carriers will become available and it is tempting to envisage future images from anglerfishes (Figure 9) and deep-sea squid going about their normal business (but this would, of course need tiny light sources as well).

Putting it all together

Combining all these different capabilities is now a realistic option and has led to the development of Ocean

Figure 9. One of the enduring mysteries of the deep sea is how anglerfish males find the right female and attach to them (as in this pair of *Melanocetus johnsoni*; the female is 100mm long). Fanciful though it is at present, we might one day be able to attach a minicamera to a female and record her encounter with a male. (Photo: Peter Herring/ imagequestmarine.com)

Observing Systems (OOS), integrated arrays of sensors deployed to unravel the dynamics of a particular region. Sensor platforms include satellites, midwater and surface moorings, bottom-mounted instruments, free-drifting neutrally buoyant packages, surface research ships, towed vehicles, ROVs and AUVs. All of them either download their data to an intermediate hub or are hardwired to the shore. Although OOS development has so far been designed mainly to unravel the dynamic oceanography of coastal regions, the incorporation of

biological sensors and imaging systems in the arrays will greatly improve our understanding of the ocean's inhabitants and their interactions, and provide new educational opportunities [20]. Once proven, the technology will be extended into ever deeper waters.

Knowledge of the oceanic ecosystem requires time, tools, imagination and persistence. We know something about what lives in the oceans, something about the capabilities of individual species and something (though often precious little) about their interactions with each other. It is a start. Our predecessors spent long periods at sea in cramped and uncomfortable conditions, usually without communication ashore. Today's biological oceanographers may (if they are lucky) have short cruises which offer hotel catering and e-mail. They can now acquire vastly more data in a given time and have access to direct information on the inhabitants of the deep ocean that their forbears would have died for (and sometimes did). Nevertheless wherever we look there are new species to be found, new observations to be interpreted and new data sets to be reconciled. Our hope is that this will help us to recognise the processes involved in the dynamics of this critical ecosystem. Only then can we hope to play a positive part in its continuity.

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