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Prof. Dr. Broder Merkel

Dipl.-Geoökol. Mandy Schipek

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Contents

Preface	7
Brümmer, F.; Fritz, G.B.; Jaklin, A.; Pfannkuchen, M. Sponge sampling: developing a good scientific practice.....	9
Vogler, C. & Wörheide, G. Scientific diving for biodiversity research.....	12
Kuklinski, P. Diving and ecological studies in a challenging environment	13
Michiels, N.K. Red fluorescence in reef fish: A new view on colour vision in marine environments.....	20
Wagler, M. & Brümmer, F. Competitive interactions of sponges and corals in the Gulf of Aqaba	26
Bauerfeind, J. Ammelshain – diving in a former stone pit	32
Novac, A.; Surugiu, V.; Teaca, A.; Begun, T. The distributional patterns of the zoobenthos from the artificial hard substratum of the Agigea dyke (Romanian Black Sea coast): preliminary results	39
Hall-Spencer, J.; Colin, M., Keith, H. Scientific divers quantify known outbreaks of cold-water coral disease	45
Schipek, M.; Barth, G., Eckardt, E., Pohl, T., Merkel, B. Environmental assessment of a site in Sv. Marina (Istria, Croatia): Examination of morphological and chemical parameters which indicate the occurrences of certain species	49
Italiano, F. Hydrothermal fluids vented at shallow depths at the Aeolian islands: relationships with volcanic and geothermal systems	55
Amend, J. A brief review of microbial geochemistry in the shallow-sea hydrothermal system of Vulcano Island (Italy)	61
Esposito, A.; Giordano, G.; Anzidei, M. Diving in Panarea volcanic (Aeolian Islands, Italy): methodology and results	68
Hall-Spencer, J. & Rodolfo-Metalpa, R. Using scientific diving to investigation the long-term effects of ocean acidification at CO ₂ vents	72
Pichler, Th. Marine shallow-water hydrothermal systems as natural laboratories	77

Price, R. & Pichler, Th.	
Measuring toxic elements and toxicity in marine shallow-water hydrothermal systems ..	82
Sieland, R.; Steinbrückner, D.; Hamel, M.; Merkel, B.; Schipek, M.	
Geochemical investigations and gas quantification of submarine fluid discharges in the hydrothermal system of Panarea (Aeolian Islands, Italy)	87
Becke, R.; Merkel, B., Pohl, Th.	
Mineralogical and geochemical characteristics of the shallow-water massive sulfide precipitates of Panarea, Aeolian Islands, Italy	94
Martin-Bueno, M.	
The Underwater Archaeology in the 3 rd millennium: legal and technical aspects	102
Staniforth, M.	
Research in Underwater Archaeology: some challenges and approaches for the future .	106
Heinicke, J.; Italiano, F.; Maugeri, R.; Merkel, B.; Pohl, T.; Schipek, M.; Braun, T.	
Evidence of tectonic control on active arc volcanism: the Panarea-Stromboli tectonic link inferred by submarine hydrothermal vents monitoring (Aeolian arc, Italy)	111
Bauer, K.; Bauer, D.; Fütterer, W.; Kleutges, J.	
Flow rate measurement at submarine volcanic gas emissions	112
Weber, M.; Faerber, P.; Meyer, V.; Lott, Ch.; Eickert, G.; Fabricius, K.E.; de Beer, D	
DOMS: The new Diver-Operated Microsensor System	118
Leidig, M. & Barth. G.	
Underwater Temperature Measurement – the underestimated basis for various scientific projects – an example for Panarea, Italy	119
Barth, G.	
Application of a global positioning system underwater	125
Lang, M.	
Scientific Diving Program Management and Scientific Diver Education	127
Sayer, M.	
Scientific Diving in the UK: training and legal requirements	133
Pohl, Th.; Schipek, M.; Merkel, B.	
12-years scientific diving at Technische Universität Bergakademie Freiberg	137
Merkel, B.; Schipek, M.; Pohl, Th.	
Work Instruction for Scientific Diving at TU Bergakademie Freiberg	142

Preface

Scuba diving is a research tool to study the underwater environment. Scientist using this tool within their research are called scientific divers. The major objective of this international workshop is to bring together scientific divers from different countries and research fields and discuss common problems, challenges and interests. Thus the workshop will be a platform to learn about scientific diving techniques and see new aspects concerning the underwater environment. Independent from the science discipline certain techniques such as surveying, sketching, transporting equipment, photo and video documentation are used by any scientific diver and are therefore of interest for all of us.

One important aspect is the training of students and researchers who have a certain scuba diving level but then needs additional skills to be a scientific diver. Training and underwater research has to follow strict rules in order to minimize the risk which is involved in scuba diving activities. The purpose of any scientific diving standard therefore is to take into account the potential hazards and inherent risk of scientific diving and to ensure that all underwater activities are conducted in a manner that will promote the protection of scientific divers and those in training from accidental injury or illness as much as possible.

Because scientific divers are not commercial respectively professional divers health insurance during the under water work might be a problem. Certain insurances cover the health risk during scuba diving only if it is a fun or sport dive. In combination with the insurance aspect it is a suspect matter if e.g. a department manager or university teacher may order a person to perform a scientific dive because a scuba dive is linked with a certain inherent risk.

The workshop **Research in shallow marine and fresh water systems** is promoted and supported by the German Research Foundation (Deutsche Forschungsgemeinschaft, DFG), the VDST (Verband Deutscher Sporttaucher e.V.), the LVS (Landestauchsportverband Sachsen e.V.) and TAZA Tauchclub Naunhof e.V.. I'm glad to welcome more than 50 scientific divers from 9 countries including Australia and United States. I would appreciate if this workshop will be the starting signal for the formation of an international network of scientific divers sharing experiences and exchanging ideas and visions in underwater research.

Freiberg/Saxony, 10.05.2009

Prof. Broder J. Merkel
Head of the Scientific Diving Center
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Sponge sampling: developing a good scientific practice

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Abstract. Marine sponges are becoming important objects of multiple research purposes. Identification, sampling and culturing, however, are facing a multitude of problems needing an acute scientific attention. Experience in the *in situ* and *ex situ* culturing of sponges, the sampling of sponges following safety standards during diving as well as scientific purposes and a multitude of research on morphology, physiology and biotechnological aspects lead to the compilation of the “Stuttgart protocol” to establish a good scientific practice to obtain sponges.

Sponge sampling

Sponges (Porifera) are primarily sessile filter feeders. Especially in the littoral zones of marine habitats they occur in high species numbers as well as abundance. Increasing interest over the past years in their diverse second metabolites for pharmaceutical (Sipkema, et al. 2005) and e.g. their siliceous skeleton for biotechnological purposes (Schröder, et al. 2007; Schröder, et al. 2008), the knowledge on sponges and appropriate sampling methods became an important task. Due to their life forms as well as often conspicuous morphology and color, they are easily accessible, especially for SCUBA divers. However, underwater determination of species present is sometimes impeded by the phenotypic variability (Klöppel and Brümmer 2006; Klöppel, et al. 2009; Klöppel, et al. 2008; Pronzato 2004), some movement besides their primary sessile life, for example via symbiotic hermit crabs (Jaklin, et al. 2007) as well as contraction (Hamer, et al. 2007) despite the lack of nervous and muscular tissue (Nickel 2004). In addition, we face the problem of sponges, which are very similar in morphological features, are being considered same species, probably reducing the actual sponge diversity dramatically. Therefore, molecular characters are considered as a valuable tool to create additional distinguishing marks for species lacking taxonomically important morphological features (Heim, et al. 2007b; Heim, et al. 2007c).

Furthermore sponges are very sensible to disturbance such as touch, tissue injuries, as well as changes in habitat conditions such as rapid pressure changes, temperature, or salinity. They react with a variety of physiological and morphological responses which again make it difficult to identify sponge species *in situ* or to describe specimens accurately after sampling. Very drastic is, for example, the chemical reactions observed in the yellow sponge *Aplysina aerophoba* which turns blue, almost black when physically disturbed or damaged (Thoms, et al. 2006). Handling the specimens requires a lot of care already under water. Contact to air has to be prevented as well as any squeezing or injuring. Changing habitats, from *in situ* to an *ex situ* environment for example, some sponges undergo irreversible changes in shape, size and color (Klöppel and Brümmer 2006). Some functional processes are altered in some sponge species due to cultivation conditions. Such processes include for example the pumping of the sponge to obtain oxygen and nutrient supply through its filtering system. Purely *ex situ* experiments showed, that under some circumstances these pumping rates can be reduced or can cease complete, leading eventually to the death of the specimen (Hoffmann, et al. 2008). On the other hand *in situ* and very well defined *ex situ* experiments showed that sponges exhibit constant pumping, altered only by extreme disturbances such as high temperatures (Pfannkuchen, et al. 2009). This demonstrates the pivotal importance of careful and skilled underwater work in sampling to allow following experiment to retrieve information about living and healthy specimens (Brümmer, et al. 2008) and prevent the recording of merely sampling and maltreatment artifacts. Additional difficulties have to be addressed after successfully collecting specimens. Transportation and conservation methods play an important factor of subsequent successful identification and description of sponges. Life transports have to ensure sufficient oxygen supply, no temperature fluctuations and a minimum of physical disturbances. The conservation of specimens requires some additional thoughts. Conventional methods, such as ethanol or formaldehyde, reduce either color or the possibility of DNA analyses. Dry ice is mostly not available in all

places. Thus, new methods have been tested such as for example transporting samples in a saturated saline solution (Heim, *et al.* 2007a).

Benthic organisms such as crustaceans, annelids, bryozoans can be sampled following established sampling protocols (Bianchi, *et al.* 1994; Castelli, *et al.* 1994). Such detailed standards do not exist for the sampling of sponge species. The following “Stuttgart protocol” is made for the establishment of a good scientific practice to sample sponges in compliance with safety guidelines for scientific diving:

1. Sample locations have to be reported to enable the correlation of potential morphological and physiological difference to the place of origin. This requires some sort of underwater positioning system either using a digital compass in combination with depth measurement or the use of the accompanying above water GPS system.
2. Specimens have to be photographed *in situ* and *ex situ* to conserve the *in situ* aspect and allow the detection of possible changes. Color indicators, such as RAL color standards, to count for different color perceptions due to the light attenuation in the water are necessary.
3. Size and changes in size have to be measured and noted.
4. Specimens have to be handled with extreme care – under as well as above water to avoid any physical damages.
5. If necessary, culturing specimens in-between sampling and final destination has to be best done in salt water tanks with a flow-through system.
6. Any *ex situ* cultivation should attempt near *in situ* conditions in relation to at least temperature, salinity, light availability, nutrients and currents.
7. Transport has to ensure oxygen supply, low temperature fluctuations and reduced physical stress or even damage.
8. Fixation, which should take place immediately following sampling, has to be carefully chosen depending on sampling purposes. Four methods should be considered: a) ethanol for DNA analyses and further processing, b) formaldehyde for histological examinations, c) salting sponges for morphology and color analyses, and d) flash freezing for all purposes.

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Scientific diving for biodiversity research

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Abstract. Marine ecosystems are under ever-increasing pressure from anthropogenic impacts. Conserving these ecosystems requires an extensive knowledge and understanding of their biodiversity and the threats they are subjected to. Yet, contrary to terrestrial ecosystems, assessing the biodiversity in the marine realm remains a challenge.

Unfortunately, morphological methods are not always sufficient to achieve rapid species identification or determine species boundaries, especially in marine systems where cryptic speciation is a common phenomenon. Molecular methods are therefore key components for rapid biodiversity assessment and discovery.

Our research focuses on marine invertebrates, from both coral reef and Mediterranean ecosystems. Molecular analyses require collecting live tissue from the study organisms, mostly by SCUBA diving or snorkeling. Organisms that are hard to access have often brought us to challenging diving environments. Through three case studies we here present different aspects of the diving required for our research:

Diving with minimal logistic support: species boundaries in the crown-of-thorns starfish species complex (*Acanthaster 'planci'*) (Vogler et al. 2008).

Cave diving in remote locations: biomineralisation of the cave-dwelling coralline sponge *Astrosclera willeyana* (Jackson 2007, Wörheide 1998).

Diving in an industrial port: detecting marine bioinvaders by DNA barcoding.

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Diving and ecological studies in a challenging environment

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Abstract. The Arctic is renowned for its extreme environment. Not only is the Arctic extremely cold, but also there is a high degree of disturbance caused for instance by ice-bergs and strong winds. Due to these extreme conditions the acquisition of scientific data, especially in the shallow subtidal zone is very challenging. In this short account I present methods for obtaining biological samples and environmental data in the European Arctic with the use of SCUBA diving. Examples of ecological studies conducted by Institute of Oceanology are provided.

Introduction

The marine environment of the Arctic Ocean is characterized by a relatively narrow range in annual temperatures, often near the freezing point of seawater, which is slightly below zero degrees (Anderson et al. 1994). Vast areas of the Arctic Ocean are covered by ice. However, there is considerable seasonal and regional variation in ice cover, ranging from $7.0 \times 10^6 \text{ km}^2$ in September to $15.4 \times 10^6 \text{ km}^2$ in March (Parkinson et al. 1999). Other physical factors that are more intense in the Arctic than at lower latitudes, especially in shallow-water, include the presence of ice, iceberg scouring and strong currents generated by wind (e.g. Dowdeswell and Forsberg 1992) (Fig.1a-b). Because of continuous darkness in the winter and continuous daylight in the summer, there is high seasonality in biological activity with the greatest abundance of many organisms in the summer (Sakshaug 2003). These harsh environmental conditions also have implications for scientists based in polar-regions. In particular, the acquisition of scientific data that requires SCUBA diving is very challenging. Diving in extreme conditions like those found in the Arctic is a very different activity from diving in warmer waters. These complexities include the use of equipment that has to function in temperatures that may be below zero. These challenges also extend to the divers themselves and in the response of human physiology to such an environment (Angelini 2007). Persons suffering from hypothermia are more likely to make mistakes during activities such as diving, which makes the divers more prone to accidents (Mueller 2007 and references therein).

Although the Arctic is a very challenging environment, there are several reasons that make this challenge worthwhile. First of all climate change in the Arctic is progressing very fast. Of all the regions of the globe, the Arctic is one of those that is warming the fastest (Schiermeier 2007). It is therefore a great natural laboratory for studies that examine the effects of climate change. It is believed that shallow subtidal zone is the most sensitive therefore this is an area where climatic changes might be noticed first. Additionally the marine life of the Arctic Ocean is highly adapted in its history, ecology and physiology to the extreme and highly seasonal conditions of the environment (e.g., Kuklinski and Porter 2004; Kuklinski and Taylor 2006). Studying these aspects of marine biota may enable us to understand better the evolution or life histories of these organisms.

This account presents briefly some of the diving strategies utilized by researchers at the Institute of Oceanology when working in the European Arctic. Case studies of both physical and biological data acquisition with use of SCUBA diving are presented.

Diving, equipment and strategy considerations

The Marine Ecology Department of the Institute of Oceanology, Polish Academy of Sciences (IO PAS) has carried out ecological research in the Arctic with use of SCUBA diving every year since 1997. Its current knowledge was built on experience shared by Norwegian, German and British colleagues. In general, we follow rules and techniques described by Lang and Sayer (2007) in Proceedings of the International Polar

Diving Workshop, with a number of small modifications. Most of these modifications are due to practical and economical reasons.

Rubber boats are used as the primary diving platform, as these have proved to be the safest, most reliable and cost effective means of transportation we have used to date in the Arctic (Fig.1c). Research vessels have also been used as diving platforms to conduct ecological research by the IO PAS team. However, these vessels are often not able to approach diving sites in shallow water as close as rubber boats.

The majority of ecological research requiring SCUBA diving by IO PAS takes place during the summer season. Such dives in the Arctic do not differ much to the winter dives in the temperate areas. Most of the sites are ice-free however icebergs and growlers are common features at diving localities. Strong winds are often the main obstacle for diving activities during the polar summer. Wind-generated waves and currents also often lead to diving activities being abandoned for days. Waves are especially problematic, as there is a risk of the boat capsizing or, less dramatically, a bumpy ride, which can lead to equipment damage. This is especially problematic if a rubber boat is used as the sole means of transport from a land base to diving locations over long distances. Therefore to increase efficiency and reduce any risk concerning transport, two dives are usually performed by each diver during one trip. After the first dive warm drinks and high-energy chocolate bars are consumed by divers. This was proved to increase both the spirit and vitality of the divers. In polar-regions not only is the water temperature low, we also face low air temperature. To minimize any risk of hypothermia dry suits are worn at all times during fieldwork (Fig.1d).



Fig.1. Examples of challenges in the Arctic research and scientific diving: **a.** Hornsund fjord (West Spitsbergen) in the summer, strong wind is probably the most dangerous enemy for divers in the Arctic; **b.** Isfjorden (West Spitsbergen) in the summer, the presence of ice may prevent access to the dive/study locality; **c.** the safest and most cost effective dive platform; a rubber boat (photo: J.Dobroszek); **d.** safety first, at all times both on and under water a dry suite is worn to prevent hypothermia (photo: J.Dobroszek).

To date the regulators of three different companies have been used by our team; Poseidon (Cyclon 5000, Jet-stream), Sherwood (Maximus) and Apex (various types). All proved to be reliable in low-temperature waters and we have not experienced any problems with regulators freezing. The only point that should be raised here is the fragility of the second stage of Sherwood Maximus regulators. These often could not withstand the harsh conditions of transportation in the rubber boats to the diving locality and damage has been observed.

Dives never exceed 30 m depth and usually last until all the work planned for that given dive is done (usually up to 60 minutes). Extra pony tanks are used for deeper dives (below 15 m depth), otherwise we use a single tank with one regulator on it. This makes the whole set lighter, more comfortable and therefore safer. No full-face masks are used.

Acquisition of environmental data

To fully understand what drives marine community developmental patterns, population structure or even organismal behavior acquisition of physical environmental parameters including water temperature, salinity, light intensity is required. Today's modern ecological studies demand acquisition of such data over

many seasons or even years. This is especially true in polar environments where we observe huge seasonality in sunlight supply, variability in the presence of ice, fresh water discharge from melting glaciers or biological production. Such data acquisition would require systematic dives over the whole year and the subsequent need to overcome the great logistic obstacles caused by polar night and all the environmental factors associated with it. However with the use of modern technology we can overcome such inconveniences. Currently there are number of cost effective environmental data loggers available on the market which enable long-term (one year or longer) data acquisition. IO PAS currently have deployed loggers in the Arctic recording water temperature, light intensity, pressure of water column and substrate movement frequency. Although such technology can greatly minimize the work-load, proper deployment of the loggers in situ still requires the involvement of divers.



Fig.2. Study investigating influence of substrate dynamics on biota colonization. Machined rocks of different shapes, each shape representing different dynamics of substrate. The metal cube frame has a mounted movement logger inside.

Moreover many environmental factors have localized influence therefore loggers have to be deployed as close as possible to investigated biota or community in question. Diving is the only technique that enables proper and secure deployment of the loggers and their retrieval at the completion of data gathering. In our case loggers are attached to a large metal frame fixed to the substrate which in turn supports plastic panels, which we use to investigate the processes of biological colonization and succession (Fig.4).

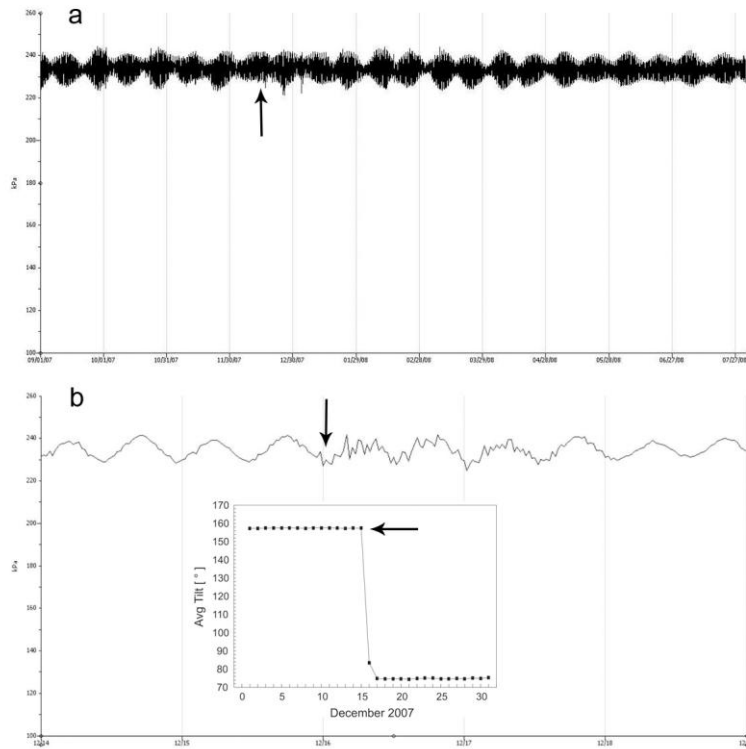


Fig.3. Examples of data provided by loggers; **a.** water level data showing the pattern of tides over one year; **b.** water level data at higher resolution showing tide pattern and waves action indicating stormy weather, inserted smaller figure show movement data (cube with logger, see Fig 2), in this case movement was generated by storm indicated by water level logger (arrow mark the moment when the cube was moved) .

The other platform for loggers installation is a much smaller, moveable frame (Fig.2). This second method of installing loggers is used in a study where we investigate the influence of substrate dynamics on benthic organism settlement (Fig.2). By having these loggers recording the position of the small, moveable frame every 30 minutes we are capable to detect disturbance intensity over the year as well as in different environments (for example at shallow and deep locations). Together with data from water level (pressure) logger we are able to identify which factors are most influential on polar rocks movement and therefore on the disturbance of biota colonizing them. Fig. 3 illustrates that the main forces that turn or disturb rocks in the Arctic are storms rather than any type of tidal forces. Acquisition of such unique data involves both deployment of loggers and their retrieval, both of which would be impossible to conduct without the involvement of SCUBA divers.

Acquisition of biological samples

Many research vessels, especially those that are capable of operating in high seas, have a draft exceeding 5 m depth. This means that the subtidal zone close to the shore is often out of the range of sampling equipment deployed from the ships. Yet this is often one of the most productive areas in the Arctic and is the focus of most of our studies.



Fig.4. Hard bottom Arctic cryptic biota development project. Metal frame on which plastic panels are mounted mimicking hard substrate. Number of environmental data loggers attached to the construction.

Understanding the ecological processes of the whole polar system requires sampling at all depths. Many scientific scopes of benthic ecology especially those conducted on the rocky bottom would be impossible without the use of SCUBA diving. Any sort of grabs cannot collect the biological life of the rock and do not close properly if a large rock blocks the wings of the grab. With dredges, sampling on the rocky bottom can be conducted, but it is not quantitative or it is semi-quantitative and often it is very selective. Although these methods can all be useful in shallow coastal waters there are number of studies where a human presence at the bottom is necessary, and at such times SCUBA diving is the only way of making progress. There are number of studies conducted by IO PAS where SCUBA diving has proved to be essential for achieving the goals of a given investigation. For example the investigation of fauna associated with kelp. Arctic kelp, with its complex three-dimensional structure, provides shelter for species rich assemblages of both sessile and mobile organisms. Quantitative sampling of over 400 algae with the use of specially designed nets were done on the western coast of Spitsbergen in 2003 (Wlodarska et al. 2009). Divers enclose the whole plant with the net and detach the holdfast from the bottom. This way sampling of both sessile and mobile associated biota is possible. More than 200 species were determined during that study. Other sampling techniques might result in collections of algae with only sessile associated fauna (small anchor) or studies that might sample mobile fauna but not provide quantitative results (dredges). Another study where SCUBA diving was used to provide quantitative samples was the collection of hermit crabs from rocky bottoms (Barnes et al. 2007, Kuklinski et al. 2008). In this case the only method that could be used was hand-picking of the animals by the diver from a known surface area of the bottom. Another example of an investigation where SCUBA diving was essential is the study of Arctic cryptic assemblages with the use of especially machined homogenous settlement panels (Barnes and Kuklinski 2005). This study is aiming at understanding how cryptic rocky assemblages are developing and interacting in space and time. Homogenous settlement panels provide good substrate for settlement, which enables comparisons to be made between different locations and excludes the potential influence of substrate properties on colonizing biota (Figs.4, 5). One set of panels is changed annually and remaining ones are photographed underwater. Each set of panels retrieved after a year provide us with information on the arrival of new biota. The photographs of the panels generate unique knowledge about how assemblages and particular biota develop over the years (Fig.5). No other sampling technique can provide such robust quantitative data as this method, which involves SCUBA diving.

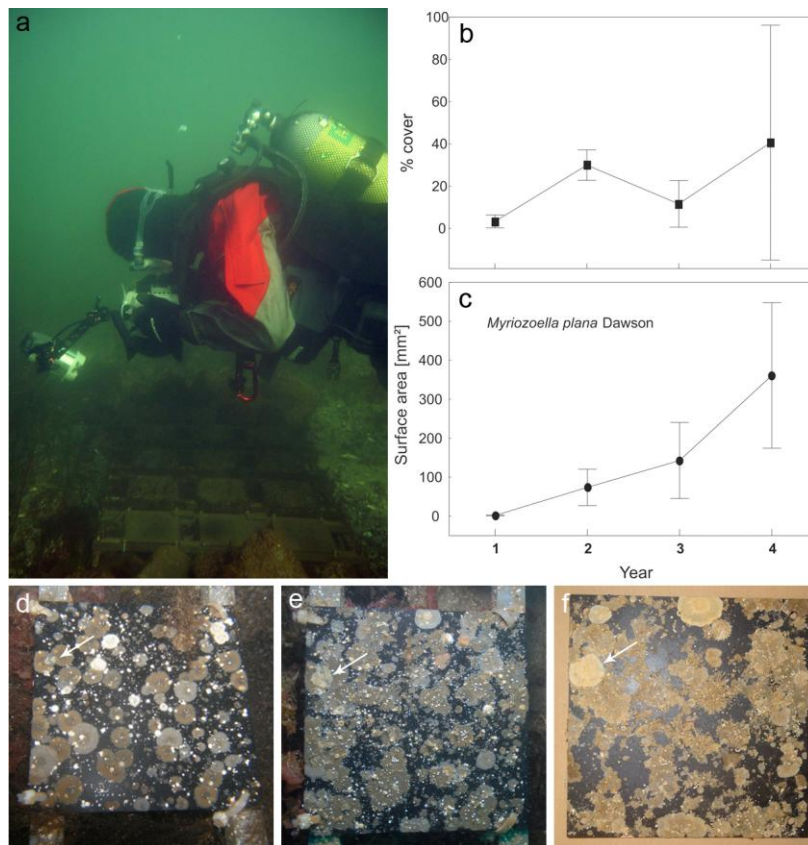


Fig.5. Hard bottom Arctic cryptic biota development project; **a.** diver is taking image of panels enabling biota assemblage analysis (photo: A.Sokolowski); **b.** example of data generated from images of the panels taken over four years period in this case percentage cover of the panels (mean \pm SE); **c.** growth rate of bryozoan *Myrriozoella plana* Dawson generated from the images of panels taken over four years period (mean \pm SE), (growing colony of *M. plana* on panels indicated by white arrow, see Figs.5d-f); **d.** two years old panel (image taken in situ); **e.** three years old panel (image taken in situ); **f.** four years old panel (image taken after retrieval).

Conclusions

Conducting research in the Arctic with use of SCUBA diving is very challenging especially due to the harsh environment. However in the light of rapid current environmental changes there is more need of investigation in polar regions than ever before. Understanding the influence of global climate change on biota from polar areas may provide the first insights in how to protect this environment and predict how climate changes will influence biota elsewhere where signs of change are not as rapid as in the Arctic. All these changes seem to occur first in the most sensitive shallow coastal zones, which is influenced both by ongoing changes in marine and terrestrial ecosystem. SCUBA diving is often a perfect and sometimes the only tool for conducting research studies in this zone. The combined experience of many nations diving and investigating polar regions and the use of common sense make it a safe tool and enable acquisition of unique scientific information which may lead to a better understanding of this vanishing beautiful world.

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Red fluorescence in reef fish: A new view on colour vision in marine environments

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Abstract. This contribution questions established views on marine fish color vision and claims that long-wavelength visual light (“red” light) plays an adaptive role for coral reef fish, also at depths where red light from the sun cannot penetrate. It follows from our discovery of red fluorescence in reef fishes, which transforms incoming blue-green light into red light independent of depth. Initial observations indicate that these fish can see red light and use it for signaling as well as camouflage. These findings warrant in-depth investigation of (1) the adaptive function and evolution of red fluorescence in marine fish and (2) histological, microbial and molecular analyses of the sources of these pigments.

Introduction

Red light is considered irrelevant to marine fish because (i) long solar wavelengths are absorbed within 10-15 m of the water column (Loew and Zhang 2006a; Marshall et al. 2003), and (ii), most fishes possess only two color sensitive retinal cones, sensitive for blue-green light, but not for red (Losey et al. 2003; Marshall et al. 2006; Siebeck et al. 2006). The generality of this view, however, needs to be questioned for various reasons:

1. Coral reefs harbor many fluorescing sedentary organisms many of which emit red light (cyanobacteria, corals, algae, sponges, pers. obs.) (Field et al. 2006; Fradkov et al. 2000; Limbaugh and North 1956; Mazel and Fuchs 2003). Hence, local light transformation results in more red in micro-habitats at depth than predicted from downwelling sunlight alone.

2. Studies in fish visual ecology routinely use average light availability at depth to correct spectral sensitivity measurements for natural light conditions (Job and Shand 2001; Lythgoe et al. 1994; Marshall et al. 2003). This procedure, however, is blind for local patches of red fluorescence, leading to an underestimate of the availability of red in the environment and, consequently, poor red light sensitivity in fishes.

3. Spectral sensitivity has been determined in a non-random subset of fishes belonging to common families that live in the open water (e.g. Pomacentridae, Labridae) (Marshall et al. 2006). Most small gobiids or blenniids have largely been ignored, as have many other benthic fish families.

4. Fish spectral sensitivities are inferred from ocular transmission and absorbance spectra from the retinal cones. This ignores neuronal post-processing. Behavioral tests represent a more integrative, comprehensive assessment of light perception. They have already allowed to demonstrate the ability of reef fish to see UV in training experiments (Siebeck 2004), which could not be derived from eye measurements alone. Such experiments are as yet lacking for red.

5. Some deep sea fish produce and see red light (Bowmaker et al. 1988; Douglas et al. 2000; Douglas et al. 1998), illustrating that the presumed lack of sensitivity for red in marine fish is not absolute. Quite some shallow-water (0-5 m) reef fishes show red patterns that can be interpreted as signals, suggesting that marine fish may actually see and use red more often than is generally assumed. One suggestive example is the toxic devil scorpion fish *Scorpaenopsis diabolus*, which lives in shallow water (1-10 m) and flashes the red inside of its pectoral and dorsal fins when disturbed (Allen et al. 2003), pers. obs.). This is without doubt a warning display, but for whom, if no-one can see red?

6. Rapid absorbance of red in water offers advantages to those that can see it. For instance, the relative amount of red light from the sun is a reliable depth gage in the upper 20-30 m of the water column and is used by plankton (Figueroa et al. 1998). Although many fish use their swim bladder to measure depth and control buoyancy (Evans and Damant 1928), one could speculate (!) that those without a swim bladder may rely on the red gradient in the water to assess current depth. It is an intriguing coincidence that benthic fish often lack a swim bladder (Amy R. McCune 2004) and often show red fluorescence (Michiels et al. 2008). Moreover, because red is quickly absorbed with distance, it represents the ideal color for private (local) communication in the neighborhood.

These arguments summarize why red light/color may be more relevant to reef fish than what established wisdom tells us. My views are instigated by our discovery of red fluorescent signals in reef fish. We recently found that quite some reef dwelling fish (5 families, 16 genera) show red fluorescent structures in the field (Michiels et al. 2008). Most of these species are small, benthic and cryptic, with distinct red fluorescent markings suggestive of visual communication. We showed that at least one goby, *Eviota pellucida*, is also able to see its own fluorescence (others await testing).

Implications

1. Red fluorescence is active color/contrast enhancement. Hence, it allows for being red, even at depth, where incoming red light is missing. It strongly indicates that red has a visual function – within or between species. This pattern is entirely different from reflective red coloration, which merely reflects incoming long wavelengths. Until now, all reds in fish were assumed to be reflective and, consequently, considered invisible and functionless at depth. Artificial, broad-spectral light (torch, camera flash) brings these “inexistent” reds to life, but at the same time makes distinguishing them from red fluorescence impossible.

2. Studies of fish color vision must be reconsidered. If fish send out red light, it is more than likely that there is a receiver who can see it. If true, many of the previous spectral sensitivity measurements indeed underestimate sensitivity for red, as suspected. Rather than using average spectra to normalize spectral sensitivity, maximal intensities at each wavelength on the reef should be used to determine the ability to see local spots of red.

3. Fluorescence is often explained as a mechanism to re-emit damaging sunlight in sensitive organisms such as corals (Salih et al. 2000). However, if (red) fluorescence is visually perceived, as our study indicates, it may have functions beyond “sun blocking”, such as attracting prey, camouflage or enhancing communication – not just in fish. Most red fluorescent organisms on reefs are filter feeders that grow in shaded places (pers. obs.).

4. Red fluorescence on reefs is underestimated. Fluorescence on reefs is typically revealed using UV light. However, given the spectral distance between UV (< 380 nm) and red (> 600 nm), UV is best for green and yellow, but not for red emission. We use blue or green for excitation. In the field, this is obtained by diving below 15 m, turning the sun into a blue-green light source. By using red masks, reefs transform into red fluorescent scenes under natural illumination and at a spatial scale inaccessible to other methods. This is how I discovered the first red fluorescent fish, the goby *Bryaninops natans* (Fig. 1).



Fig. 1. The goby *Bryninops natans* (3 cm) has red fluorescent eyes. Left: Natural illumination at 22 m depth (white balance corrected for depth). Right: Same, through red filter, hiding in *Acropora* coral head (NK Michiels 2008).

Open questions

Why do some fish fluoresce and others do not (or only weakly)? Michiels et al. (2008) list arguments for a function in intra-specific communication. They are based on anatomical structures as well as behavioral and ecological similarities across red fluorescent species. Other functions could be camouflage or prey attraction.

How widespread is red fluorescence among marine fish? Since submission of the paper, we discovered seven other fish families with red fluorescence: Scorpaenidae, Soleidae, Gobiesocidae, Callionymidae, Trachinidae, Synanceiidae and Ballistidae. Most fit the picture of cryptic, substrate-dwelling species. Many more species are expected to fluoresce. We have also found yellow fluorescent fish in other families.

Where does red fluorescence come from? We suspect that fish sequester the pigments from the environment and incorporate them into guanine-crystals (Levy-Lior et al. 2008) or bony tissue (labrids only). We also isolated bacterial samples from the gobies *Ctenogobiops tangaroai* and *Eviota pellucida*, yielding two red fluorescent strains. 16s rRNA sequences allowed provisional identification as *Enterovibrio coralii* and *Pseudoalteromonas* spec.

Why is (red) fluorescence in fish of broader interest? For the scientific community, isolating and characterizing fluorescent pigments is not only important to understand the function and mechanisms of fluorescence. It also opens avenues towards the establishment of novel fluorescent markers that are non-toxic to vertebrates. This year's Nobel Prize in Chemistry for the characterization of Green Fluorescent Protein (GFP) illustrates this. For the general public, the attractiveness lies in the discovery of a new visual communication mechanism that has been overlooked, despite the high numbers of people diving on reefs. The most fascinating aspect is that it is visible to all using something as simple as a red mask.

Future goals

What is the adaptive function of red fluorescence in fish?

Behavioral experiments will confirm whether fish can see their own red fluorescence or that of others and use it in territoriality, courtship, camouflage or for other functions. We shall also address the possibility that they use it as a depth gage, which can help to explain why fish without a swim bladder seem to use red fluorescence more often than others (in addition to other, ecology-based explanations). We shall test non-fluorescent species as this will help to explain why some predatory fish (e.g. stonefish) seem to be using red fluorescence for camouflage (pers. obs.) which is redundant if their prey cannot see red.

In the laboratory, experiments will involve offering individual fish visual access to other fish under conditions in which red fluorescence is visible or not, by using depth-simulating illumination. In some experiments, visual cues will be provided through a video system in ways similar to playback methods in fish ethology (Prof. Dr. TCM Bakker's group in Bonn (Rick and Bakker 2008a; Rick and Bakker 2008b; Rick et

al. 2006)), allowing control of the red component of the signal. Experiments will focus on species in which visual signaling is well-documented and red fluorescence is strong, such as *Corythoichthys* pipefish (Fig. 2). Other promising species are *Enneapterygius triplefins*. Training will be specific for age class, sex and reproductive stage where possible.

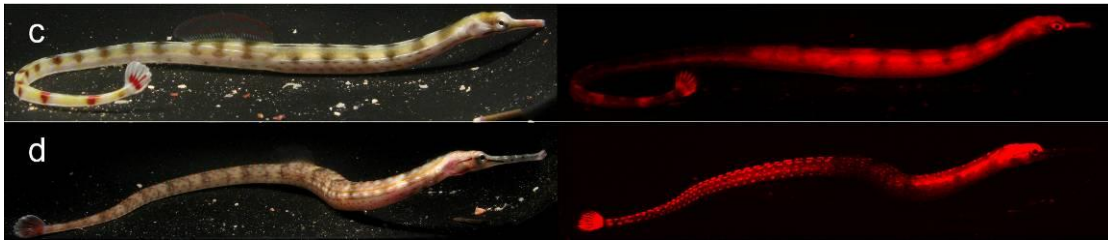


Fig. 2. Red fluorescence in *Corythoichthys* pipefish from the Red Sea under broad-spectral light (left) and blue light with red filter (right) in the laboratory. c = *C. flavofasciatus*, d = *C. schultzi* (from Michiels et al. 2008).

In the laboratory and on reefs we shall train fish to distinguish between hues of red and many other colors under a variety of illumination conditions using food rewards. A small pilot project (Red Sea, Oct 2008) revealed that pomacentrid and labrid fishes (1) can be trained quickly to pick red using food rewards and (2) preferred red in the test phase (without food or food smell). From this (preliminary) experiment we learned that effective data collection is possible under natural conditions – a very valuable option.

Who fluoresces, who does not? I want to significantly expand the list of red fluorescent fishes. To this end, we shall make use of live and preserved collections (fluorescent pigments resist storage in EtOH), the aquarium trade and field trips to selected sites in the Red Sea, Indopacific and Mediterranean. Mapping fluorescence and its associated anatomical, behavioral, life history and ecological context on the fish phylogeny will allow for a comparative analysis which is expected to yield broad insight into the origin and function of red fluorescence in fish.

What are the underlying mechanisms of red fluorescence in fish?

What pigments are involved? Where do they come from? How are they embedded in the fish? How do fish control their expression? This part will be carried out (1) in species already used for behavioral tests, (2) in representative species from each red fluorescent family, and (3) in species that can be cultivated (sterile culture experiment, see below). A representative set of related, non-fluorescent species will be studied in the same way and serve as a control.

Video, microscopy and microspectroscopy will be used to describe the locations and control of fluorescence. Fluorescent pigments are associated with guanine crystals in Gobiidae, Trypterygiidae, Blenniidae and Syngnathidae. In one wrasse (Labridae) it is linked to bony tissue. An analysis of anaesthetized fish and skin biopsies will show whether active control over fluorescence is possible, as suggested by these unpublished observations:

- The triplefin *Enneapterygius destai* expands its melanophores and turns black when anaesthetized, hiding its fluorescence. This suggests melanophore-based control.
- In pipefish, we discovered that fluorescent pigments are located in cells similar to melanophores. We call them “fluorophores (Fig. 3) and expect that they can change shape in a way similar to melanophores.
- The goby *Ctenogobiops maculosus* has bright fluorescing eye rings. However, in the field, individuals sometimes show black pigmentation on the eye rings, covering the fluorescence. Whether they can switch this “mask” on or off remains to be shown.

We shall use standard methods known to relax or contract melanophores and check for a similar effect in “fluorophores”. Recordings of live animals will confirm individual control.

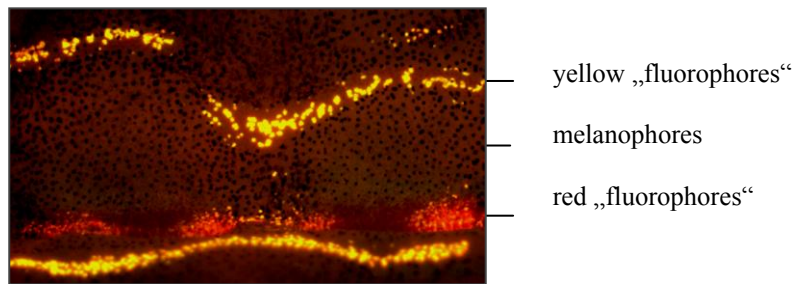


Fig. 3. Fluorescence microscopy of the skin of the pipefish *Corythoichthys haematopterus* shows black melanophores and yellow and red “fluorophores” (blue excitation, orange filter) (M. Wucherer).

As indicated above, the fluorescent pigments found in reef fish may originate from microorganisms, analogous to bacteria-induced bioluminescence in deep sea fishes (Kodric-Brown 1998). We shall systematically isolate bacterial strains from fish and their environment, select for (red) fluorescence and generate axenic cultures for identification. They will be grown in marine broth for marine bacteria, which worked successfully in the pilot experiment. We shall then search for the possible location of such bacteria in or on the fish (in situ hybridisation, electron microscopy, 3D reconstruction of fluorescent structures and body parts). In addition to collaborating with colleagues on campus, we shall also share strains with foreign laboratories for species confirmation. Strains of red fluorescent bacteria will be used to isolate and characterize their fluorescent pigments. For this aspect, I can rely on a broad methodological expertise on our campus (Attachment 2). This includes a full range of physico-chemical, and molecular tests, including detailed protein analyses in our proteomics department. We shall do the same for pigments isolated directly from fish tissue and compare them to pigments isolated from bacterial strains. If they are identical, we shall carry out infection and disinfection experiments in those red fluorescent fish that can be cultivated (e.g. some gobies and pipefish). This will allow us to prove the necessity of these bacteria for a normal development of fluorescence. We do not exclude a priori that some fish produce their own fluorescent pigments. In order to complement behavioral tests, we shall also isolate and characterize opsin and rhodopsin genes from focal fishes using standard protocols (Helvik et al. 2001; Loew and Zhang 2006b).

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Competitive interactions of sponges and corals in the Gulf of Aqaba

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Abstract. This study describes the spatial competition and interaction structure of sponges and corals in reef habitats in Dahab, Egypt. In total 890 epireefal sponges were observed. Overall 62.4% of sponges occurred alone, 37.6% in interactions with corals. Sponges in aggressive contact interactions acted rather as epibionts (19%) than basibionts (12.2%) or in stand-off interactions (1.3%), although the abundance and species diversity of epireefal sponges is by far surpassed by corals. These results suggest that sponges may be the better competitors but corals are the better colonizers.

Introduction

Space and access are the most limiting factors in benthic coral reef communities (Lang 1973). Consequently the competition for space and light makes up the biggest part of intra- and interspecific competition of sessile organisms on coral reefs. Seeing as coral reefs have a high diversity of species, their communities are inclined to develop complex competitive networks (intransitivity) rather than hierarchical structures (transitivity), (Jackson & Buss 1975, Jackson 1977). Complex networks in community structures are the result of (1) competition modification either through drawback or reorientation, (2) standoff interaction through a balance of competitive abilities of two species and (3) disturbance for example through selective predation (Tanaka & Nandakumar 1994). Dominance of species is prevented and the coexistence of species is promoted because of interspecific tradeoffs between competitive abilities and dispersal abilities and susceptibility to disease and predation (Tilman 1994). The better competitor is the worse colonizer (growth rate, mortality, generation time) and vice versa.

In consequence of the harsh competition for space one species can overgrow the other, resulting in either reduction of fecundity, and growth or death of the overgrown organism (Connell & Keough 1985). This can be induced through chemical or physical means and is an important form of space competition (Lang 1973, Buss 1980). However, the most important type of interspecific interactions in a tropical reef is the standoff interaction (Russ 1982, Sebens 1985, Karlson 1980). The standoff interaction is an interaction without a clear outcome or winner (Buss & Jackson 1979). While circumventing the growth of the interaction partner at one point with a standoff interaction, the organism can colonize free substrate in another direction (Karlson & Jackson 1981, Russ 1982, Sebens 1985).

The three most abundant groups of organisms in a typical red sea reef are scleractinia, alcyonaria and benthic algae (Benayahu & Loya 1977). Although the Red Sea accommodates high densities of sponges, most of them are cryptic and only occur in crevices (Richter et al 2001). Percentage cover of epireefal sponges in the Red Sea and Indo-Pacific is relatively small, less than 10% (Hodgson 1999) compared to the Caribbean, where they could eclipse corals in abundance and diversity (Aerts 1998, Suchanek et al 1983, Zea 1993). The abundance of coral surpasses by far the abundance of epireefal sponges hence most of interspecific sponge interactions will be coral/sponge interactions.

Hard corals have developed different mechanisms to actively harm space competitors like sweeper tentacles, mesenteric filaments and shading (Lang & Chornesky 1990, Chornesky 1983, Chadwick 1987, Lang 1973).

Soft corals can exude secondary metabolites, especially terpenes that do have allelopathic effects on neighbouring corals and can deter larvae settlement of stony corals without touching (Sammarco et al. 1983, 1986, Maida et al. 1995). These terpenes also protect against predators, infections and injuries and are therefore jointly responsible for the good regeneration ability of soft corals. Soft corals do have the ability to actively move over substrate or to change their orientation and growth patterns (La Barre & Coll 1982).

Connected with a high growth rate and the capability to form stolones it makes them opportunistic colonizers (Benayahu & Loya 1987, Cornell & Karlson 1996).

Sponges are a rich and widely researched source of secondary metabolites, which may have potential to be processed as pharmaceuticals. Many of those substances show antibiotic, antitumoric, anti-inflammatory and cytotoxic effects (Rinehart et al. 1981, Sibkema 2005). Secondary metabolites also play an important role in deterring predation (Pawlik et al. 1995, Thoms et al. 2004, Assmann et al. 2000, Burns et al. 2003) and fouling (Hirota et al. 1998). They also do have an allelopathic effect on other sponges (Engel & Pawlik 2000) and coral symbiosis in situ (Pawlik et al. 2007) and can cause necrosis in neighbouring sponge or coral tissue (Jackson & Buss 1975, Porter & Target 1988, Turon et al. 1996, Thacker et al. 1998, Turon et al. 1996). Allelopathic compounds may play a key role in the competitive capability of reef sponges. In most coral/sponge interactions the sponge is the dominant partner or rather corals are overgrown by sponges (Jackson & Buss 1975, Suchanek et al. 1983, Sullivan et al. 1983, Porter & Targett 1988, Aerts & van Soest 1997).

Subject of this study were (1) to measure the competitive ability of epireefal sponges. (2) Whether competitive abilities of sponges are influenced by growth form and size.

Methods

Data on growth form, size, percentage cover and competitive interaction were obtained by SCUBA diving to a depth of 24 m, during October 2007 and April 2008 in Dahab, Gulf of Aqaba, Egypt.

Sponge collection and identification: Samples of reefal sponges were collected during SCUBA diving. They were put in separate bags and immediately stored at 5 C° in a hypertonic salt solution (Heim et al. 2007). Furthermore, photographs were taken with a Canon Powershot A720 IS and a Canon Powershot S70. Morphological identification of sponge species was based on spicule and tissue characteristics.

Mean Percentage Cover: Estimates of mean percentage cover of substrate were determined based on the line intercept method with 20 m transect lines laid out at two different depths (10m and 16m) on the reef (Hodgeson et al. 2006).

Four different types of interactions were defined: (1) Loss: sponge was overgrown by another organism; (2) Win: sponge did overgrow another organism; (3) Stand-off interaction: no clear win or loss of the interacting organisms could be determined at the time of observation; and (4) Touch: peripheral touch of tissues without physical attachment

Study site: Research was carried out in Dahab, Egypt, Gulf of Aqaba (Fig.1). It is characterized by fringing reefs that grow north of the normal limit for reefs in the northern hemisphere.

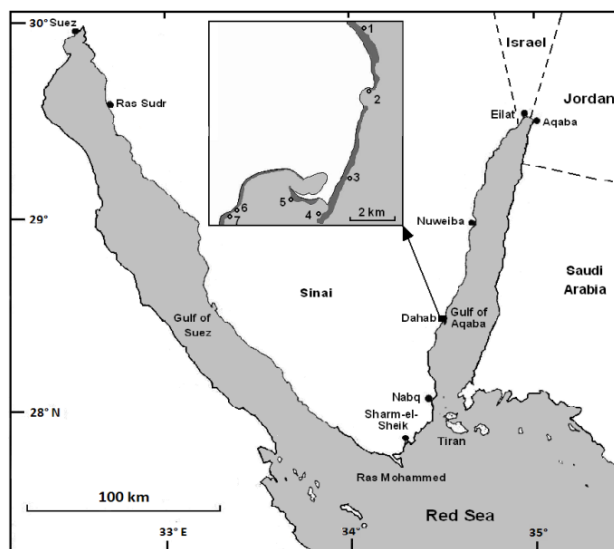


Fig.1. Map of study area in Dahab, the northern Red Sea and Gulf of Aqaba with sampling locations 1 to 7

To measure strength of association between growthform, size and interaction, the Pearson's contingency coefficient was utilized.

Results

Substrate cover at the dive sites was clearly dominated from corals although hard corals were more abundant than soft corals. Sponges made up the smallest proportion of substrate cover at all dive sites (Table 1).

Table 1. Average substrate cover of hard corals, soft corals and sponges at the study sites "1" to "7".

Study site	"1"	"2"	"3"	"4"	"5"	"6"	"7"
Hard coral cover in%	38.1	38.1	36.8	13.8	28.1	28.1	28.1
Soft coral cover in%	23.8	12.5	6.3	31.9	16.3	6.3	18.8
Sponge cover in%	1.3	1.3	0.6	5.6	1.9	3.8	6.9
Benthic algae cover in%	1.9	5.0	9.4	8.1	2.5	8.1	8.8

In total 890 sponges organisms were observed made up of 16 sponge species. Only 37.6% of the observed sponges did have one or more interactions (823). The by far most abundant sponge species was *Hemimycale arabica* (243) and *Crella cyathophora*. *Crella cyathophora* is an abundant sponge and opportunistic colonizer of massive growth form, which can be dominant in disturbed habitats (Perkol-Finkel & Benayahu 2005). *Hemimycale arabica* is a facultative cryptic sponge with a layerlike growth form. In sponge/interaction these two species mostly were able to overgrow the corals (Fig.2)

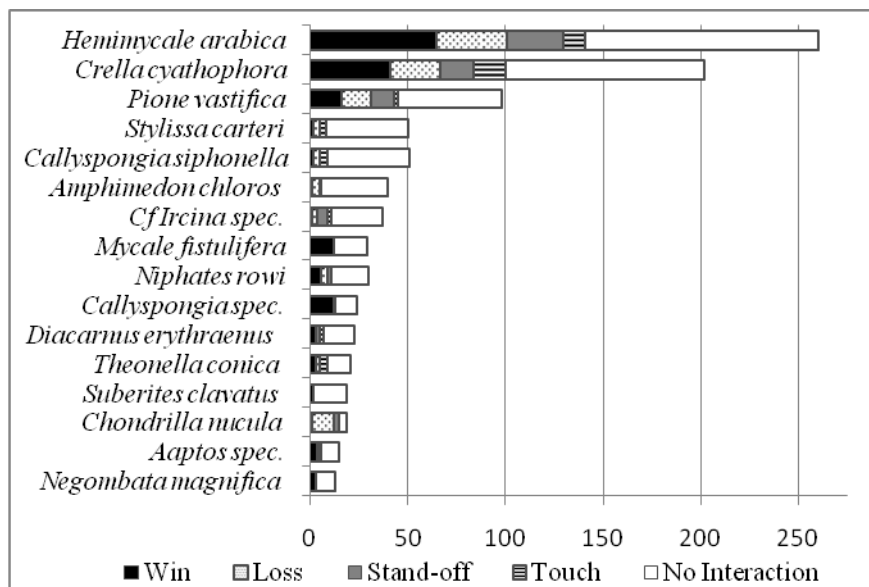


Fig.2 Total numbers of specimen and interaction categories for each sponge species.

Most coral/sponge interactions occurred with alcyonarians (204) and 155 with scleractinians. Only 23 of the interactions observed were sponge/sponge interactions. In most cases 19% poriferans occurred as epibionts, whereas 12.2% of the sponges were basibionts. Touch interactions made up 4.9% of sponge/coral interactions and only 1.3% stand-off interaction between poriferans and corals were recorded (Fig.3). The alcyonaria make up the biggest part of organisms able to overgrow poriferans and are least overgrown. The majority of hard corals were basibionts. Only 1.2% of the scleractinians observed were able to overgrow poriferans (Fig.3).

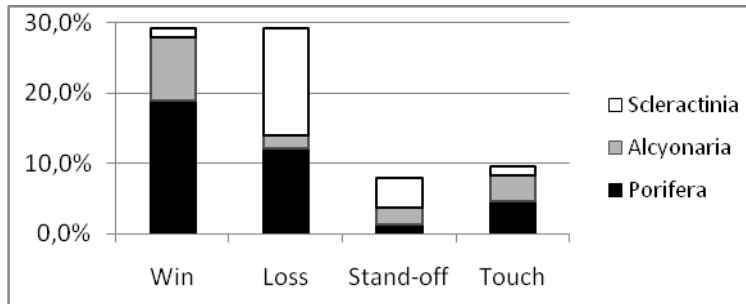


Fig.3 Percentage of scleractinians, alcyonarians and poriferans for each category of sponge/coral interactions.

Size and growth form of sponges had an evident influence on the interaction of sponges and corals (Pearson contingency coefficient $C_{\text{growth form}} = 0.26$ and $C_{\text{size}} = 0.21$ with $C_{\text{max}} = 0.71$)

The growth form of the sponges was mostly layer-like (42.6%) and compact (39.6%). Only (17.7%) exhibited a branching growth form (Poriferans with a branching growth form had the smallest proportion of interactions (24.4%) whereas 54.8% of layer-like growing sponges occurred in interactions and 42.5% of massive sponges).

With exception of the very small sponges ($\leq 25 \text{ cm}^2$) the abundance of sponges decreases with increasing size. But the proportion of occurrence in interactions increases clearly with growing size (Fig.4 B).

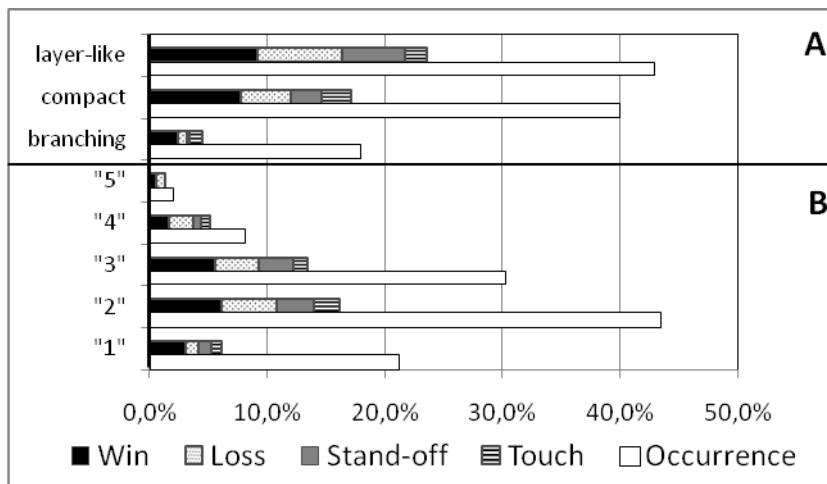


Fig.4 A Percentage of interaction categories for the three different growth forms of poriferans (layer-like, compact, branching); **B** Percentage of interaction categories for the size categories of sponges ("1" $\leq 25 \text{ cm}^2$, "2" $\leq 225 \text{ cm}^2$, "3" $\leq 625 \text{ cm}^2$, "4" $\leq 1225 \text{ cm}^2$, "5" $\geq 1225 \text{ cm}^2$)

Discussion

The growth form of the opponents is important for interactions (Lang 1973). That was also verified in this study. Most interactions were observed with layer-like growing sponges (54.8%). They have the largest surface compared to their volume and therefore present the bigger target for interaction partners. Compact growing sponges showed fewer interactions (42.5%). Branching sponges exhibited the least interactions (24.4%). They have less contact with the substrate and therefore have possibility to grow height (Meesters et al. 1996).

The size of sponges varies with age, nutrient supply and environmental conditions. Negative growth rates courtesy of predation, partial necrosis and shrinkage are not unusual. Incrusting sponges of the family clionidae for example can grow up to 13 cm per year (Aerts 2000; Rützler 2001). The percentage of interacting sponges increases with growing size. Larger sponges do have a bigger surface and periphery, where they can encounter interaction partners or can be colonized.

In several studies, sponges showed the greater competition potential in coral/sponge interactions (Jackson & Buss 1975, Suchanek et al. 1983, Sullivan et al. 1983, Porter & Targett 1988, Aerts & van Soest 1997). This could be also confirmed in this study. In 19.5% of the investigated sponges interacted as aggressor and thus did overgrow the other organisms and 12.7% were overgrown by another sponge or coral. Stand-off interactions were only a small proportion of interactions (8.2%), contrary to several other studies in marine benthos communities. For Russ (1982) the high fraction of standoffs signalled the lacking of clear dominance or intransitivity. Schmidt and Warner (1986) also thought the great occurrence of standoff interactions (67%) linked with an equal amount of wins and losses as a clear sign of intransitivity. A study on the quantification of sponge-coral interactions in Columbian reefs showed a high percentage of standoff interactions (32.5%) and just a marginally part of aggressive interactions like overgrowth (2.5%), (Aerts & van Soest 1997).

Problematic is the small abundance of poriferans compared to corals in the Red Sea. Also the degree and quality of the interactions (growth rate, allelopathy) are unknown. The results of this study lead to the conclusion that sponges are the better competitors but corals are the better colonizers (higher abundance).

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Ammelshain – diving in a former stone pit (introduction to fieldtrip location)

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Abstract. Since the 1970s the former stone pits of Ammelshain are used as a local recreation location by divers all over Germany. It seems to be interesting that the bigger eastern former stone pit evolved itself to a perfect ecosystem including a distinctive underwater biodiversity. By using the slogan “Ammelshain – Diving in a nature protection area” the residentiary dive club wants to advert divers for visiting and enjoying the natural dive spot.

So even scientist and scientifically interested laymen are trying to analyze the “Hazel Hill” area, where three water filled stone pits are located. However, next to the representative history of the area and anthropogenic influences we can bear in mind that geological and morphological facts cause the biosphere of the pit and its biodiversity. Through the specific mining a step like structure emerged. According this the structure includes mainly shallow water areas, deeper areas and precious few transition areas. Logically, this leads to a step like structured biosphere order.

Accordingly we can claim that the largest part of biological life is happening in the shallow water areas fewer than five meters. There we can find a manifold sample of micro- and macroorganism. By analyzing a watermilfoil sample, we would be able to find a lot of different little creatures like larvae, leeches or crustaceans. But by periodical diving also fishes, jellyfishes and rare creatures can be seen.

By analyzing the biodiversity and the different influences and conditions at which the ecosystem is based I could not help but see that subtle interminable influences are much more destructive than expected. It underlines the need, referring to ecosystems like the “Hazel Hill”, for being protected due a clear legislation. But nevertheless it also reveals the need for more social engagement in protecting the local environment and the interest in its biodiversity.

Furthermore there no denying the fact that, although the stone pits water has a very low nutrient concentration, Ammelshain provides an outstanding naturally ecosystem with a distinctive underwater biodiversity.

Historical Background

The “Hazel Hill”, which includes the stone pits, is primary made of quartz porphyry. Its origination goes back into the Perm - 290 million years ago - when explosive volcanoes reformed the German Midlands. Whereas in the 1870s the hazel hill was determined worthless, R. Ebert-Ammelshain knew it better when he started mining porphyry in 1887 in this area. After all, in the 1950s the porphyry production stopped completely. After all useful remaining machines were removed the pit was fulfilled by ground- and rainwater. By 1978 the water level of today was reached. Image and oral sources proof that divers started exploring the water filled stone pits during the 1970s. And even today divers all over Germany are visiting the pits, since its nice underwater visibility.



Fig.1. "Hazel Hill" stone pit still non water filled (Arrow marks water level today)

Characteristics

Geological and morphological conditions

Like already mentioned, the stone pits are located on the "Hazel Hill", which is a hill made of quartz porphyry. This brittle volcanic stone has a light to dark brown color and it's preferred as gravel and ballast in road and rail constructions. Additionally there is the need to put the regional groundwater level into consideration. It is based on the "Muldentaler glacial valley", which is characterized as a very high water level ground water occurrence. Combined with the brittle stone constant slow-going water flow through the pit seems to be a realistic assumption. Supportive facts for this claim are distinctive ice free regions on the surface during the winter season. A furthermore prove is the observable flickering water near to steep faces which indicates a water exchange between two different temperatured water occurrences. Underneath the water surface firmly bonded hard white deposits are observable.

In regard to Figure 1 you should be able to understand that the structure of a stone pit is different. In Ammelshain there are primary steep faces. This leads to a big shallow water area (<5m) in the northern part of the pit and the pit sides. Furthermore, it also leads to a large 15m deep area in the centre, as well to a 25 meter deep area in the South. (Fig.10.)

Biological situation referring to the analytical biosphere watching program 2008.

An interminable experiment series, which includes soil and plant sampling, sample analyzing and compiling an index, led to an objective overview of occurring creatures.

Constant occurring species were:

Ephemera larva (*Ephemeroptera spec.*)

It is an indicator for water with high oxygen content and it feeds upon alga and dead biomass. After three years larval stage the adult Ephemera leaves the water for several hours of reproduction and following dying. It is a preferred food for fishes and an indicator of a water quality between one and two.



Fig. 2. Ephemera larva

Nonbiting Midge larva (*Chironomidea spec.*)

The widely spread Non-biting midge larva also known as chironomids makes for a high part the food of the fishes. Their color and behavior are variable. Bloody red versions can be found in the mud and green ones in open waters. Their color depends on their behavior of eating just like their way of surviving. There are Nonbiting midge larvae, which are even able to build nets or to hunt zooplankton.



Fig.3. Chironomid

Rail Fly larva (*Trichoptera spec.*)

The Rail fly larva appears in two different forms. Without rail it tends to predatorily behavior. But with a rail it feeds upon dead biomass and is an indicator for very high quality water with high oxygen content. Furthermore it is an indicator for slightly acid water.



Fig. 4. Rail fly larva

Freshwater Acari

With their size of a few millimetres they seem to be tiny arachnids, but actually also their behaviour of eating depends on the situation. It feeds upon dead biomass and living creatures. They can feed upon fishes and bigger organisms parasitically.



Fig. 5. Freshwater Acari

Results

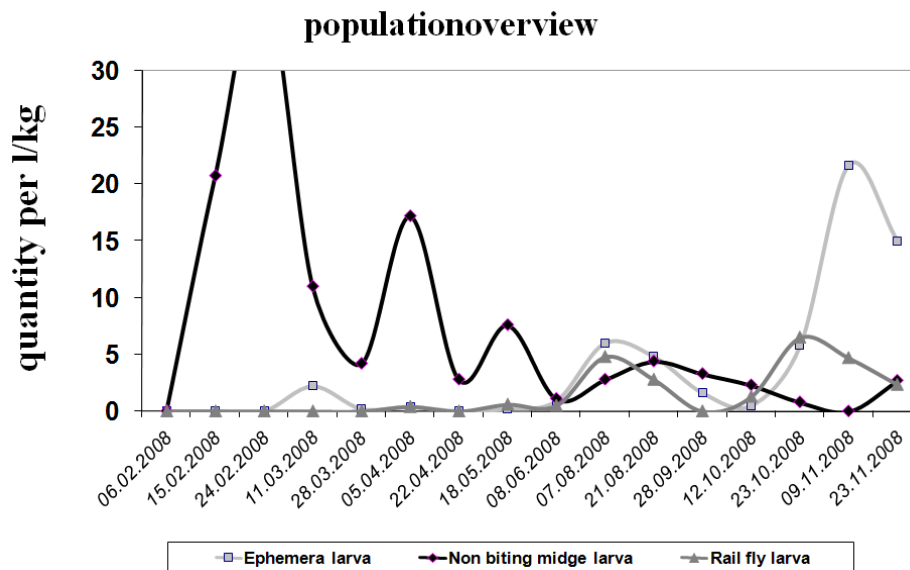


Fig. 6. Population overview

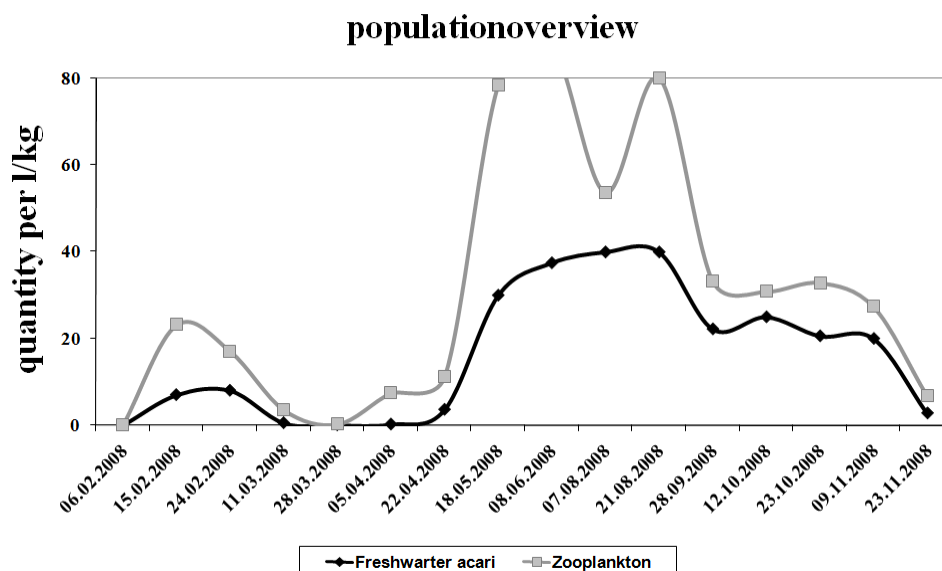


Fig. 7. Population overview

By comparing Fig. 6. and Fig. 7. a similarity to Fig. 8. can be noticed. Temperature and the general developing of the population line are looking similar. Warm and eutrophic water, especially in the summer season, supports the developing of biodiversity in the upper water layers. Considering the temperature high of 28°C on the 07th August 2008 and the population highs of the Freshwater acaris and the Zooplankton we can notice a shift of one month before or after a temperature change. So it is not a question about how high the temperature of the water is but rather which weather trends can be expected for the next days and weeks. So by noticing of temperature increasing the organisms start to increase their reproduction rate as well. Through thermal tolerance every organism has its individual living needs and limits. Therefore the population line of the zooplankton goes much more similar to the temperature line than the population line of the ephemera larvae. This leads to the assumption that more complex organisms like larvae are able to tolerate much higher temperature differences than microorganism like plankton are able to tolerate.

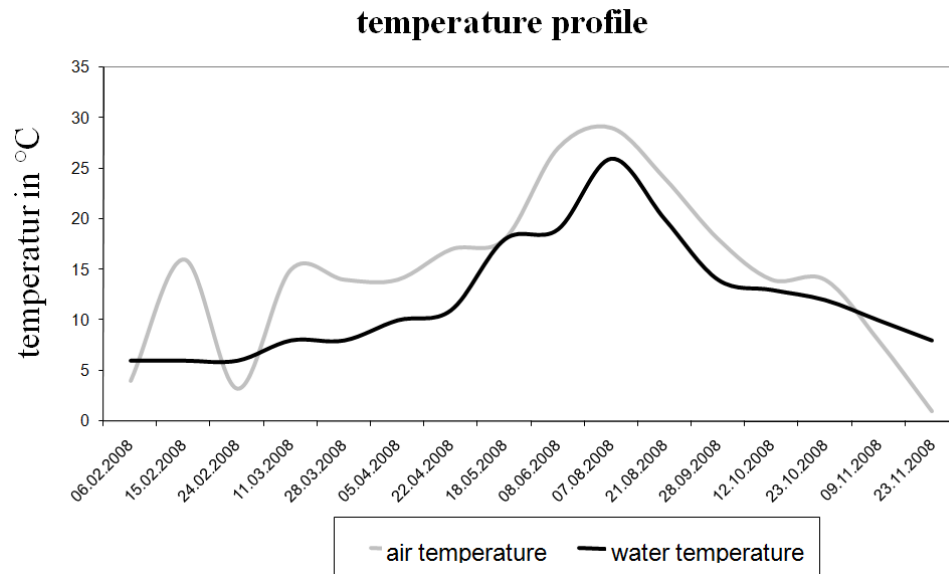


Fig. 8. Temperature profile

In contrast to the temperature aspect stands the fact that distinctive population pikes can be seen in Fig. 6. This can not be explained by arguing with the temperature aspect. Considering the predator-prey relationship these pikes can be explained. As noted before Nonbiting Midge larvae can be considered as main food source of fishes. Empirical observations of young fish shoals leaving the deeper water layers on the 16th February might explain the first distinctive population minimum in March. Logically predators adjust to their feeding conditions. By an increasing number of potential prey animals they are changing their behaviour of getting food. Even the population highs of the Ephemera larvae and the Rail Fly larvae might be caused by fishes leaving the upper water layers in autumn. To put in a nutshell there are manifold influences which are causing a population change. Often these influences are very subtle but especially by being long time effective, much more population relevant than expected. In regard the influences there should be considered the fact that a ecosystem has a enormous complexity and that even by constant watching not every single influence can be noticed and considered.

2.3. Further biologic information about the stone pits biodiversity.

Generally there are two typical hydrophytes. In areas from 7 to 15m Stoneworts can be found in concise population sizes. Stoneworts gain their stability by using Ca^+ ions for building an inner framework-like structure, which makes them an indicator for water with a high Ca^+ concentration.

Lower areas above 6m are largely populated by watermilfoils, which are preferred for hiding by microorganisms and more complex animals like fishes.

Usual central European fishes like Carp, Red-Eye, Catfish, Pike, Perch and Tench are living in the pit. Much more interesting is the fact, that some special creatures made the pit their resident. The exotic Freshwater Jelly, for example, contributes to the beatification of the stone pit although its roots are going back to china. Tiny Freshwater Adenoids, which are factually able to clone their selves through pullulating, are rewarding everybody who's searching beauty in detail. In Addition, we should also consider the alien Zebra Mussels, which are contributing through their extreme efficient filtration ability to the increasing visibility. The appearance of the rare Freshwater Spongilla is indicating high clean water qualities of the pit.

It is the basis for the distinctive appearance of microorganism like dragon fly larvae, ephemera larvae, fish leeches, and caddis fly larvae.



Fig. 9. Stoneworts



Fig. 10. Watermilfoil

Chemical characteristics and water quality

Table 1. chemical values

water temperature	smell	visibility	depth	NO ₃ ⁻	PO ₄ ³⁻	NH ₃	NO ₂ ⁻	pH	Water Hardness
10 °C	neutral	clear to slightly dull	surface	0 mg/l	0 mg/l	0.05 mg/l	<0.02 mg/l	7.4	12 dh°

¹Values from 11.11.2006| 12.00, used materials: Aquanal water test

Chemical analysis proof, that the water has excellent low concentrations of ammoniac, nitrate, phosphate, nitrite ions and a passably balanced, slightly basic pH value of 7.4. But contrary that low concentrations, the water has a distinctive value of water hardness, which may refers to a high concentration of Ca⁺ ions. The already mentioned occurrence of Stoneworts and their need for Ca⁺ ions underlines the assumption of a high Ca⁺ concentration. Also by having a look at the geological conditions this assumption can be confirmed. The white hard deposits, which are firmly bonded to the stone surface underneath the water surface, can be considered as lake marl. This is a chemical deposit which is created through the combination of carbonic acid and a high concentration of calcium ions. Carbonic acid is a common acid, which is created naturally in Freshwater lakes. Through the diffusion of carbon dioxide out of the air into the lake water Hydrogencarbonate is created. Combined with a calcium ion a hardly soluble white deposit is created, also know as lake marl.

The distinctive good water quality may be explained by the geological conditions. Due the constant flow of fresh ground water into the pit the water does not totally stand still but can be refreshed.

Map of the eastern stone pit of Ammelshain – located on top of the “Hazel Hill”

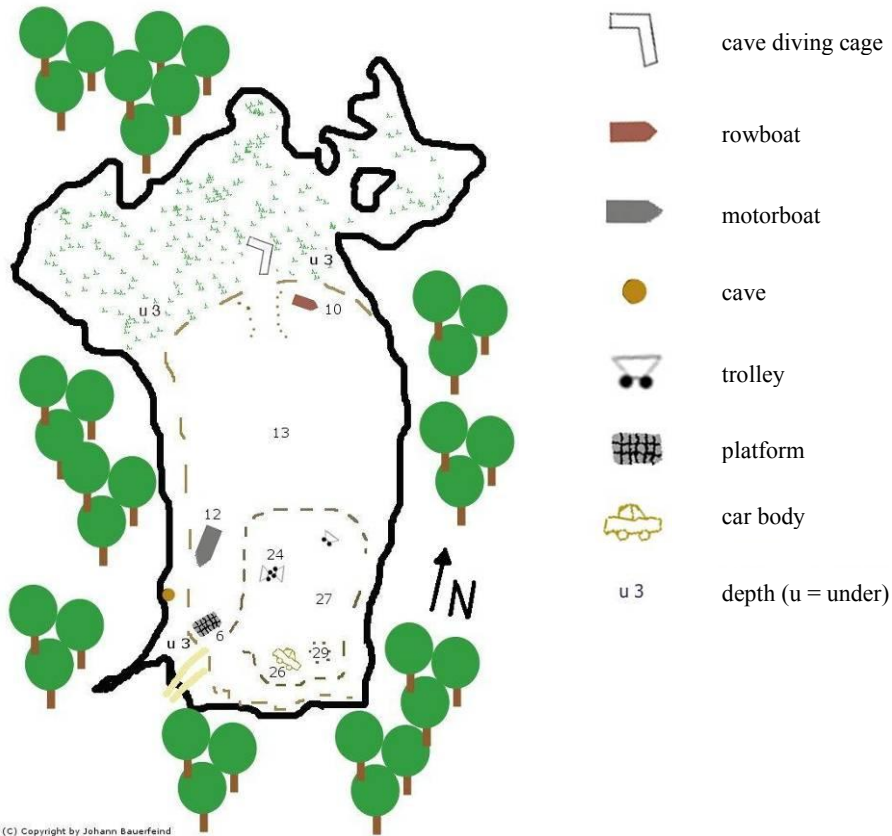


Fig.10. A map of the pit 1: 25000

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The distributional patterns of the zoobenthos from the artificial hard substratum of the Agigea dyke (Romanian Black Sea coast): preliminary results

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Abstract. The aim of the present study is to reveal the structure and distribution of benthic assemblages established on artificial hard substratum of the midlittoral fringe and of the upper sublittoral of the Agigea dyke. On the basis of 52 samples collected seasonally by diving in 2004 and 2005 between 0 m and 16 m depth a total of 88 species belonging to 22 systematic groups were found. The crustaceans were the dominant group as number of individuals, whereas the biomass was dominated by the bivalve molluscs. The average density of the fauna from rocky mussel bio-coenosis was found to be 1,690,100 ind.m⁻² and the biomass 25,323.94 g.m⁻².

Introduction

The hard substratum occupies off the Romanian Black Sea coast an area of approximately 70 km², which represents only 0.3% of the total surface of the Romanian continental shelf. Nevertheless, the rocky seabed has a particular scientific and economic importance. Thus, due to its irregularities, cracks and cavities, the rocky substratum presents an extraordinary habitat complexity, offering favourable conditions for the development of a rich and diversified life. The hard substratum harbours many more species than the relatively homogenous soft sediments in their vicinity, being veritable hotspots for marine biodiversity. Rocky substratum represents the preferred biotope for almost all sessile and encrusting fauna, forming rich epibioses, which at their turn lead to even bigger differentiation of the ecological niches and microhabitats. Boring organisms induce, by the galleries they perforate, an even more accentuated 3-dimensionality of the structure of this type of sediment. Additionally, seaweeds grow only on hard substratum, increasing by themselves the surfaces available for the settling of epibenthic organisms (Băcescu et al. 1971). In addition to natural rocky seabed, hard substratum is also represented by man-made structures such as docks, piers, jetties, breakwaters and shipwrecks.

Despite the fact that hard substratum is characterized by such extraordinary biodiversity the quantitative studies of this type of sediment are still limited in number. This can be explained by the fact that remote sampling using different types of trawls, dredges and grabs, are inappropriate to sample hard substrata or lead to serious underestimations of the number of individuals. Difficulties arise also in collecting all organisms inhabiting cavities and fissures of rocks and stones and in extrapolating the number of collected organisms per square metre, taking into account that this type of sediment is extremely rugged in structure and whose surface is hard to estimate. The only method for quantitative study of organisms from rocky substratum is that of underwater visual census and *in situ* sampling using SCUBA.

At the Romanian littoral the first record regarding the use of autonomous divers for the sampling of hard substratum is that of Băcescu et al. (1963). This paper represents the first quantitative study upon zoobenthos of the rocky facies, which took into account the macrofauna as well as the microfauna. Although this study refers to unusual environmental conditions created by massive settling out of fine suspensions derived from throwing into the sea of a rocky cliff, in order to enlarge the beach from Agigea, it can be considered as a reference paper. In the same period, Gomoiu and Müller (1962) made up the first direct observations by diving with SCUBA in the area of Cape Tăbăcarie (Constanța), as a result of which, between 1-4 m depth, they have established the existence of a sandy-clay outcrops, densely inhabited by the boring mollusc *Barnesia candida*. The only method for quantitative research of this substratum, with compact consistency, was the sampling and *in situ* census of organisms using diving techniques employed by the authors themselves.

The same techniques were applied for the assessment of the abundance and biomass of seaweeds and animals associated to *Cystoseira barbata* canopies and other macroscopic algae (Müller *et al.* 1969; Țigănuș 1972, 1977, 1991-1992) or from artificial hard substrata (Gomoiu 1982, 1986; Gomoiu and Țigănuș 1974, 1976, 1981). Among quantitative studies regarding the hard substratum from Romanian seacoast, which employed autonomous divers for collecting the samples, can be mentioned that of Kühlmann (1960), Băcescu (1965), Dumitrescu (1973), Andriescu (1977), Țigănuș (1979, 1981), Mustață *et al.* (1998), Surugiu (2005), Teacă *et al.* (2006a, 2006b, 2008), Surugiu and Novac (2007) *etc.*

The aim of this study is to update the species inventory, to assess the abundance and the biomass of organisms inhabiting the artificial rocky substratum of the Romanian Black Sea coast and to provide information on their seasonal and depth distribution patterns within this type of habitat.

Material and methods

The analysis of the fauna associated with hard artificial substratum is based on samples collected seasonally (16-06-2004, 1-09-2004, 3-10-2004, 16-04-2005, 26-06-2005 and 23-10-2005) by SCUBA diving from the outer part of the southern dyke of Agigea seaport (Fig.1). Quantitative sampling was carried out along a transect perpendicular to the dyke at 9 different depths (0, 2, 4, 6, 8, 10, 12, 14, and 16 m). At each depth the epibiosis from a randomly chosen area of 400 cm² was scraped off with the aid of a knife. The knife blade served at the same time as a measuring reference for 20 × 20 cm quadrats. In order to prevent the loss of the material, the scraped samples were placed *in situ* into a bag net with 0.125 mm mesh size and with the hatch fastened through a binder. At the surface each sample was immediately transferred into plastic bags, properly labelled and preserved in 4% buffered formalin. In the laboratory the samples were washed through a series of sieves (1 mm, 0.5 mm and 0.125 mm) in order to separate macro- and meiofauna from the sediment. The organisms retained on each sieve were transferred into jars containing 70% ethanol. The organisms were sorted by taxonomical groups, identified as possible to the species level, counted and weighted.

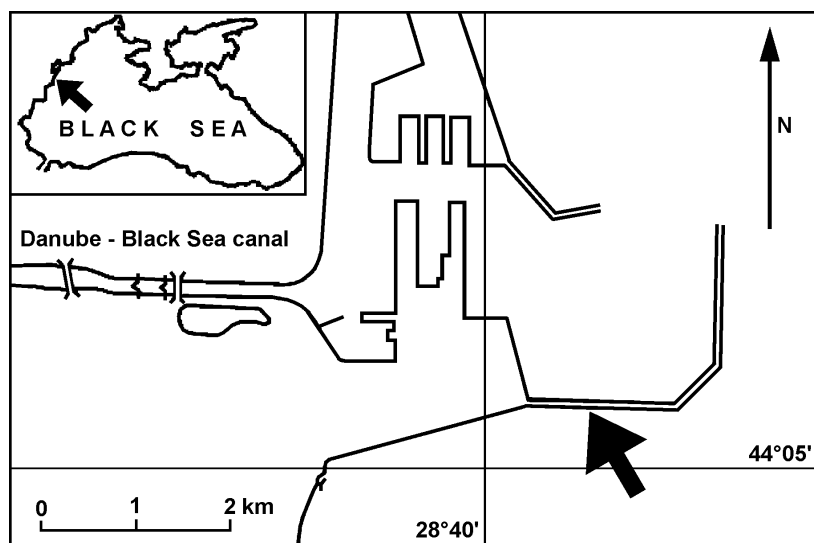


Fig.1. The map of the Constanța South - Agigea harbour, indicating the location of the sampling area (black arrow)

Results

The faunistic analysis of the 52 samples revealed the presence of 97 taxa, from which 88 were identified to the species level. Dominant groups as number of species were Polychaeta (18 species), Amphipoda (15 species), Hydrozoa (8 species), Decapoda (7 species) and Turbellaria (5 species), which accounted for more than 50% of the total number of the species identified. Crustaceans were the most abundant group as the number of individuals with 1,076,243 ind.m⁻², representing 63.68% of the mean total density (Fig. 2). These were followed by worms (530,487 ind.m⁻² or 31.39%), molluscs (47,084.58 ind.m⁻² or 2.79%), coelenterates (34,628.73 ind.m⁻² or 2.05%) and other small taxa (1699.42 ind.m⁻² or less than 0.1% of the general mean density).

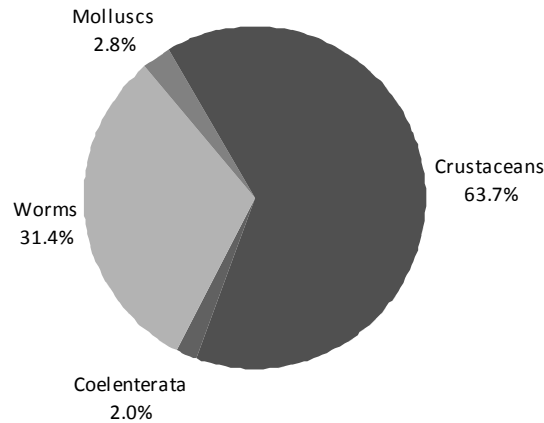


Fig.2. Percent values of the mean density of the main groups of benthic organisms in the Agigea dyke area.

As biomass dominant in all samples were molluscs (24,408.56 g.m⁻² or 96.39% of the total biomass), due to the presence of the leading species on hard seabed – *Mytilus galloprovincialis*, which in the midlittoral and superior sublittoral was associated with *Mytilaster lineatus* (Fig.3). These were followed by crustaceans (821.77 g.m⁻² or 3.2%), worms (28.77 g.m⁻² or 0.11%), coelenterates (16.65 g.m⁻² or 0.07%) and other smaller groups (48.2 g.m⁻² or 0.19%).

The most frequent species on the artificial hard substratum were *Platynereis dumerilii*, *Mytilus galloprovincialis*, *Mytilaster lineatus*, *Balanus improvisus* (all with an occurrence of 100%), *Polydora cornuta*, *Melita palmata*, *Microdeutopus gryllotalpa*, *Stenothoe monoculoides* (which occurred in 97.67% of the samples), *Neanthes succinea* (93.02%), *Jassa oca*, *Pilumnus hirtellus* (both with 88.37%) and *Pisidia longicornis* (86.05%).

The highest species richness was observed in the 2-6 m depth horizon (in the *Mytilus* – *Actinia* subcoenosis) during the autumn and the lowest at 8-16 m depth horizon (the typical *Mytilus galloprovincialis* subcoenosis) during the spring. The total number of species generally increased from spring to autumn. The maximum mean density was found at 2 m depth in the summer (11,172,104 ind.m⁻²), while the minimum was recorded also in the summer at 16 m depth (109,713 ind.m⁻²) (Fig.4). The mean total density of benthic organisms increased from 1,390,210 ind.m⁻² in the spring to 2,288,053 ind.m⁻² in the summer and then decreased again in the autumn (1,159,689 ind.m⁻²). The biomass values showed almost the same trend, with a minimum of 6,506 g.m⁻² at 14 m in the spring and a maximum of 45,412 g.m⁻² at 0 m also in the spring. The mean biomasses of zoobenthos increased gradually from 22,292 g.m⁻² in the spring, to 25 334 g.m⁻² in the summer, and reached the maximum level in the autumn (26 255 g.m⁻²).

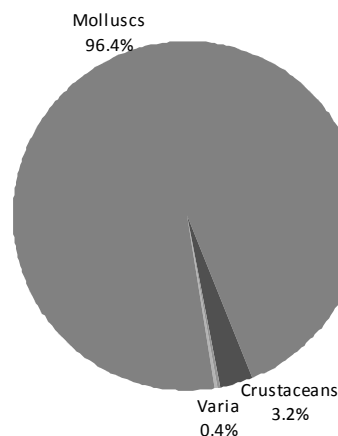


Fig.3. Percent values of the mean biomass of the main groups of benthic organisms in the Agigea dyke.

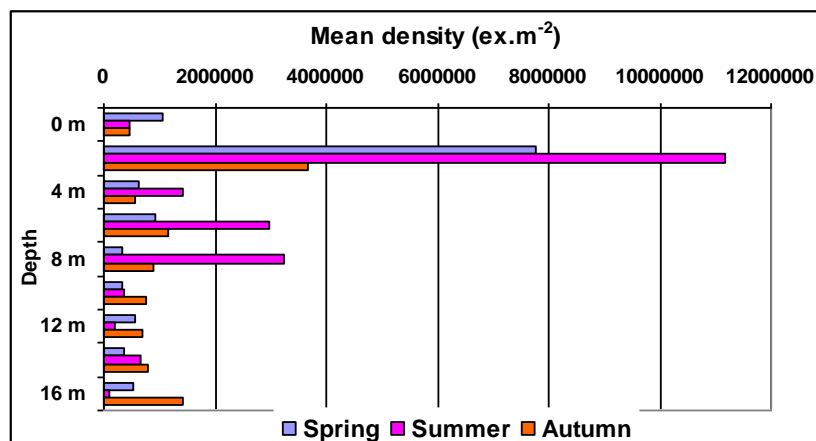


Fig.4. Vertical seasonal variation of total mean density (ind.m⁻²) of the zoobentos in the Agigea dyke area.

Discussion and Conclusions

Agigea marine area was traditionally considered as a “hotspot” for ecological diversity due to the various types of substrata present here (Andriescu 1977). Thus, Băcescu et al. (1963) reports 120 taxa from the rocky mussel’s beds of Agigea. In the 70-80’s this area was subjected to severe man-made disturbance as a consequence of the enlargement in the immediate vicinity of the Constanța Sud – Agigea seaport, the construction of the Danube – Black Sea Canal and of the increased levels of eutrophication and pollution. The macrozoobenthos from this area has experienced loss of biodiversity and major changes in the community structure and functioning, being characterized by the reduction of number and abundances of crustaceans and by increase of abundances of different groups of worms. For this period Țigănuș (1979) indicates at Agigea only 28 taxa. This low number of taxa is explained also by the fact that many benthic groups (Nematoda, Polychaeta, Halacarida, Ostracoda, Copepoda and Chironomidae) were not identified to the species level. Starting with the 90’, due to the socio-economic recession in the riparian Black Sea countries, there is a gradual recovery of the communities associated with hard substratum. Mustață et al. (1998) cites for Agigea 61 taxonomic groups of benthic invertebrates. For other rocky bottoms, Teacă et al. (2006a, 2006b, 2008) reveals the occurrence of 141 species and 31 supraspecific taxa. A series of species considered rare or absent some 20 years ago in faunal assemblages of the rocky bottoms, such as *Opercularella lacerata*, *Ventroma halecioides*, *Tergipes tergipes*, *Limapontia capitata*, *Erichthonius difformis*, *Jassa ocia*, *Siriella jaltensis jaltensis*, *Athanas nitescens*, *Palaemon adspersus*, *Pisidia longicornis*, *Eriphia verrucosa*, have increased their frequency and abundance.

The mean density of benthic organisms from Agigea dyke found during this study was estimated to be 1,690,100 ind.m⁻². This value exceeds the densities reported for natural rocky seabed from Agigea well before the anthropogenic impact on the sea (which ranged from 11,641 ind.m⁻² and 432,400 ind.m⁻²) (Băcescu et al. 1963) or when the human pressure on the benthic fauna was at its maximum (mean abundance value of 250,000 ind.m⁻²) (Țigănuș 1979). However, our data are comparable to that of Teacă et al. (2008), which indicate for natural mussel beds, situated between Cape Midia and Vama Veche, densities comprised between 1,300,000 ind.m⁻² and 2,100,000 ind.m⁻².

The average biomass of the fauna associated with rocky artificial substratum from Agigea was found to be 25,323.94 g.m⁻². Băcescu et al. (1963) found for natural rocky bottom from the same area much lower biomasses (142.48-897.77 g.m⁻²). Nevertheless, the present study support the data provided by Teacă et al. (2006a, 2006b) for the southern sector of the Romanian Black Sea coast (16,500 g.m⁻²) or for Mamaia and Mangalia sectors (19,437.20 g.m⁻²).

The comparison of mean densities of benthic invertebrates at different depths and in different seasons indicates similar variations (increase in density at 2, 4, 6 and 8 m depth, then the decrease at greater depths) in all seasons, but generally with greater values in summer. The total biomass values show the same trend as the density, with the biggest values occurring at depths smaller than 10 m. This can be explained by favourable conditions for the development of epibenthic fauna at these depths: attenuated wave action, but still sufficient light for the development of the macrophyte algae.

The results of this study show that the community structure on the artificial hard substratum from Agigea dyke is very similar to that of the natural rocky seabed from Agigea or from other areas off the Romanian

Black Sea coast. Irregularities of the seabed, luxuriant epibioses and high density of the organisms all contribute to the fact that hard substratum represents the place for living, for hiding from predators, for feeding and for reproduction for many animals. Teacă et al. (2008) shows that, at the Romanian Black Sea coast, the epibenthic community matrix generally is composed of approximately 18-20 major invertebrate taxa. However, hard bottoms are highly dominated by only three sessile species: *Mytilus galloprovincialis*, *Mytilaster lineatus* and *Balanus improvisus* (Băcescu et al. 1971).

Regarding the vertical distribution of species it can be observed a net separation of midlittoral benthic fauna from that of the sublittoral, due to the peculiar type of formation of epibenthic associations as a result of severe wave action. Benthic fauna from greater depths is differentiated by an increased number of species common on sedimentary bottoms.

The results obtained during this study have enlarged our knowledge on the distributional patterns of the fauna associated with hard substratum and may serve as a baseline for future research and for establishment of an integrated ecological monitoring at the regional and/or national level.

Acknowledgements

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Scientific divers quantify first known outbreaks of cold-water coral disease

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Abstract. Coral diseases are widely reported in the tropics but the first incidence of cold-water coral disease was not noted until 2002 when divers recorded an outbreak at 10-28 m depth off Lundy in a NE Atlantic marine protected area. The seafan *Eunicella verrucosa* exhibited coen-chyme necrosis and subsequent diving surveys of >600 colonies at 13 sites since revealed that disease outbreaks were widespread in SW England to depths of 50 m from 2003-2008, possibly caused by infection by *Vibrio* bacteria at high temperatures.

Introduction

There is a growing incidence of disease in warm-water corals with widespread concerns over the deleterious knock-on effects on biodiversity, tourism and coastal defence (Harvell et al. 2002, Bruno et al. 2003, Linares et al. 2005, Ritchie 2006). Hall-Spencer et al. (2007a) reported the first studies of diseases in cold-water corals at a time when there is growing awareness of the ecological importance of habitat-forming corals in temperate and deep-sea environments (Hall-Spencer et al., 2002, 2007b; Davies et al., 2007; Roberts et al. 2009).

We reports *in situ* and laboratory observations of a disease outbreak in cold-water populations of *Eunicella verrucosa* following preliminary reports of the disease at a well-monitored site at Lundy, a Marine Protected Area in SW England. This gorgonian has a limited distribution around the British Isles and is one of the few marine species fully protected under the Wildlife and Countryside Act, 1981 and listed in the UK Biodiversity Action Plan. It is also listed as vulnerable on the International Union for Conservation of Nature and Natural Resources 'red list' of threatened species (IUCN, 2006). The three main aims of the present study were to (1) evaluate whether an outbreak in 2002 was an isolated incident, (2) to examine the nature of the disease and (3) to investigate the hypothesis that pathogenic microbes were responsible.

Methods

Field observations

Destructive sampling was kept to a minimum because of the protected status of *E. verrucosa*. Surveys of disease incidence were undertaken using digital video camera during SCUBA dives along depth-stratified belt transects at sites known to support dense populations of the gorgonians based on interviews with local divers. Thirteen sites were surveyed between 1-7 times in the English Channel, and one site on Lundy No Take Zone, Bristol Channel from 2003-2008 allowing an analysis of the incidence of disease in >600 separate gorgonian colonies.

The camera was used to record all gorgonians encountered in a 25 min period along belt transects within depth contours of ± 1.5 m. Still pictures were taken from the video showing perpendicular photoquadrats of each gorgonian next to a ruler for scale. The photographs were analysed to record prevalence of necrosis and the degree of fouling by epibionts was recorded on a three rank scale (0 = no overgrowth, 1 = <10% of gorgonian overgrown, 2 = >10% of gorgonian overgrown) to test the hypothesis that fouling organisms were more prevalent on gorgonians at sites where necrosis was noted. Repeat surveys of the same gorgonians were made at Knoll Pins, Lundy using obvious features of the submerged cliff and boulder field to re-

locate individuals whereas different areas were sampled on repeat surveys at the other sites to ensure that the same individuals were not surveyed twice.

Divers collected the first 21 gorgonians encountered from a patch of specimens in various states of disease and overgrowth to identify their associated macrobiota at 19-22 m below CD on Mewstone Ledges (50°18.15'N 4°06.74' W) in accordance with a licence granted by English Nature. Each gorgonian was encased in a labelled plastic bag before using garden secateurs to cut through the stem and tying the bag closed. Samples were preserved in 4% seawater formalin and rinsed over a 0.2 mm sieve to retain larger epibiota. Attached species were removed under a dissecting microscope, identifying the various species present.

Methods used to describe the effects of the disease on the histology and microbiology of the sea fans, plus infection challenge experiments are given by Hall-Spencer *et al.* (2007).

Results

Video transects allowed subsequent analyses of 634 gorgonian colonies around SW England, of which 9% had tissue necrosis. In some areas off Plymouth, UK this species forms coral forests (Figure 1).



Fig. 1. Dense stand of *Eunicella verrucosa* with shoal of poor cod at 24 m depth on the wreck of the *Persier*, Plymouth. Photograph by Sally Sharrock. Insert, diseased necrotic sea fans overgrown by fouling organisms.

Affected colonies had patches of necrotic tissue which were soft and white (Fig. 2A) and could easily be rubbed off by hand to expose an underlying black gorgonian skeleton. Healthy gorgonians and unaffected parts of colonies with necrotic tissue damage had a tough, typically orange-pink coenochyme which was firmly attached to the skeleton. Gorgonian necrosis was recorded at 7 out of 13 sites surveyed between on both the north and south coasts of SW England. The disease was noted from May-October at temperatures of 12.1-15.3°C and depths of 6-28 m but was not found at depths of >30 m or on the 99 colonies examined in winter. Repeat observations made of diseased colonies at Knoll Pins, Lundy revealed a progression whereby necrotized tissue was sloughed allowing epibionts to colonize exposed skeleton (Fig. 2B) with remaining tissue taking on a thin, wasted appearance with a grey hue.

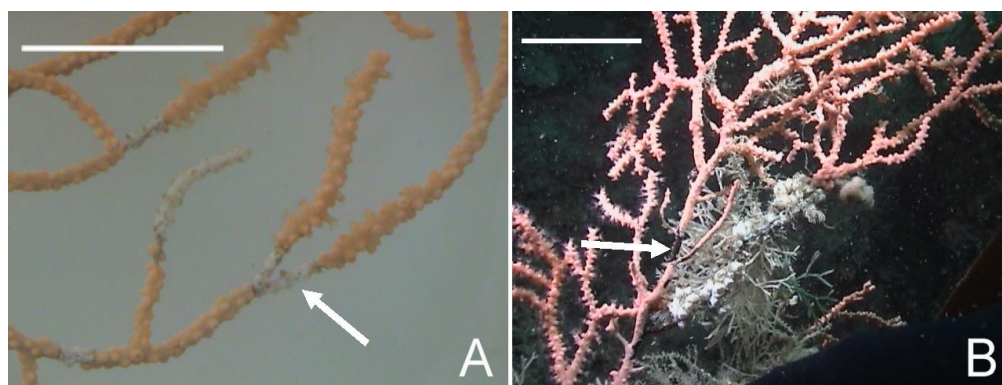


Fig. 2. *Eunicella verrucosa* at 21 m depth at Knoll Pins, Lundy, (A) early onset of coenchyme necrosis (arrow), (B) post-necrotic exposure of gorgonin skeleton (arrow) with fouling community of hydroids, barnacles (*Solidobalanus fallax*) and bryozoans (*Cellaria* sp.). Scale bars = 40 mm.

No fungi were isolated from any of the *Eunicella verrucosa* tissue samples tested, despite using culture media that were selective for fungi. *On MSA media, however, far more bacteria were cultured from diseased than from healthy tissue and most of these were part of the large group of V. splendidus strains. Eunicella verrucosa* clippings maintained at 15°C did not develop tissue necrosis regardless of which mixtures of vibrios they were inoculated with. After 15 days at 20°C necrosis was evident in 60% of samples inoculated with a mixture of vibrios originating from diseased colonies and 80% of samples inoculated with vibrios isolated from healthy specimens.

Conclusions

Corals are highly diverse and widespread in cold-water areas such as high-latitude and deep-sea regions of the oceans (Roberts et al., 2009). Scleractinian and gorgonian corals in particular can form dense stands in cold-water regions, increasing the structural complexity of habitats and thereby contributing strongly to their biodiversity, just as they do on tropical coral reefs. Current threats to cold-water coral communities include trawling (Hall-Spencer et al. 2002) and climate-driven changes in ocean chemistry (Davies et al., 2007), but the effects of disease on such communities are unknown.

An outbreak of *Eunicella verrucosa* disease, first recorded at Lundy marine reserve in 2002, was not an isolated incident but occurred again in the summer months of 2003-2005, both in the Bristol Channel and at six English Channel sites. It seems unlikely that previous outbreaks of *E. verrucosa* disease would have gone unnoticed in recent years at Lundy marine reserve as the gorgonians are monitored by divers, although outbreaks could have been overlooked in other areas. A widespread die-back of *E. verrucosa* that may have been related to disease was recorded during standard trawls of the Marine Biological Association UK in 1924 when it was noted that “a great amount of *Eunicella* brought up in the Plymouth area was dead, many colonies brought in were partially dead, none in such good condition as in the previous July” (Plymouth Marine Fauna, 1954).

The nature of *E. verrucosa* disease was similar to that reported during outbreaks of gorgonian disease in the Caribbean and the Mediterranean (Cerrano et al. 2000; Kim & Harvell 2004). As in those regions, the disease manifested itself as patches of necrosis; the coenchyme became soft and lost structural integrity sloughing away from the gorgonin skeleton and allowing fouling organisms to colonise. Fouling communities were diverse but dominated by hydroids, bryozoans and *Solidobalanus fallax* barnacles, an invasive southern species only recently recorded in SW England (Southward et al., 2004). Low incidences of fouled colonies were found at sites where necrosis was not seen, suggesting that disease might not be the only trigger for fouling communities to colonise gorgonians. However, numbers of unfouled colonies were significantly higher at sites where necrosis had not been seen.

The nearest reports of disease in other gorgonian species are from the NW Mediterranean, with mass mortalities recorded in 1999 during a period of unusually high sea-water temperatures (Cerrano et al. 2005; Linares et al. 2005). Increased incidences of coral disease have also been linked to high temperature events in the tropics which may be attributable to the effects of global warming (Harvell et al. 2002; Rosenberg & Ben Haim, 2002), although nutrient enrichment is also known to increase the severity of tropical coral diseases (Bruno et al., 2003). As with NW Mediterranean outbreaks, we only recorded gorgonian disease in warmer months and in shallow-water populations although maximum *in situ* temperatures recorded around the gorgonians (to 15.4 °C) were not unusual for the area.

The disease outbreaks recorded in SW England have so far not been as devastating to gorgonian populations as the mass mortality events recorded in the Caribbean in 1994 and in the NW Mediterranean in 1999. *Eunicella verrucosa* recruitment was noted in areas affected by disease, despite outbreaks being recorded each summer in 2003-2005, and many of the colonies affected in SW England showed regeneration around damaged parts of affected colonies. There were also numerous sites in the region that showed no evidence of disease outbreak. However, both Linares *et al.* (2005) and Cerrano *et al.* (2005) noted that sublethal effects of gorgonian disease can have long-term effects on regional benthic ecology. Effects can be long-lasting because gorgonian corals are typically slow-growing, high-biomass organisms that contribute to the three dimensional complexity of the benthic habitat. Clearly, our first observations of localized disease outbreaks in *Eunicella verrucosa* indicate that coral diseases may be widespread in cold-water regions of the planet and that vigilance is required to assess their causes and effects.

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Environmental assessment of a site in Sv. Marina (Istria, Croatia): Examination of morphological and chemical parameters which indicate the occurrences of certain species.

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Abstract. Within the scientific diving project of the TU Bergakademie Freiberg an environmental assessment study was conducted showing the complex interacting of different ecological habitats. The exceptionally situation of the Adriatic Sea is due to its submarine karst discharge and the anthropogenic influence factors (tourism, coastal industries, lack of appropriate waste water treatment). Biodiversity, water chemistry and accumulation of trace elements were investigated and evaluated.

Aim of investigation

Aim of the investigation was an estimation of the anthropogenic influence on a complex marine ecosystem. Geological and morphological characterization gives an overview about the site. Biological mapping was used to define indicator species which reflect site-specific conditions of each area. Hydro- and geochemical investigations of environmental compartments (water, sediment, and biota) was used to estimate the extent of anthropogenic impact.

Investigation area

During 2003 to 2005 submarine sites at the eastern peninsula Sv. Marina on the Istrian peninsula (Croatia) were investigated during 3 diving campaigns. The area of interest is characterized by steep slopes and steep faces up to a depth of 60 m. The geology is determined mainly by karstified limestones. Fresh water from the mainland, as well as seawater can move through submarine karst features. Coarse debris occurs directly at the coastline followed by fine sediments from 3 m water depth and solid rocks (steep face) in the deeper part of the coastal area.

A waste water pipe draining the sanitary waste water of the camping area sitting on the peninsula was of special concern.

Methods

Mapping

Primarily, the investigation started with the 3 transects right from the waste water pipe. Transects were laid out by using ropes with markers in a distance of one meter. The width of a transect was defined by arm's length to both sides. The location of each transect was determined with a GPS floating at the water surface. Weights were used to prevent buoyancy or drift of the rope. Each transect was divided in zones by the type of substrate, slope, etc.

The coverage of each individual species was determined in the various transect zones by using the Braun-Blanquet-index.

Sampling

Water sampling was done for reading the in-situ-parameters pH, electrical conductivity, redox potential, and O₂, determination of NO₃⁻, NO₂⁻, NH₄⁺, Fe²⁺, Fe_{total}, and PO₄³⁻ by means of photometry in a field laboratory. Furthermore samples were taken for total inorganic carbon, major anions and cations (ion chromatography) and trace elements (ICP-MS).

Additionally, sediment and biota were sampled in order to estimate the anthropogenic influence caused by waste water discharge. Determination of trace elements in sediment samples was performed using microwave extraction and ICP-MS. Trace elements in biota samples were determined as well by ICP-MS after pressurized liquid extraction.

Analytics

The cation determinations were conducted with a LC-System L 6000 (Merck-Hitachi) using an L 6000 A Interface, a L 5025 Column heater, a L 6200 A HPLC-Pump, and a L 3720 conductivity detector. A RT LiChrosil[®] IC CA 2 from Merck (length 125 mm, internal diameter 4.6 mm, Sorbens: spherical silica gel with polymer cation exchange phase, particle size 5 µm) was used as analytical column. A RT LiChrosil[®] IC CA 2 from Merck (length 10 mm, internal diameter 4.6 mm) served as a pre column.

An Eppendorf Biotronik instrument IC 2001 in combination with a Shimadzu Liquid Chromatograph LC-10 As was available for the determination of anions. The suppressor column used was a FGC BTS AG-P. Measurements of trace elements in sediment and biota samples were carried out using the ICP-MS-instrument ELAN DRC-E (Perkin Elmer, Sciex Inc.).

Main Results

Morphological characterization

An obvious correlation of the investigated transects (I'03, I'04 and II'04) by morphological characteristics and their geological location was found. Clearly visible are the individual layers and their striking. Depending on their exposure also differences in erosion and crevasse formation were noted (e.g. zone B on transect I'04 is due to their proximity to an existing fault zone more fractured than transect II'04).

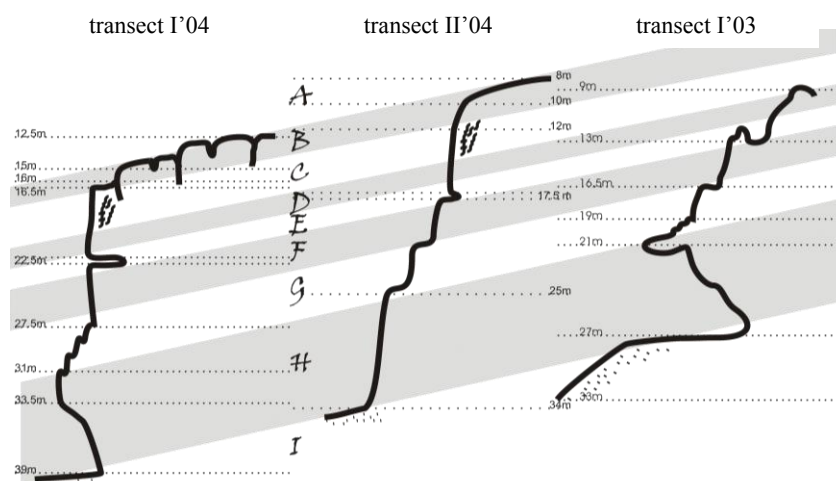


Fig.1. Correlation of transect I'04, II'04 and I'03 by morphological and geological parameters. Grey bars indicate similar zones.

The upper surface of transect I'04 and II'04 is relatively flat, divided by sinks and fractures in various plateaus which are covered by sediments (A). B describes the first section of steep face, which is strongly

fissured and features many caves and niches. These karst features emerges particularly strongly in Zone C. The steep face is divided by a large cave (E). After continuous decrease of the steep slope a staircase-shaped slope follows. These features are caused by artifacts of limestone. Zone H runs at transect II'04 almost perpendicular to the bottom, while at transect II'04 an overhang exists. The steep face is limited by a shallow sloping surface with few centimeters of sediment cover.

Diversity of species

Depending on morphological structures different species could be identified.

Figure 2 illustrates the most common species in the zones of the investigated transects.

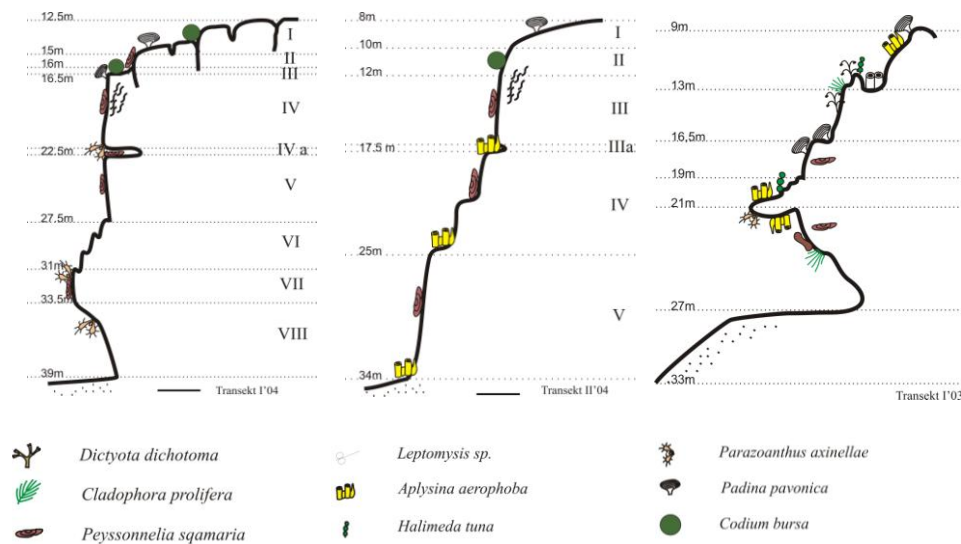


Fig.2. Biodiversity on transect I'04, II'04, and I'03

During mapping one species was characterized as shade-preferring species: *Peyssonnelia squamaria*. It was found in shade to shady areas, as well as in caves. However, a preference for hard-ground areas was observed. Nearly no existence on sediment-covered areas occurred (e.g. zone A and B). This can be reasoned by 2 factors: light and sediment.

Another inhabitant of hard ground was *Codium bursa*. In contrary to *Peyssonnelia squamaria*, *Codium bursa* prefers light habitats. It was found mainly in the upper hard-ground zones A and B. In contrast, *Parazoanthus axinellae* and *Leptopsammia pruvoti* only exist in cave entrances, edges of overhangs or cave ceilings (zones E and H). *Padina pavonica* was found up to a depth of 16.5 m (zone B). Due to few sediment areas only sporadic incidence was seen for *Cerianthus membranaceus*. Only one zone was characterized as hard-ground with sediment coverage (zone G). *Derbesia lamourouxi* seems to be a species adapted on a variety of environmental conditions. Incidence was observed for light and shaded areas, hard-ground with and without sediment covering.

Anthropogenic influence caused by waste water discharge

Water chemistry

Electrical conductivity was used as an indicator for natural freshwater discharges, as well as for anthropogenic impact.

Conductivity profiles at various locations within the study area Sv. Marina are shown in Fig. 3. Locations from left to right are waste water pipe, dive-site "Felsentor", dive site "Porta Longa" (a bay in the vicinity of Sv. Marina). Different graphs represent measurements on different days.

The increase in conductivity with increasing water depth presents the influence of near-surface fresh karst water and the special position of the northern Adriatic Sea as an isolated basin with little water exchange and thus the highest salt concentration of the entire Mediterranean Sea.

In the left (light grey graph) the influence of the waste water pipe can be identified by the abrupt decrease of electrical conductivity in a depth of 45 m.

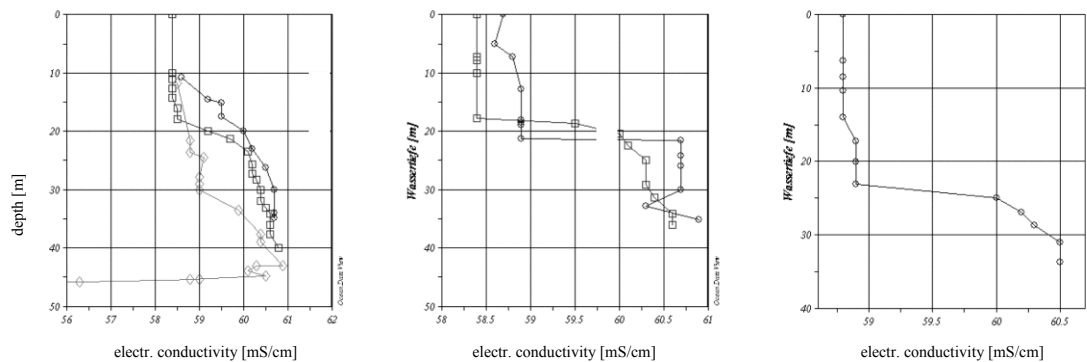


Fig.3. Profiles of electrical conductivity. From left to right: waste water pipe, “Felsentor”, neighboring bay “Porta Longa”

Accumulation of trace metals

Sediment samples were investigated with respect to arsenic, lead, cadmium, mercury, chromium, copper, and zinc. Table 1 shows main results of trace metal contents in comparison with literature.

Table 1. Trace metal content in sediment samples.

Element	Concentration [$\mu\text{g/g DM}$]	Caves [$\mu\text{g/g DM}$]	Harbour area [$\mu\text{g/g DM}$]	Literature*, ** [$\mu\text{g/g DM}$]
Arsenic	6 – 31	6 - 11	28.3 (median)	5 - 15
Lead	14 - 48	18 - 48	14 - 20	5 - 30
Cadmium	0.13 – 0.28	0.15 - 0.28	0.13 – 0.20	0.1 – 0.6
Mercury	< 0.01 – 0.29	0.12 – 0.26	0.04 - 0.11	0.2
Chromium	20.5 – 67.8	30 – 51	20.5 – 67.8	50 - 100
Copper	10.5 – 26.9	17 – 26.9	10.5 - 18	10 - 50
Zinc	19.8 – 69.6	52 – 69.6	20 – 46	-

* Neff 2002

** Salomons & Förstner 1984

No significant anthropogenic influence on the sediment was observed. Concentrations of investigated trace metals were in a range as described in literature.

Samples of *Aplysina aerophoba*, *Peyssonnelia squamaria*, and *Padina pavonica* were investigated with respect to arsenic, lead, cadmium, chromium, copper, and zinc.

Figure 4 shows the total copper content in the investigated algal and sponge samples. Elevated levels in tissues were seen for *Aplysina aerophoba* (light grey). As cited in literature, macro-algae show lower copper concentrations than Periphyton (NEFF 2002). This can be reasoned by lime incrustation processes and the associated contamination with sediment particles. Peryphyton algae are represented by *Peyssonnelia squamaria* (black).

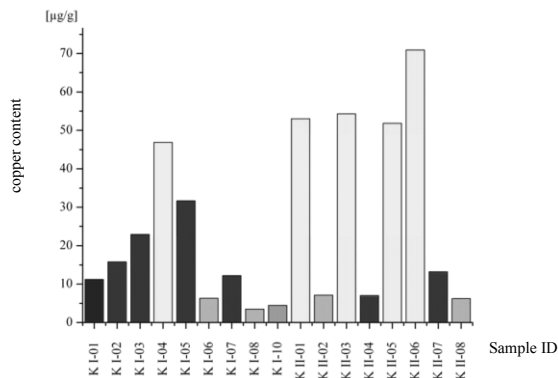


Fig.4. Total copper content of biota samples.

The following trend can be noted: *Aplysina aerophoba* as main representative of sponges accumulated arsenic, copper, and zinc in significant amounts. Smaller accumulation rates were found for *Peyssonnelia squamaria*. The Brown algae *Padina pavonica* didn't show significant concentrations of trace elements at all.

Conclusions

Due to the geological situation (karst) and the local marine regime Sv. Marina represents a special locality in the Adriatic Mediterranean. It showed a high biodiversity.

The random survey and the high degree of dilution made it difficult to quantify the influence of the waste water pipe. The local flow regime seems to be of great importance; slow water currents and calm areas (e.g. caves) play an important role.

The trace element content of the sediment cores showed little variations compared to uncontaminated sediments from the literature.

Species differences in the accumulation of trace elements in algae and sponge samples were detectable and comparable with literature data.

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Hydrothermal fluids vented at shallow depths at the Aeolian islands: relationships with volcanic and geothermal systems.

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Abstract. Scuba diving investigations carried out over the last two decades at the Aeolian islands revealed the existence of submarine magmatic and late-magmatic hydrothermalism at all the islands, despite the absence of on-shore activity at some of the islands. The results gained by diving activities provided useful information to evaluate the volcanic and geothermal activity and to manage the volcanic crisis occurred on November 2002 off the island of Panarea.

Introduction

The Aeolian islands are a ring-like volcanic arc consisting of 7 islands and 10 seamounts and constitutes about 200 km of the inner side of the Peloritani-Calabrian orogenic belt (e.g. Calanchi et al., 2002 and references therein). The dynamics of the arc is controlled by still active faults (Lanzafame and Rossi, 1984). All the Aeolian islands are characterized by a subduction-related magmatism that has taken place from 400 ka to present (Calanchi et al., 2002), and is still active along the arc. One of the main regional faults moving from the main land (NW-SE strike slip fault; Fig. 1) splits the arc into two sectors with different levels of volcanic activity. The eastern sector (including Vulcano, Lipari, Panarea and Stromboli) is characterized by active volcanism and deep-focus earthquakes depicting a narrow NW-dipping Benioff-Wadati plane. Contrastingly, the western sector, including the islands of Salina, Alicudi and Filicudi, is seismically quiet compared to the eastern side and the volcanism is considered as extinct since a long time. Scuba diving investigations carried out from middle 80's, had shown that despite the absence of on shore volcanic manifestations, submarine hydrothermal activity is recognizable at shallow depth around all the Aeolian islands related either to volcanic and geothermal activity. The investigations had been carried out by scuba diving up to a depth of 40 meters and many different methods have been set over the time to allow measurements, sample collection and prospection in such a new working environment (e.g. Italiano and Nuccio, 1991; Gugliandolo et al., 1999; Caracausi et al, 2005a, b).

This paper accounts for the chemical and isotopic composition of the venting gases with the aim to evaluate the existence of geothermal bodies and relationships with the volcanic activity.

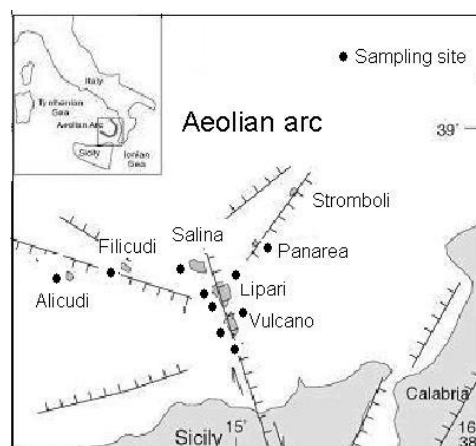


Fig.1. Sketch map of the Aeolian arc and the main tectonic lines. The main fault splitting the Aeolian islands in two sectors is shown. Shallow vents have been found around all the islands; the black dots show the sampling sites.

Gas geochemistry

The gases vented at the sea bottom had been collected driving attention to the uncommon sampling activity that was carried out in diving. A reasonable equilibrium was attained to combine the necessity to take good samples, to work in a safe way, to work even in bad weather conditions (when necessary for civil protection purposes), to speed up the sample collection as much as possible, to carry as lighter sampling equipments as possible. As such, most of the samples were made of gases, collected in 30-50ml glass bottles by an inverted funnel (Fig. 2). The sampling equipment was filled by marine water to avoid any atmospheric contamination, then the funnel was placed over the venting gas and the sample quickly collected by water displacement.

The collected samples were analyzed in the laboratory by gas-chromatography and mass spectrometry: the former to establish the chemical composition, the latter to determine the isotopic signature of carbon and helium.

Results from samples collected over a 20years time span are listed in Tables 1 (chemical composition) and 2 (isotopic composition of helium and carbon). The chemical composition of the sampled gases shows how all of them are CO₂-dominated with low oxygen amounts and reactive gases (H₂, CO, CH₄ and H₂S) with concentrations ranging from a few ppm to some mole percent (Table 1). Sometimes significant N₂ amount are detectable together with high helium contents (see Panarea 1, Table 1). Samples having low CO₂ content besides relevant N₂, O₂ and He amounts are the consequence of CO₂ dissolution in sea-water due to gas-water interactions (GWI) occurred before the sample collection.



Fig.2. Venting gases sampled off the island of Panarea. The pyrex bottle was filled by sea-water before diving. The gas coming from the inverted funnel displaced the water allowing the collection of a gas sample without atmospheric contamination and with a pressure equilibrated at the sampling depth (overpressure).

The high CO₂ solubility (878 ml/l, T=20°C, P=1bar) may, in fact, decrease the CO₂ content in the venting gases (e.g. 21.5%, sample Panarea 1), thus increasing the concentrations of the less soluble species (e.g. He 8 ml/l, CO 23 ml/l and CH₄ 33.8 ml/l) in the gas mixture. Such a process might occur at any level, however because of the slow water circulation in deep sediments, CO₂ is able to saturate the circulating sea-water. As a consequence it is favored the hypothesis of an occurrence of relevant GWI during the final path where, due to the circulation of air-saturated sea-water (ASSW) in the shallow sediments. The atmospheric-derived oxygen is probably stripped from the sea-water at that level.

The isotopic composition of carbon displays a small range of values while helium isotopes are in the range of $4.1 < R_{a_c} < 7$ (Table 2). Table 2 lists also the ⁴He/²⁰Ne ratios useful to take indications on the atmospheric contamination of the venting gases. All the recorded ratios are above the atmosphere (⁴He/²⁰Ne=0.318) and, considering the occurrence of gas-water interactions (CO₂ dissolution), it has to be observed that the ⁴He/²⁰Ne is also well above the ASSW ratio of 0.285. The ⁴He/²⁰Ne ratio is used to calculate the pristine ³He/⁴He ratio assuming that all the neon is of atmospheric origin. The calculated values are listed in Table 2 in the R/R_{a_c} column.

Hydrothermal fluids and submarine geothermal systems

Despite the gases come from both active and extinct Volcanoes, their chemical composition is similar. Contrastingly the isotope composition of helium shows a large heterogeneity with values spanning from 4.1 to 7 Ra_c. The highest isotopic ratios are surprisingly measured at the extinct volcanic islands in the western sector, with lower values detected in venting gases from active volcanoes (e.g. Vulcano and Panarea, Table 2).

Table 1. Chemical composition of the submarine venting gases of the Aeolian islands. Concentrations in mole%. The estimated equilibrium temperature is reported (last column). See text for details.

Site	He	H ₂	O ₂	N ₂	CO	CH ₄	CO ₂	H ₂ S	T°C
Alicudi	8,8E-04	1,0E-04	0,7	2,8	7,1E-05	2,1E-03	96,4	1,5E-02	214
Alicudi	8,3E-04	1,0E-03	6,4	23,3	6,3E-04	5,6E-04	70,3	n.d.	314
Alicudi	6,9E-04	1,0E-04	0,3	2,1	6,1E-05	1,6E-03	97,5	1,5E-02	211
Filicudi	2,0E-02	2,1E-03	2,4	43,9	2,2E-04	1,2E-02	53,7	1,1E-02	250
Salina 1	1,0E-04	1,0E-04	0,6	1,1	4,0E-05	1,7E-04	98,3	1,0E-02	215
Salina 2	8,6E-04	1,0E-04	1,1	2,3	7,1E-05	2,3E-03	96,5	1,0E-02	213
Vulcano 1	n.d.	1,7E-03	n.d.	1,8	3,5E-04	1,1E-01	96,1	2,0E+00	233
Vulcano 2	2,1E-04	9,9E-05	1,1	3,0	4,5E-04	3,2E-04	95,9	9,9E-03	296
Vulcano 3	2,4E-03	1,6E-03	0,8	20,1	1,7E-03	1,1E-02	79,1	n.d.	322
Vulcano 4	2,1E-04	1,1E-02	0,1	1,3	3,3E-04	1,5E-03	98,6	n.d.	268
Lipari 1	2,1E-03	0,0E+00	2,3	26,6	1,5E-03	2,4E-02	71,1	n.d.	311
Lipari 2	1,6E-03	3,0E-03	0,2	35,0	4,6E-05	1,8E-02	64,8	9,3E-03	194
Lipari 3	6,8E-03	4,1E-03	n.d.	31,6	5,0E-05	4,7E-02	68,4	1,0E-02	189
Panarea 1	7,7E-02	3,1E-03	13,4	63,4	3,7E-03	1,6E+00	21,5	n.d.	345
Panarea 1	9,0E-04	4,7E-04	0,2	2,4	2,6E-04	1,2E-02	97,4	n.d.	241
Panarea 2	1,3E-03	6,0E-04	n.d.	2,0	1,7E-03	6,6E-04	97,0	1,0E+00	347
Basiluzzo	7,6E-04	1,0E-04	n.d.	2,5	4,1E-05	3,6E-04	97,5	n.d.	210

The explanation of such a difference is not related to the volcanic activity at all, but to the parent mantle that in the western side looks to be less contaminated compared to the eastern side. Crustal contamination has been invoked by several authors (Ellam et al., 1989) as the main factor that caused the dramatic ³He/⁴He decrease. Results of helium isotope composition on gases extracted by crushing on solids from the Aeolian islands (Martelli et al., 2008) support this hypothesis. Although the parent mantle produced magmas with different isotopic signature, the gas phase looks similar (Table 1). To explain the results of the chemical analyses it is proposed that similar deep boundary conditions (pressure, temperature, oxidation level) act as buffers for the chemical composition of the venting gases. With the aim of investigating their origin, estimations of the deep equilibration conditions have been carried out. The reactive compounds detected in the sampled gases, largely used for geothermometric and geobarometric considerations of hydrothermal fluids (Giggenbach 1980; Italiano and Nuccio 1991 and references therein), are here used in a system based on the CH₄-CO-CO₂ contents assuming the presence of a boiling aqueous solution.

The equilibrium constants of the adopted reactions are a function of temperature and oxygen fugacity, being the latter buffered by the mineral assemblage of the host rocks (see Italiano and Nuccio, 1991 for details). Due to the similarity in the chemical composition of the gases vented at all the islands, the theoretical model developed to interpret the chemical composition of the gases released at Panarea during the last volcanic crisis (Caracausi et al., 2005a), is here applied. The results of the estimated equilibration temperatures and pressures are listed in Table 1. The distribution of the samples on Figure 3 shows that geothermal boiling systems are detectable at all the islands with temperatures up to 350°C. The adopted geothermobarometric system is more sensitive to the contents of CO and CH₄ than that of CO₂, implying that although GWI induce modifications in the chemical composition, the estimated equilibrium temperatures do not change very much for variations of the CO₂ content in the range of several volume percent, thus, whether or not the gaseous mixture underwent GWI. Moreover, the slow reaction kinetics of CO and CH₄ allow them to keep the deep equilibrium conditions during uprising and the similar solubility (see above) does not alter their abundance ratios.

Table 2. Isotopic composition of helium and carbon in submarine venting gases of the Aeolian islands. All the $^3\text{He}/^4\text{He}$ ratios (R) have been normalized to the atmosphere ($^3\text{He}/^4\text{He} = 1.39 \times 10^{-6} = \text{Ra}$) and corrected for the effects of atmospheric contamination (R/Ra_c) using $(\text{R/Ra}_c) = \{(R/\text{Ra})X-1\}/\{X-1\}$ where X is the air- He/Ne ratio (Hilton et al, 1998). Carbon isotopic ratios expressed in ‰ versus the PDB international standard. n.d. = not detected.

Site	$^3\text{He}/^4\text{He}$	He/Ne	R/Ra_c	$\delta^{13}\text{C}$
Alicudi	$8,9 \times 10^{-6}$	43,4	$6,4 \pm 0,0036$	-3,7
Alicudi	$8,9 \times 10^{-6}$	36,1	$6,4 \pm 0,0056$	n.d.
Alicudi	$9,7 \times 10^{-6}$	49,2	$7,0 \pm 0,0060$	n.d.
Filicudi	$6,8 \times 10^{-6}$	31,0	$4,9 \pm 0,0036$	n.d.
Salina 1	$4,9 \times 10^{-6}$	1,8	$4,1 \pm 0,143$	n.d.
Salina 2	$8,2 \times 10^{-6}$	32,2	$5,9 \pm 0,041$	-2,2
Vulcano 1	$6,5 \times 10^{-6}$	26,5	$4,7 \pm 0,069$	-1,2
Vulcano 2	$6,2 \times 10^{-6}$	2,7	$4,9 \pm 0,040$	n.d.
Vulcano 3	$5,8 \times 10^{-6}$	19,3	$4,2 \pm 0,049$	n.d.
Vulcano 3	$5,8 \times 10^{-6}$	14,2	$4,2 \pm 0,037$	n.d.
Vulcano 4	$5,7 \times 10^{-6}$	5,0	$4,3 \pm 0,072$	n.d.
Lipari 1	$6,6 \times 10^{-6}$	9,8	$4,8 \pm 0,111$	-3,0
Lipari 2	$5,5 \times 10^{-6}$	3,1	$4,3 \pm 0,046$	n.d.
Panarea 1	$6,1 \times 10^{-6}$	362,0	$4,4 \pm 0,041$	-3,2
Panarea 1	$5,7 \times 10^{-6}$	19,7	$4,1 \pm 0,065$	n.d.
Panarea 1	$6,1 \times 10^{-6}$	362,0	$4,4 \pm 0,041$	n.d.
Panarea 2	$5,7 \times 10^{-6}$	19,7	$4,1 \pm 0,065$	n.d.
Basiluzzo	$5,9 \times 10^{-6}$	76,2	$4,3 \pm 0,035$	n.d.

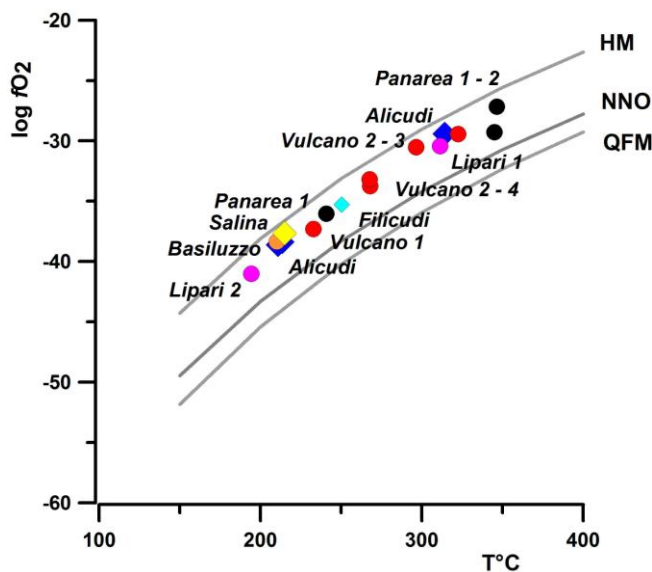


Fig.3. Temperature vs oxygen fugacity (expressed as $\text{Log } f\text{O}_2$) graph. The solid buffers quartz–fayalite–magnetite, nickel–nickel oxide and hematite–magnetite are plotted as reference. The equilibrium conditions estimated by the $\text{CH}_4\text{-CO-CO}_2\text{-H}_2\text{O}$ system are shown. Circular symbols refer to gases from the eastern sector (pink=Lipari; black=Panarea; orange=Basiluzzo; red=Vulcano); squared marks show samples from the western sector of the Aeolian arc (blue=Alicudi; yellow=Salina; light blue=Filicudi)

Hydrothermal fluids and submarine volcanic activity

The first application of scientific diving for active volcano monitoring purposes was carried out at Panarea during the last volcanic crisis (2002-2003). As a response to a magmatic input, fluids normally released from hydrothermal systems, exhibit an abrupt change in both their geochemical features and releasing rate. Since the occurred event was a low-energy submarine explosion, followed by a huge release of

volcanic gases (Caracausi et al., 2005b), it was clear that the capability of a system to discharge huge amounts of fluids throughout fractured rocks becomes a key condition in determining the occurrence of violent explosive events or smoother gas emission. Gas output evaluations allowed to estimate the energy for crater formation by considering the vertical shift of the involved rock volume. Measurements of gas output inside the crater at the crisis onset lack as the strong water flux due to the massive degassing rate, caused serious troubles for divers' safety and a gas output estimation for that time was based on a motion picture of the main emission, resulting about two orders of magnitude higher (about $10^6 \text{ m}^3 \text{ d}^{-1}$) than the first direct measurements carried out two weeks later (ranging from 10^4 to 10^5 moles per day).

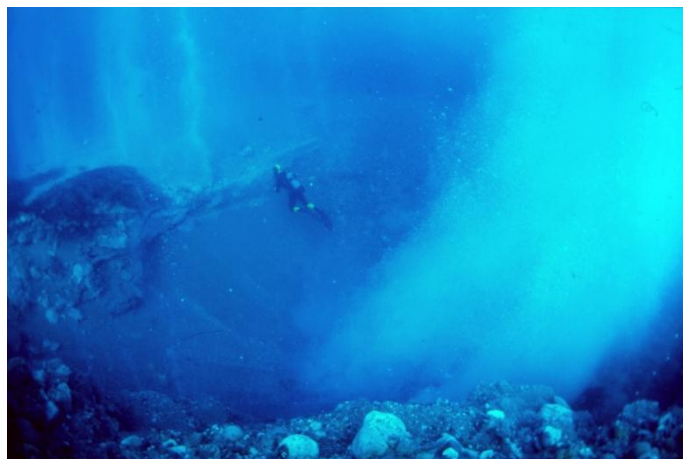


Fig.4. Picture of the huge degassing from the crater formed on November 2th, 2002 off Panarea island. The crater was 8 meters deep by 20 meters in diameter and caused by low-energy explosion occurred at shallow depth. Only diving activity allowed to recover scientific information useful to constrain the occurred volcanic event

Combining the results of the output estimations with the interpretation of the geochemical features of the released gases interpreted by the above mentioned theoretical model (Caracausi et al., 2005a), it was clear that having a so high gas output, a few seconds had been enough to perform the required work of crater formation.

Considering the capability of the geothermal system of Panarea to dramatically transfer energy and fluids towards the surface and the suggested tectonic link with the active volcano of Stromboli (Heinicke et al., 2009) able to transfer magmatic volatiles, the possibility of future crises involving energetic phreatic and/or phreato-magmatic events has to be taken into account.

The diving activity, that gave insight on the existence of geothermal bodies along the whole Aeolian islands, is also the only possible approach to evaluate the volcanic activity of that system and to provide tools to minimize the volcanic risk.

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A brief review of microbial geochemistry in the shallow-sea hydrothermal system of Vulcano Island (Italy)

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Abstract. The shallow-sea vents, heated sediments and geothermal wells at Vulcano are home to a diverse composition of heat-loving Archaea and Bacteria. Their potential energy sources include hundreds of organic and inorganic redox reactions that yield up to ~120 kJ per mole electrons transferred. The dominant biogeochemical processes in the anoxic sediments appear to be fermentation and sulfate reduction.

Introduction

Deep-sea hydrothermal systems with black smoker sulfide chimneys and photogenic macrofauna have garnered much scientific attention since their discovery in 1977 on the Galapagos rift (Corliss et al. 1979). That same year, Archaea (formerly Archaeobacteria)—see Table 1—were identified as the third domain of life, joining the Bacteria (formerly Eubacteria) and Eukarya (Woese and Fox 1977). Perhaps because many of the cultured Archaea are very high temperature organisms—hyperthermophiles—their discoveries and geochemical investigations of hydrothermal ecosystems have been invariably linked. It should be noted, however, that shallow-sea hydrothermal systems, and not their deep-sea counterparts, have yielded the majority of the cultured hyperthermophiles known to date. In fact, one system, the vent waters and heated sediments at Vulcano (Aeolian Islands, Italy) is the ‘type locality’ for 10 of the circa 25 known genera that include hyperthermophiles (Amend et al. 2003a; Deckert et al. 1998; Fiala and Stetter 1986; Fischer et al. 1983; Hafenbradl et al. 1996; Huber et al. 1986; Stetter 1982; Stetter et al. 1987; Zillig et al. 1983). These genera belong to the Euryarchaeota (*Thermococcus*, *Pyrococcus*, *Palaeococcus*, *Archaeoglobus*, *Ferroglobus*), the Crenarchaeota (*Pyrodictium*, *Staphylothermus*, *Thermodiscus*) and the Bacteria (*Aquifex*, *Thermotoga*).

Table 1. Glossary of terms

Aerobe: an organism that uses O ₂ as its terminal electron acceptor.
Anaerobe: an organism that uses oxidants other than O ₂ as its terminal electron acceptor.
Archaea: one of the three phylogenetic domains, in addition to the Bacteria and Eukarya.
Autotroph: an organism that uses CO ₂ (or CO) as its carbon source.
Bacteria: one of the three phylogenetic domains, in addition to the Archaea and Eukarya.
Chemotroph: an organism that gets energy from chemical disequilibrium, not solar energy.
Crenarchaeota: one of the two main phyla (or kingdoms) of Archaea.
Eukarya: one of the three phylogenetic domains, in addition to the Archaea and Bacteria.
Euryarchaeota: one of the two main phyla (or kingdoms) of Archaea.
Fermentation: the disproportionation of organic compounds, usually producing CO ₂ , H ₂ , and small-chain fatty acids or alcohols; no TEAs are involved in the redox reactions.
Heterotroph: an organism that uses organic compounds as its carbon source.
Hyperthermophile: a thermophile organism with an optimum growth temperature ≥80°C.
Lithotroph: an organism that gets its energy from inorganic redox reactions.
Operational taxonomic unit (OTU): definition or name of the taxa included in phylogenetic analyses; often equated with ‘species’.
Phylogeny: the ordering of species into higher taxa and the evolutionary relatedness among various groups of organisms.
Physiology: the mechanical, physical, and biochemical functions of living organisms.
Respiration: the oxidation (respiration) with a TEA of reduced compounds.

Terminal electron acceptor (TEA): the oxidant in a net respiration reaction.

Thermophile: an organism with an optimum growth temperature $\geq 50^\circ\text{C}$.

16S rRNA gene: a segment of DNA (gene) that encodes for a part of the ribosomal RNA; commonly used for constructing phylogenetic trees.

Pyrodictium occultum, isolated from heated marine sediments on Vulcano, was the first organism in pure culture with an optimum growth temperature above 100°C (Stetter 1982). It is an obligate chemolithoautotroph that gains metabolic energy from the reduction of elemental sulfur (S^0) and dihydrogen (H_2). The other Vulcano hyperthermophiles include aerobes and anaerobes, heterotrophs and autotrophs, fermenters and respirers. In laboratory cultivations, the autotrophs can fix CO_2 into biomass by several different biochemical pathways. Among the Vulcano heterotrophs, on the other hand, some metabolize simple volatile fatty acids, and others require complex proteinaceous organic matter. The anaerobic respirers shuttle electrons to nitrate (NO_3^-), sulfate (SO_4^{2-}), ferric iron phases (Fe^{III}), and other terminal electron acceptors (TEAs). *Aquifex*—the water maker—catalyzes the ‘knallgas’ reaction ($\text{H}_2 + \frac{1}{2}\text{O}_2 \rightarrow \text{H}_2\text{O}$). Some of the organisms thrive at circumneutral pH, but others are acid-loving (acidophiles) (Simmons and Norris 2002). Numerous laboratory characterization studies have hinted at the physiologic variability among the hyperthermophiles, especially the archaea, and these studies have set the stage for detailed investigations of the chemo-physical parameters that define the ecosystem, the phylogenetic and metabolic diversity afforded by the environment, and the in situ biogeochemical processes that dominate.

It is no surprise that the archaeal and bacterial diversity in the thermal environment of Vulcano far exceeds that represented by the cultured organisms mentioned above. DNA extracted from heated sediments and from thermal fluids points to phylogenetic groups that consist almost entirely of uncultured Archaea and Bacteria (Rogers 2006). For example, in a geothermal well (Pozzo Istmo), more than a dozen archaeal lineages were identified that have no cultivated representatives (Rogers and Amend 2005). In this brief communication, I will summarize and synthesize what has been learned over the past decade or so regarding the geochemical energy sources for microbial metabolism, the archaeal and bacterial composition in hydrothermal fluids and heated sediments, and the dominant biogeochemical processes in the high temperature ecosystem at Vulcano.

Redox energetics

All life requires energy, which is obtained from electron transfer (redox) processes. The dominant net metabolisms near the surface of the Earth are oxygenic photosynthesis—driven by an input of solar radiation energy—and aerobic respiration of a wide range of organic and inorganic electron donors. However, photosynthesis is limited to temperatures below $\sim 73^\circ\text{C}$ (Blankenship 2002), and TEAs other than O_2 become increasingly important in the subsurface and in high temperature environments. Hydrothermal systems, including, and perhaps in particular, the shallow-sea variety, provide a tremendous array of redox couples that can serve as metabolic energy sources for high temperature microorganisms. To evaluate the energy yields from net metabolic reactions, Gibbs energies (ΔG_r) were calculated in accord with

$$\Delta G_r = \Delta G_r^\circ + RT \ln Q_r,$$

where ΔG_r° denotes the *standard* Gibbs energy of reaction, which is evaluated from *standard* Gibbs energies of the individual reactant and product species (ΔG_i°), R and T are the gas constant and temperature in Kelvin, respectively, and Q_r stands for the activity product of the reaction. Values of ΔG_i° as a function of temperature and pressure were generated with established equations of state (see (Amend and Shock 2001) and references therein). Evaluating Q_r requires activities or fugacities of the reactant and product species at the environmental conditions of interest, which, in turn, requires corresponding compositional data. Such data from 10 shallow-sea vents, heated sediment seeps, and geothermal wells at Vulcano were incorporated into a thermodynamic framework of redox energetics (Amend et al. 2004; Amend et al. 2003b; Rogers and Amend 2006).

In Table 2, 90 inorganic redox reactions are given, and their values of ΔG_r at the Vulcano environmental conditions are plotted in Fig. 1. These reactions are potential net metabolisms for chemolithoautotrophs, but only a relatively small sub-set of these reactions are known to be catalyzed by Vulcano hyperthermophiles. These include, for example, Reac. 8 (by *Aquifex*), Reac. 65 (by *Pyrodictium*), and Reac. 69 (by *Arc-haeoglobus*). Note in Fig. 1 that these inorganic redox reactions yield a wide range of energies from ~ 0 to ~ 120 kJ per mole of electrons transferred. It should be pointed out that while there is a general correlation between energy-yield and TEA (O_2 before NO_3^- before Fe^{III} before $\text{SO}_4^{2-}/\text{S}^0$ before CO_2), there also are numerous exceptions and a significant amount of overlap. For example, the top 3 and 5 of the top 6 energy yielding inorganic reactions at Vulcano feature NO_2^- and not O_2 as the TEA. It is hypothesized that some of the identified but uncultured Archaea and Bacteria in the Vulcano hydrothermal ecosystem may obtain their

cellular energy from some of the exergonic redox reactions in Table 2 and Fig. 1 that, to date, have not been attributed to microbial net metabolisms.

Table 2. Inorganic redox reactions that represent putative net metabolisms for chemolithoautotrophs at Vulcano. The reactions are listed in order of decreasing energy yield (increasing value of ΔG_r) as shown in Fig. 1. All species are aqueous, except S^0 and Mt, which represent elemental sulfur and magnetite, respectively

1	$2NO_2^- + 3CO + 2H^+ = N_2 + 3CO_2 + H_2O$	46	$CO + Mt + 6H^+ = CO_2 + 3Fe^{2+} + 3H_2O$
2	$2NO_2^- + 3H_2 + 2H^+ = N_2 + 4H_2O$	47	$NO_3^- + 4H_2S + 2H^+ = NH_4^+ + 4S^0 + 3H_2O$
3	$8NO_2^- + 3CH_4 + 8H^+ = 4N_2 + 3CO_2 + 10H_2O$	48	$4NO_2^- + 3H_2S + 2H^+ + 4H_2O = 4NH_4^+ + 3SO_4^{2-}$
4	$CO + 0.5O_2 = CO_2$	49	$Mt + H_2 + 6H^+ = 3Fe^{2+} + 4H_2O$
5	$NO_2^- + NH_4^+ = N_2$	50	$NO_2^- + 3H_2S + 2H^+ = NH_4^+ + 3S^0 + 2H_2O$
6	$2NO_2^- + CH_4 + 2H^+ = N_2 + CO + 3H_2O$	51	$NH_4^+ + 1.5O_2 = NO_2^- + 2H^+ + H_2O$
7	$2NO_3^- + 5CO + 2H^+ = N_2 + 5CO_2 + H_2O$	52	$CH_4 + 4Mt + 24H^+ = CO_2 + 12Fe^{2+} + 24H^+$
8	$H_2 + 0.5O_2 = H_2O$	53	$2NH_4^+ + 3Mt + 16H^+ = N_2 + 9Fe^{2+} + 12H_2O$
9	$2NO_2^- + S^0 = N_2 + SO_4^{2-}$	54	$NH_4^+ + 2O_2 = NO_3^- + 2H^+ + H_2O$
10	$8NO_2^- + 3H_2S + 2H^+ = 4N_2 + 3SO_4^{2-} + 4H_2O$	55	$CH_4 + 3Mt + 18H^+ = CO + 9Fe^{2+} + 11H_2O$
11	$2NO_3^- + 5H_2 + 2H^+ = N_2 + 6H_2O$	56	$5NO_2^- + 2H^+ = 3NO_3^- + N_2 + H_2O$
12	$2NO_2^- + 3H_2S + 2H^+ = N_2 + 3S^0 + 4H_2O$	57	$S^0 + 3Mt + 16H^+ = SO_4^{2-} + 9Fe^{2+} + 8H_2O$
13	$CH_4 + 2O_2 = CO_2 + 2H_2O$	58	$NO_3^- + 3Fe^{2+} + 3H_2O = NO_2^- + Mt + 6H^+$
14	$2NH_4^+ + 1.5O_2 = N_2 + 2H^+ + 3H_2O$	59	$H_2S + Mt + 6H^+ = S^0 + 3Fe^{2+} + 4H_2O$
15	$8NO_3^- + 5CH_4 + 8H^+ = 4N_2 + 5CO_2 + 14H_2O$	60	$NO_2^- + 0.5O_2 = NO_3^-$
16	$CH_4 + 1.5O_2 = CO + 2H_2O$	61	$S^0 + CO + H_2O = H_2S + CO_2$
17	$S^0 + 1.5O_2 + H_2O = SO_4^{2-} + 2H^+$	62	$NO_3^- + 12Fe^{2+} + 13H_2O = NH_4^+ + 4Mt + 22H^+$
18	$H_2S + 2O_2 = SO_4^{2-} + 2H^+$	63	$3NO_3^- + NH_4^+ = 4NO_2^- + 2H^+ + H_2O$
19	$3NO_3^- + 5NH_4^+ = 4N_2 + 2H^+ + 9H_2O$	64	$SO_4^{2-} + 4CO + 2H^+ = H_2S + 4CO_2$
20	$6NO_3^- + 5CH_4 + 6H^+ = 3N_2 + 5CO + 13H_2O$	65	$S^0 + H_2 = H_2S$
21	$6NO_3^- + 5S^0 + 2H_2O = 3N_2 + 5SO_4^{2-} + 4H^+$	66	$SO_4^{2-} + 3CO + 2H^+ = S^0 + 3CO_2 + H_2O$
22	$8NO_3^- + 5H_2S = 4N_2 + 5SO_4^{2-} + 2H^+ + 4H_2O$	67	$4CO + 2H_2O = 3CO_2 + CH_4$
23	$H_2S + 0.5O_2 = S^0 + H_2O$	68	$N_2 + 3CO + 2H^+ + 3H_2O = 2NH_4^+ + 3CO_2$
24	$2NO_3^- + 5H_2S + 2H^+ = N_2 + 5S^0 + 6H_2O$	69	$SO_4^{2-} + 4H_2 + 2H^+ = H_2S + 4H_2O$
25	$NO_3^- + CO = NO_2^- + CO_2$	70	$NO_2^- + 9Fe^{2+} + 10H_2O = NH_4^+ + 3Mt + 16H^+$
26	$NO_2^- + H_2O = NO_3^- + H_2$	71	$N_2 + 5Mt + 28H^+ = 2NO_3^- + 15Fe^{2+} + 14H_2O$
27	$4NO_3^- + CH_4 = 4NO_2^- + CO_2 + 2H_2O$	72	$2NO_2^- + 2H^+ = N_2 + 1.5O_2 + H_2O$
28	$3NO_3^- + CH_4 = 3NO_2^- + CO + 2H_2O$	73	$SO_4^{2-} + 3H_2 + 2H^+ = S^0 + 4H_2O$
29	$NO_3^- + 4CO + 2H^+ + H_2O = NH_4^+ + 4CO_2$	74	$4S^0 + CH_4 + 2H_2O = 4H_2S + CO_2$
30	$3NO_3^- + S^0 + H_2O = 3NO_2^- + SO_4^{2-} + 2H^+$	75	$CO + 3H_2 = CH_4 + H_2O$
31	$4NO_3^- + H_2S = 4NO_2^- + SO_4^{2-} + 2H^+$	76	$N_2 + 3H_2 + 2H^+ = 2NH_4^+$
32	$NO_2^- + 3CO + 2H^+ + H_2O = NH_4^+ + 3CO_2$	77	$2NH_4^+ + 3S^0 = N_2 + 3H_2S + 2H^+$
33	$NO_3^- + 4H_2 + 2H^+ = NH_4^+ + 3H_2O$	78	$4S^0 + 4H_2O = SO_4^{2-} + 3H_2S + 2H^+$
34	$2NO_2^- + 9Fe^{2+} + 8H_2O = N_2 + 3Mt + 16H^+$	79	$SO_4^{2-} + CH_4 + 2H^+ = H_2S + CO_2 + 2H_2O$
35	$NO_3^- + H_2S = NO_2^- + S^0 + H_2O$	80	$CO_2 + 4H_2 = CH_4 + 2H_2O$
36	$NO_2^- + 3H_2 + 2H^+ = NH_4^+ + 2H_2O$	81	$3S^0 + CH_4 + H_2O = 3H_2S + CO$
37	$NO_3^- + CH_4 + 2H^+ = NH_4^+ + CO_2 + H_2O$	82	$4SO_4^{2-} + 3CH_4 + 8H^+ = 4S^0 + 3CO_2 + 10H_2O$
38	$3Fe^{2+} + 0.5O_2 + 3H_2O = Mt + 6H^+$	83	$CO + H_2O = CO_2 + H_2$
39	$3NO_3^- + 4CH_4 + 6H^+ = 3NH_4^+ + 4CO + 5H_2O$	84	$8NH_4^+ + 3SO_4^{2-} = 4N_2 + 3H_2S + 2H^+ + 12H_2O$
40	$4NO_2^- + 3CH_4 + 8H^+ = 4NH_4^+ + 3CO_2 + 2H_2O$	85	$N_2 + 2.5O_2 + H_2O = 2NO_3^- + 2H^+$
41	$H_2S + 4Mt + 22H^+ = SO_4^{2-} + 12Fe^{2+} + 12H_2O$	86	$4N_2 + 3CH_4 + 8H^+ + 6H_2O = 8NH_4^+ + 3CO_2$
42	$3NO_3^- + 4S^0 + 7H_2O = 3NH_4^+ + 4SO_4^{2-} + 2H^+$	87	$3SO_4^{2-} + 4CH_4 + 6H^+ = 3H_2S + 4CO + 8H_2O$
43	$NO_3^- + H_2S + H_2O = NH_4^+ + SO_4^{2-}$	88	$2NH_4^+ + SO_4^{2-} = N_2 + S^0 + 4H_2O$
44	$NO_2^- + CH_4 + 2H^+ = NH_4^+ + CO + H_2O$	89	$SO_4^{2-} + CH_4 + 2H^+ = S^0 + CO + 3H_2O$
45	$NO_2^- + S^0 + 2H_2O = NH_4^+ + SO_4^{2-}$	90	$2NH_4^+ + CO = N_2 + CH_4 + 2H^+ + H_2O$

There is a similar scenario for the chemoheterotrophs and the organic redox reactions that support their growth (data not shown). The aerobic and anaerobic respiration of numerous sugars, amino acids, and fatty acids also yield approximately 0-120 kJ/mol e^- in the different Vulcano thermal environments (Rogers and Amend 2006). These Gibbs energy calculations relied on available aqueous organic analyses (Amend et al. 1998; Skoog et al. 2007; Svensson et al. 2004). Again, only a small sub-set of the reactions are known net metabolisms of the Vulcano hyperthermophiles, but perhaps some of the other reactions support the unculturable archaea and bacteria identified to date only by their 16S rRNA gene sequences.

Community composition

The “molecular revolution” that started in the late 1970’s enabled a careful cataloging of microbial species (or often more accurately, operational taxonomic units (OTUs)) without the need for culturing (Woese and Fox 1977). In theory, all that is required is a sample (e.g., water, sediment, biomass) from which DNA can be extracted (Pace et al. 1986). It is now common practice to use 16S rRNA gene surveys to generate relatively accurate pictures of ‘who is there’, and this approach now has been applied to many hydrothermal environments (Brinkhoff et al. 1999; Huber et al. 2003; Jackson et al. 2001; Meyer-Dombard et al. 2005; Reysenbach et al. 2000; Sievert et al. 2000; Simmons and Norris 2002). When combined with classic and newly developed culturing techniques and with quantification methods (e.g., fluorescent in situ hybridization (FISH)), this picture of the community composition can be further enhanced. Most organisms are detected, many can be enumerated, and some are cultured and their physiologies characterized.

One of the first gene surveys of the Vulcano hydrothermal system targeted the archaeal diversity in a geothermal well fluid (Pozzo Istmo) at 56 °C and pH 5.8. A library of ~250 clones yielded 17 OTUs, with unique crenarchaeal (13), euryarchaeal (3), and korarchaeal (1) sequences. The majority of the crenarchaeal sequences were from a novel, deeply-branching clade, and 2 of the euryarchaeal sequences represent phylotypes of a novel genus. The third euryarchaeal phylotype was 99% identical to *Palaeococcus helgesonii*, a rare aerotolerant, hyperthermophilic fermenter previously isolated from the same well (Amend et al. 2003a). This highly focused gene survey (1 site, 1 domain) served as a pilot project that evolved into a larger investigation of numerous hydrothermal sediments and fluids collected from submarine vents and on-shore seeps, with archaeal and bacterial clone libraries constructed from 16S rRNA genes (Rogers and Amend 2009). A subset of archaeal phylotypes belonged to the well-known thermophilic and hyperthermophilic orders Archaeoglobales, Thermococcales, Thermoproteales, and Desulfurococcales; other phylotypes were from the uncultured Korarchaeota, the ubiquitous “Marine Group 1” Crenarchaeota, and the deep-sea hydrothermal vent euryarchaeal group. A majority of the bacterial phylotypes were from the Proteobacteria, mostly the ϵ -subdivision, but also included were some from the α -, β -, γ -, and δ -subdivisions. In addition, sequences closely related to the Aquificales—known for their catalysis of the knallgas reaction—Planctomycetes, Firmicutes, and Actinobacteria were identified.

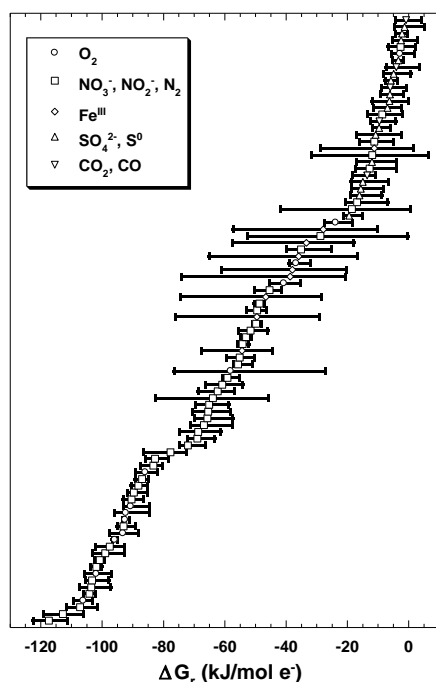


Fig.1. Values of ΔG_r for the reactions given in Table 1, symbol-coded by groups of TEAs. The horizontal bars represent the range of values determined for the 10 different sites investigated at Vulcano, and the symbols denote the average values.

To complement the gene surveys and to enumerate some of the key thermophilic phyla at Vulcano, quantitative FISH studies were carried out. To date, such studies in high temperature systems have been rather rare (Harmsen et al. 1997). Owing to poor coverage of hyperthermophilic Bacteria and certain key hyperthermophilic Archaea, new oligonucleotide probes for the Aquificales, Thermococcales, and Arc-

haeoglobales were designed and evaluated (Rusch and Amend 2004). With these and existing FISH probes, all major groups of (hyper)thermophiles, except methanogens, were detected, with up to 19% Crenarchaeota, 22% Thermococcales, 14% Archaeoglobales, 8% Aquificales, 12% *Thermotoga/Thermosipho*, 12% *Thermus*, and 12% thermophilic *Bacillus*.

Biogeochemical processes

Gene surveys and culturing studies identified many of the microorganisms that inhabit the Vulcano hydrothermal system, and Gibbs energy modeling showed which metabolisms are possible and which are energetically most profitable. However, just because an organism is there does not mean it is active, and just because a redox reaction is thermodynamically favorable does not mean it supports cellular growth. To best understand how shallow-sea hydrothermal ecosystems function, the dominant biogeochemical processes must be elucidated. Several indicators suggest that sulfur redox plays an essential role in the Vulcano ecosystem—the ghastly stench of hydrogen sulfide is pervasive; mineral precipitates include pyrite (FeS_2), elemental sulfur (S^0), and alunite (a hydrated potassium aluminum sulfate); many of the cultured thermophiles reduce sulfur as their principal metabolism (or show enhanced growth in the presence of S^0); and low pH fluids, resulting from sulfur oxidation to sulfuric acid, are ubiquitous (Amend et al. 2003b; Sedwick and Stueben 1996; Simmons and Norris 2002).

Anaerobic metabolism by natural hyperthermophilic communities from Vulcano was investigated in incubation experiments (Tor et al. 2003). Marine sediments from the surf zone of the Baia di Levante were incubated anaerobically ($\text{N}_2:\text{CO}_2$ headspace) at 90 °C with a variety of ^{14}C -labeled substrates and artificial seawater. The analytical data showed that sulfate reduction was the dominant terminal electron accepting process at 90 °C in these marine sediments. A variety of organic electron donors, including acetate and glucose, were oxidized to $\text{CO}_2/\text{HCO}_3^-$ in the process. When microbial sulfate reduction was inhibited by the addition of molybdate, the anaerobic degradation of organic matter was apparently limited to fermentation, resulting in the accumulation of the metabolites acetate and H_2 . Methanogenesis—acetoclastic or chemolithotrophic—was not observed in the incubation studies.

A schematic of the primary biogeochemical reaction network is depicted in Fig. 2. Complex organic matter can be respired aerobically and anaerobically (e.g., with S^0), or fermented to CO_2 , H_2 , and simple fatty acids (e.g., acetic acid). The H_2 and acetic acid can serve as electron donors in the reduction of sulfate to sulfide. However, H_2 (with CO_2) and acetic acid also can serve as substrates in methanogenesis, but the near-surface marine sediments apparently are too oxic to permit this highly oxygen-sensitive metabolism to proceed. At greater depths, however, methanogenesis may be a factor in the biogeochemical cycling of carbon. The model in Fig. 2 is consistent with subsequent FISH analyses of the microbial communities across the oxic/anoxic interface at this sediment site (Rusch et al. 2005). Thermophiles that gain metabolic energy from the fermentation of organic matter (Thermococcales, *Thermotoga/Thermosipho* spp., *Bacillus* sp.) were detected in significant abundances, as were thermophiles capable of oxidizing VFAs with SO_4^{2-} (e.g., Archaeoglobales).

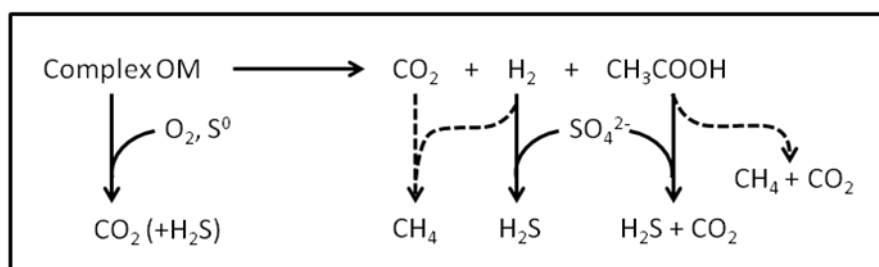


Fig.2. Schematic of the dominant biogeochemical processes (solid arrows) in the Vulcano hydrothermal ecosystem, including fermentation, aerobic respiration, and anaerobic respiration (with S^0 and SO_4^{2-} as TEAs). Methanogenesis (dashed arrows) may be occurring at depth, where the environment is less oxic.

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Diving in Panarea volcanic (Aeolian Islands, Italy): methodology and results.

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Abstract. A submarine gas eruption started in November 2002 offshore of Panarea volcano (Aeolian Islands, Italy). We planned a scuba diving campaign using a high resolution Marine Digital Terrain Model to study the exhalation centres and the geological, morphological and structural features and to clarify the relationships between gas vents distribution, submarine volcanological structures of this submerged area. We drew the first geological and structural maps of the seafloor surrounding the exhalative area and establish the role of NE- and NW- trending fractures as the main pathways for the 2002 gas exhalation.

Introduction

Panarea is a largely submarine Quaternary volcano located 20 km southwest of Stromboli (Aeolian Islands arc, southern Italy, Fig.1). In November 2002, a huge submarine high-Temperature gas eruption started offshore 3 Km east of Panarea island on top of a shallow rise of the 2.3 km² seafloor surrounded by the islets of Panarelli, Lisca Bianca, Bottaro, Lisca Nera and Dattilo between -2 m and -30 m of depth. A local seismic swarms was recorded contemporaneous to the gas eruption (Saccorotti et al., 2004). Moreover the degassing of Panarea on November 3rd followed a sequence of geophysical and volcanological events which occurred in the southern Tyrrhenian zone (Anzidei et al. 2003b; Esposito et al., 2006): the offshore earthquake (Ml=5.6) between Palermo and Ustica Island on September 6th, the onset of the paroxysmal eruption at Mt Etna on October 27th and the eruption at Stromboli that started on December 28th (Fig. 1 a). The gas eruption event at Panarea produced new concern on a volcano generally considered extinct, even if minor exhalative activities due to a shallow hydrothermal system were well known since historical times.

Volcanological setting

Panarea Island and its archipelago of small islets (~ 3.3 km²) are the emergent tips of a larger submarine stratovolcano more than 2000 m high and about 20 Km across. According to available chrono-stratigraphic data, most of the Panarea activity was focussed between 150 and 54 ka. The youngest deposits at Panarea are pyroclastic deposits of exotic provenance (Lucchi et al. 2007) between 20.5 ± 0.2 ka and 8.6 ± 1.5 ka (Punta Torriente formation, Calanchi et al. 1999a-b, Punta Torriente lithosome, Lucchi et al., 2007, Dolfi et al., 2006). The fumarolic areas of La Calcara at Panarea, Punta Levante at Basiluzzo and to a lesser extent on the less accessible stretch of seafloor facing the islets of Dattilo, Panarelli, Lisca Bianca, Bottaro e Lisca Nera (Italiano and Nuccio, 1991; Calanchi et al., 1995) have been known.

Methodology

Bathymetric data were acquired, after the 2002 gas eruption, with multibeam technique. The pace of acquisition allowed for 9 million data points to be obtained in 2.3 km². The bathymetry has a high resolution of 0.5 m/pixel. A 3D Marine Digital Elevation Model (MDEM) has been extracted for quantitative and semi-quantitative geomorphological analyses (Anzidei et al., 2003, 2005).

Scuba-dive sites were selected to maximise information on the nature of the seafloor and geology at gas eruption centres (Fig. 1) and to validate the morphological interpretations of the bathymetry. The orientation of faults and fractures has been measured with underwater compasses. The seafloor has also been in-

investigated via snorkelling along a pseudo-rectangular grid during days of good visibility, in order to extend the direct observations (Fig. 1).

The dives were performed between December 2002 and September 2003 at depths between -5 and -30 m.

Results

We have recognised three lithostratigraphic units and we have been able to reconstruct their stratigraphic relationships (Fig. 1).

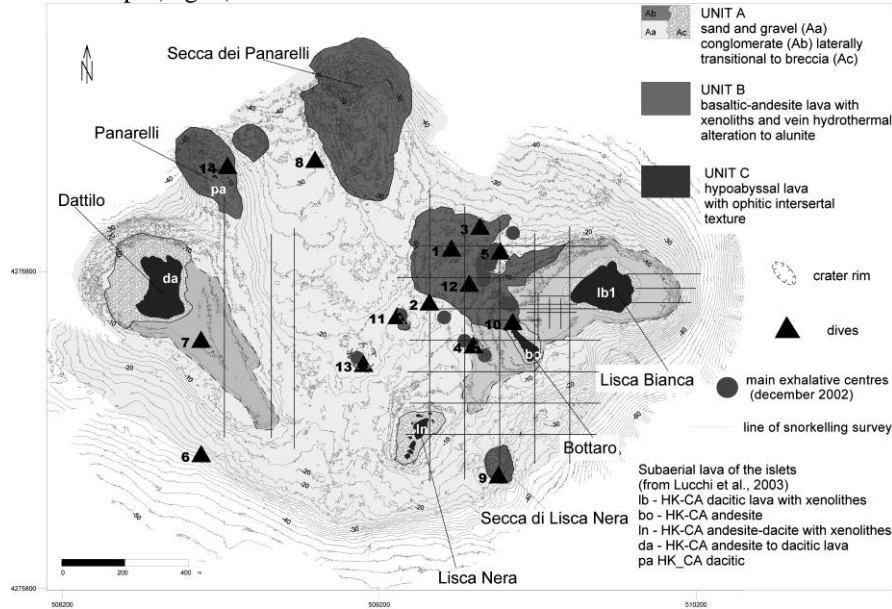


Fig.1. Geological sketch map of the seafloor around the emission centres.

The seafloor around the islets above the -30 m b.s.l. is characterised by both circular and linear features. The lineaments have been drawn using the shaded relief map illuminated from different azimuths (a) and inclinations (e) [a135-e65; a45-e68; a52-e67; a90-e52; a00-e60; a-135-e65; a178-e50] (Fig. 2).

NE trending lineaments are mostly located in the eastern part of the rise, N- to NNW-trending lineaments run through the central part, and NW-trending lineaments are located mostly in the western sector. E-trending lineaments define the north offshore area of Lisca Bianca.

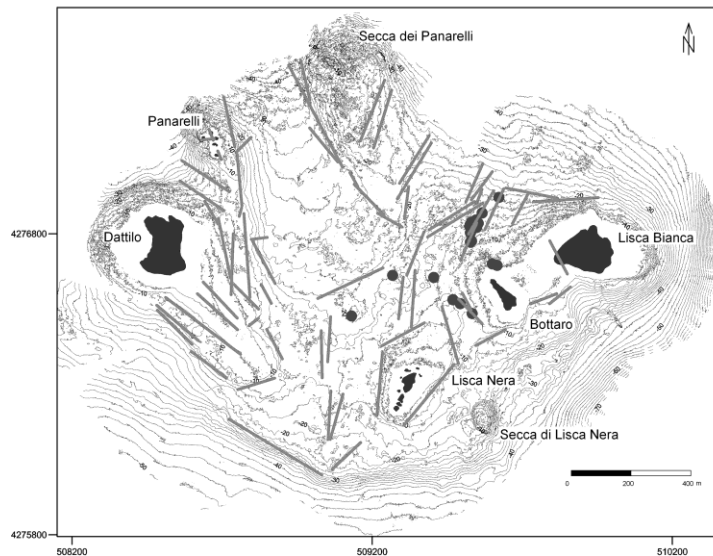


Fig.2. Map of lineaments identified using the shaded relief illuminated from different azimuths (a) and inclinations (b) main exhalative craters (full black circle).

A total of 606 EC was detected and classified by intensity and significance level (Fig.3). These latter criteria were expressed by the number of soundings that identified each EC from multiple overlapping swaths (0-1-2 levels) and from their height above the seafloor (A-B-C levels).

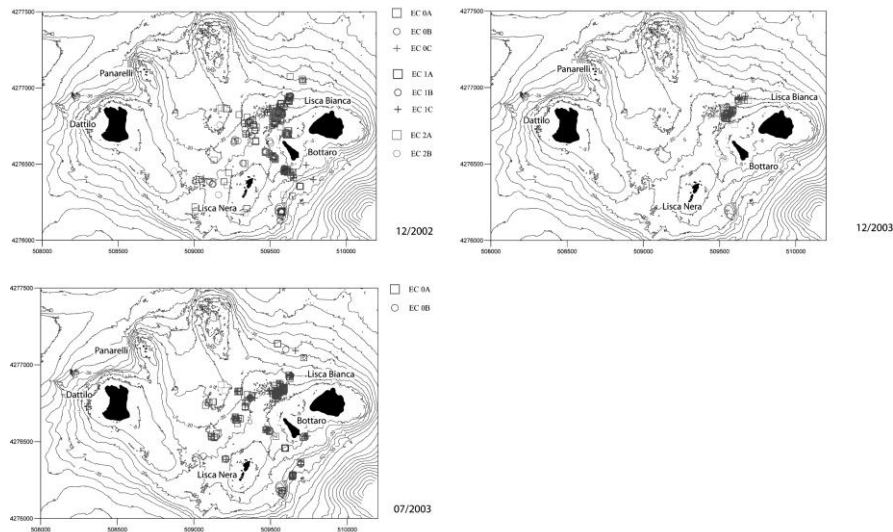


Fig.3. Emission Centers distribution in time span December 2002 – December 2003

The most frequent gas bubble alignments show two main trends, NE- and NW-oriented. The fractures on Unit B show the same NE-, and NW-trends system, also with a quantitatively important N-trending system. Fractures are generally opened and may be filled with alunite or crusts of native sulfur.

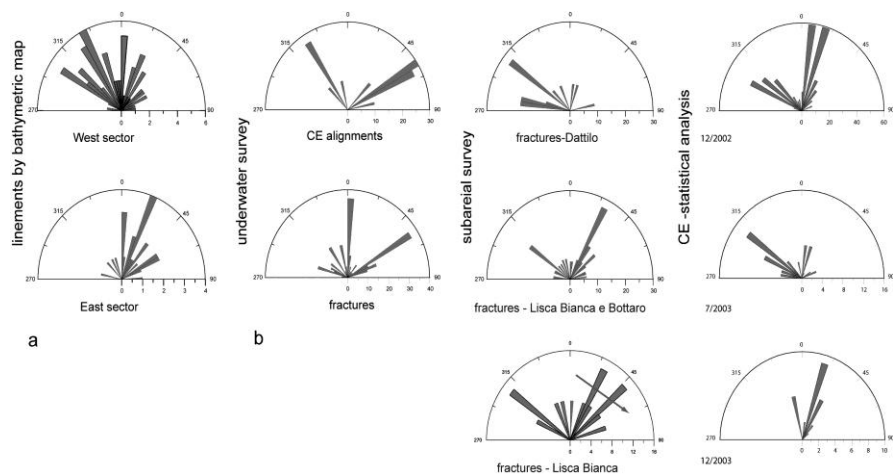


Fig.4. Rose diagrams of structural analysis

The gas vents formed in November 2002 are NE-SW and NW-SE aligned. These two alignments are the same measured for the fractures, statistical analysis. The NE-SW and NW-SE systems are and have been the main pathways for the upwelling of hydrothermal fluids. These NE-SW and NW-SE fracture systems are parallel to the main regional tectonic structures (De Astis *et al.*, 2003).

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Using scientific diving to investigate the long-term effects of ocean acidification at CO₂ vents.

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Abstract. We are using SCUBA to document ecosystem responses to long-term ocean acidification at volcanic CO₂ vents. There are 30% reductions in biodiversity at average pH 7.8-7.9 (minimum pH 7.4), compared with areas with normal pH (8.1-8.2). Some groups (seagrasses and many algae) are tolerant of the increased CO₂ levels but others (corals, sea urchins and calcified algae) are removed from the ecosystem. Transplant experiments show dissolution of calcareous organisms and our study demonstrates, for the first time, what happens to coastal marine ecosystems when key groups of species are killed due to rising CO₂ levels.

Introduction

The global oceans currently absorb over 25 million tons of CO₂ every day. This has caused surface waters to become 30% more acidic since wide-spread burning of fossil fuels began. As well as lowering pH, increased CO₂ levels are altering surface water chemistry, causing a decline in carbonate ions, an increase in bicarbonate ions and lowering calcium carbonate saturation states. Falling calcite and aragonite levels are a concern since these are the building-blocks of shells for a range of marine organisms from tiny coccolithophores to giant coral reefs. Research into the marine environmental effects of increased oceanic CO₂ levels is mainly being carried out using short-term-shock experiments whereby pH or CO₂ levels are manipulated in aquaria and enclosures over short timescales (Hall-Spencer et al. 2008).

To examine long-term ecosystem effects of ocean acidification researchers have begun to exploit natural gradients in pCO₂, such as differences in plankton community composition in the Baltic vs. Black Seas (Tyrrell et al. 2008), the depth distribution of cold-water corals in relation to the aragonite and calcite saturation horizons (Guinotte et al. 2006), tropical coral calcification strength in the high carbonate Caribbean vs the low carbonate Galapagos (Manzello et al. 2008), or changes in gastropod shell length and dissolution along gradients of estuarine pH (Marshall et al. 2008). Deep sea vents are also usually acidified, which may explain their characteristic lack of echinoderms (Van Dover 2000), and recent studies show that CO₂-rich vents in the Okinawa Trough indicate strong coupling of faunal patterns and pH (Boetius et al. unpublished). Our ongoing research programme is utilizing natural gradients in pH to investigate the effects of ocean acidification at coastal CO₂ vent sites. We have begun with comparisons of community structure and the condition of individual species along these gradients providing strong evidence that environmental hypercapnia has both negative and positive effects on the local species pool (Hall-Spencer et al. 2008, Martin et al. 2008).

Methods

Vents are being studied on the north and south sides of Castello Aragonese, Ischia, Italy (40°043.84'N; 013°57.08'E) adjacent to steeply sloping rocky shores (Fig. 1).

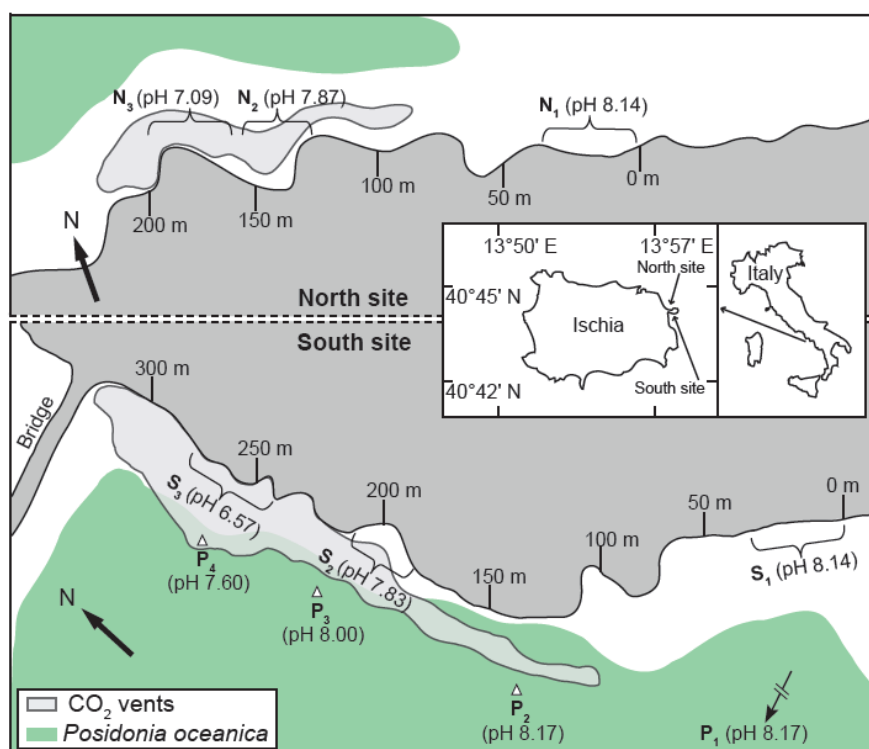


Fig.1. Map of CO₂ vent sites north and south of Castello Aragonese. Mean surface pH is shown at 35-m wide stations N1-N3 and S1-S3. Mean subtidal pH is shown at stations P1-P4 together with distributions of the gas vents and sea-grass meadow.

Physical measurements

Gas emission is monitored using SCUBA divers who deploy 1 m² plastic funnels connected to 250 ml bottles placed on the seabed in areas of high (>10 vents m⁻², constant flow), medium (5-10 vents m⁻², constant flow) and low (<5 vents m⁻², intermittent flow) gas emission rates. There is a video of this at

<http://news.bbc.co.uk/2/hi/science/nature/7437862.stm>.

Gas is collected for analysis in evacuated 50 ml glass tubes, partially filled with 20 ml of 0.1M Cd(OH)₂ + 4N NaOH suspension and connected to a plastic funnel positioned over the rising bubbles. A YSI/25 FT pH (NBS scale) meter is used for rapid assessment of salinity, temperature and pH and surface and bottom water samples are taken for more precise measurements of the spatial and temporal variability in pH (in total scale) measured immediately after water sample collection using a meter accurate to 0.01 pH units (Ion 6, Acorn Series, EUTECH Instruments, Singapore) and calibrated using TRIS/HCl and 2-aminopyridine/HCl buffer solutions. Irradiance and temperature are monitored using Hobo Onset Computer® data loggers.

Seawater samples are passed through 0.45 µm pore size filters (GF/F Whatman) and poisoned with 0.05 ml of 50% HgCl₂ (Merck, Analar), to avoid biological alteration, then stored in the dark at 4°C. A titration system composed of a pH meter with an ORION pH electrode (calibrated using NBS standard solutions) and a 1 ml automatic burette (METHROM) is used to analyse 20 ml samples at 25°C. The pH is measured at 0.02 ml increments of 0.1 N HCl. Total alkalinity is calculated from the Gran function applied to pH variations from 4.2 to 3.0, as a function of the added volume of HCl. Carbonate system parameters (*p*CO₂, CO₃²⁻, HCO₃⁻ and DIC concentrations, saturation states of calcite and aragonite) are calculated from pH (in total scale), total alkalinity, temperature and salinity using a program by Lewis & Wallace.

Biological parameters

On each field visit, extensive SCUBA surveys are made to photograph and sample macroorganisms present within and adjacent to vent areas to build a voucher collection of the species present and record their distributions. Abundances of macroorganisms are quantified in gridded 0.25 m² quadrats placed at 5

m intervals along transects. Point counts are used to determine macroalgal cover and each quadrat is censused fully for macrofaunal abundances, taking specimens for laboratory identification when necessary. Larger organisms, such as sea urchins are counted each 30 m² (10 m length x 3 m depth) throughout 200 m long (north shore) and 300 m long (south shore) vent areas. Transplant and colonization experiments are now underway and will be the subject of future publications.

Results

Vents off Castello Aragonese acidify seawater through gas emissions comprising 90.1-95.3% CO₂, 3.2-6.6% N₂, 0.6-0.8% O₂ 0.08-0.1% Ar and 0.2-0.8% CH₄ (no sulphur). Salinity (38‰) and total alkalinity (2.5 mEq kg⁻¹) are homogenous between survey stations and temperature matches ambient seasonal fluctuations (13-27°C). On the southern side of the island gas is emitted at 1.4 x10⁶ l day⁻¹ in an area of about 3,000 m² (mainly >5 vents m⁻²); on the north side gas is emitted at 0.7 x10⁶ l day⁻¹ in an area of about 2,000 m² (mainly <5 vents m⁻²). No seasonal, tidal or diurnal variation in gas flow rates was detected in 2006-2009. The pH and saturation states (Ω) of calcite and aragonite vary with sea state, being lowest on calm days, and exhibited large decreases as pCO₂ levels increased from ca 300 to > 2000 μatm through the venting gas fields. Tables 1-2 present the macroalgal and macrofaunal species recorded along 300 m stretches of shore on north and south sides of Castello Aragonese in spring 2007.

Table 1. Macroalgal presence (dots) along 35 m sections of rocky shore in spring 2007 at mean pH 8.14 (stations N₁, S₁), 7.87 (N₂), 7.83 (S₂), 7.09 (N₃) and 6.57 (S₃) adjacent to CO₂ vents off Ischia, Italy. Calcifiers are shown in bold, note reduced numbers of species at N₂, S₂ and their absence at N₃ and S₃. Total numbers of taxa at each station are given at the end of the table.

Taxa		N1	S1	N2	S2	N3	S3	Taxa	N1	S1	N2	S2	N3	S3
Phlorophyceae	<i>Acerabularia acetabulum</i>		•					<i>Codium effusum</i>	•	•	•	•		
	<i>Bryopsis plumosa</i>	•	•					<i>Codium vermilara</i>	•		•	•		
	<i>Caulerpa prolifera</i>	•	•	•	•			<i>Udotea petiolata</i>	•	•	•	•		
	<i>Caulerpa racemosa</i>	•	•	•	•		•	<i>Halimeda tuna</i>	•	•				
	<i>Chaetomorpha</i> sp.	•	•	•	•	•	•	<i>Pedobesia simplex</i>	•	•				
	<i>Cladophora prolifera</i>	•	•	•	•			<i>Rhizoclonium tortuosum</i>	•	•	•	•		•
	<i>Cladophora rupestris</i>	•	•	•	•	•	•	<i>Valonia utricularis</i>	•	•	•	•		
	<i>Codium bursa</i>	•	•	•	•	•		Other Corallinaceae	•	•	•	•		
	<i>Acrosorium ciliolatum</i>	•	•					<i>Gulsonia nodulosa</i>	•	•	•	•		
	<i>Aglaothamnion tenuissimum</i>	•	•					<i>Hildenbrandia rubra</i>	•	•	•	•	•	•
Rhodophyceae	<i>Amphiroa rigida</i>	•	•					<i>Jania longifurca</i>	•	•				
	<i>Pterothamnion plumula</i>	•	•		•			<i>Jania rubens</i>	•	•				
	<i>Apoglossum ruscifolium</i>	•	•	•	•			<i>Laurencia obtusa</i>	•	•	•	•	•	•
	<i>Asparagopsis taxiformis</i>	•	•	•	•	•	•	<i>Lithophyllum incrustans</i>	•	•				
	<i>Ceramium</i> sp.	•	•	•	•			Mesophyllum lichenoides	•	•				
	<i>Ceramium virgatum</i>	•	•	•	•			<i>Peyssonnelia squamaria</i>	•	•	•	•		
	<i>Chondracanthus teedei</i>	•	•	•	•		•	<i>Polysiphonia cf. stricta</i>	•	•		•		
	<i>Corallina elongata</i>	•	•											
	<i>Corallina officinalis</i>	•	•											
	Phaeophyceae	<i>Cladostephus spongiosus</i>	•	•	•	•	•	•	<i>Halopteris filicina</i>	•	•	•	•	•
<i>Colpomenia sinuosa</i>		•	•		•		•	<i>Kuckuckia cf. spinosa</i>	•	•				
<i>Cutleria</i> sp.		•	•	•	•	•		<i>Padina pavonica</i>	•	•				
<i>Cystoseira amentacea</i>		•	•	•	•			<i>Ralfsia verrucosa</i>	•	•	•	•	•	•
<i>Cystoseira compressa</i>		•	•	•	•			<i>Sargassum vulgare</i>						
<i>Diclyopteria polydoides</i>		•	•					<i>Sphacelaria ciliata</i>	•	•	•	•	•	•
<i>Diclyota dichotoma</i>		•	•	•	•	•	•	<i>Sphacelaria fusca</i>	•	•	•	•	•	•
<i>Diclyota fasciola</i>		•	•				•	<i>Stypocaulon scoparium</i>	•	•	•	•	•	•
<i>Diclyota spiralis</i>	•	•	•	•	•	•	<i>Turf</i>	•	•	•	•	•	•	
Total number of taxa									49	51	34	36	15	20

Table 2. Macrofauna recorded (dots) along 35 m sections of rocky shore in spring 2007 at mean pH 8.14 (stations N1, S1), 7.87 (N2), 7.83 (S2), 7.09 (N3) and 6.57 (S3) adjacent to CO₂ vent sites at Castello d'Aragnese, Italy. Organisms with external shells or skeletons shown in bold. Total numbers of taxa at each station are given at the end of the table, note reduced numbers of species at N2 and S2 and further reductions at N3 and S3.

Taxa		N1	S1	N2	S2	N3	S3	Taxa	N1	S1	N2	S2	N3	S3
Sponges	<i>Ageias oroides</i>		*					<i>Dysidea</i> sp.		*		*	*	*
	<i>Cacospongia</i> sp.	*	*	*	*	*		<i>Haliclona mediterranea</i>	*	*	*	*	*	*
	<i>Chondrilla nucula</i>	*	*	*	*	*		<i>Ircinia variabilis</i>	*	*	*	*	*	*
	<i>Chondrosia reniformis</i>	*	*	*	*	*		<i>Spirastrella cunctatrix</i>	*	*	*	*	*	*
Cnidarians	<i>Actinia equina</i>	*	*	*	*	*	*	<i>Balanophyllia europaea</i>	*	*				
	<i>Aglaophenia pluma</i>	*	*	*	*	*	*	<i>Caryophyllia smithii</i>	*	*				
	<i>Anemonia viridis</i>		*	*	*	*	*	<i>Cladocora caespitosa</i>	*	*				
	<i>Eudendrium</i> sp.	*	*	*	*	*	*	<i>Parazoanthus axinellae</i>	*	*			*	*
Amni- lids	<i>Pomatosceros triquetus</i>	*	*	*	*	*	*	<i>Serpulidae</i>	*	*	*	*	*	*
	<i>Sabella spallanzanii</i>		*	*	*	*	*	<i>Spirorbidae</i>	*	*	*	*	*	*
Crusta ceans	<i>Balanus perforatus</i>	*	*	*	*	*	*	<i>Pachygrapsus marmoratus</i>	*	*	*	*	*	*
	<i>Chthamalus stellatus</i>	*	*	*	*	*	*	<i>Palaemon serratus</i>	*	*		*	*	*
	<i>Euraphia depressa</i>	*	*	*	*	*	*							
Molluscs	<i>Acanthochitona crinita</i>	*	*	*	*	*	*	<i>Lima lima</i>	*	*				
	<i>Aplysia depilans</i>		*	*	*	*	*	<i>Melaraphe neritoides</i>	*	*	*	*	*	*
	<i>Arca noae</i>	*	*	*	*	*	*	<i>Mytilus galloprovincialis</i>	*	*	*	*	*	*
	<i>Astrea rugosa</i>	*	*	*	*	*	*	<i>Ocenebrina edwardsi</i>	*	*		*	*	*
	<i>Anomia ephippium</i>	*	*	*	*	*	*	<i>Ostraea</i> sp.	*	*				
	<i>Brtilium reticulatum</i>	*	*	*	*	*	*	<i>Ostlinus turbinata</i>	*	*	*	*	*	*
	<i>Buccinulum corneum</i>	*	*	*	*	*	*	<i>Patella caerulea</i>	*	*	*	*	*	*
	<i>Cerithium vulgatum</i>	*	*	*	*	*	*	<i>Patella rustica</i>	*	*	*	*	*	*
	<i>Elysia timida</i>	*	*	*	*	*	*	<i>Patella ulysiponensis</i>	*	*	*	*	*	*
	<i>Fiabellina</i> sp.	*	*	*	*	*	*	<i>Serpulorbis arenarius</i>	*	*				
	<i>Fasciolaria lignaria</i>	*	*	*	*	*	*	<i>Thais haemastoma</i>	*	*	*	*	*	*
	<i>Jujubinus striatus</i>	*	*	*	*	*	*	<i>Tricolia pullus</i>	*	*	*	*	*	*
	<i>Haliotis</i> sp.	*	*	*	*	*	*	<i>Octopus vulgaris</i>	*	*	*	*	*	*
	<i>Hexaplex trunculus</i>	*	*	*	*	*	*	<i>Vermetes triquetus</i>	*	*				
<i>Lepidochitona cinerea</i>	*	*	*	*	*	*								
Echino- -derms	<i>Arbacia lixula</i>	*	*	*	*	*	*	<i>Holothuria tubulosa</i>	*	*	*	*	*	*
	<i>Coscinasterias tenuispina</i>	*	*	*	*	*	*	<i>Holothuria forskali</i>	*	*	*	*	*	*
	<i>Echinaster sepositus</i>	*	*	*	*	*	*	<i>Paracentrotus lividus</i>	*	*	*	*	*	*
Fish	<i>Chromis chromis</i>	*	*	*	*	*	*	<i>Sarpa sarpa</i>	*	*	*	*	*	*
	<i>Diplodus annularis</i>	*	*	*	*	*	*	<i>Scorpaena porcus</i>	*	*	*	*	*	*
	<i>Labrus bimaculatus</i>	*	*	*	*	*	*	<i>Symphodus roissali</i>	*	*	*	*	*	*
	<i>Labrus viridis</i>	*	*	*	*	*	*	<i>Trypterion tripteronotus</i>	*	*	*	*	*	*
	<i>Muraena helena</i>	*	*	*	*	*	*							
Total number of taxa									57	69	40	49	15	11

One of the most obvious effects of the CO₂ vent system is the dissolution of shelled organisms and their absence from waters with a mean pH <7.1 (Figure 2).

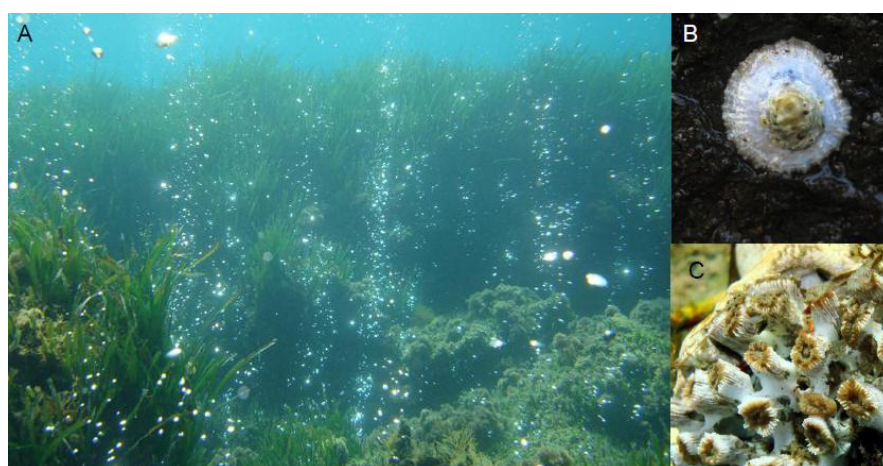


Fig. 2 A) General view of *Posidonia oceanica* meadow with CO₂ vents at 2 m depth, May 2009. B) *Patella caerulea* living naturally at mean pH 7.4 showing shell dissolution. C) *Cladocora caespitosa* transplanted to mean pH 7.4 showing skeletal dissolution.

Discussion

We have shown that natural CO₂ vents can provide insights into which species are tolerant of long-term high CO₂ levels and can be used to test predictions based on modelling and laboratory work, such as what levels of CO₂ exposure restrict the ability of marine organisms to build shells (Fig 2). Although lush stands of seagrasses thrived at increased CO₂ levels (Fig 2), major groups such as corals, sea urchins and bivalves were removed from the ecosystem and replaced by algae such as *Sargassum* sp. and *Caulerpa* spp. (Tables 1&2). In brief, our work shows:

- major ecological tipping points along a gradient of increasing CO₂ levels
- acidification dissolved the shells of calcified species such as corals, sea urchins and snails, which were absent in areas with a pH less than 7.4
- high CO₂ favoured the production of seagrass and removed its calcareous epiphytes
- the amount of calcified algae, which bind coral reefs together in the tropics, fell from more than 60 per cent cover outside the vent areas to zero within these areas
- invasive alien species, which cause damage to ecosystems worldwide, may thrive at high CO₂ levels

Scientific diving work at natural CO₂ vents (Hall-Spencer et al., 2008, Martin et al., 2008, Rodolfo-Metalpa et al. in prep) is providing the first data on what happens to coastal marine ecosystems when key groups of species are killed due to rising CO₂ levels. Having proven our concept with preliminary studies, we recommend that our approach is adopted and supported more widely given the need to gain a sound understanding of the socioeconomic implications of ocean acidification. We are now undergoing the fastest rate of ocean acidification the Earth has seen for at least the past 20 million years so this study adds urgency to the international policy drive to reduce CO₂ emissions.

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Marine shallow-water hydrothermal systems as natural laboratories

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Abstract. Marine shallow-water hydrothermal systems were discovered at more than 40 locations. The discharge of hot mineralized fluids into near shore marine environments creates dramatic physical, chemical and biological gradients. This, combined with easy accessibility, makes them excellent “natural” laboratories to study a wide range of chemical, physical, and biological processes. Studies can be performed in the form of passive thought experiments or actively by inserting experiments into the system. These types of experiments possess the potential to overcome several of the limitations of „normal“ laboratory environments.

Introduction

Research on seafloor hydrothermal activity has focused primarily on deep-sea black smoker-type locations, which are found along volcanically active portions of the mid-ocean ridges and in deep back-arc basins. Submarine hydrothermal activity, however, is not confined to deepwater environments. Hydrothermal vents have been documented on the tops of seamounts, on the flanks of volcanic islands, and in other near-shore environments characterized by high heat flow (e.g., Dando and Leahy, 1993; McCarthy et al., 2005; Prol-Ledesma et al., 2004). Their easy accessibility, relative to deep-sea hydrothermal systems, makes them excellent “natural” laboratories to study a wide range of chemical, physical, and biological processes.

The purpose of this contribution was not to give an all-encompassing account of the use of marine shallow-water hydrothermal systems as natural laboratories. Rather the intent is to introduce the idea and to demonstrate their utility on hand of select examples. In addition some basic information about the relationship between, geology, water source, heat source and discharge depths are given to aid in the search of marine shallow-water hydrothermal systems with desired physico-chemical conditions.

Abundance of marine shallow-water hydrothermal systems

Approximately 40-50 sites are presently known. However considering that many are found in remote, poorly explored areas of the globe it is likely that for every known there will be several unknown localities. Their occurrence is closely controlled by geologic and tectonic conditions, because a large heat differential is needed to initiate hydrothermal circulation (Fehn and Cathles, 1986). This heat differential is generally caused by heat released from intruding magma bodies or by the generation of frictional heat along fractures. As a result, most marine shallow-water hydrothermal systems are found near ocean island volcanoes (mantle hot spots), island arc volcanoes (subduction zones) and large active faults (transform faults).

Characteristics of marine shallow-water hydrothermal systems

Unfortunately there is no clear definition of what marine shallow-water hydrothermal systems are, nor is there any clear agreement how to call these phenomena. This author defines them as: the submarine discharge of a hydrothermal fluid into the shallow ocean (<200 m); they can have characteristics of sub aerial hot springs or that of deep-sea hydrothermal systems; the hydrothermal fluid source can include any fraction of meteoric water; phase separation (boiling) during or immediately prior to discharge is likely; they discharge into the photic zone and may contain appreciable amounts of dissolved oxygen.

The term “marine shallow-water hydrothermal systems” was used consistently throughout the manuscript and conforms to what is currently used in the scientific literature (Aliani et al., 1998; Ishibashi et al.,

2008; Miwako Nakaseama, 2008; Pichler et al., 2006; Price et al., 2007; Price and Pichler, 2005). This terminology seems to be the most appropriate to avoid possible confusion. The term marine is necessary to separate these systems from their lacustrine counterparts. The term shallow-water, hyphenated, is necessary to indicate that these systems are at a shallow water depth, rather than that their subsurface circulation is shallow.

A simple approach to classify marine shallow-water hydrothermal systems could be based on the chemical composition of their liquid and gaseous discharge, geologic setting and physico-chemical parameters, such as temperature, pH, etc. Climate, however, may also play an important role. For example, their occurrence in a desert climate such as Bahia California should prevent the presence of meteoric water in the hydrothermal system (Forrest et al., 2005; Prol-Ledesma et al., 2004). In other areas, such as Dominica (McCarthy et al., 2005), large annual variations in rainfall may force large annual variations in salinity of the hydrothermal fluid.

Gas chemistry

The chemical composition of gas in marine shallow-water hydrothermal systems is closely controlled by a combination of heat source and host rock. Nevertheless, generally CO₂ is the main component (Botz et al., 1996; Capasso et al., 1997; Forrest et al., 2005; McCarthy et al., 2005; Pichler et al., 1999). Systems in volcanic areas may have a chemical composition very close to that of volcanic gases discharging directly from active volcanoes in the vicinity. The source of such gases is likely phase separation in the magma chamber. Such gases can be high in H₂S, SO₂ and He (Giggenbach, 1996). Marine shallow-water hydrothermal systems whose heat source is frictional may be much higher in N₂ or CH₄, for example (Forrest et al., 2005). Large concentrations of CO₂ and CH₄ may also indicate carbonate rocks in the subsurface (Pichler et al., 1999).

Solution chemistry

The source and therefore, initial chemical composition, of a hydrothermal fluid is an important factor controlling the final chemical composition. While the determination of fluid sources is relatively straightforward in deep marine and inland settings, the subject becomes more complex in coastal regions (offshore and on-shore) where marine and terrestrial sources may both be involved (Pichler, 2005). A fluid in a hydrothermal system may be derived from any, or any combination, of the following sources: meteoric water, seawater, connate water, magmatic water and juvenile water. Mixing of waters from different sources affects many aspects of the geochemistry of a hydrothermal system, such as its chemical composition, isotopic composition, temperature profile or gas content.

Marine shallow-water hydrothermal systems, which circulate seawater or mixed seawater-meteoric water, have abundant complexing ligands in the form of Cl⁻ and SO₄²⁻ to transport metals, such as Pb, Zn, Cu and Cd (Vidal et al., 1978). On the other hand, fluids from marine shallow-water hydrothermal systems, which circulate mainly water of meteoric origin generally transport metals, which form oxyanions, such as As, Sb, Tl, etc. (Pichler et al., 1999).

Physico-chemical parameters

The two most important physico-chemical parameters to categorize marine shallow-water hydrothermal systems are temperature and pH. The discharge temperatures of hydrothermal fluids range from just above ambient up to approximately 130°C. They largely depend on water depth (i.e., the pressure curve) gas content (i.e., CO₂ curve) and salinity (i.e., the NaCl curve) (e.g., Henley and Ellis, 1983; Nicholson, 1992). Therefore the fluid with the least gas content, highest salinity and deepest discharge point has potentially the highest discharge temperature.

The pH values can range from as low as 1-2 up to 8-9. This range is controlled by a combination of water-rock interaction, initial buffer capacity and near surface conditions. Based on pH and chemical conditions we can group many marine shallow-water hydrothermal systems using the same classification used for on land thermal springs: acid sulfate, bicarbonate and neutral chloride (Giggenbach, 1997).

Examples of “natural” laboratory studies

The use of marine shallow-water hydrothermal systems as natural laboratories is comprised of two fundamentally different approaches, passive or active. The difference is that the passive approach relies on already generated data, while the active approach generates new data as part of the experiment.

A passive approach consists of a thought experiment, which can be tested with data generated as part of the chemical, physical, biological, etc. description of the system. An active approach consists of inserting an experiment into the system and observing a reaction, change or response.

The passive approach involves the following steps: (1) generation of a thought experiment (identification of a scientific problem), (2) complete description of the site – or at least of those parameter, which pertain to the problem, (3) extraction of data from desired parts or locations of the system, and (4) data evaluation and interpretation. The active approach on the other hand consists of: (1) design of an experiment, (2) complete description of the site – or at least of those parameter, which pertain to the experiment, (3) insertion of the experiment at desired location(s), (3) extraction of data from the experiment, and (4) data evaluation and interpretation. Concrete examples for both types are given below.

Determination of oxidation rates (passive approach)

Mixing of the hot mineralized hydrothermal fluid with ambient seawater causes chemical and physical gradients in sediments surrounding marine shallow-water hydrothermal systems and in the water column above. If these gradients can be mapped, they present an exceptional opportunity to study a wide variety of reactions. Particularly the influence of redox and temperature changes on precipitation, dissolution and adsorption. The gradients can be readily mapped with the help of flow meters and the careful determination of mixing between hydrothermal fluid and seawater (see also the section on Isotopes below).

The scientific problem to be studied is that the oxidation rates of iron and arsenic in seawater proceed at different rates; Fe(II) being much faster oxidized by seawater derived O₂ than As(III). To cover a wide range of physico-chemical conditions (i.e., flow rate, temperature, salinity, oxygen content) the four sites at Tanga (Pichler, unpubl. data), Ambitle (Pichler et al., 1999), Bahia Concepcion (Prol-Ledesma et al., 2004) and Dominica (McCarthy et al., 2005) are ideal “natural” laboratories. Although physico-chemically quite different, all of these sites have elevated arsenic and the precipitation of hydrous ferric oxide (HFO) in common.

A first step would be to sample the hydrothermal discharge at five locations stepping away from the vent orifice. This will provide a time sequence and concentration gradient, similar to the sampling along an out flow channel from an on-land hot spring. The measurement of flow velocity and change in Fe(II)/Fe(III) and As(III)/As(V) will allow to compute an approximate oxidation rate. The computation of oxidation rates ($R = \partial\text{Fe}^{2+}/\partial t$) for these field experiments can now follow the approach of Johnson and Pilson (1975).

Determination of oxidation rates (active approach)

The active approach to measure oxidation rates would be to force precipitation at a controlled site and to measure precipitate accumulation. To achieve this several small (1 cm²) ceramic plates can be positioned at the vent orifices directly in the hydrothermal discharge as close as possible to the locations of ferrihydrite deposition. Ceramic plates are an ideal substrate for ferrihydrite precipitation. By installing them on the first day of fieldwork, one can collect precipitation information for the duration of fieldwork and by installing another set at the end of fieldwork, one may be able to collect precipitation information for the duration between two field seasons. The amount of precipitation can be determined by weighing the plates before and after the experiment. In conjunction with a measurement of hydrothermal flow, chemical composition of the vent fluid (Fe(II)/Fe(III) and As(III)/As(V)), and chemical composition (Fe and As) of the ferrihydrite, this will allow the calculation of an approximate *in situ* oxidation rate for Fe and As.

Determination of isotope fractionation factors (passive approach)

One of the dominant precipitate in the hydrothermal system at Ambitle Island is aragonite. Its precipitation occurs due to a change in Eh, pH, temperature and chemical composition when the hydrothermal fluids discharge and mix with ambient seawater. The aragonite, however, is not homogenous and shows a distinct layering of different colors, crystal sizes and chemical/isotopic composition. It can be hypothesized that the layering is a result of changing physico-chemical conditions during precipitation, particularly discharge rate

and thus seawater mixing. This has an impact on precipitation temperature, saturation and precipitation velocity. The amount of seawater-hydrothermal fluid mixing during precipitation was calculated using strontium isotopes (Pichler, 2001):

$$\left(\frac{{}^{87}\text{Sr}}{{}^{86}\text{Sr}}\right)_M = \left(\frac{{}^{87}\text{Sr}}{{}^{86}\text{Sr}}\right)_{HF} \left(\frac{\text{Sr}_{HF} * (1-x)}{\text{Sr}_M}\right) + \left(\frac{{}^{87}\text{Sr}}{{}^{86}\text{Sr}}\right)_{SW} \left(\frac{\text{Sr}_{SW} * x}{\text{Sr}_M}\right)$$

where M is the mixture, HF is the hydrothermal fluid, SW is seawater and x is the degree of mixing (or fraction of hydrothermal fluid present in the mixture). Sr_M has to be calculated for each mixture using equation (1). Based on the ${}^{87}\text{Sr}/{}^{86}\text{Sr}$ -mixing model, the aragonite layers precipitate at different temperatures and precipitation ceases at a seawater admixture of 11%, which corresponds to a temperature drop of up to 7°C. Thus, the lamination of aragonite signal changes in the physical and chemical conditions at and near vents.

The $\delta^{13}\text{C}$ of Tutum Bay hydrothermal aragonite ranges from 1.9 to 2.2‰ (VPDB). This range of values is in good agreement with experimental data (Bottinga, 1968; Kim and O'Neil, 1997), indicating that C-13-equilibrium has been reached during its formation. Values for $\delta^{18}\text{O}$ range from 14.2 to 14.7‰ and calculated isotopic equilibrium temperatures are approximately 20°C lower than directly measured vent fluids and those temperatures obtained from fluid inclusion measurements and the ${}^{87}\text{Sr}/{}^{86}\text{Sr}$ mixing model. This indicates that either oxygen isotope equilibrium was not attained or that the calcite-water fractionation factor for oxygen isotopes is not applicable to the precipitation of Tutum Bay hydrothermal aragonite. Apparently, carbon isotopic equilibrium was reached, while for oxygen complete equilibrium was not attained.

An alternative explanation for the discrepancy between the measured and calculated could be that the assumption of identical fractionation factors for aragonite and calcite at higher temperatures is not correct. Assuming that the aragonite in Tutum Bay precipitated in equilibrium with its parent fluid, the isotope values here can be used for a $\delta^{18}\text{O}$ aragonite-water fractionation factor between 90 and 100°C.

Determination of arsenic toxicity (active approach)

Laboratory experiments measured the effects of $[\text{As}^{3+}]$ and $[\text{As}^{5+}]$ on foraminiferal growth and survivorship. Exposure of foraminifers to As^{3+} and As^{5+} at concentrations of 0 to 1000 µg/kg showed that As^{3+} is approximately 2.2× more toxic to foraminifers than As^{5+} , that concentrations of 600-1000 µg/kg As^{3+} is sufficient to kill or severely impair *Amphistegina gibbosa* specimens on approximately two-week timescales, and that concentrations of 1000 µg/kg As^{5+} or 200 µg/kg As^{3+} are sufficient to retard their growth (McCloskey et al., 2009).

To test these results in a more realistic (natural) setting growth experiments were positioned at increasing distances from the marine shallow-water hydrothermal systems in Tutum Bay, PNG, thus exposing foraminifers to a range of As^{3+} concentration. The foraminifers, together with some natural substrate were, enclosed in mesh bags and positioned at the sediment-seawater interface. Following extraction the data collection procedures were identical to those of the laboratory experiment.

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Measuring toxic elements and toxicity in marine shallow-water hydrothermal systems

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Abstract. Marine shallow-water hydrothermal systems are often enriched in biologically toxic elements. Here, we report our investigation of the bioavailability, uptake, and bioaccumulation of hydrothermally-derived arsenic into several coral reef organisms from the arsenic-rich marine shallow-water hydrothermal system of Tutum Bay, Ambitle Island, in northeastern Papua New Guinea. Hydrothermal venting provides bioavailable As by two major pathways throughout Tutum Bay: 1) easily-exchangeable As from hydrothermally influenced sediments to as far away as 200 m from focused venting, and 2) in surface seawaters, which may allow for biological uptake by phytoplankton and transfer up the food web. The soft coral *Clavularia sp.*, the calcareous algae *Halimeda sp.*, and the tunicate *Polycarpa sp.* collected from the hydrothermal area displayed distinctly higher total arsenic (2 to 20 times) compared to the control site, with increasing trends while approaching focused hydrothermal venting.

Introduction/Rationale

Marine shallow-water hydrothermal vent fluids often contain elevated concentrations of potentially toxic elements such as As, Sb, Se, Cr, Co, Pb, Cd, Ag, Cu, Tl, Zn, Hg, and S, as well as possible limiting nutrients such as Si and Fe (Dando et al., 1999; Price and Pichler, 2005; Varnavas and Cronan, 1988; Vidal et al., 1978). Thus, the discharge of fluids from shallow marine hydrothermal systems can have a considerable impact on not only the chemical, but also the biological composition of the surrounding coastal environment. These systems are therefore compelling natural analogs for coastal anthropogenic pollution, and 'ecotoxicological' research of this type are critical to advancing the state of knowledge concerning coastal pollution.

In an effort to clearly define the potential toxicity, biogeochemical cycle, and bioaccumulation patterns in a coastal area affected by arsenic-rich hydrothermal discharge, we focus on the shallow marine hydrothermal system off Ambitle Island, Papua New Guinea (Fig. 1). As previous research has shown, as much as 1.5 kg of arsenic are estimated to be discharged into Tutum Bay on a daily basis (Pichler and Dix, 1996). Arsenic is the only potentially toxic element which is highly enriched in this system, thus allowing for the first time the investigation of the effect a single toxin has on a coastal coral reef ecosystem. We present here the bioavailability of arsenic from the inorganic compartment of the hydrothermal system, as well as the total bioaccumulated arsenic present in the tissue of several coral reef organisms collected along a transect moving away from focused hydrothermal venting (Figure 2 A and B). For the purposes of these proceedings, the discussion of organoarsenic speciation is left out of this manuscript, but will be presented during the workshop.

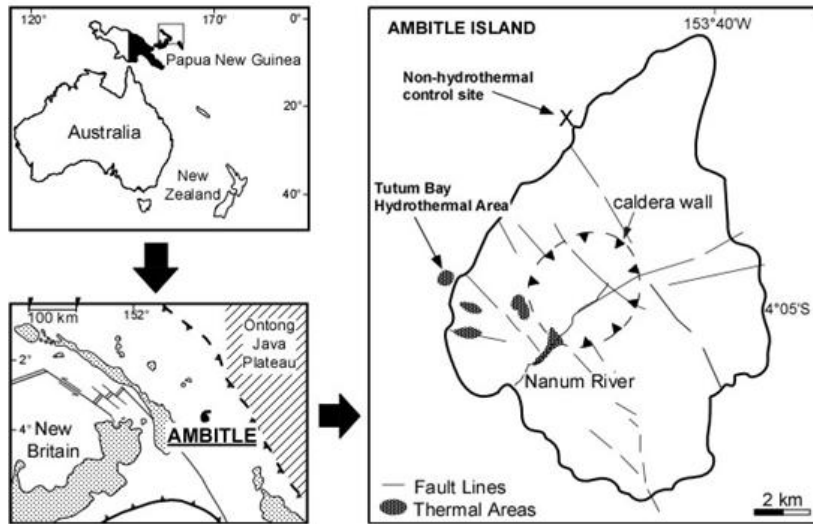


Fig. 1. Location of Ambitle Island and the marine shallow-water hydrothermal vents investigated in this research. Tutum Bay hydrothermal area and the control sites are indicated (modified from Pichler and Dix, 1996).

Methods

Field

Sediment, seawater, and tissue samples were collected by SCUBA diving along a roped, metered transect beginning at an area of focused venting and extending it out to 300 m, well beyond the influence of hydrothermal activity (Fig. 2). A non-hydrothermal control site was sampled approximately 1.6 km north of the main vent area (Fig. 1). The control site was located just off Picnic Island, and is a pristine coral reef.

Seawater and sediment samples were preserved following Price and Pichler (2005). Once biological samples were on board the research vessel, the organisms were dissected and washed thoroughly with milli-Q water, placed in 1.5 mL centrifuge tubes and kept frozen during transport. Each sample was freeze dried and taken to the Water Quality Centre at Trent University for arsenic abundance and arsenic speciation measurements.

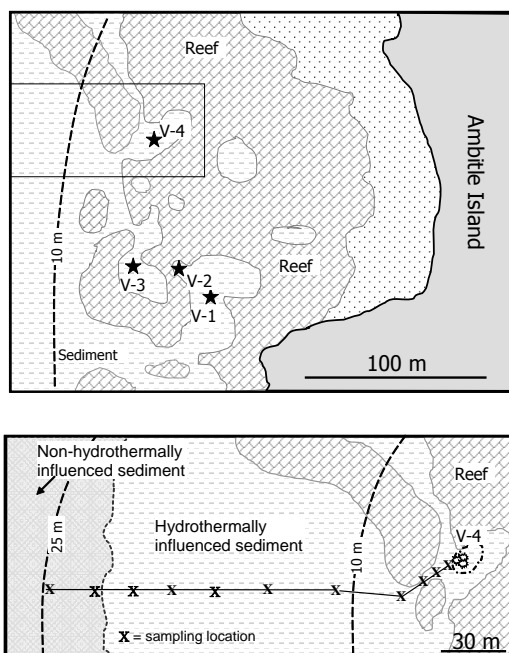


Fig. 2. (A) Location of the main venting areas of the hydrothermal system at Ambitle Island. The box in the upper left is enlarged in (B) (modified from Pichler et al., 1999c). (B) Location of transect along which tissue samples were collected for this study.

Laboratory

The bioavailable arsenic from surface sediments collected along the roped transect was extracted through a cation-exchange reaction using a solution of $\text{KH}_2\text{PO}_4/\text{K}_2\text{HPO}_4$ at a pH of 7.2, following Gleyzes et al. (2002). Arsenic abundance and speciation in seawater was assessed following Price and Pichler (2005).

Complete digestion of tissues was carried out by weighing freeze-dried tissue (50 – 200 mg) into 50 mL digestion vials, adding 1 mL HNO_3 (16 mol/L) and 0.5 mL H_2O_2 (10 mol/L), and letting the reaction proceed for 1 h at room temperature, followed by 1 h at 80 °C in a water bath. This two-step procedure was employed to reduce sample foaming during digestion. After cooling, the digest was diluted to 50 mL with Milli-Q water and filtered to < 0.2 μm . A second dilution was necessary to normalize carbon content (Larsen and Sturup, 1994). All reagents used for this study (e.g., HNO_3 , H_2O_2) were ultrapure grade and were obtained from Fisher Scientific and High Purity Standards.

Total arsenic (TAs) in the tissue digests was determined by inductively-coupled plasma-dynamic reaction cell-mass spectrometry (ICP-DRC-MS) using O_2 to eliminate the $^{40}\text{Ar}^{35}\text{Cl}^+$ interference on $^{75}\text{As}^+$ (Bandura et al., 2001). ^{193}Ir was used as an internal standard to compensate for other matrix interferences.

Results and Discussion

Bioavailability of Arsenic

Diffuse venting seems to play a critical role on the distribution of As throughout Tutum Bay surface sediments, which have a mean As concentration of 527 ppm while excluding the vent precipitates (range=1483 to 52 ppm). Up to 54 ppm As were extracted from the easily extractable fraction of surface sediments (mean=19.7 ppm), using a $\text{K}_2\text{HPO}_4/\text{KH}_2\text{PO}_4$ buffer at pH=7.2. Arsenic from this fraction is considered to be the most available for biological processes, and therefore the most dangerous for biota. However, sequential extraction shows that a mean of 93.3% in surface sediments (range=88.2% to 96.3%), is coprecipitated with the hydrous ferric oxide (HFO) fraction. Thus, the bulk of the As being discharged into Tutum Bay is scavenged by the HFO, and should remain stable unless the physicochemical conditions surrounding the oxides change. In surface seawaters of Tutum bay, we found as much as four times the average seawater concentration of As (8.4 $\mu\text{g}/\text{L}$ compared to ~ 2 $\mu\text{g}/\text{L}$). The abundance of As in seawater just above the sediment/water interface is near normal, although As(III) in both surface and bottom seawater throughout Tutum Bay is substantially enriched compared to average seawater.

Hydrothermal venting therefore provides bioavailable As by two major pathways throughout Tutum Bay: 1) easily-exchangeable As from hydrothermally influenced sediments to as far away as 200 m from focused venting, and 2) in surface seawaters, which may allow for biological uptake by phytoplankton and transfer up the food web.

Bioaccumulation of Arsenic into Biota

Total arsenic concentrations in *Clavularia* tissue samples from the hydrothermally-influenced sites ranged from 2.9 to 20.9 mg/kg dry weight. Values are elevated in samples collected closer to focused hydrothermal venting (Fig. 3 A), although some data scatter occurs. This scatter is possibly a combination of several factors, including specimen age (increased time to accumulate more As), and sampling of some organisms which were not in direct contact with hydrothermal fluids. The average TAs for samples from the hydrothermal transect is 10.6, thus giving an enrichment factor of ~ 5.0 , on average, compared to the control site sample (i.e., 2.1 ± 0.1 mg/kg). Given the fact that these values are well above analytical uncertainty, it is probable that bioaccumulation to up to 10 times above background concentrations can occur in *Clavularia* from Tutum Bay as a result of hydrothermal venting of arsenic-rich fluids (e.g., the highest concentration was 20.9, thus 9.8 times enriched compared to the control site).

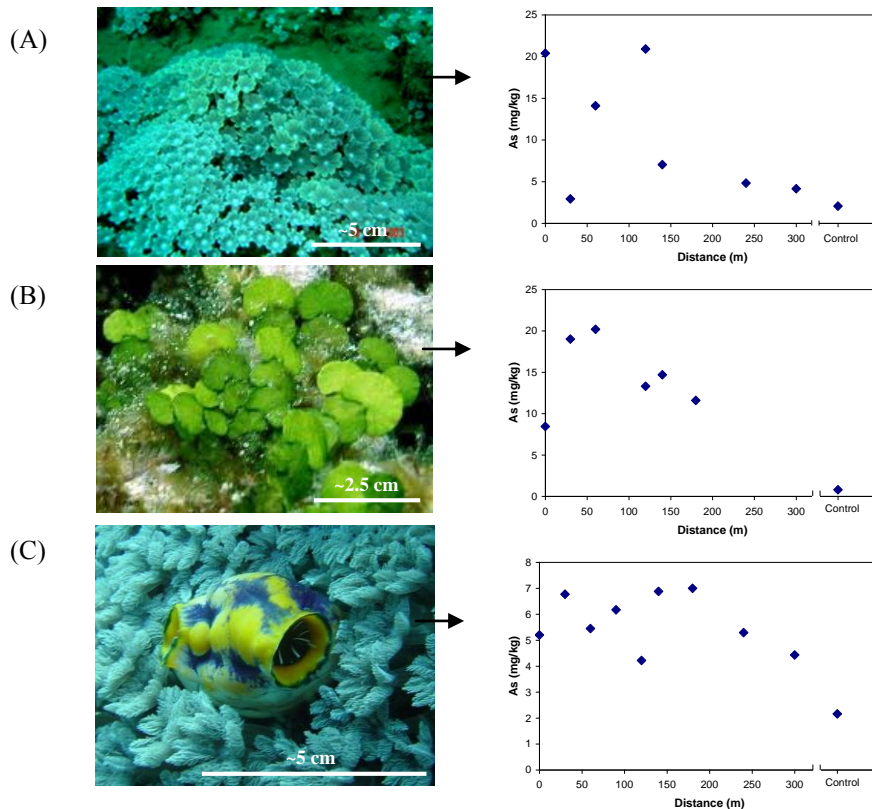


Fig. 3. Underwater photographs and associated total arsenic concentration of coral-reef organisms collected from Tutum Bay. A) *Clavularia sp.* B) *Halimeda sp.* C) *Polycarpa sp.* surrounded by *Clavularia sp.*

Total arsenic concentrations in the *Halimeda* collected along the hydrothermal transect ranged from 8.5 to 20.2 mg/kg dry weight (Fig. 3 B). All samples collected from the hydrothermal area contained elevated concentrations of TAs compared to those collected from the control site. The average TAs concentration measured in *Halimeda* collected along the hydrothermal transect was 14.5, giving an average enrichment factor of 18.4 over the concentration of TAs in *Halimeda* collected from the control site, while an enrichment factor of 25.4 is possible.

Arsenic abundance in the tunicate *Polycarpa* collected along the roped transect ranged from 4.2 to 7.0 mg/kg dry weight (Fig. 3 C). There is no clear increasing trend in TAs for samples collected closer to focused hydrothermal venting. However, the average concentration for samples collected along the hydrothermal transect is 5.8 versus 2.2 in the control site sample. This calculates to an enrichment factor of 2.7, with a maximum of 3.2. Thus, arsenic may be bioaccumulating into tunicates surrounding hydrothermal venting. *Polycarpa* only grow on the reef and on coral mounds above the sediment/water interface, thus the opportunity for enhanced bioaccumulation as a result of direct contact with hydrothermal fluids discharging through sediments is less. However, it is very likely that the *Polycarpa* in the hydrothermal area are uptaking arsenic by feeding on falling plankton, which may contain elevated TAs, and thus may explain elevated TAs in *Polycarpa*.

Summary and Conclusions

Arsenic in Tutum Bay is bioavailable to surrounding organisms by two major pathways: 1) easily-exchangeable As from hydrothermally influenced sediments to as far away as 200 m from focused venting, and 2) in surface seawaters, which may allow for biological uptake by phytoplankton and transfer up the food web.

This is the first clear evidence of arsenic bioaccumulation along a gradient of increasing arsenic concentrations in sediments and waters surrounding a coral reef, and the first time TAs have been presented for *Clavularia*, *Halimeda*, and *Polycarpa*. TAs in all 3 organisms investigated in this study is very elevated for samples from the hydrothermal area compared to control-site samples.

Contaminants effecting coastal marine ecosystems are of increasing concern as human populations expand adjacent to these communities. It is therefore very important to understand the natural, coastal biogeochemical cycle of arsenic and other contaminants. This knowledge will allow us to better anticipate changes arising from human activity, and evaluate the best procedures for dealing with toxins in coastal marine environments.

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Geochemical investigations and gas quantification of submarine fluid discharges in the hydrothermal system of Panarea (Aeolian Islands, Italy)

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Abstract. Geothermal water discharging submarine east of Panarea Island was investigated during several scuba diving campaigns in 2007 and 2008. Among the acidic, high-mineralized fluid samples two distinct water types could be identified. The end-member composition was calculated using $^{87}\text{Sr}/^{86}\text{Sr}$ ratios and geothermometers (Na/K, K/Mg). Additionally, attempts were made to quantify the total gas exhalation of the hydrothermal system. The total gas release was estimated with about $2.3 \cdot 10^7$ L/day.

Introduction

Monitoring active volcanoes nowadays is predominant a routine procedure but very important for risk assessment analysis of volcanic areas around the world. Less attention was paid so far to submarine volcanic activities because it is much more difficult to get samples and / or take readings from fumaroles and water discharging points at the seafloor. One of the first investigations in this direction was performed by Italiano and Nuccio (1991). From 2006 on the Scientific Diving Research Group of the Technische Universität Freiberg started scuba diving activities at Panarea island in order to improve submarine sampling and measuring techniques on the one hand and to investigate the gas and water discharging in depth between 8 and 30 meters on the other hand.

Investigation area

The volcanic island Panarea is located in the Tyrrhenian Sea in the north of Sicily. Together with six further islands and several seamounts it belongs to the Aeolian Archipelago, which is in total of volcanic origin. The formation of the Aeolian Arc as well as its volcanic activity is related to the active subduction of the African continental plate below the Eurasian plate (Dekov and Savelli 2004). The investigation area is located about 2.5 km east off the coast of Panarea on a submarine plateau up to 30 m water depth. This area of about 2.3 km² is surrounded by several islets, which represent the remnants of a former crater rim (Esposito et al. 2006, Gabbianelli et al. 1990). The recent volcanic activity is characterised by significant exhalations of hydrothermal waters and gas escaping at numerous sites from the seafloor. Within the crater six locations with interesting conditions were identified: Bottaro West, Bottaro North, Black Point, Hot Lake, Point 21 and Area 26 (Fig. 1). These sites are close to each other with only few hundred of meters in between.

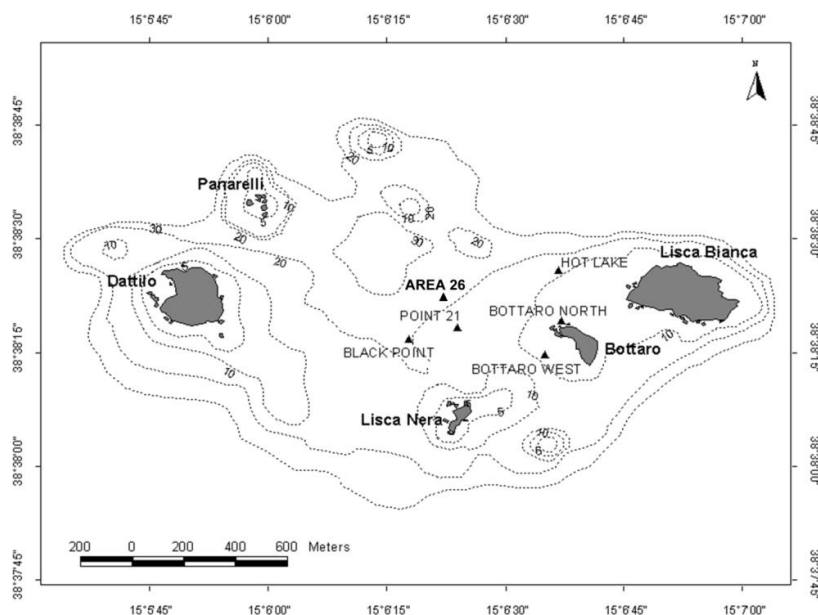


Fig. 1. Investigation area east of Panarea within several small islets (modified from Rohland 2007). Sampling sites are shown as triangles.

Methods

Geochemical analyses

Temperature, pH value, redox potential and electric conductivity were taken either in situ or immediately after sampling. Water samples were analysed for their chemical and isotopic composition by photometry and ion-sensitive electrodes in a field lab and by ion chromatography and ICP-MS within 2 to 3 weeks after sampling.

For submarine sampling the most important aspect is to obtain hydrothermal fluids with as less as possible intermixing with seawater. In one case a Teflon lance was installed using a drop hammer device. By means of special rods it was possible to penetrate the sea bottom to a depth of 3 m in a small crater filled with loose debris (Hot Lake). Water samples were taken using syringes which were connected to the Teflon lance via a Teflon hose and a self sealing coupling. At another site (Black Point) a commercial drilling machine (compressed air) was used to open a fracture to a depth of 50 cm in order to capture the water as deep as possible and thus minimize mixing with seawater.

Gas quantification

To quantify the gas output of the submarine venting area, at five sites the total gas flow rate of all gas emanations was determined during a diving campaign in September 2008.

Gas flow rates of small and medium sized submarine fumaroles were measured by means of water displacement in a bottle with a total volume of 1.2 L. The gas was captured by a stainless-steel funnel (diameter: 19.3 cm, height: 45 cm) which was hold over the emission point. Via the bypass of the funnel the water in the bottle was replaced and the time recorded in duplicates. In most cases the outlet gas temperature was measured additionally with a digital thermometer (GMH 3350, Greisinger electronics) which was encapsulated in a home made container. In order to compensate different gas temperatures, measured flow rates were referenced to the standard ambient temperature of 25 °C using the ideal gas law. The latter was also applied to correct the results with respect to hydrostatic pressure.

Since each site is featured by several hundreds of fumaroles, it was practically impossible to measure all gas flow rates. The fumaroles were clustered into four classes depending on their intensity of gas release, which was observable for divers by the extent of the bubbling gas columns. The gas emanations were

counted and multiplied with the measured average flow rate of each class (Table 1). This yielded the total gas output.

Table 1. Classes of gas output based on the measurement of 48 fumaroles

class	A	B	C	D
range of gas flow rate [L/min]	< 2.1	2.1 - 3.6	3.6 - 7.2	> 7.2
number of vents	23	8	6	11
mean [L/min]	1.06	3.04	4.86	9.89
standard deviation [L/min]	0.53	0.47	1.18	2.13

At least 10 fumaroles in the Panarea area discharge greater amounts of gas (> 40 L/min) and could therefore not be determined with the above described technique. For these type of fumaroles a much larger funnel and a flow through cell measurement device was developed, and is described elsewhere (Bauer et al. 2009). Two fumaroles at Point 21 site were measured with the technique. Based on the results the gas output of some vents with similar intensities of gas release was estimated by comparison of photographs.

Results and discussion

Geochemistry of waters

The submarine water discharges were characterized by temperatures ranging between 44° and 135°C and pH values ranging between 2.9 and 5.9. Samples from Hot Lake and Black Point showed conductivities significantly higher (up to 100 mS/cm) than those from the other sampling sites (mean: 56 mS/cm, 19 samples) or normal seawater (Table 2). Most fluid samples were typified by reducing redox conditions. But samples from Black Point differed by significantly higher redox values (Eh) up to about +300 mV (pH ~ 3). This indicates only partly reducing conditions which might be caused by a higher input of magmatic gases such as SO₂, HCl or HF (Capaccioni et al. 2007).

Table 2. On-site parameters (mean ± standard deviation) of sampled water exhalations from different sites

Site	#	pH	EC [mS/cm]	Eh [mV]	O ₂ [mg/L]	T _{in-situ} [°C] ^e
Hot Lake	5	4.9 ± 0.25	93.7 ± 10.56	-39.6 ± 18.47	0.8 ± 0.32	96
Black Point	3	3.1 ± 0.25	71.7 ± 4.97	303.7 ± 47.88	6.2 ± 1.40	135
Point 21	3	5.2 ± 0.26	53.4 ± 1.29	-3.54 ± 12.70	1.9 ± 0.22	71
Bottaro North	2	5.8 ± 0.14	56.0 ± 0.71	-8.4 ± 38.07	3.1 ± 2.30	46
Bottaro West	2	5.4 ± 0.21	55.6 ± 1.41	33.03	4.2 ± 0.11	44
Seawater	-	8.2 ^a	54 ^b	500 ^c	6.0 ^d	27 ^f

- number of samples, ^a Millero 2006, ^b Gugliandolo et al. 2006, ^c Merkel & Planer-Friedrich 2002, ^d Brown et al. 1995, ^e maximum temperature directly measured in the point of the fluid discharge, ^f average seawater temperature during the investigations in September 2008

A cluster analysis (Ward algorithm, squared Euclidean distance) on the chemical composition of 36 submarine water samples from six different sites taken in 2007 and 2008 ended with three distinct groups of samples. One group comprised only samples taken at Black Point, another consisted of all samples from Hot Lake and the last group summarized the remaining samples from the other diving sites.

Samples from the latter group were dominated by seawater mixing either due to subsurface mixing or during the sampling procedure. On the contrary, the fluid samples from Black Point and Hot Lake were enriched with respect to seawater in Na⁺, K⁺, Ca²⁺, Cl⁻, SiO₂ and Mn²⁺, but distinctly depleted in Mg²⁺ and SO₄²⁻. Therefore, they are supposed to be strongly influenced by typical hydrothermal processes.

The acidic, high mineralised fluids are further enriched in several minor and trace elements such as Li, Br, B, Fe, Mn, Pb or Zn up to 8 orders of magnitude in relation to normal seawater (Fig. 2).

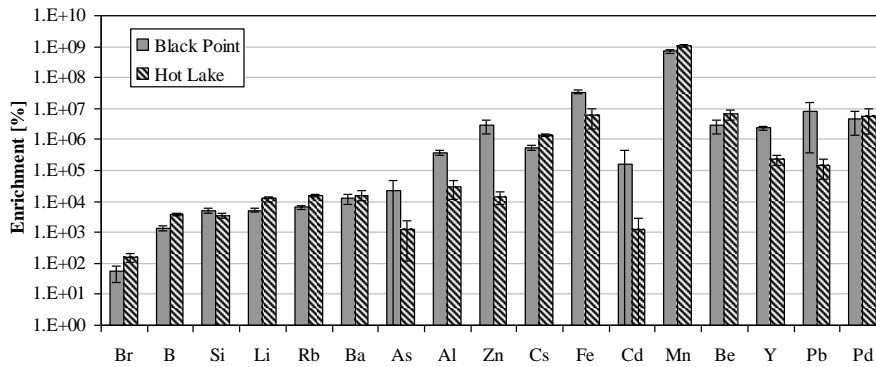


Fig. 2. Relative enrichment of selected elements in the fluid samples from Black Point and Hot Lake in relation to normal seawater (reference data for seawater were taken from Brown *et al.* 1995)

The biggest problem in sampling of hydrothermal fluids in submarine environments is a more or less intensive mixing with surrounding seawater. To evaluate the uninfluenced fluid composition, seawater contribution was determined with the help of strontium isotopic ratios of fluids and rock samples applying a two component mixing model. Representative rock samples were taken from the surrounding subaerial islets. The $^{87}\text{Sr}/^{86}\text{Sr}$ ratios versus the reciprocal strontium contents of the samples plot along a straight line ($R^2 = 0.9184$, Fig. 3) which can be interpreted as a mixing line between the rock and the seawater isotope pattern (Stosch 2004).

For water samples from Black Point and Hot Lake the lowest seawater contributions were calculated to be 33.1 and 37.2 %, respectively. In comparison a sample from Point 21 resulted in a seawater share of 80.0 %.

Another approach to estimate the seawater contribution is based on the application of two different geothermometers. The Na/K and K/Mg geothermometers developed by Giggenbach (1988) were used to estimate the reservoir temperature. For this, chemical equilibrium conditions were assumed for ion-exchange reactions between the involved fluid and rock phase at depth. It was further assumed that the fluids are only a mixture of one hydrothermal end-member and seawater. The analysed Na, K and Mg concentrations of the fluid samples were corrected semi-numerically by removing different proportions of seawater. These corrected concentrations were used to calculate the reservoir temperature with both geothermometers. Those Na, K and Mg concentrations, for which both temperature estimations were almost equal, were assumed as the final seawater contribution of the fluid samples.

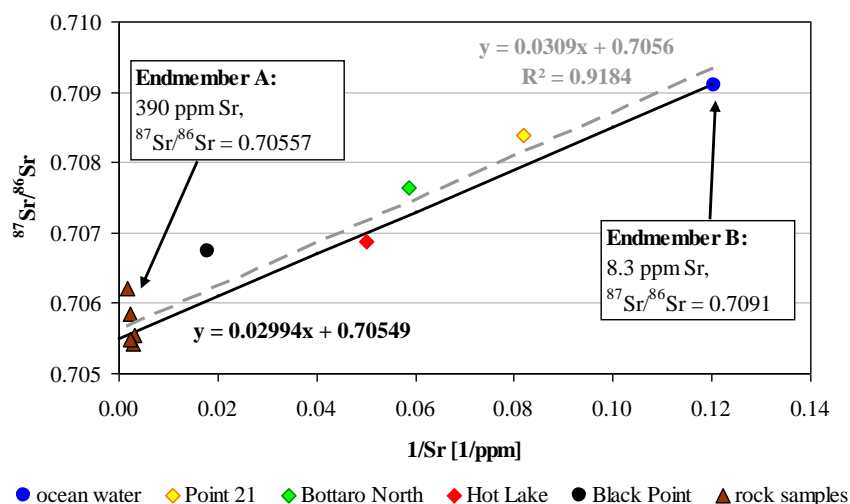


Fig. 3. Two-component mixing model between rock-derived strontium isotopes (End-member A) and seawater strontium isotopes (End-member B): drawn line indicates two-component mixing line, dashed line represents regression line.

According to the geothermometers the Hot Lake fluid contains on average 71.1 % of seawater whereas the Black Point fluid showed a contribution of about 68.7 %. The estimated reservoir temperature ranges between 310°C for the Black Point fluid and 345°C for the Hot Lake fluid. To verify the assumptions of this approach a ‘maturity index’ in accordance with Giggenbach (1988) was calculated (MI should be

higher than 2). It followed that the Hot Lake fluid had been in full equilibrium during water-rock interactions in the reservoir (MI = 2.82). On the contrary, the Black Point fluid did not fulfil the required conditions (MI = 1.59).

Apparently, great differences for the calculated seawater contributions exist between these two approaches. One possible reason might be the existence of a third source of strontium in relation with the dissolution of marine evaporates which were formed during the Messinian Salinity Crises in the late Miocene (Mueller & Mueller 1991). This is confirmed by $^{87}\text{Sr}/^{86}\text{Sr}$ ratios and $\delta^{34}\text{S}$ data of gypsum, carbonates, halite or dolomite reported by Stein et al. (2000) which are similar to data of this study. In this case, the two-component model does not adequately explain the seawater contribution of the fluid samples. Another difficulty exists in the determination of the strontium end-member ratio for the rock basement. Taken rock samples were in part influenced by alteration and weathering processes. It might be possible that the strontium isotopic ratio of the reservoir rock is more similar to the ratio of fresh oceanic crustal material ($^{87}\text{Sr}/^{86}\text{Sr} = 0.7029$, Spooner 1976). Assuming this, a two-component mixing model would lead to higher seawater contributions.

In sum, the most reliable results derive from the semi-numerical approach. Mg^{2+} and SO_4^{2-} are almost completely removed in both end-member fluids, but Na^+ , K^+ , Ca^{2+} and Cl^- are highly enriched (Fig. 4). It is conspicuous that the Hot Lake end-member is characterised by main ion concentrations approximately twice as high as the Black Point end-member (Fig. 4). These large variations may indicate phase separation processes. Boiling of the fluids under high temperatures and low pressure conditions in a shallow hydrothermal system leads to the formation of a vapour phase and a high-density liquid phase (Foustoukos and Seyfried 2007). The latter is thought to be the main reason for the high mineralization especially of the Hot Lake fluid.

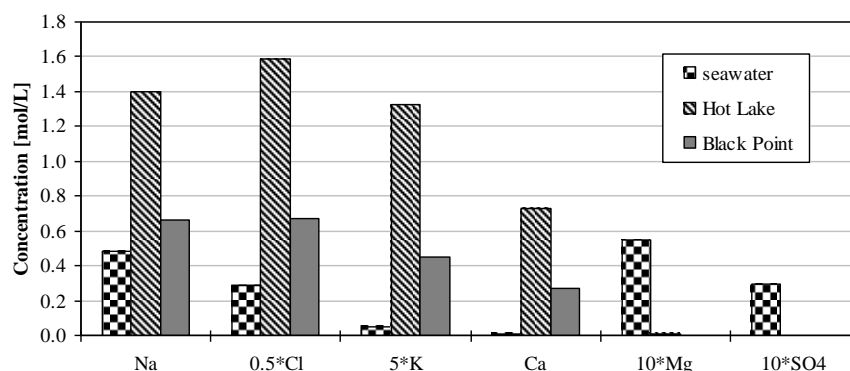


Fig. 4. End-member composition of the Hot Lake and Black Point fluids in comparison to seawater.

Quantified gas output

The five investigated sites differed with respect to the number of observed gas exhalations (Table 3). Most of them occurred at Fumaroles Field, Black Point and Bottaro West. Due to the smaller area of the Bottaro North site and a quite different morphology in respect to the other locations only 180 vents were counted. Fumaroles fell predominantly into class A representing the lowest intensity of gas output. They accounted for about 75 – 92 % of all fumaroles at every investigation site. Classes of higher gas flow rates followed with decreasing frequency of individual vents (Table 3). The total gas output that resulted from the four classes ranges from about 560 to 3520 L/min for the appropriate submarine site (Fig. 5).

Table 3. Number of the classified vents

investigation site	class			
	A	B	C	D
Bottaro West	591	39	9	2
Bottaro North	144	11	14	6
Fumaroles Field (close to Hot Lake)	549	90	37	4
Point 21	297	61	24	10
Black Point	546	36	8	2

Very strong exhaling vents, examined separately and measured by Bauer *et al.* (2009), only occurred at Point 21 and Bottaro North - five vents at each site. The individual gas flow rates of these fumaroles ranged between about 210 and 870 L/min (Bauer *et al.* 2009). Hence, these few very strong fumaroles contributed in a considerable proportion to the total gas output: at Point 21 by 57 % and at Bottaro North even by 78 % (Fig. 5). For that reason the Point 21 site was emitting gas in the highest rates with about 4620 L/min, followed by Bottaro West (3520 L/min). The sites of Bottaro North, Fumaroles Field and Black Point had almost an equal total gas output of about 2490 to 2740 L/min (Fig. 5).

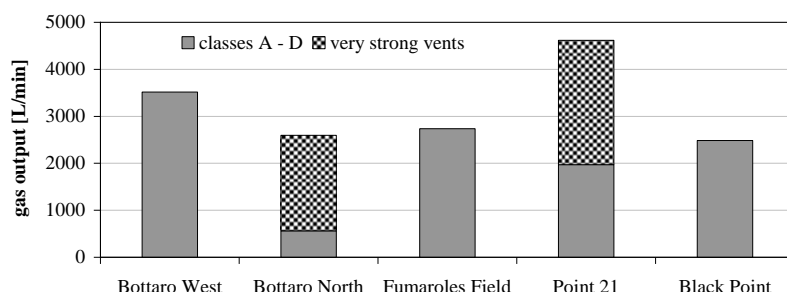


Fig. 5. Results of the gas quantification for the individual submarine sites.

In total, the entire submarine hydrothermal area of about 2.3 km², which is represented by the five investigated locations, emitted 15,951 L/min. It equals a daily gas output of $2.3 \cdot 10^7$ L/day predominantly made up of CO₂ (more than 93 vol.% in the gaseous emissions).

Since the number of estimates was much higher than direct measurements of the individual gas flow rates, the determination of the total submarine hydrothermal gas output should be deemed as semi-quantitative. Errors resulted from measurements and estimations as well as unconsidered vents have to be taken into account. Nevertheless, the authors suppose a good quality of the quantification. Even though an error of 100 % is possible, the order of magnitude of 10^7 L/day remains the same.

To compare with previous investigations, the determined total submarine gas emission of $2.3 \cdot 10^7$ L/min is more than twice as high as the emission rate of about $0.9 \cdot 10^7$ L/day, first determined in the mid 1980's by Italiano and Nuccio (1991). This rate remained almost constant until November 2002, when a submarine gas burst led to an anomalous degassing event (Caliro *et al.* 2004). The gas output rose by more than two orders of magnitude (Caracausi *et al.* 2005b) and was gradually decreasing after November 2002 to about $2 \cdot 10^7$ L/day in July 2003 (Caliro *et al.* 2004). The latter value comes very close to the gas output estimated in September 2008.

Conclusions

The geochemical investigations of the submarine hydrothermal system of Panarea revealed the existence of two distinct fluid types discharging at the locations « Black Point » and « Hot Lake ». They differ with respect to their mineralisation, redox potential as well as their minor and trace element enrichment. Possible reasons for these differences are thought to be phase separation processes under high temperature (up to 350°C) and low pressure conditions. Thereby, a high-salinity brine phase is formed which is thought to dominate the Hot Lake fluid. The Black Point fluid, however, is supposed to be more influenced by the contribution of magmatic gases.

Measurements and estimations of gas flow rates from fumaroles resulted in a total gas release within the investigation area of about $2.3 \cdot 10^7$ L/day. This is of a similar order of magnitude compared with the situation in 2003 when the system has stabilised following the crisis of 2002. All in all, it might be supposed that the hydrothermal system remains constant from 2003 until 2008.

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Mineralogical and geochemical characteristics of the shallow-water massive sulfide precipitates of Panarea, Aeolian Islands, Italy

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Abstract. Massive hydrothermal sulfide precipitation were observed along submarine gas and fluid exhalations in shallow-water depths of the submerged volcanic complex in the east of the island of Panarea, Aeolian archipelago. During two scientific diving fieldtrips in 2007 and 2008, hosted by the Scientific Diving Center, TU BA Freiberg, these precipitates were sampled and investigated for a better understanding of the genesis of shallow-water massive sulfide deposition in back-arc basins.

Galena, sphalerite, pyrite, and strontio-barite are the most common minerals. They occur both in form of porous sulphide crusts and as sediment-hosted, disseminated ore aggregates. Furthermore the appearance of Pb-As sulfosalts was proved. Iron- and manganese-rich crusts were also found and investigated. In this case the identification failed and it is assumed, that these crusts own complex characteristics.

The elements Zn, Pb, Ba, As, Sr in the sulfide crusts and Fe, Mn in the iron- and manganese crusts show the highest contents in the precipitates. The elements Cu, Sn, Au, and Ag occur only in minor concentrations.

Introduction

The evolution and deposition of submarine, hydrothermal and volcanogenic mineral associations stands since the discovery of hydrothermal springs on the seafloor of the Galapagos rift system in the late 1970s (Corliss et al. 1979) in the focus of marine geological research. Since that time the research evolved further and so it is well known today, that Volcanic-hosted Massive Sulfide (VHMS) deposition may occur on several geodynamic positions, for instance along middle ocean rift systems, within island arc systems, intraplate hot-spot systems and within back-arc basins.

In the Mediterranean Sea hydrothermal massive sulfides were first discovered in 1984 at the Palinuro seamount (Minniti and Bonavia, 1984). After that, several expeditions were carried out to explore the mineralogical and geochemical characteristics and occurrence of VHMS deposits in the back-arc basin of the Tyrrhenian Sea (e.g. Marani et al. 1997, Savelli et al. 1999). An overview of 30 years of hydrothermal research in the Tyrrhenian Sea is given by Dekov & Savelli (2004).

The Scientific Diving Center of the Technische Universität Bergakademie Freiberg (TU BAF) started its research of the hydrothermal system of Panarea in 2006. Since that time, 4 further scientific diving campaigns in 2007 and 2008 were undertaken. During these 3 years several volcanogenic and hydrothermal features, including the deposition of VHMS-associations, were found. The purpose of this article is to present the first results of geochemical and mineralogical analyses of these VHMS-associations, which were sampled in extreme shallow water depths in the east of the main island Panarea.

Geological setting

The island of Panarea is part of the Aeolian Volcanic District (AVD), a complex of seven volcanic islands and seven seamounts located in the back-arc basin of the Tyrrhenian Sea. Panarea consists of a main island and six minor islets (Basiluzzo, Dattilo, Bottaro, Lisca Bianca, Lisca Nera and Panarelli) eastward of the main island (De Astis et al. 2006). The whole complex of Panarea owns a surface area of 240 km² and an average diameter of 17.5 km (Favalli et al. 2005). It represents the emergent relicts of a connected submarine stratovolcano rising from a depth of about 1700 m b.s.l. to 421 m a.s.l. on Punta del Corvo, Panarea.

The volcanic evolution of Panarea is marked by seven eruptive epochs. The activity started in approximately 150 ka BP and reached his first summit till the end of the fifth eruptive epoch between 118 - 105 ka BP. In this space of time the basement of Panarea and the minor islets, except for Basiluzzo, was formed (De Astis et al. 2006). The following epochs were characterized by periods of quiescence, which were disturbed by periods of low volcanic activity. The last eruptive products of the volcanic complex of Panarea were produced during the sixth eruptive epoch between 70 - 8 ka BP. In this epoch the formation of Basiluzzo and the emplacement of pyroclastic products (i.e. Brown Tuffs) on Bottaro and Lisca Bianca took place (De Astis et al. 2006).

Special features 2.5 km east of the major island Panarea are five small islets. These five islets represent the emergent remnants of former lava domes and border a submarine plateau in the middle of them. The submarine plateau covers an area of about 2.3 km² and reaches a maximum water depth of ~ 30 m b.s.l. (Esposito et al. 2006). There, recent hydrothermal gas exhalations and fluid discharge can be observed on several submarine exhalation fields. Esposito et al. (2006) described a total of 21 exhalative fields, whereas three of them were the major locations of the submarine gas eruption in 2002 and 2003. One of them, we called "Black point", presents the working area, where the fieldwork and rock sampling for this report was done.

Materials & Methods

All specimen discussed in the following were sampled during two scientific diving field campaigns in the years 2007 and 2008, organized by the Scientific Diving Center of the TU Bergakademie Freiberg.

Field work

All investigated rock samples were sampled from the diving site "Black point", which represents a former submarine explosion crater in the dimensions of 26 m (N-S-direction) and 24 m (E-W-direction) and in a water depth of 23.5 m. Over the whole area hot fluid and gas discharges were observed at several spots. The location is named after a conglomeratic rock complex which is located in the southeast part of the crater and covered with black sulfide and manganese incrustations. Directly in a discharge point of the rock complex a water temperature of 134.8°C was measured.

All samples were collected with conventional scuba dive equipment in working groups of 2 to 4 scientific divers. For rock sampling, chisel and hammer as well as a drilling machine driven by pressured air were used.

An overview about the fieldwork and the appearance of special geological features is given in Fig. 1.

Geochemical investigations

For the geochemical and mineralogical determination of the VHMS mineral association following techniques were used: ICP-MS and XRF analyses to determine the geochemical element distribution of the specimens and Powder X-Ray Diffraction (XRD) and SEM-EDX analyses to determine the mineralogical characteristics of the specimens.

For the bulk geochemistry a total of 10 samples were analyzed. Thereby the elements Pb, Zn, Fe, Mn, Ba, As and Sr were measured by means of XRF analyses and the elements Cu, Au, Ag, Cd, Mo, Sb, Ti and V by means of ICP-MS analyses. The analyses were performed in the department of analytical chemistry at the Helmholtz-Center of Environmental Research Leipzig (UFZ).

The Powder-XRD analyses were performed at the Institute for Geophysics und Geology, University of Leipzig with an X-ray diffractometer (Rigaku Miniflex). Here, a total of 6 samples were analyzed. The SEM-EDX analyses were performed at the Institute for Geology, TU Bergakademie Freiberg with a com-

bined REM-EDX device (JEOL JSM 6400 Noran Vantage). Thereby, 9 specimens without a previous preparation were analyzed.

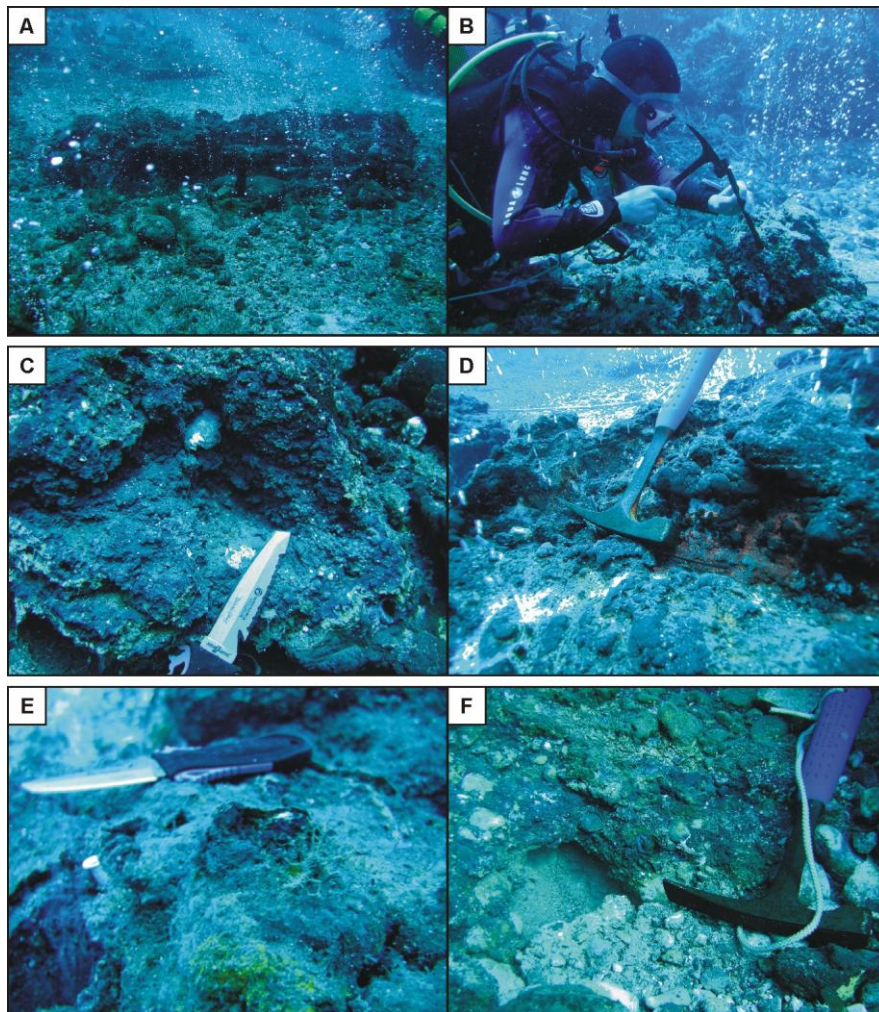


Fig.1. Underwater photographs of special geological features of “Black point”: (A) overview of the whole conglomeratic rock complex; (B) rock sampling with hammer and chisel; (C) fine layered disseminated mineral precipitates on the frontwall; (D) psilomelane-like crust covers the surface of the rock complex; (E) former inactive hydrothermal chimney on the top of the complex; (F) disseminated sulfide precipitates fill the interstices between the conglomeratic components at the base of the rock complex.

Results

Macroscopic appearance of the precipitates

On the whole surface as well as within the conglomeratic rock complex “Black point” a massive, dense mineral incrustation can be observed. Thereby, 3 different zones of mineralization can be distinguished, which will be termed in the following as mineral associations (MA).

The internal layer starts with disseminated lead-grey aggregates, which fill the interstices between the conglomeratic components (Fig. 2-B). Because of its characteristic color this layer can be described as lead-grey MA. Probably in direct contact of the hot hydrothermal fluid to the cold seawater, the next zone was formed. This layer shows orange-rusty colors and owns only a very low thickness of about 0.5 - 2mm (Fig. 2-A). It can be named as oxidized MA and probably represent the zone, where a rapid change in the pH and

redox conditions could be observed. The external layer is characterized by a black colored, fine porous psilomelane-like MA (Fig. 2-A,B). This layer is in immediate contact to the seawater.

As reported in the next section, the element distribution within these three zones varies completely.

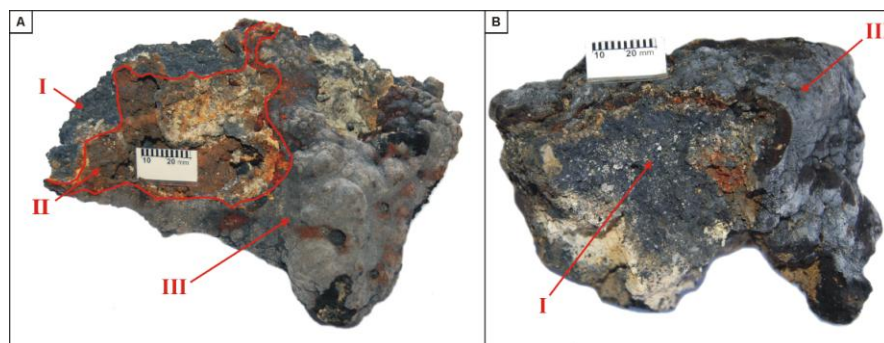


Fig.2. Rock samples of the conglomeratic rock complex “Black point”: Three distinct zones can be identified on the rock samples: **I** represent the lead grey MA, which fills cavities between the conglomeratic components, **II** represent the oxidized MA, which reached only a very low thickness and may represent the reaction zone between the hot hydrothermal fluid and the cold seawater, **III** represent the outer layer and is characterized by black, porous, psilomelane-like aggregates.

Bulk geochemistry

Polymetallic enrichments occur in all investigated samples but differ in the distribution of the individual elements in the particular MA (Table 1).

In the lead-grey MA a high enrichment of Pb and Zn for the formation of metal sulfide minerals (e.g. galena and sphalerite) and Ba and Sr for the enrichment of sulfate minerals (e.g. strontio-barite) can be noticed (Tab. 1). Furthermore an enrichment of Cd, Sb, Se and Tl is visible. In the oxidized MA Fe and Mn own the highest element contents in reference to the formation of ore minerals. Here in comparison of all samples As and V show the highest values. In the psilomelane-like MA Mn shows the highest content. Furthermore the concentrations of Fe and Mo are increased. The contents of Pb, Zn, Ba, and Sr are in both, oxidized and psilomelane-like MA, depleted.

The contents of Ag, Au and Cu, those elements who often occur in economical amounts in VHMS deposits of middle ocean rift systems, are insignificant.

Table 1. Bulk geochemistry of the sulfide and manganese crusts of “Black point” analyzed by means of XRF analyses and ICP-MS analyses.

sample	Pb	Zn	Fe	Mn	Ba	Sr	As	Cd	Sb
XRF results	wt %	wt %	wt %	wt %	wt %	wt %	wt %	ppm	ppm
PAN-20 (MA I)	7,79	31,50	0,50	0,03	5,20	0,70	0,18	750	100
PAN-25 (MA I)	13,20	37,30	0,57	0,20	5,26	0,80	0,18	630	50
PAN-13 (MA I)	12,09	29,37	1,77	1,85	7,38	1,25	1,35	790	100
PAN-26 (MA II)	5,05	2,51	15,16	14,05	1,39	0,27	2,53	39	20
PAN-18 (MA III)	2,29	1,63	2,83	32,50	0,36	0,07	0,26	< LOD	< LOD
PAN-23 (MA III)	2,80	4,09	5,03	26,00	1,56	0,26	0,65	40	< LOD
PAN-04 (MA III)	3,31	3,65	1,63	31,03	0,55	0,09	0,21	< LOD	< LOD
PAN-11 (MA III)	2,00	3,42	2,27	35,00	0,26	0,05	0,24	< LOD	< LOD
PAN-07 (MA III)	3,34	9,05	1,27	24,00	1,91	0,25	0,15	410	< LOD
PAN-14 (MA III)	3,23	6,17	1,49	28,00	1,53	0,25	0,19	165	< LOD

ICP-MS results	Ag ppm	Au ppm	Cu ppm	Mo ppm	Se ppm	Tl ppm	V ppm
PAN-20 (MA I)	2	<4	6,6	29	24	110	<4
PAN-25 (MA I)	1	<4	8	100	23	120	26
PAN-13 (MA I)	2	<4	6,4	65	18	300	100
PAN-26 (MA II)	<0,1	<4	2,2	490	2	18	2300
PAN-18 (MA III)	<0,1	<4	3,8	580	2	43	700
PAN-23 (MA III)	<0,1	<4	3,7	350	4	11	540
PAN-04 (MA III)	<0,1	<4	5,4	410	2	20	240
PAN-11 (MA III)	<0,1	<4	3,6	450	3	13	330
PAN-07 (MA III)	0	<4	4,6	470	5	27	270
PAN-14 (MA III)	0	<4	4,4	440	4	40	280

Mineralogy

XRD analyses were performed on samples of all three different MA. The analyses of the lead-grey MA show that galena, sphalerite and strontio-barite are the major components. Analyses carried out on samples of the oxidized and the psilomelane-like MA reveal non meaningful results. It is supposed that the precipitates of these layers exhibit amorphous character.

By means of SEM-EDX analyses the results of the XRD analyses could be verified. The interpretation of SEM-microphotographs shows that galena, sphalerite and strontio-barite appear partly in perfect idiomorphic shape. Thereby the galena aggregates exhibit in octahedral, the sphalerite aggregates in spheroidal and the strontio-barite aggregates in tabular habits. In addition galena aggregates which are massive ingrown with each other can be observed. Special features represent on the one hand the columnar and dendritic shape of some Pb-S aggregates and on the other hand the existence of arsenic in some galena aggregates.

Figure 3 shows selected SEM-microphotographs in an overview. The identification of aggregates of the psilomelane-like MA by means of SEM-EDX analyses failed. The data of the EDX analyses carried out on aggregates illustrated in Fig. 3 are shown in Table 2.

Table 2. EDX data of in Fig. 3 presented SEM-microphotographs (MP = measuring point)

MP	Pb wt %	As wt %	S wt %	Zn wt %	S wt %	BaO wt %	SrO wt %	SO ₃ wt %
1	85,68	3,14	11,18					
2	69,88	13,56	16,56					
3				60,17	38,76			
4				62,79	36,89			
5				63,59	36,13			40,57
6						50,76	5,89	
7	73,89	6,79	19,32					
8	87,06	-	12,94					
9	90,54	-	9,46					
10	92,38	-	7,62					
11				63,35	36,65			
12						53,42	7,14	39,44
13						58,72	4,01	37,27

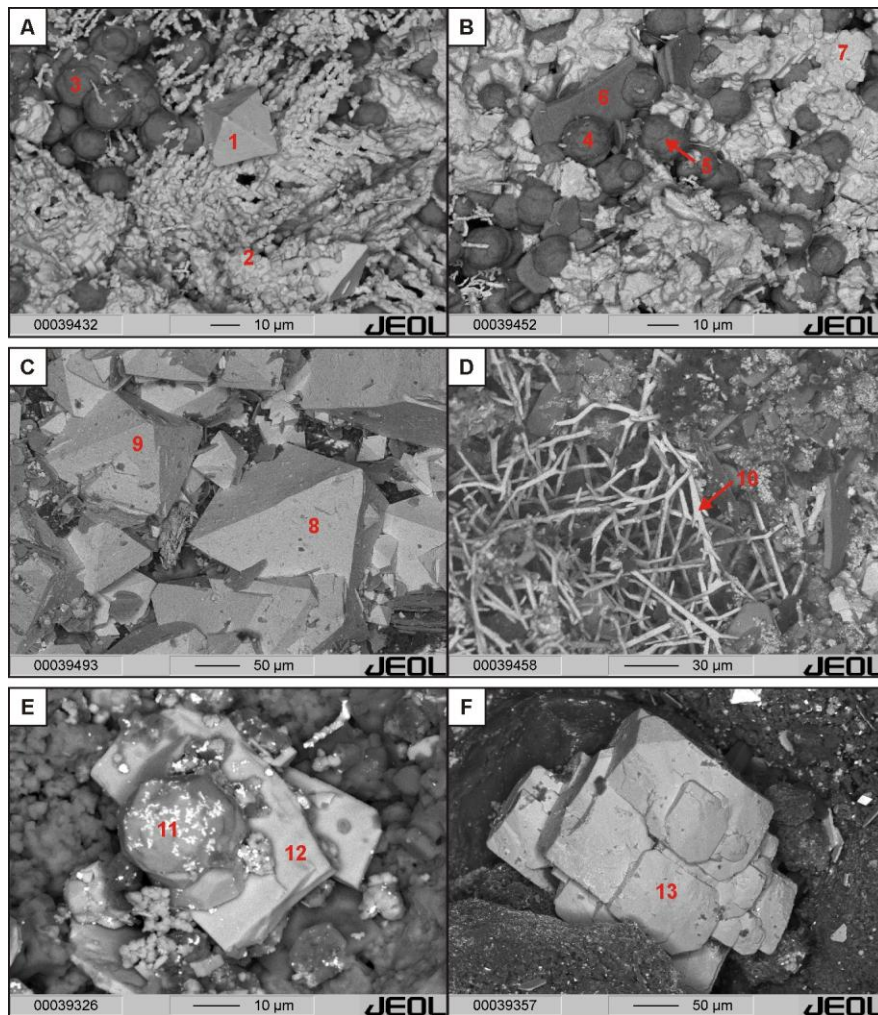


Fig.3. SEM microphotographs of specimen sampled at rock complex “Black point”, the red numbers illustrate the measuring point of the EDX-analysis: (A) octahedral galena aggregates (lightgrey) besides globular sphalerite aggregates (darkgrey); (B) massive ingrown galena crystals next to globular sphalerite and tabular strontioarite aggregates; (C) perfect idiomorphic and octahedral galena crystals; (D) columnar Pb-S-aggregates in dendritic shape; (E) idiomorphic sphalerite aggregate grown up on a tabular strontioarite aggregate; (F) perfect idiomorphic strontioarite aggregate in tabular shape.

Conclusions

Recent VHMS formation can be observed in shallow-water depths on a submarine plateau eastward of Panarea. The ore forming processes are limited to areas where hydrothermal fluids are discharging. These discharges are characterized by water temperatures reaching a measured maximum of 134.8°C. The chemical composition is described elsewhere (Sieland 2009).

The rock complex of “Black Point” can be subdivided into 3 distinct mineral associations which are zonally arranged. The inner lead-grey mineral association is made up of typical VHMS minerals such as galena and sphalerite. Strontioarite as gangue mineral is socialized with these minerals. The middle oxidized layer and the outer psilomelane-like layer are typified by high Mn and Fe contents. Due to their amorphous character no precise predication about their mineralogical composition can be done.

Finally it is concluded, that the massive sulfide deposition of Panarea is controlled by epithermal ore forming processes.

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The Underwater Archaeology in the 3rd millennium: legal and technical aspects.

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Abstract. For Underwater Archaeology the XX century means a new era for the approval of the UNESCO Convention of 2001 (coming into force on February, 2009) for the protection of Underwater Cultural Heritage. International opinion acquires conscience of the necessity of preserving this heritage for future generations; at the same time, the techniques applied for its study, recovery and preservation have experienced an enormous progress. Eventually the capacity to diffuse quickly the information and the existence of mass-media aware with that necessity of protecting the heritage shows us that we are in disposition of getting an educational, cultural and social utility from it.

General Aspects

The big changes experienced by the Archaeological Underwater Heritage in the last decades have been fundamental in every aspect, those which have incidence in scientific development, as well as those which influence in a significant way legal and administrative aspects, and those of management or of the specialized personnel's training.

This situation does not have the same importance in other scientific fields that have a direct relationship with underwater world, as Geology, Biology or marine Environment or of inner waters; but it does in a special way in the Cultural Underwater Heritage that is lately acquiring a special prominence among the sciences that are in charge of the research in this underwater field.

Until the 80's, Underwater Archaeology, former simply known as Submarine Archaeology, played a linear and simple development linked to the progress in the field of Land Archaeology on one hand and to those of the marine sciences and the development of the autonomous diving on the other one.

It had been incorporated in a marginal way to the scientific underwater research like an appendix of the land Archaeology and with a very stereotyped view by administrations and society. There was also a simplistic image, related with adventure, with fantastic treasures and with recoveries of objects and materials that were, in many cases, far from any scientific planning.

Some spectacular discoveries, as the Viking ships of Roskilde (DK), Vasa (SW), Mary Rose (UK), Yassi Ada or Ulu Burun (TK), Madrague of Giens (FR), Commacchio (IT), Culip IV (SP), were gradually creating a different conscience about this underwater heritage and opening the doors to their incorporation like historical science once and for all. On the other hand, they were the interventions of groups of firms, those commonly known as treasure hunters, whose merely commercial interests caused, and are still causing, serious damages to a common heritage of the world that needs a special protection and a civic conscience more understanding and committed in their preservation.

Nevertheless, these difficulties and the beginnings experienced previously, from the early XX century, which are always difficult in any science, marked a deciding inflection that only a quarter of century later have reached a positive inflection point that doesn't have going back.

Legislative interventions

In those years and as a consequence of the Convention of the United Nations on the Right of the Sea of Montego Bay, the international community opened the doors to the necessity of editing a specific convention by the UNESCO to be in charge of establishing an international document able to protect and to regulate the activities in connection with that important Underwater Cultural Heritage which the Convention of United Nations itself had studied in a not very deep way, due to the huge quantity of topics it had to regulate and to the specificity of the problems that outlined the Underwater Cultural Heritage.

The Council of Europe carried out a first attempt in the 80's. A document was drawn up, a Convention that once approved, didn't go into effect because of the blockade of some countries (Greece and Turkey).

Later on ICOMOS approved and published the famous Charter for the Protection of Archaeological Underwater Heritage, a mainly technical document but of great utility. Eventually the UNESCO concluded in December, 2001 and approved by majority the Convention for the Protection of the Underwater Cultural Heritage, not without big difficulties and the remaining opposition of many countries. The Convention has finally come into effect in February, 2009, after the ratification of the twenty member states, among them Spain. In the current moment they are twenty-four the countries that have ratified the Convention and the quick incorporation from most of the states that have interests in this field is expected.

We can say that as a consequence of this important legislative measure, the countdown for the gradual disappearance of the companies of rescue of treasures has begun, as one of the main objectives of the Convention is the total prohibition of those activities which have among its objectives the commercial exploitation of the Cultural Underwater Heritage.

The adoption of an international legal instrument, at its highest level, as this UNESCO Convention of 2001, has as immediate consequence the adaptation to it of numerous national laws or the creation of other specific ones in the member states as well as the creation in the member states of administrative structures or adaptation of the existent ones to the created new reality.

The international cooperation and the training

In the same way, the increase of international cooperation in this field for the protection as well as for the investigation or the transfer of knowledge and applicable technologies to that investigation to facilitate its access to the less developed countries, but with an abundant heritage in their waters.

After legal protection, specialized personnel training is, without a doubt, the high-priority objective. The new situation can quickly create a need of qualified personnel that at this precise moment the committed states with this investigation and those that will consent immediately, are not under conditions of offering.

Some countries: France, Spain, Italy, Germany, Denmark, Sweden, Switzerland, Holland, United Kingdom, Portugal, Canada, USA, Turkey and Australia can be able to face up to a project of some importance, but unfortunately other do not, mainly because of lack of material infrastructures and of human and technical resources needed for this aim.

Nowadays scientific training in this field is one of the high-priority challenges and the necessary channels should be established to facilitate that training and the exchange of experiences and training programs at different levels, which are necessary, from the scientists of high preparation level, to the intermediate technicians, specialists in special works, qualified operatives, restorers, specialists in complementary works, etc.

Training should be taught at scientific institutions of high level like universities, faculties and colleges, as well as at institutions and public or private organisms of other levels. The old concept of underwater works should be ruled out as regards archaeological heritage only carried out by archaeologists, nowadays scientific teams should be multidisciplinary and with specialists' intervention with different training levels.

In the field of training for underwater researches special attention should be paid to the training in scientific diving techniques. At the present time the Scientific Diver according to the CMAS Standards, worldwide

recognized and developed within the U.E. (Division IX), constitutes the best guarantee for that necessary training required to the scientists, technicians and specialists that have to develop part of their work under the water. The training program in Scientific Diving in two levels for the diver and two levels for the instructor, established by CMAS, guarantees that essential training for the good operation of the investigation teams which develop a part of its activity under the waters.

Adjustment of administrative and material infrastructures

At the present time the proliferation of situations that require urgent interventions or programmed investigation projects are forcing the public administrations that negotiate the underwater heritage to adapt their structures to be capable of administrating research projects that may come from scientific institutions. It implies the creation of administrative services and qualified technicians, for the evaluation of projects and the later necessary inspection. It is here where nowadays in most of the countries we appreciate remarkable lacks that prevent from an effectiveness of necessary management according to the UNESCO Convention, 2001 (in force since February, 2009).

If personnel infrastructure is important the economic one is too, the available means to face up to the projects, public financing as well as private one or both, and certainly the infrastructure in the field of conservation and recovery of underwater objects and structures or conserved in situ. Laboratories and special installations due to its economic cost should be rationalized and shared setting up national or regional centres and even international referenced centres.

One of the problems of permanent relevance is that there does not exist enough installations for treatment and preservation of archaeological materials coming from the seas. On one hand that lack causes the immediate limitation of projects authorized by the competent administrations in the matter and at the same time they prevent new techniques from developing to advance general process.

In this field we are almost the same as four decades ago when some of the most important projects were developed as Vasa, Mary Rose, Mainz, Skuldelev, Marseille, etc. Later on they have decreased for economic and technical reasons, but mainly because of the indifference of the responsible institutions to face projects of economic high cost.

Underwater Cultural Heritage and Society

One of the current challenges is to pass on the new reality in this matter to international society; the necessity of having a scientific knowledge and some material resources and enough technicians as to be able to protect and to recover this heritage.

Society in general has an incomplete and biased knowledge of the reality of the interest of this heritage for research, education and cultural enjoyment by the world. It is essential nowadays that scientific institutions and the associations implied intercede in the appropriate spreading of messages in the correct sense, that it is about a heritage that should be preserved for the future and that it is a heritage dedicated to education and general cultural enjoyment, never to get economic benefit and less still to allow their privatization or trade.

In this sense the disseminating campaigns, social awareness and spreading in the different levels, from scientific magazines and publications of high level to the simplest, of general spreading, exhibitions, programs of primary education, etc., they should be applicable resources in each case to facilitate those common objectives that the UNESCO Convention, 2001 proposes in benefit of the World.

All implied institutions, state or regional organisms, scientific and educational institutions, civic associations and the media will work jointly in education campaigns and spreading of the necessity of having this heritage for the educational, cultural and social mentioned aims, but never for their commercial and economic exploitation.

At the present time, when the world is going through a strong economic crisis, we should look for innovative solutions that allow having means to apply them in the protection and recovery of this underwater cultural heritage and be conscious of the important source of richness it can mean, from research, introduction

to the public, cultural tourism promotion, etc. It is a source of richness and employment that should not be forgotten because it can help in a very active and important way in this global fight against the economic depression and the unemployment.

Research in Underwater Archaeology: some challenges and approaches for the future

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Abstract. Abstract: This paper suggests that rather than changes in methods or advances in technology it is changes at an International level as a result of the bringing into force of the 2001 *UNESCO Convention on the Protection of the Underwater Cultural Heritage* that will provide the greatest challenges to the way in which research in underwater archaeology will be conducted in the future. By providing a framework of standards of practice and encouraging nations to collaborate and co-operate in underwater archaeological research and training, the UNESCO Convention 2001 will fundamentally change the ways in which underwater archaeological research is conducted in many countries of the world.

Introduction

Underwater archaeology can be defined in terms of the environment in which archaeology, or the systematic study of past human life, behaviors, activities and cultures using physical (or material) remains (including sites, structures and artifacts) and the relationships between them, is undertaken. It is where archaeological evidence is found in the underwater (or submerged) environment and such evidence may exist beneath fresh (or inland) waters or beneath salt (or marine) waters. It may be visible on the bed of the water body (i.e. seabed) or it may be buried beneath sediment. Research on underwater archaeological sites can include the remains of ships (shipwrecks), boats (boat finds), other watercraft or vessels and aircraft underwater as well as cultural material that was accidentally dropped, lost overboard or deliberately deposited into the water body. Archaeological evidence can also include the remains of structures that were originally built wholly or partly underwater (such as fish-traps, crannogs, bridges, piers, jetties and wharves). In addition underwater archaeology may examine the remains of human activity that originally took place on dry or marshy land that has subsequently been inundated (or submerged) by processes such as rising water levels or by marine (or fluvial) erosion (Adams 2002; Bowens 2009:2-10; Delgado 1997; Delgado & Staniforth 2002; Flatman & Staniforth 2006; Staniforth 2007; Staniforth & Nash 2006).

Methods in Underwater Archaeology

The development of the Self-Contained Underwater Breathing Apparatus (SCUBA) and its expanding use after WW2 also saw the emergence of underwater archaeology as a sub-discipline of archaeology. SCUBA diving using compressed air allowed archaeologists to enter the underwater environment and by the 1950s scholars were beginning to recognize the archaeological potential of underwater sites such as shipwrecks. It was the 1960s that saw the development of many of the now standard methods and techniques used for archaeological work underwater by researchers such as Dr. George F. Bass and his colleagues working on Classical period shipwrecks sites in the Mediterranean (Bass 1966, 1972). Underwater site surveying, recording, and excavation methods using air-lifts and water-dredges have become widely adopted and well established over the past half century and are used by underwater archaeologists around the world (Bowens 2009; Green 2004). Perhaps the greatest changes to methodology in underwater archaeology in recent years have been in the areas of establishing site position such as the use of Global Positioning Systems (GPS), the replacement of traditional film cameras with digital photography and other activities conducted after the diving is over (or above water) employing computer technology such as Geographical Information Systems (GIS) and Site Recorder software (see, for example, <http://3hconsulting.com/>).

Over the past fifty years SCUBA diving has had, and still has, limits and working underwater is commonly restricted to shallow water depths of less than about 40 meters. The development over the last twenty

years of "mixed gas" diving using different mixtures of nitrogen, oxygen and helium such as NITROX and TRIMIX has allowed underwater archaeologists to dive to greater depths as well as to spend longer on underwater sites. Nevertheless, Occupational Health and Safety (O,H & S) regulations, coldwater temperatures and water depths beyond 10 meters usually severely restrict the time that can be spent on an underwater site to a fraction of the time each day that terrestrial archaeologists can spend on a comparable land site. Many of these factors, together with the associated costs, are proving to be significant, and enduring, limitations on underwater archaeological research and it is unlikely that comprehensive, technologically-based answers are likely to be found in the short-term future.

Regulating Underwater Archaeology

The most common mechanism by which underwater archaeological sites throughout the world are protected is by using cultural heritage legislation (Bowens 2009:45-52). In 1982 the United Nations Convention of the Law of the Sea (UNCLOS) provided that 'States have the duty to protect objects of an archaeological and historical nature found at sea and shall cooperate for this purpose' (Fletcher-Tomenius & Forrest 2000). National and state or provincial governments have the right to enact and enforce legislation and regulations for the protection and preservation of underwater cultural heritage lying in or under their internal waters, territorial seas and Exclusive Economic Zone (EEZ), in some cases as far out as the edge of the Continental Shelf. Unfortunately some countries have chosen not to protect their underwater cultural heritage and continue to allow treasure-hunting to take place within their jurisdictions. Furthermore, in most cases, there has been little or no regulation of underwater archaeology beyond the limits of state territorial jurisdictions.

In addition, many national governments retain title to (or ownership of) ship and aircraft wrecks that once formed a part of that nation's military forces (Army, Navy or Air-force). These property rights are not lost to that government due to the passage of time and apply whether the vessel or aircraft was lost in national, foreign or international waters (Dromgoole 1996; Williams 2001). Some countries such as the UK and USA have enacted specific legislation which addresses and controls activities aimed at the military vessels component of underwater cultural heritage even when that cultural heritage lies in International waters or within the jurisdiction of another nation.

In more recent years, multilateral attempts to control activity on, and to create standards for the conduct of archaeological research on, underwater cultural heritage sites, such as shipwrecks, have led to the creation of firstly the ICOMOS *Charter for the Protection and Management of Underwater Cultural Heritage* 1996 see:

http://www.international.icomos.org/charters/underwater_e.htm

secondly, and perhaps more significantly, the UNESCO *Convention on the Protection of the Underwater Cultural Heritage* 2001

http://portal.unesco.org/en/ev.php-URL_ID=13520&URL_DO=DO_TOPIC&URL_SECTION=201.html

Challenges for the future

The 2001 *UNESCO Convention on the Protection of the Underwater Cultural Heritage* (UNESCO Convention 2001) entered into force on 2 January 2009. This milestone was achieved after the deposit of the 20th instrument of ratification, acceptance or approval by Member States of UNESCO. On 2 October 2008, Barbados became the 20th State Party to accept the 2001 Convention, As each subsequent country ratifies, after a three month waiting period, the Convention will also apply to that country (as of 25 April 2009 24 countries have ratified the Convention).

UNESCO classifies the world into five regional areas: Africa, Arab States, Asia and Pacific, Europe and North America, and Latin America and the Caribbean. According to these regional divisions, the 24 countries that have joined the 2001 *UNESCO Convention on the Protection of the Underwater Cultural Heritage* are: Africa (3) – Libyan Arab Jamahiriya, Nigeria, and Tunisia; Arab States (2) – * Libyan Arab Jamahiriya [Note this country is classified in two regional areas], and Lebanon; Asia and Pacific (1) - Cambodia; Europe and North America (11) – Croatia, Spain, Lithuania, Bulgaria, Portugal, Ukraine, Romania, Montenegro, Slovenia, Slovakia, and Albania; and Latin America and the Caribbean (8) – Panama, Mexico, Paraguay, Ecuador, Saint Lucia, Cuba, Barbados, and Grenada. It should be noted that three-quarters of the signatories are from Europe, Latin America and the Caribbean.

The inaugural meeting of States Parties to the 2001 *UNESCO Convention on the Protection of the Underwater Cultural Heritage* took place on 26–27 March 2009 at the UNESCO Headquarters in Paris.

Present at the meeting were 19 of the 20 States Parties to the Convention with the right to vote, while Albania, Grenada, Slovakia and Tunisia attended as observers (they ratified the Convention less than three months before the meeting). Attending as observers were 71 States non-Parties to the Convention, 5 inter-governmental organizations, and 23 non-governmental organizations (NGOs). See the UNESCO website at: http://portal.unesco.org/culture/en/ev.php-URL_ID=33966&URL_DO=DO_TOPIC&URL_SECTION=201.html

New approaches

The coming into force of the 2001 UNESCO Convention marks the beginning of a process whereby it will become an important international instrument for the protection and management of the world's fragile, finite and irreplaceable underwater cultural heritage. The Convention provides clear guidelines for the protection of underwater cultural heritage in the form of the Annex (rules) to the Convention as well as minimum standards for the training of underwater archaeologists in Article 21 which states:

- States Parties shall cooperate in the provision of training in underwater archaeology, in techniques for the conservation of underwater cultural heritage and, on agreed terms, in the transfer of technology relating to underwater cultural heritage.

Even when a nation or state has not ratified the Convention, many are adopting the Annex to the Convention as guidelines for practice and the preservation of underwater cultural heritage. One of the new approaches that is likely to increase as a result of the implementation of the UNESCO Convention will be in the area of international cooperation and collaboration that is advocated in Article 19 on Cooperation and information-sharing, which states:

- States Parties shall cooperate and assist each other in the protection and management of underwater cultural heritage under this Convention, including, where practicable, collaborating in the investigation, excavation, documentation, conservation, study and presentation of such heritage.

The international nature of shipping, where a vessel may be built in one country, be sailing from a second country to a third country and have a crew made up of citizens of many countries, often leads to situations where there may be more than one nation with interests in a particular underwater archaeological site and its contents. Finding multi-lateral ways of recognizing different countries' interests is going to require new approaches which accept, and embrace, notions of shared heritage rather than, say, concepts of 'national' heritage (see, for example :

http://www.ikuwa3.com/documents/shared_heritage_seminar.pdf

The UNESCO Convention 2001 will provide the greatest challenges to the way in which research in underwater archaeology will be conducted in the future. By providing a framework of standards of practice and by encouraging nations to collaborate and co-operate in underwater archaeological research and training, the UNESCO Convention 2001 will fundamentally change the ways in which underwater archaeological research is conducted in many countries of the world. Over time, it should result in the reduction of treasure-hunting and an increase in the quality and quantity of underwater archaeological research throughout the world.

An Australian case study

The Department of Archaeology at Flinders University is currently the only university department in the Asia-Pacific region to develop, and regularly conduct, an integrated program of teaching in maritime archaeology from undergraduate to research higher degree level called the Maritime Archaeology Program (MAP) (Staniforth 2008a, 2008b). One model for the implementation of Articles 19 and 21 of the UNESCO Convention 2001 that has been developed by MAP is the 2009 *Flinders University Intensive Program in Underwater Cultural Heritage Management (UCHM)*. This program, largely funded by the AusAID Australian Leadership Awards-Fellowship Program, brought 10 mid-career professionals involved in maritime archaeology from the Asia-Pacific region (represented nations were Sri Lanka, Thailand, Cambodia, Indonesia and the Philippines) to Australia for a six week intensive training program from 19th January 2009 to 2nd March 2009. The Australian Leadership Awards - Fellowships are a component of a regional program that aims to develop leadership and build partnerships and linkages with the Asia-Pacific. They are intended for those who are already leaders or have the potential to assume leadership roles that can influence social

and economic policy reform and development outcomes, both in their own countries and in the Asia-Pacific region.

While in Australia, the ALA Fellows undertook a one-week intensive topic in vessel construction and analysis, a one-week intensive topic in underwater cultural heritage management and a two-week intensive field school at Mount Dutton Bay, South Australia. These topics can provide ALA Fellows the credentials needed to achieve a Graduate Certificate in Maritime Archaeology (GCMA) from Flinders University. The program also allowed the ALA Fellows to receive AIMA/NAS Part 1 Training and DAN Asia-Pacific First Aid and O2 certification. Following the field school, ALA Fellows undertook a two week placement (internship) with a museum, underwater cultural heritage agency or related organization. Partner agencies included Heritage Victoria, Heritage South Australia, Australian National Maritime Museum, New South Wales Heritage Office, Western Australian Maritime Museum, and the Department of Environment & Heritage, Canberra. The 2009 Flinders University Intensive Program in UCHM was managed by Flinders Partners, the Maritime Archaeology Program (MAP) in the Department of Archaeology, and Flinders University of South Australia, in partnership with the Australian host organizations and the ALA Fellows overseas counterpart organizations.

Conclusion

The bringing into force of the 2001 *UNESCO Convention on the Protection of the Underwater Cultural Heritage* will provide the greatest challenges to the way in which research in underwater archaeology will be conducted in the future. By providing a framework of standards of practice and encouraging nations to collaborate and co-operate in underwater archaeological research and training, the UNESCO Convention 2001 will fundamentally change the ways in which underwater archaeological research is conducted in many countries of the world. The Maritime Archaeology Program (MAP) at Flinders University has developed the 2009 Flinders University Intensive Program in Underwater Cultural Heritage Management (UCHM) as a model for the implementation of Articles 19 and 21 of the UNESCO Convention 2001. It is hoped that this model will be adopted by other countries and organizations in the Asia-Pacific region resulting in increased numbers of ratifications of the Convention and as the basis for effective collaboration and co-operation in both the training of underwater archaeologists and in conducting underwater archaeological research.

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Evidence of tectonic control on active arc volcanism: the Panarea-Stromboli tectonic link inferred by submarine hydrothermal vents monitoring (Aeolian arc, Italy)

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Abstract. A continuous gas flow monitoring carried of the submarine hydrothermal vents off the island of Panarea has shown contemporaneous modifications of the gas flow rate at Panarea and the seismic activity at Stromboli. The possibility that a tectonic link joins the two volcanic edifices of Panarea and Stromboli (Aeolian islands, Italy) fits with the contemporary recording of the same $3\text{He}/4\text{He}$ ratios at both the islands during the 2002-2003 crisis. The almost contemporary beginning of the last eruptive activity at Stromboli in 2007 is here considered as a possible evidence for an interconnected magmatic fluid reservoir which feeds also the Panarea edifice. The contemporary anomalous release of magmatic fluids from deep geothermal reservoirs of both volcanoes implies a pressure signal triggering via a $\text{N}40^\circ\text{E}$ normal fault system visible by a large number of submarine crater-shaped structures which links both volcanoes.

The long term records at Panarea of outlet temperature of submarine vents, the degassing rate and the sea bottom pressure from September 2006 to April 2007 as well the geochemical data from the discontinuous-based monitoring by sampling methods, in particular the analysis of $3\text{He}/4\text{He}$ ratios, will be discussed here.

Flow rate measurements at submarine volcanic gas emissions

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Abstract. In November 2002 the marine sector surrounding the island of Panarea (Aeolian Islands, Italy) has been affected by strong gas eruptions. Since then this area has been in increasing focus of interest concerning research of volcanic activities. However, the existence of submarine gas emissions has been known since the Roman age. The aim of this paper was to investigate the unsteady gas discharges at selected fumaroles in order to clarify a relationship between gas flow and geothermal activities. Therefore a new device for the acquisition of submarine gas emissions was developed and tested during scuba dives. The first results of the measurements are presented herein.

1 Introduction

The Aeolian Islands consist of 7 major islands where Panarea is the smallest of these. Panarea is located 20 km southwest of Stromboli and 20 km northeast of Lipari and Vulcano (Fig.1). Together with the small islets Dattilo, Panarelli, Lisca Nera, Bottaro and Lisca Bianca, Panarea forms a large caldera of a formerly active volcano (Fig.2). Inside this caldera, the small islets were recognized as the remnants of a crater rim (Gabbianelli et al., 1990).

In November 2002 gas eruptions offshore the Island Panarea triggered many geological investigations as well as the study presented in this paper. Caliro et al. (2004) made the assumption of a new magmatic input in this area. Comparing the degassing activity to the situation before 2002 the appearance of five new areas of intense submarine degassing was recognized. However their activity decreased during the subsequent days.

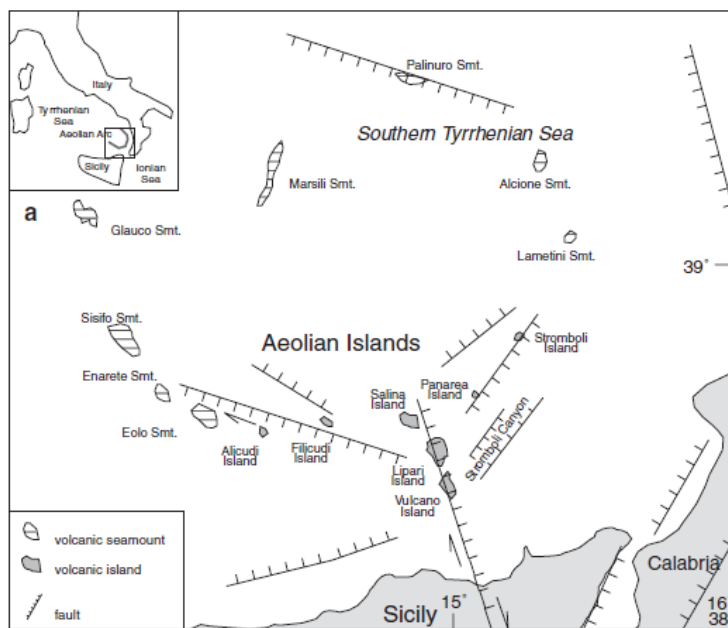


Fig.1. Location of Aeolian Islands, Chiodini et al. (2006)

Exhalative activity at Panarea has been well known since historical times as reported in Mercalli (1883); Romano (1973); Gabbianelli et al. (1990); Italiano and Nuccio (1991); Calanchi et al. (1995) and Anzidei (2000) although gas eruptions of this intensity have not previously been observed. The most intense gas flux was measured immediately to the west of the islet of Bottaro, where just one vent was erupting at 108–109 l/day (Caracausi et al., 2004; Caliro et al., 2004), about two orders of magnitude more than the average total gas flux previously measured in the area. Italiano and Nuccio (1991) also investigated the submarine degassing near Panarea before 2002 in detail. They identified gas emissions down to the depth of 150m by sonar. The gases generally come out from open fractures of rocks belonging to the main tectonic directrices. Geochemical analyses evidenced constant high He/N₂ ratio suggesting that all fumaroles are fed by the same deep hot fluids. The main gas component is CO₂ with a portion of more than 90% at all fumaroles. Additionally all fumaroles were marked by snow like deposits of colloidal sulphur. They further measured an average flow rate of several fumaroles in order to receive information about the thermal energy dissipated by the geothermal system.

For our investigations flow rate measurements were also in the focus of interest. However besides the total amount of gas emitted by the fumaroles we were interested in the changes of flow rate over time. Therefore a new device suitable for submarine flow rate measurements had to be developed. Most important was the material resistance against these aggressive conditions caused not only by saline water but also by the high acidity level and hot temperatures. Temperatures measured in selected fumaroles were in the range between 30 and 100°C. Additionally, sediment of bacteria and growth of plant within the working section had to be excluded. First measurements were used to prove the feasibility of the measurement technique and the material resistance. The measurement system was set up during scuba dives in the area 2.5km east of Panarea at Point 21 (see Fig.2).

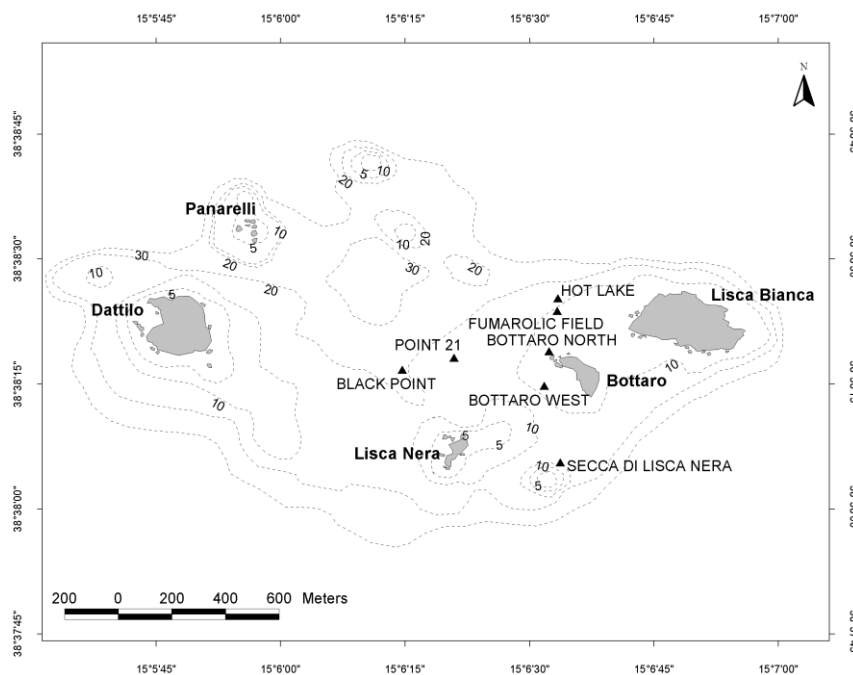


Fig.2. detailed map of diving area (Rohland 2007)

2 Measurement technique

As mentioned above due to the aggressive submarine conditions standard flow rate measurement techniques could not be used and a new set-up had to be developed.

A flexible funnel with a maximum diameter of 0.65m was mounted on top of a fumarole at a distance of about 0.3m from the seafloor (Fig.3). To guarantee the stability of the set-up the funnel was fixed by 5 weights of 12 kg each on the sea floor. The weight was necessary to compensate the buoyancy of the gas entering the funnel.

The diameter of the funnel had to be large enough to cover a complete fumarole. Since at the exit of the fumarole the gas is mixed with water the gas had to be separated from the water first. The pure gas is then guided through a straight tube in which an impeller is driven by the gas flow. The rotation frequency was acquired by Neodymium magnets mounted on top of the impeller blades and inducing a voltage in a coil mounted outside of the tube very close to the rotor blades. This frequency of the induced voltage corresponds to the rotation speed of the impeller. The data were stored on a flashcard during the measurements by a computer (Blackfin, Bluetechnix). The rotation speed is proportional to the mass flow rate through tube which could be derived after calibration. If the gas flow was not completely separated from water, bacteria living in the water are transported to the measuring section. Here they are given the chance to settle on several parts, e.g. the impeller. Consequently the measurements are distorted. Fig.3 shows the principle of the separation of water. By a the tube system the flow is first forced down where the remaining water leaves the system. The pure gas flow will rise up to the measuring section.

The material used for the set-up was chosen to resist the adverse conditions at the area of investigation. Mainly materials made of plastics and Plexiglas were used. However the shaft and the bearings at which the impeller was mounted had to be made of more mechanically robust material. Therefore, the shaft was made of carbon and glass bearings were used.

Very important is the autarkic power supply which was a 3.3Ah, 12V battery. Together with the computer for data storage the measurement duration was limited to only 12 hours. It was known that for a significant interpretation of the measurements this duration is not large enough but sufficient for the prove of feasibility. According to the measurement duration the frequency of saving data could be adapted.

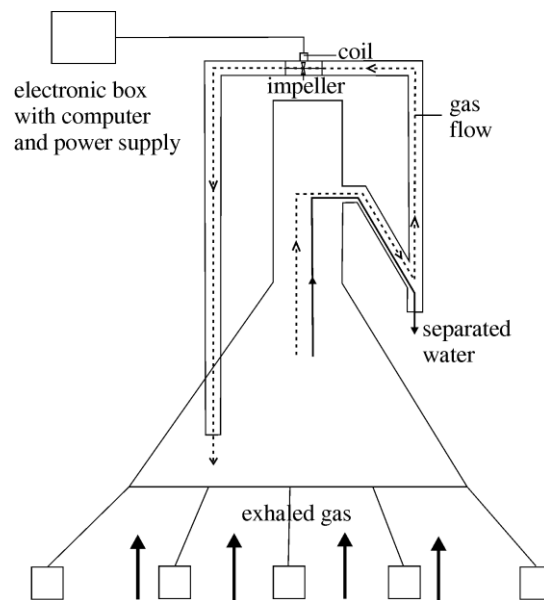


Fig.3. schematical set-up of measurement system

3 Field Measurements

As mentioned above large exhalation fields were found at Bottaro. However recent dives have shown that their intensity has decreased. In order to receive a reliable signal the minimum flow rate had to be 40 l/min. Below, the rotor drag could not be overcome. Hence, the location called Point 21 has been chosen (see Fig.2). The number indicates the depth. This location is characterized by strong fumaroles located close to a steep face (see Fig. 4). Flow rate measurements could be performed at two different fumaroles. Their exact location is shown in Fig.4 (left). The fumarole at position 1 is at an exact depth of 20m, its temperature was measured at 71°C. The second fumarole investigated is at a slightly deeper level (20.6m) but its temperature is only 31°C.

The recording frequency of the voltage which was induced by the rotating magnets in the coil was set to 4 per minute, the duration is 1s. The computer allows a sample rate of 96kHz. Hence, one record consists of 96000 values for the voltage.

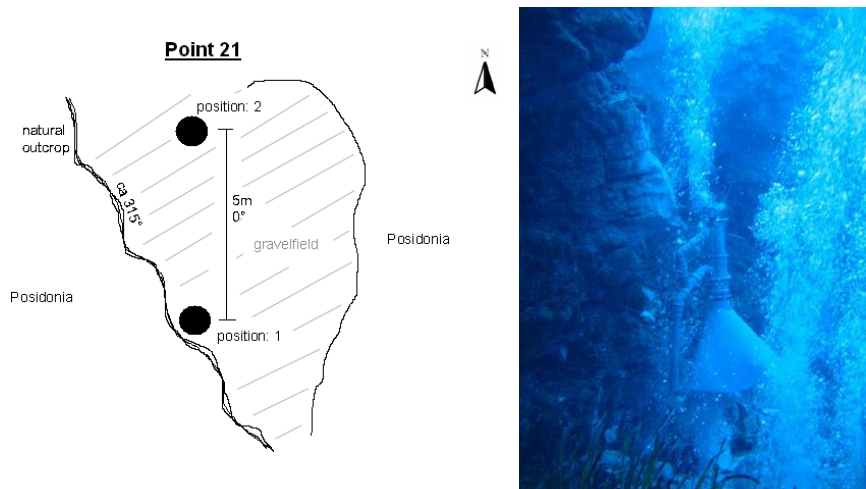


Fig.4. location of the fumaroles (left), photograph of the set-up mounted on top of the fumarole at position 1 (right)

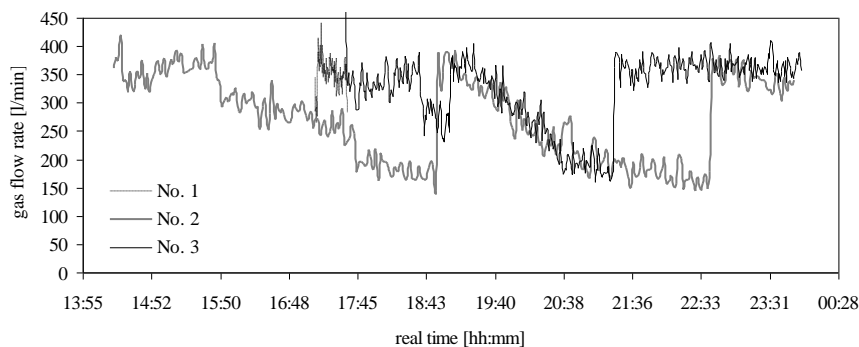
4 Results

As mentioned above, with the current set-up a recording time with a maximum of only 12h could be reached due to the high power consumption of the computer. Nevertheless data for evaluation could be achieved. First measurement tests even were of shorter duration since it had to be checked whether the data acquisition works properly.

At the fumarole of position 1 the flow rate varies between 150 and 400l/min as indicated by measurement No. 2 shown in Fig.5a). The flow rate decreases continuously from 400l/min down to 150l/min during a time of about 4 hours, then the flow rate suddenly increases again up to 400l/min during a time of less than one minute. This behavior seems to have periodic character as suggested by measurement 2 and 3. However, measurement 3 is of even shorter duration and the periodicity less obvious. After the steep increase at 21:20h the flow rate seems to remain at the constant high level. Altogether three measurements have been carried out at position 1 at different days but similar times during the day.

The observations at fumarole at position 2 were in contrast to the findings of position 1. Besides fluctuations the flow rate remains at a constant level of about 300l/min (Fig.5b).

a) position 1 | temperature: 71°C | pressure: 3 bar



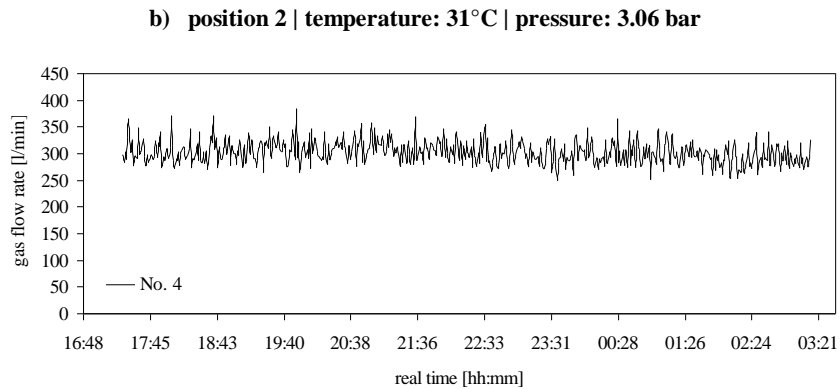


Fig.5. Gas flow rate over time at the fumaroles of position 1(a), and position 2(b)

5 Discussion

The measurements have proven the feasibility of the measurement technique. Data could be acquired and they are in a plausible range. Of course this technique needs strong improvement concerning the power supply. It has to be ensured that long term measurements, which are the final aim of this project, can be realized. The results have shown that the duration of the measurements can not provide statistically firm results. A periodicity of the flow rate at position 1 can be derived however a constant frequency can not be determined. Therefore longer measurements of at least 48h are mandatory. The same holds for position 2.

Indication for periodic gas flow has also been observed during earlier measurements (Görlitz, 2007). Here, the periodic time was in accordance with the tides, even though the flow rate was phase shifted. Consequently the tides themselves could not cause the change in flow rate.

However, hints for this kind of periodicity could not be found for our measurements. Consequently, other mechanisms have to be responsible for the varying gas flow rate observed at position 1. It was observed that after a maximum has been reached the gas flow decreases continuously. This could be explained by deposits or varying permeability of the gas channel of the fumarole. As soon as a critical pressure due to accumulating gas in the channel has been exceeded the deposits are discharged by rising gas. The flow rate suddenly increases and is starting to fall again due to new sedimentations or displacement effects within the sediment. The measurements at the fumarole of position 2 have shown that this does not hold for all fumaroles.

Acknowledgements

We thank Thorsten Hegewald for his technical support.

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DOMS: The new Diver-Operated Microsensor System

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Microsensors are powerful tools for microenvironment studies. One can investigate with high spatial and temporal resolution microorganisms, their physical and chemical environment, metabolic processes and interactions. Their application in shallow water ecosystems has been restricted to the lack of adequate equipment for in situ deployments. The new diver-operated motorized microsensor system allows underwater flexible field operations in fragile ecosystems such as coral reefs. The system has been tested to depths of 25 m, and the microsensors have a resolutions of up to 5 μm . Fine scale chemical and physical gradients of pH, hydrogen sulfide, oxygen, and light can be resolved in very short response times between 0.5 and 20 seconds, depending on the type of sensor. Two sensors can be applied simultaneously in a distance of 50 μm .

The *in situ* measuring system was already successfully applied in recording oxygen depletion and decreasing pH in sediments accumulated on sensitive scleractinian corals of the Great Barrier Reef in Australia, oxygen distribution in sand ripples and oxygen dynamics in a sedimentation experiment on red algae in the Mediterranean, Elba, Italy (Weber et. al, 2007). The new system provides a manual and very precise positioning, the choice of motorized and autonomous profiling. However, the diver has the control over all functions in order to interact with the system by a logger with a signal display. The external battery supplies can last for 24 hours to several days, as they can be changes via a Y-cable. Data logging is possible at 0.1 second interval over 74 days with a compact flash card (max. 64 million data points). Data transfer and programming is provided via USB and data can be assessed through the logger. Two different stand have been designed and can be applied depending on the divers needs. The whole set up fits in a small Zarges box and has a total weight of 10-20kg. For operation underwater a UTZ suitcase is used.

M. Weber, P. Färber, V. Meyer, C. Lott, G. Eickert, K.E. Fabricius, and D. de Beer (2007) *In situ* applications of a new diver-operated motorized microsensor profiler. ES&T, 41:6210-6215.

Underwater Temperature Measurement – the underestimated Basis for various Scientific Projects – an Example for Panarea, Italy

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Abstract. In the year 2002 a volcanic unrest north of Sicily led to a fairly massive submarine gas output. Since that time various scientific researches on different fields have been observing the gas and thermal water exhalations and the resulting environmental conditions. This could be useful to predetermine the time and intensity of seismic and volcanic events. The investigated area, part of the Aeolian Arc, is located next to Panarea. This is a neighbour island of the active volcano Stromboli. The work presented indicates the importance of temperature measurements as the basis for many scientific researches in general and in that particular, geologically, active area.

Motivation

The underwater temperature measurements carried out during the diving field trip in Panarea, Italy, mainly in 2007 are the basis for many further investigations in this area.

They are important for projects in different scientific fields. In geology related researches, temperature measurements can be used to compare with the results obtained through the application of geothermometer (e.g. Sieland 2009). In biology, measurements of temperature are urgently needed to find and identify possible living areas for any kind of flora, fauna and bacteria (e.g. Gebhardt 2006 and Lucas 2007). Besides, knowing only the pure living conditions of bacteria for further laboratory experiments the temperature was used as one factor in the mapping of micro-organism (Lucas und Nietzsche 2007). In chemistry, temperature measurements are needed for general analytics, especially water properties but also for gas analytics (e.g. Hamel 2007). In analytics, water and gas, pH-value and various solution equilibriums depend on temperature. In addition to that results obtained from temperature measurements have been used especially for sampling and analysis of fumarolic gases occurring at the sea-bottom near Panarea (e.g. Hartwig 2006, Steinbrückner 2009).

Furthermore, it is relevant in the large field of researches in geo-sciences. Knowing hot spots in temperature is essential and required for the identification of thermal-water and gas exhalations and the related mapping (e.g. Geier 2007). Based on these information one can draw conclusions on submarine geological features and structures.

Investigation Area

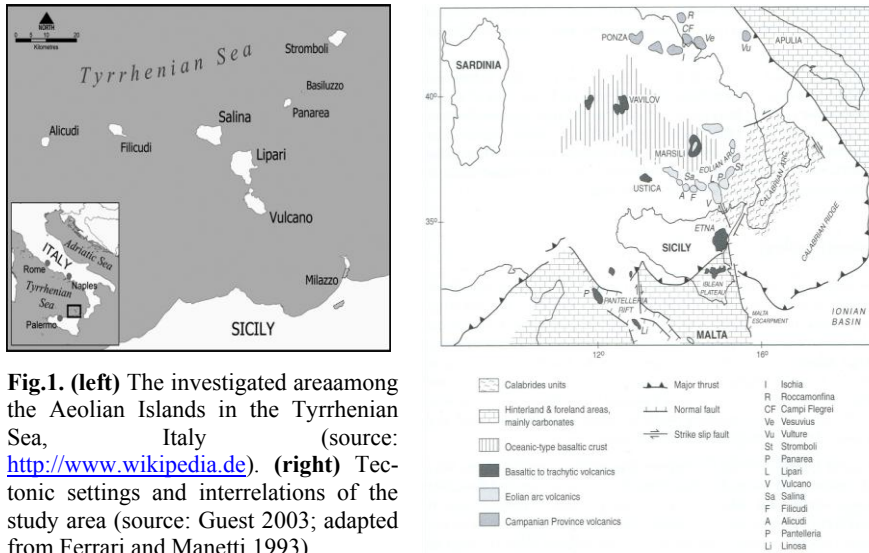


Fig.1. (left) The investigated area among the Aeolian Islands in the Tyrrhenian Sea, Italy (source: <http://www.wikipedia.de>). **(right)** Tectonic settings and interrelations of the study area (source: Guest 2003; adapted from Ferrari and Manetti 1993).

Characteristics of the Diving Spots

During the diving excursion 2007 temperature measurements were carried out at the three locations: “Hot Lake”, “Point 21” and “Black Point”.

Black Point

The location Black point is named relating to a characteristic black stone where a vent releases blackish hydrothermal water. The stone can be found at the edge of a gravel-field. Among this gravel-field there are various gas exhalations of different size and temperature.

Hot Lake

The location Hot Lake is named after a deepening of the seabed giving the impression of a small lake. The diameter of this structure can go up to 7 meter. Temperatures on the ground are higher than at the surrounding seafloor. Nonetheless, gas outlets in form of gas-bubbles could not be observed.

Point 21

This location to the west of Bottaro got its name from the depth of about 21 meter. The spot is characterized by a 4 meter high and 8 meter long stone wall where 5 big fumaroles can be found. Next to these five large spots several smaller ones with gas exhalations can be found.

Methods

First Approach

The submarine hydrothermal CO₂- emissions of the observed area are determined and observed with respect to the fact that these emissions predominantly consist of CO₂. Temperature measurements have been carried out by putting the measuring sensor into the sediment or rock cracks of the gas exhalations. These tempera-

ture values are the gas exhalation temperatures of the respective fumaroles. The volume flow rate determinations are combined with the temperature measurements. Afterwards the temperatures are used to calculate a reference temperature. This leads to the possibility to compare the spots with gas exhalations with one another. In addition to that the gas exhalations have been corrected with the average water depth for a better and easier comparison.

In general: the in-situ temperatures have been used and needed to correct the volume flow rates in relation to their temperatures and the corresponding hydrostatic pressure (Steinbrückner 2009).

Second Approach

Preparation

All the investigated locations are fairly easy to access and in depths in the range of 17 to 24 meter. Among these three locations we focused on the sediment field at “Black Point”. We carried out measurements in different depths. While we measured in depths of 25, 50 and 100 cm in “Hot Lake” to obtain a depth profile, we measured only in about 10 to 15 cm below ground at “Black Point” and “Point 21” due to the rock cover or too stony ground. In addition to that, we determined the temperature at some submarine fumaroles. These additional measurements have been done to identify changes in temperature and intensity compared to previous excursions and to give a basis for future researches.

Equipment

Basis for our measurements is a GMH 3350 measurement device with Alarm and Logger function (Fig. 2).

Technical Specifications

- Name: GMH 3350
- Surface temperature input T2: NiCr-Ni, Type “K” with a temperature drift of 0.01%/K
- Temperature range: -80°C to 250°C
- Logger function
- Probe:
 - diameter: 2 mm
 - length: up to 1 m

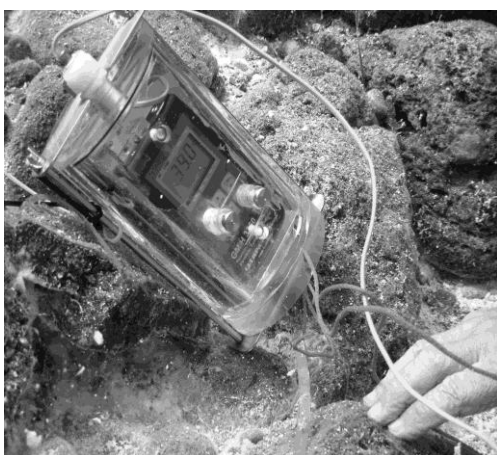


Fig.2. Temperature measurement

Using bigger probe diameter would result in too long response times which would finally result in a decrease of measurements per dive. The 2 mm diameter was identified as the ideal ratio of response time and stability for measurements even in stony sediments. For the identification of interesting measurement spots

and as orientation along the measurement transects (to have always the same measurement distance) specially prepared golf balls have been used as markers.

Implementation

The accomplishment of these measurements is a joint effort. The task of one diver was to write down all the data, while the other diver carried out the measurements and searched the spots. To make the measurements reproducible and to put the acquired data in the correct context, measurements on larger spots like "Black Point" were performed along transects. These transects had a distance from each another of 1 meter. For documentation purposes and to make each dive straight forward and effective the transect line was marked in an interval of 1 meter.

Beside the mentioned aim of fast, straight forward dives and measurements, the second huge advantage is the reproducibility. The use of transects, starting from a unique and in observed timescales unchangeable points, allows to redo the measurements in any future campaigns. Furthermore, it made us independent from promising but not yet trustful (due to its developing status) underwater GPS measurements (e.g. Rohland et al. 2006, 2007).

Immediately after each dive we transferred all data from the list to a hand drawn map before transferring data (true to scale) into the computer. For first analysis and visualisation we used MatLab®.

Results

The temperature measurements indicated the occurrence of hot and cold gas and water exhalations. At some particular locations temperatures above 90° C have been acquired within only some cm below the surface, while next to these "hot spots" temperatures in the range of the surrounding seawater have been measured.

At the location "Black Point" (the stone itself) temperatures of about 134°C have been monitored. That is close to the absolute maximum which is physically possible under the given (pressure) conditions.

Graphical analysis of the performed measurements was made by using a MatLab® script (Leidig 2007). Some examples of the findings can be found in Figure 3 and Figure 4.

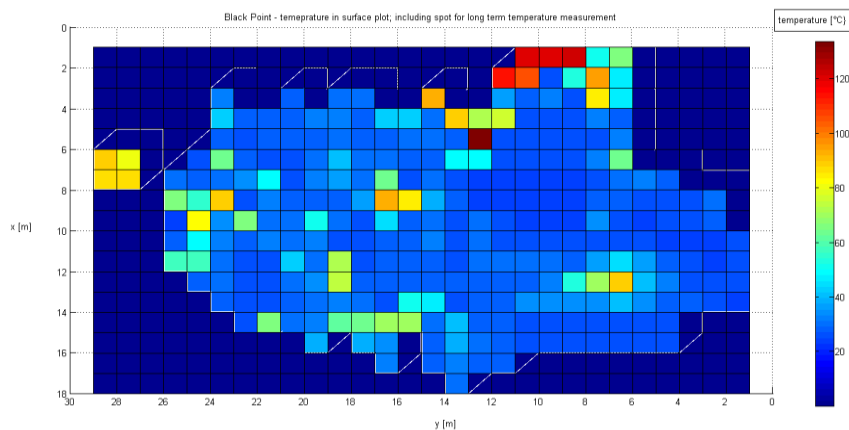


Fig.3. 2d view of the temperature readings at the location „Black point“. The highest temperature was monitored at the black stone. Temperatures on the gravel-field reach from about 25°C to about 93°C and even more at some fumaroles.

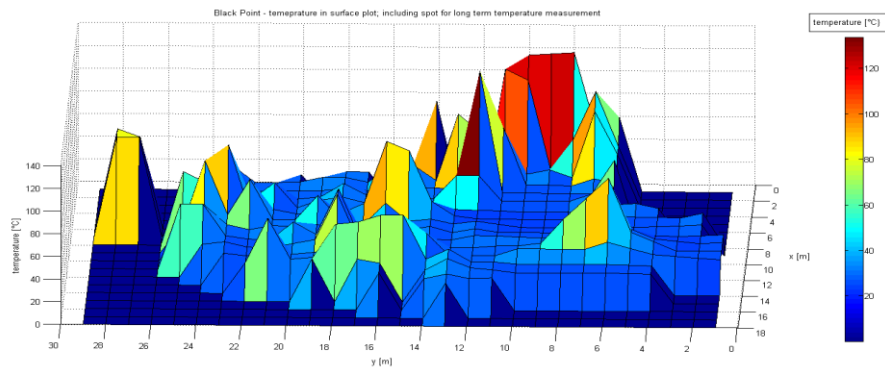


Fig.4. 3d plot of the temperature measurement at the location Black Point.



Fig.5. Temperature measurements in a depth of 50 cm at Hot Lake.

Due to the stony sediment it was not possible to take a depth profile at the Black Point location, but it was possible at Hot Lake. Measurements took place in depths of 25, 50 and 90 cm (Figure 5). Depending on the depth of measurement, temperatures varied in the range of 32°C and 94°C. Using transects with markers allowed a reproducible monitoring and the exact localization of the measurement points and the generation of maps.

Conclusion

It has been shown that temperature readings are a challenging tool. They are necessary and required for a wide range of other investigations. The measurements have been essential in identifying spots of thermal-water and gas exhalations. Temperature is a factor to consider due to the fact that it is a parameter which is most rapidly changing in geothermal systems. A close and detailed observation of this factor is advised in many fields of underwater investigations.

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Application of a global positioning system underwater

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Abstract. The precise determination of the geographical location under water requires high technical effort and is thus very expensive. For the scientific diving group of the TU Bergakademie Freiberg such facilities were not available. On the other side, the use of commercially available GPS devices underwater is not possible due to the fact that GPS satellite signals can not be received below the water surface.

Goal therefore was to develop an inexpensive device based on a normal GPS (Garmin 276C) and applicable to a maximum depth of 40 m. The concept for this low cost system was water tight encapsulating the hand held GPS on the one side and floating the GPS antenna at the surface directly above the scuba diver on the other side. Antenna and hand held are connected via a cable. However, conventional antennas have only weak receiving power and cables have a high dissipation loss. The special antenna used had therefore a higher antenna gain than usual and the special cable used owned a very low impedance. With the help of waterproof connectors the GPS in the watertight housing is connected with the antenna floating in a buoy on the water surface. For the operation of the GPS system two divers are required to keep the buoy as far as possible straight above the diver which is only possible without significant currents and winds. Estimating the angle of deviation in combination with a compass reading for the direction a proxy for correcting the GPS coordinate can be deduced.



Scientific Diving Program Management and Scientific Diver Education

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Abstract. A diving safety manual and a diving control board are the two primary management tools for operating scientific diving projects at a university or research institution. Scientific diver certifications and authorizations include diver-in-training, scientific diver and temporary diver. Diving specialty authorizations include diver education in decompression diving, surface supplied diving, drysuit diving and working in overhead environments. Training curricula include scuba skills, diving first aid, and dive computer use for determination of decompression status.

Introduction

The purpose of a research diving project is the advancement of science. Scientific divers, based on the nature of their activities, use scientific expertise in studying the underwater environment and, therefore, are scientists or scientists-in-training. The tasks of a scientific diver are those of an observer and data gatherer who uses scuba diving as a research tool. Information and data resulting from a scientific project usually are disseminated in a technical document or peer-reviewed research publication. *Scientific diving* is “diving performed solely as a necessary part of a scientific, research, or educational activity by employees whose sole purpose for diving is to perform scientific research tasks.” It does not include performing any tasks usually associated with commercial diving such as: placing or removing heavy objects underwater; inspection of pipelines and similar objects; construction; demolition; cutting or welding; or, the use of explosives. Heine (1999) describes scientific diving projects as occurring in tropical and subtropical seas, temperate waters, fresh water lakes, polar environments, blue-water (open ocean) and submarine canyons, estuaries, and offshore platforms. Lang (2001a) and Lang and Baldwin (1996) present a wide variety of methods and techniques of underwater research in various habitats and geographical ranges, representing scientific disciplines in most of the natural sciences.

The scientific diving programs in the United States can be broadly categorized into three groups: research institutions (predominantly research), public and private universities, museums and aquaria (predominantly education and teaching, and research), and consulting companies (predominantly contractual environmental, geological and archaeological investigations). The current scientific diver population in the United States is estimated at 4,800 individuals. A minority of these are long-term, career scientific divers (*e.g.*, federal employees, university professors) who may be considered in the 40+ average age category. At the university level the turn-over of scientific divers can be rather high as evidenced by undergraduate students enrolled in diving courses, research technicians on grant funds or students in Master’s degree or Ph.D. curricula. This population tends to be in the 18-34-age category. An upper age limit for scientific diver certification does not exist; the lower limit is generally 18 years of age. Of the total scientific diver population, approximately one fourth is estimated to be women.

Scientific diving standards

An employer is responsible for publishing a diving safety manual and appointing a Diving Control Board (DCB) that consists of a majority of active scientific divers who have autonomous and absolute authority over scientific diving program operations in accordance with procedures covering all diving operations specific to the program; procedures for emergency care, including recompression and evacuation; and, criteria for diver training and certification. At the institutional level, scientific diving projects are peer-reviewed by

the Diving Control Board and supervised by the Diving Officer (DO). Federal regulations are promulgated by the U.S. Department of Labor (2008).

The American Academy of Underwater Sciences (AAUS) publishes *Standards for Scientific Diving Certification and Operation of Scientific Diving Programs*. The purpose of this document is to ensure that all scientific diving is conducted in a manner that will maximize protection of scientific divers from accidental injury and/or illness, and to set forth standards for training and certification that will allow a working reciprocity between organizational member institutions that adhere to these standards. This document sets minimal standards for AAUS-recognized scientific diving programs, the organization and conduct of these programs, and the basic regulations and procedures for safety in scientific diving operations. The AAUS standards are generally considered the standard of practice for scientific diving in the U.S.

The University National Oceanographic Laboratory System (UNOLS) Research Vessel Diving Safety Standards (1996) amplify the AAUS standards for the special considerations and applicable legislation of diving from a research vessel platform. The UNOLS operates the U.S. academic research vessel fleet, funded primarily by the National Science Foundation.

Flemming and Max (1996) published a review of international scientific diving legislation and regulations. Several nations issue scientific diving licenses (occasionally under commercial diving regulations) and have national registries. In recent years there has been a move towards self-regulation and peer-promulgated standards (codes) of practice that replace government-mandated regulations. The Confédération Mondiale des Activités Subaquatiques (CMAS) established diving qualification equivalencies and certification for scientific divers. National diving federations belonging to CMAS may avail themselves of these certifications and standards. Since 2008 the European Scientific Diving Committee is a panel of the European Science Foundation's Marine Board that promotes scientific diving excellence, safety and standardized training across Europe.

Diver training

Scientific diving authorizations

There are three types of scientific diving authorizations.

1. Diver-in-Training. This authorization signifies that the diver has completed entry-level training requirements through a nationally or internationally recognized scuba certification agency (*e.g.*, PADI, NAUI, SSI, and BSAC) or scientific diving program.
2. Scientific Diver. This certification is a permit to dive with compressed air within no-decompression limits. This permit is valid only while it is current and for the depth and specialty intended (see below).
3. Temporary Diver. This authorization is issued only following a demonstration of the required proficiency in diving and if the person in question can contribute measurably to a planned dive. Temporary diver authorization is restricted to the planned diving operation under the host institution's auspices and complies with all other scientific diving policies, regulations and standards, including medical requirements.

Depth certifications

The scientific diving community has long adhered to an incremental experience-gathering schedule through depth certifications. The scientific diver certification authorizes the holder to dive to the maximum depth indicated on the scientific diver card and DO-approved dive plan, which may be exceeded by one step only if accompanied by a diver certified to a greater depth. Diving with compressed air is not permitted beyond a depth of 58 msw (190 fsw).

1. Certification to 9 msw (30 fsw) depth is the initial certification, approved upon the successful completion of the 100-hour training course.
2. Certification to 18 msw (60 fsw) depth can be obtained after successfully completing, under supervision of a scientific diver certified to that depth or greater, 12 logged training dives to depths between 31 and 60 fsw, for a minimum total time of 4 hours.
3. Certification to 30 msw (100 fsw) and 40 msw (130 fsw) depths can be obtained after successfully logging four dives near each of these maximum depths, and successfully completing a check-out dive approved by the DO.
4. Certification to depths over 45 msw (150 fsw) and 60 msw (190 fsw) can be obtained by logging four dives near each of these maximum depths, and successfully completing a check-out dive approved by the DO. Dives are planned and executed under close supervision of a scientific diver certified to this

depth. The diver also needs to demonstrate knowledge of the special problems of deep diving, and of special safety requirements.

Diving specialties

Diving specialties require additional training and approval by the DO. Scientific diver certification is a prerequisite for engaging in the following specialties: decompression diving, surface-supplied diving, mixed-gas or oxygen-enriched air (nitrox) diving, semi- or closed circuit rebreather diving, lock-out and saturation diving, blue-water diving, dry suit diving, overhead environment (ice, cave or wreck) diving, and altitude diving.

Swimming evaluation

The applicant for training performs the following tests without swim aids: an underwater swim for a distance of 25 m without surfacing, a 400 m swim in less than 12 min, a 10-min water tread (or 2 min without the use of hands), the transport of another person of equal size for a distance of 25 m in the water.

Scuba training

1. Practical Training

At the completion of training, the trainee is tested on his/her ability to perform the following, as a minimum, in a pool or in sheltered water: water entry with full scuba equipment; face mask clearing; air sharing (including both buddy breathing and the use of an alternate air source as both donor and recipient, with and without a face mask); alternate between snorkel and scuba while kicking; underwater signs and signals; simulated in-water mouth-to-mouth resuscitation; rescue and transport, as a diver, a passive simulated victim of an accident; removal and replacement of scuba equipment while submerged; and, an in-water level of competence that is acceptable to the DO.

2. Open Water Evaluation

The trainee must demonstrate at least the following in open water: surface dive to a depth of 3 msw in open water without scuba; air-sharing proficiency (including both buddy breathing and the use of an alternate air source, as both donor and recipient); open water or surf entry and exit, or leaving and boarding a diving vessel, while wearing scuba gear; a 400-m surface kick while wearing scuba gear without breathing from the unit; judgment adequate for safe diving; the ability to maneuver efficiently in the environment, at and below the surface; a simulated emergency swimming ascent; mask and regulator clearing while submerged; maintenance of neutral buoyancy while submerged; ascent at a rate not to exceed 10 m/min (30 fsw/min) and a hovering stop; self- and buddy rescue; underwater navigation; dive planning and execution with a buddy; and, completion of 12 supervised open-water dives in a variety of dive sites for a minimum cumulative bottom time of 6 hours.

3. Theoretical training

Theoretical aspects beyond the Diver-in-Training level (minimum cumulative time is 100 hrs) include principles and activities appropriate to the intended area of scientific study. Topics include, but are not limited to, data gathering techniques, collecting, common biota, behavior, installation of scientific apparatus, use of chemicals, site selection and relocation, animal and plant identification, ecology, tagging, underwater photography, scientific dive planning, dive rescue and accident management, diving first aid (Lang et al. 2007), coordination with other agencies, appropriate governmental regulations, small boat operation, and diving specialties. The theoretical aspects are tailored to the individual scientific diver based on his/her academic background and research methodologies. This theoretical knowledge is documented through the satisfactory completion of a written examination.

Continuation of scientific diving certification

During any 12-month period, each certified scientific diver must log a minimum of 12 dives, two of which are within the certified depth range. Divers certified to 45 msw (150 fsw) or deeper may satisfy these requirements with dives over 40 msw (130 fsw). If no dive is made for a 6-month period, a check-out dive must be made. Once the initial scientific diver certification requirements are met, divers whose depth certification has lapsed due to lack of activity may be requalified through work-up dives. If a scientific diver's certification expires, is suspended or revoked, he/she may be recertified after complying with a refresher program.

Diving safety and training

The scientific diving community has a traditional proactive record of furthering diving safety. The first scientific diving safety program was established at Scripps Institution of Oceanography in 1952 in preparation for the Capricorn Expedition to the South Pacific. This program pre-dated the national recreational scuba training agencies. Most scientific diving programs today trace their ancestry to common elements of the original Scripps diving program.

Diving safety programs can be generalized as fulfilling a two-fold purpose. The first being a research-support function, which assists the diving scientist with specialized underwater equipment, advice, and diver support to assist in fulfilling the scientific objectives of the diving project. The second is a risk management function that protects the safety and health of the individual scientist, and the employing organization from excessive liability exposure, by providing state-of-the-art diving equipment, breathing air, training and medical surveillance programs (Lang 2003).

More recently, ongoing scientific diving safety research has been conducted to consider a more effective means of decompression status monitoring using dive computers (Lang and Hamilton 1989). DCBs approve specific makes and models of dive computers that may be used as a means of determining decompression status. Each diver relying on a dive computer to plan dives and indicate or determine decompression status must have his/her own unit and pass a practical and written training session. On any given dive, both divers in the buddy pair follow the most conservative dive computer. If the dive computer fails at any time during the dive, the dive is terminated and appropriate surfacing procedures are immediately initiated. A scientific diver is not allowed to dive for 18 hours before activating a dive computer to control his/her diving, and once in use, it is not switched off until complete outgassing has occurred. Multiple deep dives and/or decompression dives with dive computers require careful consideration.

Lang and Egstrom (1990) investigated the slowing of ascent rates and performance of safety stops to provide scientific divers with a greater margin of decompression safety. Before certification, the diver demonstrates proper buoyancy, weighting and a controlled ascent, including a "hovering" stop. Ascent rates are controlled at a maximum of 10 msw/min from 20 msw and at the rate specified for the make and model of dive computer if a variable ascent rate from depth. Scientific diving programs require a stop in the 5-8 msw zone for 3-5 minutes on every dive. Scientific divers using drysuits receive additional practical training in their use. Drysuits must have a hands-free exhaust valve and buoyancy compensators a reliable rapid exhaust valve that can be operated in a horizontal swimming position. A buoyancy compensator is required with drysuit use for emergency flotation. In the case of a runaway ascent, breathing 100% oxygen above water is preferred to in-water air procedures for omitted decompression.

Multi-day, repetitive diving physiological aspects were examined by Lang and Vann (1992). Although diving is a relatively safe activity, all persons who dive must be aware that there is an inherent risk to this activity. In 1992, the risk of decompression illness in the United States was estimated at 1 incident per 1,000 dives for the commercial diving sector, 2 incidents per 10,000 dives (4 per 10,000 in 2008) for recreational diving activities and 1 incident per 100,000 dives for the scientific diving community. Scientific diving programs provide continuous training, recertification and dive site supervision, which helps maintain established safe diving protocols. Increasing knowledge regarding the incidence of decompression sickness (DCS) indicates that our ability to predict its onset on multi-level, multi-day diving is even less sensitive than our ability to predict DCS on single square-wave dives. There appears to be good evidence that there are many variables that can affect the probability of the occurrence of DCS symptoms. The ability to mitigate these variables through education, good supervision and training appears to be possible for hydration, fitness, rate of ascent, fatigue, etc., and are continuously promoted. Scientific divers are subject to a host of specific conditions that may increase risk if precautions are not taken. There is adequate technical support for the use of oxygen-enriched air or nitrox (Lang 2006) and surface-oxygen breathing in scientific diving where higher gas loadings are anticipated in multi-level, multi-day dives. We must continue to remember that DCS is generally recognized as a probabilistic event, which tends to lean the scientific diving community towards a more conservative diving position.

The order of dive profiles was investigated (Lang and Lehner 2000), in part, because of the difficulty for scientific divers to adhere to the “dive progressively shallower” rule while on projects investigating coral reefs at varying transect depths. More importantly, the genesis and physiological validity of the “dive deep first” rule was in need of examination. Historically, neither the U.S. Navy nor the commercial sectors have prohibited reverse dive profiles. Reverse dive profiles are acknowledged as being performed in recreational, scientific, commercial, and military diving. The prohibition of reverse dive profiles cannot be traced to any definite diving experience that indicates an increased risk of DCS. There is no convincing evidence that reverse dive profiles within the no-decompression limits lead to a measurable increase in the risk of DCS. Lang and Lehner (2000) found no reason for the diving communities to prohibit reverse dive profiles for no-decompression dives less than 40 msw (130 fsw) and depth differentials less than 12 msw (40 fsw).

Oxygen-enriched air (nitrox) has been used in the scientific diving community since the early 1970's. Lang (2001b) reports for entry-level, open-circuit nitrox diving, that there is no evidence that shows an increased risk of DCS with the use of oxygen-enriched air (nitrox) versus compressed air. A maximum PO₂ of 1.6 atm is generally accepted based on the history of nitrox use and scientific studies. Routine CO₂ retention screening is not necessary for open-circuit recreational nitrox divers. It should be noted that CNS oxygen toxicity could occur suddenly and unexpectedly. Based on history of use, no evidence is available to show an unreasonable risk of fire or ignition when using up to 40% nitrox with standard scuba equipment. The level of risk is related to specific equipment configurations and the user should rely on manufacturer's recommendations.

Operational guidelines for polar scientific diving operations were promulgated on a consensual basis by the senior practicing scientific divers (Lang and Stewart 1992; Lang and Sayer 2007; Lang and Robbins 2009). Solutions for advanced and deep mixed gas scientific diving operations were offered by Lang and Smith (2006).

Conclusion

The scientific diving community has very effectively used scuba as a research tool for over 45 years, since the first program was established at Scripps Institution of Oceanography. The safety record is excellent, in part, due to thorough medical, training and operational standards and programmatic supervision of diving activities. Safety considerations are of primary concern for the diving programs and regulations are promulgated by the underwater scientists who live by them.

Scientific research objectives, whether through mensurative or manipulative experiments, in many instances could not have been accomplished without scientific diving techniques, as evidenced in materials and methods sections of peer-reviewed published literature (Lang 2007). The complimentary use of diving and remotely operated vehicles (ROVs), autonomous underwater vehicles (AUVs), and remote sensing equipment has greatly advanced our underwater research capabilities in recent years. One-Man Atmospheric Diving Systems (OMADS) have also become more sophisticated and affordable and are more widely used by the marine research community. At some point in the future, decompression, dive training, and medical issues may no longer be of major concern to scientists, as emerging technologies develop. In the meantime, many topics of current scientific interest, including marine biodiversity, coral reef health, sea-level change and global warming are to a large degree dependent on placing the trained scientific eye under water to record, interpret and sample the marine environment.

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Scientific Diving in the UK: training and legal requirements

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Abstract. In the UK, scientific diving has been designated as one of the five main professional sectors of the diving industry since 1997. As such, anyone in the UK diving in support of science for their work, must comply with the 1997 Diving at Work Regulations which are a Statutory Instrument of the UK 1974 Health and Safety at Work etc. Act. This paper outlines how scientific divers in the UK work under these regulations and the minimum standards required for diving competence and medical surveillance. Following the introduction of EU directive 2005/36/EC in 2007, there is now the legal basis for the recognition of professional qualifications between member states; practical guidance on how scientific divers can work between member states, using the UK as an example, will be given.

Introduction

Scientific diving throughout the world has an extremely good safety record (Carter et al. 2005; Lang 2005; Sayer 2005; Sayer and Barrington 2005; Dardeau and McDonald 2007; Sayer et al. 2007). However, through its very nature, many national legislators view occupational scientific diving as carrying a higher than normal risk (Sayer 2004; Sayer and Forbes 2007). Because of this, many countries insist on scientific divers having varying levels of training and qualifications in order to dive as part of their work (Sayer et al. 2008).

Until the early 1980s, diving at work in the UK was largely unregulated but instead was undertaken in association with a series of industry sector codes (Fleming and Max 1996). The introduction of the 1981 Diving Operations at Work Regulations (DOWR 1981) as a statutory instrument of the 1974 Health and Safety at Work etc., Act (HSW 1974) was largely as a consequence of significant levels of diving fatalities in the offshore sector which was dominated by diving operations associated with oil exploration and exploitation activities in the North Sea (Warner and Park 1990; Limbrick 2002). DOWR 1981 was put in place through the UK Health and Safety Commission (HSC) and implemented through its Health and Safety Executive (HSE). Because of its origins, DOWR 1981 was largely targeted at the offshore sector and was necessarily prescriptive. However, the regulations were in place to cover all diving operations at work in the UK at that time and the prescribed approaches to offshore diving operations did not always fit easily with the other industry sectors. Two revisions to DOWR 1981 were made in 1985 and 1992 but for significant numbers of diving contractors, specifically those covering diving at work for journalists, scientists, archaeologists and recreational instructors, operations could only continue through the issue of exemption certificates by the HSE.

In the mid-1990s the HSE recognised that although there could be generic regulations in place to control diving operations there were large differences in approaches and needs of various sectors within the diving at work industry. The HSE, therefore, set about creating a framework under which there would be a set of generic regulations for implementing diving operations at work in the UK complemented by sector-specific codes of practice. The codes of practice were written jointly by the HSE with bodies or groups that were representative of the respective sectors. Once accepted by the HSE these became the Approved Codes of Practice (ACoPs) for each sector.

There were five ACoPs recognised representing the Offshore, Inshore, Scientific and Archaeological, Recreational and Media sectors. However, they were all constructed to be much less prescriptive than the DOWR 1981 in a way that set minimum standards for each sector and were largely self-regulated through processes of risk assessment and risk management. Following extensive consultation the new HSE Diving at Work Regulations were formally accepted as statutory instrument No. 2776 of the HSW 1974 Act in November 1997 and came into force in April 1998 (although they are known by their 1997 acceptance date and are identified as the HSE Diving at Work Regulations 1997; DWR, 1997).

Diving at work in the UK is regulated by the HSE Diving Group which is part of the Offshore Division of the Hazardous Installations Directorate (www.hse.gov.uk/diving). The HSE Diving Group sponsors two meetings a year with representatives of the various Diving Industry sectors. The scientific and archaeological sector is represented by the UK Scientific Diving Supervisory Committee (www.uk-sdsc.com).

This account presents an overview of how the DWR 1997 impact scientific and archaeological diving operations with respect to compliance and training requirements. A brief summary of how the UK regulations now link with other European systems is also given.

UK Scientific Diving Regulations

The DWR 1997 are effectively goal setting regulations that apply to all diving at work operations that are undertaken in UK territorial waters; operations from UK-registered ships are covered by the Merchant Shipping [Diving Safety] Regulations 2002). The DWR 1997 set out, in generic terms, the roles of the diving client, the diving contractor, the diving supervisor and the diver. They outline the minimum qualification and medical requirements for a person to dive at work and describe minimum dive team numbers for specific types of diving operation. Because of their generic nature, the regulations do not give detailed or specific guidelines for how a particular operation should be conducted in practical terms and state specifically that minimum standards are unlikely to be acceptable for most diving operations. However, there are specific requirements to produce and maintain written records of the appointment of the diving supervisor, a diving project plan and a diving operation record and specific reference is made to the diving project plans and operation records being based on a system of risk assessment (DWR, 1997).

More sector-specific detail is included in the relevant ACoPs; the Scientific and Archaeological ACoP covers all diving operations undertaken in support of science and archaeology (but specifically not commercial salvage), and more recently also public aquarium diving (Sci-Arch ACoP 1997). A summary of the main areas of compliance with some definition is given below; this is only a brief summary and complying with UK diving at work legislation would only be possible with access to the full regulations and specific ACoP.

Every diving operation must have a named Diving Contractor. The Contractor is normally the employer of the divers and can be the corporate body (e.g. the University). However, diving operations need to be incorporated within a transparent system of health and safety management that can influence how they are conducted. This will usually be made through employing suitably qualified staff to oversee and report on diving operations.

All diving operations must be overseen by a suitably qualified and competent Diving Supervisor. The Supervisor is often a current diver, but may also be an ex-diver with suitable competence. The Supervisor has a legal responsibility to ensure the safe conduct of the diving operation and that it complies fully with all the administrative requirements of the regulations. Although the Supervisor can dive as part of the diving operation they are supervising that could only happen in a small number of circumstances; the normal situation is that the Supervisor remains on the surface.

UK scientific, archaeological and public aquarium divers must be properly qualified for diving at work. For this sector (i.e. other diving industry sectors have different minimum levels of certification) the minimum qualifications can either be based on those given by recreational agencies or are industry based; these are detailed below under training. In addition, a diver must obtain an annual HSE Diving at work medical; this is a thorough examination of the diver's health and fitness and must be undertaken by an Approved Medical Examiner of Divers (HSE AMED). Divers should also have and maintain appropriate certified levels of First Aid competency. All divers must be competent for the tasks to be undertaken underwater; competence is usually defined as a combination of experience and training (see below).

A principal element of UK Health and Safety law in the workplace is that the employee is given equipment and clothing that is suitable for the task. For diving at work operations this will obviously include diving suits and all diving equipment. There is a legal requirement to ensure that all equipment supplied is maintained and tested in accordance with the manufacturer's guidelines or to legal standards where they exist.

Before diving operations can commence a project plan must be compiled. The project plan summarises details of the diving operations to be undertaken as part of the wider project, the actions to be taken in the event of an emergency (to include recompression considerations) and the higher-level risk assessment (see below). It should also indicate the levels of dive supervision required as well as the numbers of divers.

A Diving Operation is a dive where the tasks and locations are the same. Often in science and archaeology, the main intention of the dives is to gather data that contribute to a replicated series. To that end, a diving operation may be repeated over a variety of timescales and so may be a number of dives but in some

circumstances, a diving operation may cover a single isolated dive. It becomes a different diving operation once the task or location differs.

The DWR 1997 are based on the principles of formal and ongoing risk assessment. These principles are based on a dynamic process that integrates the required experience and numerical strength of the dive teams with the demands of the task to be undertaken and the types of environment where the dives are to happen (Sayer 2004; Sayer and Forbes 2007).

There must be a record (either paper or electronic) of all diving operations kept both by the Diving Contractor and the Diver, and both for a minimum period of two years. As well as recording the details of the operation, the record must also document any accidents, serious occurrences and/or near misses. It is a legal responsibility in the UK to report all dangerous occurrences that happen at work.

UK Scientific Diver Training

The UK DWR 1997 are based on minimum competencies without defining particular training standards. This has resulted in the development of three routes for certifying basic levels of diving competence for UK scientific and archaeological divers. The first is the professional diving industry standards on which divers can be competence-assessed at HSE-approved diving schools. These standards range in complexity depending on which sector the diver wants to work in. For science and archaeology, the entry-level certificate is “HSE SCUBA” and there are a number of attainment methods ranging from starting as a non-diver and training for 5-6 weeks, to short (2-3 day) competence assessments for experienced recreationally-trained divers. The HSE SCUBA qualification is not specific to scientific and archaeological diving and can be used in other (but not all) sectors. The competence level of the HSE SCUBA qualification is equivalent to the ISO 24801-3:2007 recreational standards which are similar in scope to the older CMAS 3star standards.

Certified prior recreational experience is the second training route available to UK scientific and archaeological divers with, again, the minimum level of competence being ISO 24801-3:2007 or CMAS 3star equivalent standards; a table of international equivalencies is given by Fleming and Max (1996). The third training standards are the European and Advanced European Scientific Diver (ESD, AESD) certified competencies which are detailed by Sayer et al. (2008) and summarised below.

In addition to basic dive training, UK scientific and archaeological divers also need to obtain (and maintain) an intermediate knowledge of first aid training (HSE First Aid at Work). This completes the basic level of training required; all other training is either for more advanced types of diving or for scientific specialisms. The following lists are not intended to be exhaustive but for diving techniques there is additional training for: surface supplied diving; full-face masks; mixed gases; rebreathers; cold-water (under ice); dive supervision; boat handling. For science-support, training is provided for; underwater photography; underwater videography; survey techniques; project planning and management. For techniques that are more specific, then the HSE will recognise manufacturer’s courses or self-certification as a form of proof of competence where no other agency route exists.

Integration of UK standards with Europe

As of 2007, European Union Working Directive 2005/36/EC imposed a legal requirement on all member states (and associate states with regard to EU science funding) to recognize and accept relevant professional qualifications gained in another member state. This directive was anticipated in the scientific diving community and common standards have been developed and are now officially ratified (Sayer et al. 2008). The development of the European and Advanced European Scientific Diver (ESD, AESD) equivalencies permits either existing national qualifications to be based on the ESD/AESD scales or for inexperienced divers to be trained to those levels.

EU member states may have different medical requirements for diving in their own health and safety at work legislation. The validity of the relevant medicals obtained in the home state will usually be time-dependent and should retain a period of acceptance before having to comply with the requirements of the host state. An example is the UK where the relevant medical certificate in the home state (where the diver must have been examined by a doctor) is acceptable if it was obtained within three months of coming to work in the UK. ASE and AESD certificates are standards which now have full recognition in UK Diving at Work legislation.

Conclusions

With the introduction of revised diving at work regulations in 1998, scientific and archaeological divers in the UK have less prescriptive diving at work legislation to comply with. This does not mean regulation has been relaxed and there are no obvious negative trends in safety of UK scientific diving. However, the legal burden of responsibility is placed heavily at the level of Diving Supervisor and on having to prove levels of competence. However, acceptance of the relevance of recreational training standards has meant that the range of competence assessors has increased. The introduction and acceptance of the ESD/AESD equivalencies has removed any restrictions for the movement of scientific and archaeological divers within Europe and the use of mixed diving teams.

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12-years scientific diving at Technische Universität Bergakademie Freiberg

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Abstract. Diving education at the Technische Universität Bergakademie Freiberg was launched in 1990. Since 12 years a course scientific diving is offered. 2007 a safety standard for scientific diving at the TU Bergakademie Freiberg was established. This combination of teaching both special scuba diving skills and theory and as well scientific techniques is unique in Germany.

History of scientific diving at TU Bergakademie Freiberg

In 1990 education in scientific work under water started with training course in snorkeling and fin swimming. Scientific camps and field trips followed and included diving activities for students and scientists. This was based on activities and courses in swimming, snorkeling, and scuba diving offered by the University's Sport Center at the Technische Universität Bergakademie Freiberg. A main objective from the very beginning was developing interdisciplinary actions in theory and practice. The scientific diving education was thus established consequentially in the studium generale offer. Activities in the past are listed in Table 1.

Table 1. Chronology of developing education in scientific diving at the Technische Universität Bergakademie Freiberg.

Year	Activities
1990	Dive camps – acid mine lake “Senftenberger See”
1993	Starting scuba diving education offered by the sport center of TUBAF
1997	Scientific diving within the framework of university sports program
2002	Scientific diving course offered by “studium generale”
2003	1. International scientific diving camp: Lanterna / Sv. Marina, Istria, Croatia
2004	Implementation of the lecture “fascination water” 2. International scientific diving camp: Sv. Marina, Istria, Croatia
2005	3. International scientific diving camp: Sv. Marina, Istria, Croatia
2006	4. International scientific diving camp: Panarea, Italy
2007	Implementation of scientific diving safety regulation Establishment of 2 modules “Scientific Diving I”, “Scientific Diving II” 5. International scientific diving camp: Panarea, Italy
2008	6. International scientific diving camp: Panarea, Italy
2009	7. International scientific diving camp: Panarea, Italy

From 2003 until 2005 Sv. Marina, Istria (Croatia) was for three times the location for diving field trips. From 2006 on the diving field trip took place in Panarea, Eolian Islands, Italy with the friendly support of Dr. Franco Italiano (INGV Palermo). Several students started their Bachelor and Master thesis during these field campaigns. In summary 8 Master, 20 bachelor and student projects, and 6 field survey reports are the result of our activities.

Table 2 shows participants statistics from Scientific Diving since 2004.

Table 2. Participants statistics from Scientific Diving since 2004.

	SS 04	WS 04/05	SS 05	WS 05/06	SS 06	WS 06/07	SS 07	WS 07/08	SS 08	WS 08/09	SS 09
Applied Science	-	2	2	1	2	1	2	1	1	-	-
Network Computing	3	2	2	2	1	1	1	-	-	-	-
Chemistry	-	-	1	1	1	1	1	2	-	1	1
Geology	1	2	2	1	3	2	2	2	2	2	-
Geoecology	9	3	5	6	6	7	8	2	4	3	2
Geotechnology/Mining	1	1	2	-	-	-	1	-	-	-	1
Mechanical Engineering	2	2	1	1	4	2	5	-	-	-	1
Mineralogy	1	1	1	1	1	-	-	-	-	-	-
Applied Mathematics	-	-	-	1	1	-	-	1	-	-	-
Process Engineering	1	1	1	1	1	1	-	1	2	1	1
Environmental Engineering	1	-	-	1	1	2	2	1	1	1	1
Business Engineering	3	-	-	1	-	1	1	1	1	1	-
Geology/Mineralogy	-	-	-	-	-	2	2	-	-	-	7
Automobile Engineering: Materials and Compounds	-	-	-	-	-	-	2	-	-	-	-
Geoinformatics	-	-	-	-	-	-	-	3	3	-	-
Materials Science and Technology	-	-	-	-	-	-	-	1	-	1	1
Postgraduate courses & Ex- ternals	-	1	2	1	2	-	3	6	1	2	2

Education modules

Integration as lecture

The lecture “Fascination water - Diving and Science” communicates theoretical and practical knowledge of several aspects concerning scientific diving. It was adapted for bachelor and master levels. For the practical part a basic scuba diving license CMAS* or equivalence is obligatory for the scientific diving module I. A CMAS ** or equivalence is required for the module II. Of course as well a valid health certificate is prerequisite.

Scientific diving module I contains the lecture “fascination water”, theory, training in advanced scuba diving skills (pool and field), 2 weekend diving camps. Students will obtain at least 10 logged dives during the course work for module I and they earn 4 ECTS credit points.

After successful participation in module I students will get - depending on their focus of research - the certificate of a CMAS Geology Diver (CMAS 2000a) or CMAS Diver Freshwater Biology (CMAS 2000b).

The Scientific diving module II contains a minimum of 10 days scientific diving field trip and 16 logged dives. Each student has to prepare himself for a certain task; he will be responsible for the special equipment needed for his task, the planning of his dives in communication with other divers and the dive master. Finally he has to write a report. Another 4 ECTS credits will be earned and as well the CMAS scientific diver certificate.

Figure 1 shows a flow chart of the educational structure of scientific diving at TU Bergakademie Freiberg.

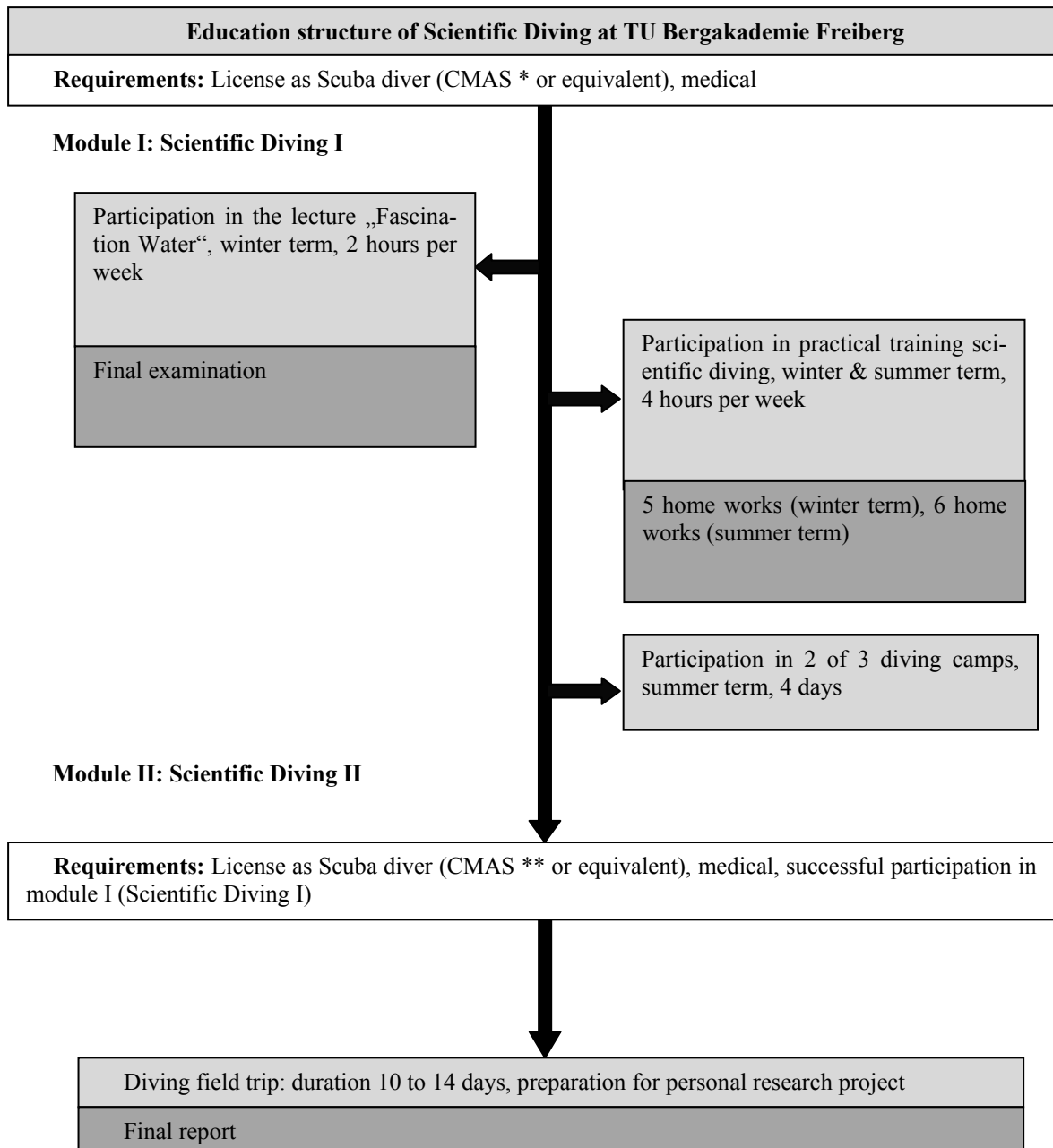


Fig.1. Education structure of Scientific Diving at the Technische Universität Bergakademie Freiberg

Content of teaching

In winter term, theoretical knowledge is imparted by the lecture “Fascination Water”. Following topics are included:

- I Oceanology
- II Applied Geo-Sciences
- III Working methods above and under water
- IV measurement-technology gathering of physical and chemical parameters
- V video and photo documentation

Basic practical training dealing with measuring devices under water, mapping, and photo-/video documentation is carried out in pool exercises in winter term. During summer term, special tasks will be specifically deepened and trained in working groups in 3 diving camps. A list of teaching contents during practical education can be found in Fig. 2.

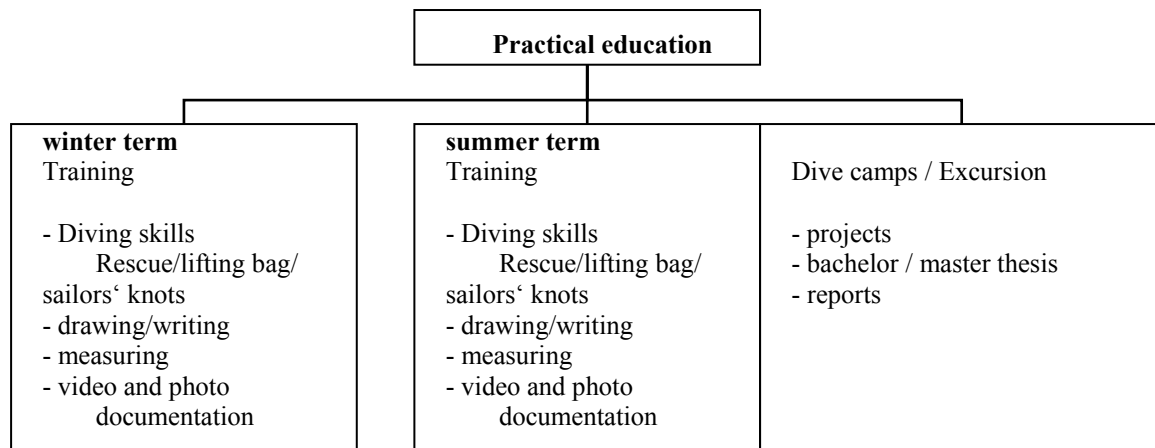


Fig.2. Educational structure of practical education

Training includes theory and practicing working techniques underwater in the following scientific fields:

- Geo-Science (Geology, Hydrogeology, Water Chemistry, Earth Systems Science, etc.)
- Biology, ecology, micro-biology, environmental science
- Mapping, documentation, and visualization
- Development of tools for in-situ measurements, sampling, drilling, documentation, positioning, surveying, automation, communication
- Modeling
- Management of ecosystems.

Using a whole range of underwater techniques our students learn:

- Underwater grid setup, visual estimation of benthic cover, grid counting
- Survey, sketching and documentation
- Lay out of a transect, photo transects, video transects
- Full survey of an underwater environment
- Transport of equipment with lift bags
- Operating a drop hammer device (e.g. sediment core sampling)
- Drilling with hydraulically powered submersible drill (core drilling in rocks, hole drilling)
- Various water sampling techniques
- Various gas sampling techniques (underwater fumaroles) and gas flow measurements
- Using in-situ probes for pH, redox potential, electric conductivity, temperature, carbon dioxide

Fig.3 is displaying some of the task students have to perform during their training

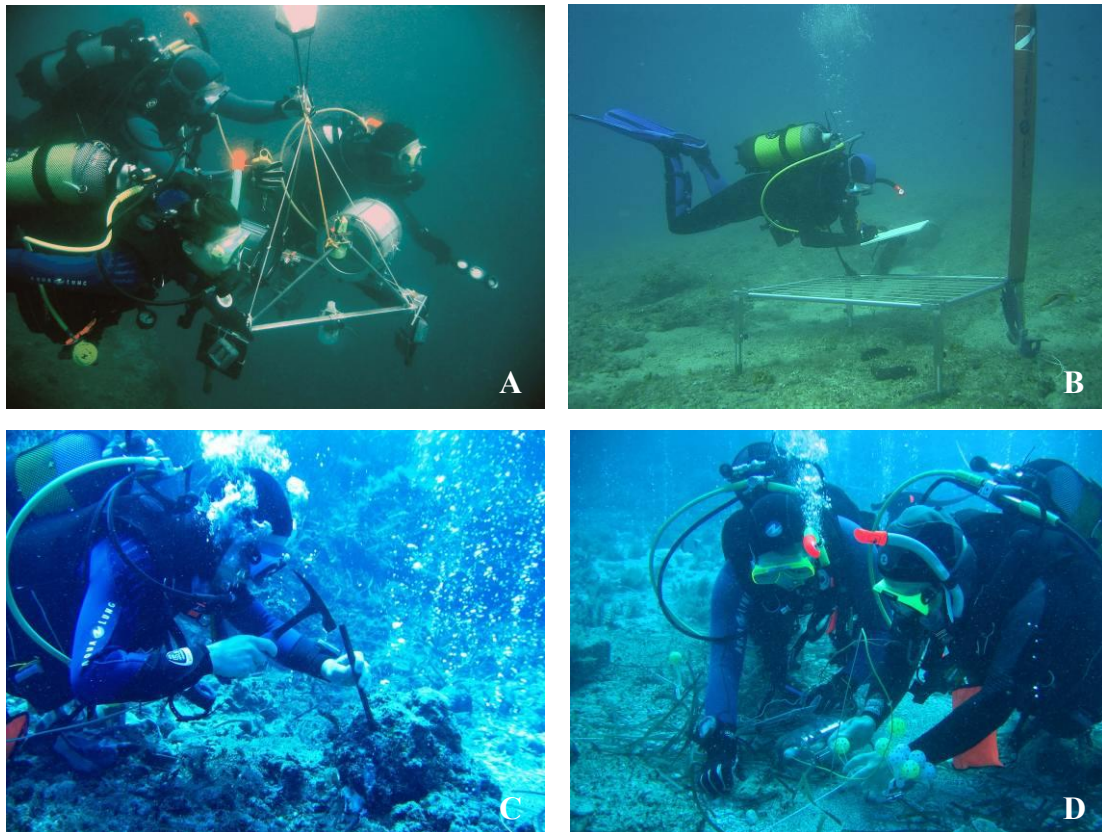


Fig.3. Teaching contents during Scientific Diving education at TU Bergakademie Freiberg. A – Transport with lifting bag. B – Mapping with grid. C – Sampling of hand-speciemen. D – Temperature measurement.

Scientific Diving Safety regulation

Scientific diving means using scuba diving as a research tool to investigate the shallow underwater environment. Our policy with respect to scientific diving is based on the standards of the CMAS (2000c) published as Guidelines for Scientific Diving.

The actual version of these guidelines (version 1.1, 08/23/2007) comprises the regulation of responsibilities, equipment, diving activities, response to accidents, rules and regulations, as well as regulations of dive planning and documentation (Merkel et al. 2009).

By application graduates and undergraduates of TU Bergakademie Freiberg are insured with the statutory accident insurance of the federal state Saxony.

References

- CMAS (2000a) Standard – Marine Geology, version 2000/00.
- CMAS (2000b) Standard – Fresh Water Biology, version 2000/00.
- CMAS (2000c) CMAS Standard for Scientific Diver, version 2.1.
- Merkel et al. (2009) Work Instruction for Scientific Diving at TU Bergakademie Freiberg.

Work Instruction for Scientific Diving at TU Bergakademie Freiberg.

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Abstract.

Scuba diving is a research tool for any kind of research of underwater environment. It is taught at the Technische Universität Bergakademie Freiberg. Since 2007 a Work Instruction for Scientific Diving is effective. This paper outlines the major objectives of the work instruction and discusses similarities and differences to other standards.

Introduction

In 1997 the Sports Centre of the Technische Universität Bergakademie Freiberg launched the course “Scientific Diving”. Effective from 2002 this course is part of the *studium generale* in cooperation with the Department of Geoscience, Geotechnique and Mining. Since winter semester 2007 two modules “Scientific Diving I” and “Scientific Diving II” are offered, giving the opportunity for students to combine science and sports and to earn the qualification CMAS scientific diver.

Scientific diving as integrative and interdisciplinary course supports learning team skills and promotes the personality development of graduates and undergraduates of the TU Bergakademie Freiberg and can be chosen as soft skill course in any Bachelor or Master program offered at TU BAF.

Requirements for students (graduates and undergraduates) at TU Bergakademie Freiberg

To enroll in the modules “Scientific Diving I and II” students must have a corresponding diving qualification and a valid medical (medical certificate: fitness for recreational scuba diving according to fitness-to-dive-guidelines of the GTÜM, Gesellschaft für Tauch- und Überdruckmedizin e.V., edition 1998). For enrolment in module I (Scientific Diving I), CMAS * (or equivalent), for participation in module II, CMAS ** (or equivalent), are required.

Insurance coverage

Insurance is an important fact during training and education: All participants in our scientific diving training are insured via the DGUV (German Social Accident Insurance). The DGUV is the umbrella association of the accident insurance institutions for the industrial and public sectors and is organized on the level of the federal states within Germany. The DGUV handles the communication with policymakers, regional, national, European and international institutions, and employers' and employees' representative bodies.

In order to maintain the requirements of the Social Accident Insurance of the federal state of Saxony a Work Instruction for Scientific Diving (WISD) was developed. According to this work instruction scientific diving and education concerning scientific diving has been conducted in a manner that will minimize the risk of accidental injury and/or illness.

The WISD [6] for the TU Bergakademie Freiberg was written taking into account both the requirements of the CMAS STANDARD FOR SCIENTIFIC DIVER [1] and the safety and health and health protection rules of the German Social Accident Insurance [2].

In addition to the DGUV insurance all students and scientists of the Scientific Diving Center at TU Bergakademie Freiberg are insured via the VDST diving insurance.

Outline of the work instruction

The work instruction is a safety manual (version 1.1, 08/23/2007) in total 35 pages including tables and forms. The manual covers the following items:

- Responsibilities
- Equipment
- Pre- and post diving checks
- Dive operations
- Checking equipment
- Procedures in case of accidents
- Laws
- Dive planning
- Decompression schedules
- Risk assessment

Several appendixes contain forms which have to be used by instructors, students, and researchers. At the very moment NITROX is not yet included but will be in a new version which is recently in preparation.

Reports

Any scientific dive no matter if it is during the education or in context with a bachelor, master or PhD research has to be performed according to the work instruction (WISD). This includes the necessary paper work. These documents are kept for at least 10 years in order to document any incidents or accidents.

Risk assessment

Each diving operation must be supervised by a competent and experienced diving supervisor. The supervisor has to prepare a risk assessment of each diving project including statements with respect to water conditions (e.g. current, ice covering, shipping traffic, tides, etc.), weather conditions (e.g. temperature, precipitation, etc.), and operating conditions. In case of particular danger the supervisor has to break off the diving operation.

Diving operation protocol

TU Bergakademie Freiberg must ensure that the responsible dive master organizes diving groups, solo diving is strictly forbidden. Responsible for this is head of the Scientific Diving Center (Prof. Merkel). Within a diving group one diving leader and one stand-in has to be nominated. Within the diving groups buddy teams are nominated. The diving protocol comprises information of

- Participating divers
- The nominated group leader and his stand-in
- Start / end of the dive (time)
- Total dive time, air pressure (start / end)
- Maximum depth for each diver.

Scientific diving logbook

Participants must keep a scientific diving logbook (“report of the day”) recording each dive with scientific background and the following information:

- Date
- Location
- Weather conditions
- Divers involved / buddy teams
- Diving equipment
- Surface time
- Dive-time, planned / real
- Maximum depth, planned / real
- Air consumption
- Decompression-stop-depths with associated decompression times

Further they have to report their

- Objectives
- Scientific equipment
- Observations
- Samples results
- Further working suggestions

Discussion

The WISD of the TUBAF [6] is compatible with the CMAS STANDARD FOR SCIENTIFIC DIVER [1] and thus as well with the STANDARDS for European Scientific Divers (ESD) and Advanced European Scientific Divers (AESD), because these two standards are more or less identical. However, so far we do not have defined the requirements for an advanced scientific diver in our standard. The other German standard for scientific divers [2] differs from the above mentioned in some points e.g. solo diving, usage of signal lines etc. This is due to the fact that the GUV R-2112 was derived in some way from the habits and rules of professional diving. No severe differences can be found by comparing the WISD of the TU Bergakademie Freiberg with the safety instructions of the STANDARDS FOR SCIENTIFIC DIVING of the American Academy of Underwater Sciences [5]. The Smithsonian Institute Diving Safety Manual [3] is in some parts more detailed e.g. defining certifications for 30, 60, 100 and 130 and over 130 foot depth. This manual includes as well HOOKAH diving, surface supplied diving, and NITROX which is the case as well for the STANDARDS FOR SCIENTIFIC DIVING of the American Academy of Underwater Sciences [5].

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