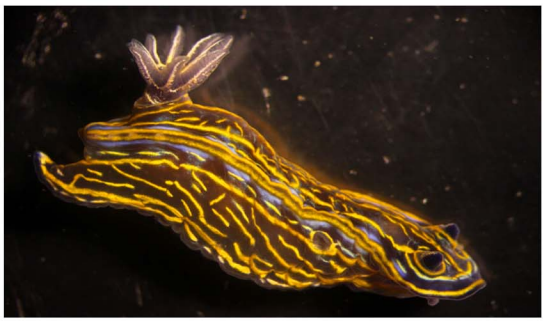
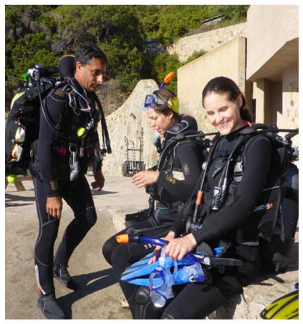





Marine Biological Excursion and Course



Calvi

2014

universität innsbruck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Marine Biological Excursion and Course

at STARESO Station, Calvi, Corsica

August 30th - September 13th 2014

Participants

Teachers

Robert Gschwentner
Reinhold Hanel
Bert Hobmayer
Iris Krainer

Attendents

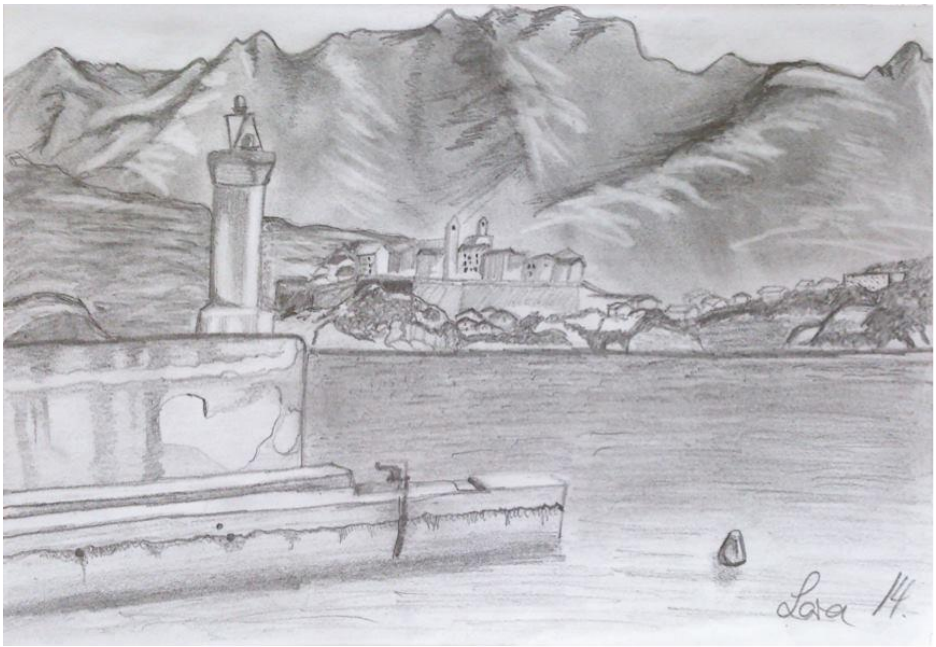
Jana Hobmayer
Sabine Gufler
Irene Schatz
Heinrich Schatz

Students Kiel

Jascha Dehm
Marie Massmig
Dana Michaelis
Henrike Wunderow
Huajing Yan

Students Innsbruck

Saskia Amann
Werner Bader
Nina Egger
Alexandra Grosbusch
Clara Hechenberger
Isabella Hilti
Michaela Hittorf
Ines Hrabie
Bianca Jansen
Stefanie Kuen
Teresa Müllauer
Verena Naschberger
Lara Schmielau
Raimund Schnegg
Raphael Strohmaier
Laura von Raffay



Contents

Introduction

Originating in 1992, the biannual marine biological excursion and course of the Universities of Innsbruck and Kiel again took place from August 30th to September 13th, 2014, at STARESO station. As in previous years, the first week of the event was dedicated to study the various marine habitats, their fauna, and the adaptations of the organisms to specific environments. The second week was dedicated to three marine biological projects (fish physiology, mollusk diversity, and sea urchin embryology) mainly carried out in the laboratories of the station.

Studying marine biodiversity involves sampling, either by hand or by using more or less sophisticated tools. Generally, it meant that basically every day students had to do extensive snorkeling. In the case of Plancton and Corralligène, however, the station boat was used. With the exception of fish, which were determined by live observation in the water, all the animals were brought to the station, determined - if possible - to the genus and species levels, and finally set back into the sea.

STARESO station offers an almost perfect environment for this course. Among the most important is the fact that the students basically live and sleep a few meters from the sea. It has immediate and protected access to the water, which makes snorkeling for the students easy to accomplish. Many different marine habitats are in close distance to the station, thus sampling can be done with limited effort. The station also has wet and dry laboratories, which are predominantly used in the second week. In order to achieve high end microscopic imaging and photography, several binoculars and a fluorescence microscope connected to a digital camera were additionally brought from Innsbruck. Furthermore, the station can provide all the equipment required for snorkeling or diving including wet suits, and every student was invited to do a test dive together with an instructor free of charge.

During the entire course, students worked in groups. They discussed their findings with the entire team at the end of every working day, and they presented the results of their second week-projects during a mini-symposium at the end of the course. All these efforts are here documented as daily and project protocols in the following pages.

Daily Reports

Boulder Field

Marie Massmig, Lara Schmielau, and Ines Hrabie

Introduction

Boulder fields are vertically structured marine habitats consisting of layers of rocks or stones. Depending on grain size and packing density, these layers form networks of crevices and caves of different sizes offering a huge diversity of microhabitats for a rich biodiversity. These habitats are further influenced by environmental factors like wave exposition and inclination, currents, tides as well as light intensity and biological effects (Hofrichter 2002).

Boulder fields do not only offer a habitat for a variety of species, they also protect coastlines during storm events (Green 2012). Thus there is ongoing research concerning the construction of artificial boulder-fields and the type of rock that should be used to enable a diverse biota in and on the new habitat. This turns out to be a real challenge, because there is always a big variability among replicate boulders (Green 2012).

The grain size via the turnover rate of rocks and stones influences the growth of algae and other sessile organisms. Smaller stones are more frequently turned over than bigger ones and so the overgrowing biota on single stones is represented in different developmental stages. Stones of intermediate size are known to be overgrown by the most diverse biota (Sousa 1979). This can be explained by the theory of intermediate disturbance, first mentioned by Connell (1978). In protected areas conditions are more stable and these areas mostly become dominated by single species in contrast to areas with quick changes of environmental factors and hence frequent repopulation by competing species (Sousa 1979). In the following observation the overall species diversity of a boulder field on the west coast of Corsica was determined.

Methods

Boulder fields in the vicinity of STARESO were sampled by snorkeling in September 2014. The average water temperature was about 22°C. Sampling took place in the harbor area directly in front of the station and on the boulder fields of the adjacent bays along the northwest shoreline of the Revellata peninsula. Organisms were collected from shallow depths (0-6 m) until slightly above the sea surface, into zip plastic bag and then transferred into dishes filled with seawater at the station. The phyla and species identification was performed with the naked eye or under a reflected stereo microscope, if necessary. The taxonomic identification was based on Hofrichter (2002), Guido (2000), Bergbauer (2008) and Riedl (1983). Nomenclature follows Riedl (1983).

Results

Overall, 40 species of 6 phyla were identified, including Cnidaria, Mollusca, Annelida, Arthropoda and Echinodermata (Table 1). The molluscs were the most abundant phylum with 56% (Figure 1) of the species diversity, represented by the three classes Gastropoda, Bivalvia and Polyplacophora. The second and third most abundant phyla were the Annelida and Echinodermata, equally represented with 13% of the total species diversity, followed by the Arthropoda with 10% and the Cnidiaria with 8% (Figure 1).

Among the molluscs, gastropods snails formed the biggest group wih 64%, followed by bivalves with 22% and chitons (polyplacophores) with 14% (Figure 2).

All collected organisms were identified to species level.

Phylum	Class	Family	Species
Cnidaria	Anthozoa	Actinidae	*Actinia equina*
			Anemonia sulcata
		Dendrophyllidae	*Balanophyllia europaea*
Mollusca	Gastropoda	Trochidae	*Gibbula umbilicalis*
			Gibbula divaricata
			Monodonta turbinata
		Haliotidae	*Haliotis tuberculata*
			Haliotis lamellosa
		Littorinidae	*Littorina neritoides*
		Patellidae	*Patella ulyssiponensis*
		Conidae	*Conus mediterraneus*
		Thaididae	*Thais haemastoma*
		Columbellidae	*Mitrella minor*
		Cerithiidae	*Gourmya vulgata*
		Muricidae	*Hexaplex trunculus*
	Bivalvia	Arcidae	*Barbatia barbata*
		Pectinidae	*Chlamys multistriata*
			Chlamys brunei
		Aecidae	*Arca noae*
		Mytilidae	*Musculus costulatus*
		Carditidae	*Cardita calyculata*
	Polyplacophora	Chitonidae	*Chiton olivaceus*
			Acanthochitona communis
		Ischnochitonidae	*Lepidochitona cinerea*
		Lebidopleuridae	*Lepidopleurus cancellatus*
Annelida	Polychaeta	Aphroditidae	*Lepidonotus clava*

		Nereidae	*Nereis pelagica*
		Terebellidae	*Nicolea venustula*
		Terebellidae	*Amphitritinae -*
Nermatoida	Nematoda	Lineidae	*Lineus geniculatus*
Arthropoda	Crustacea	Grapsidae	*Pachygrapsus marmoratus*
		Porcellanidae	*Pisidia longicornis*
		Parthenopidae	*Maia squinado*
		Xanthidae	*Xantho poressa*
Echinodermata	Ophiuroidea	Ophiuridae	*Ophioderma longicaudum*
	Echinoidea	Echinidae	*Paracentrotus lividus*
		Toxopneustidae	*Sphaerechinos granularis*
		Arbaciidae	*Arbacia lixula*

Table 1: Taxonomically identified species with family, class and phylum.

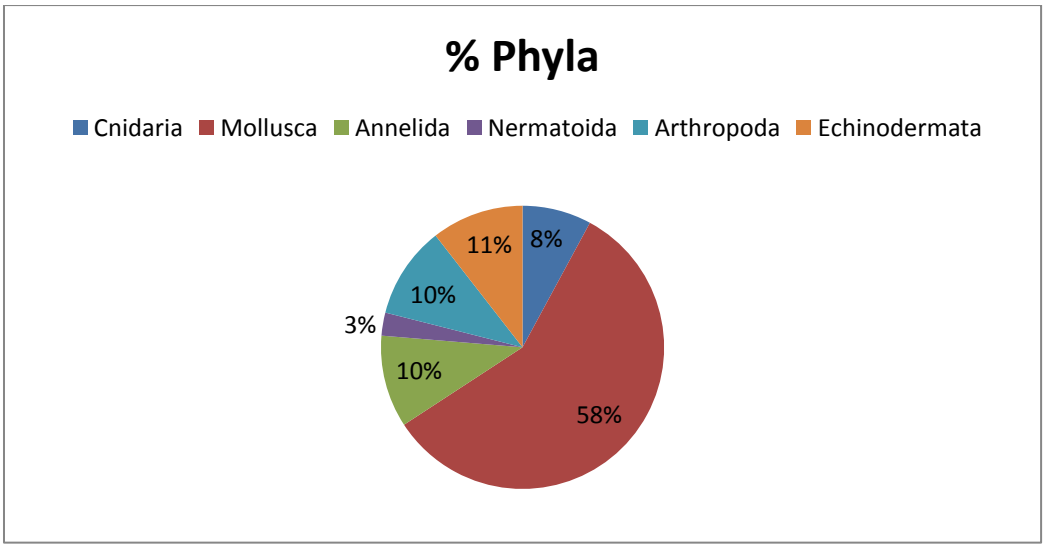


Figure 1: Percentage of phyla of the total amount of organisms found.

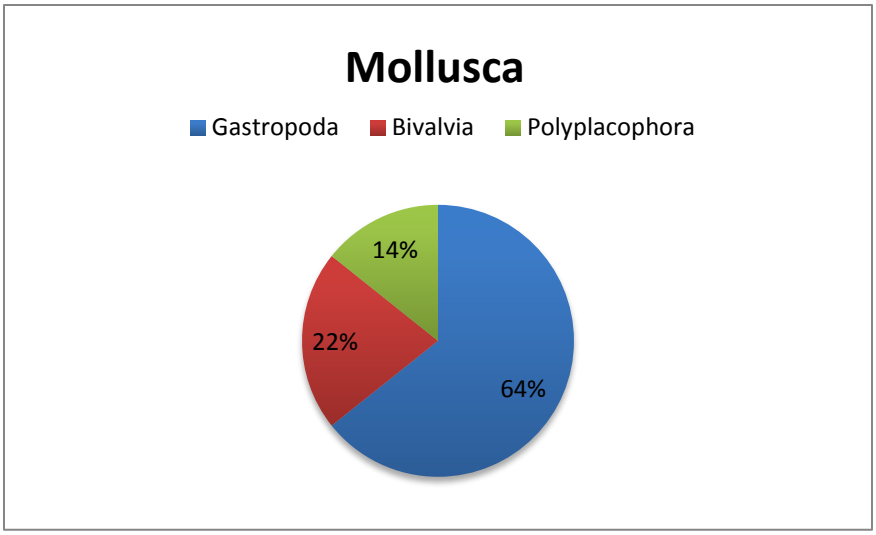
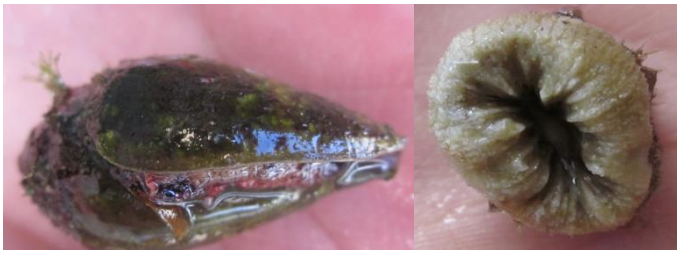


Figure 2: Classes of the molluscs.

Thais haemastoma *Gibbula umbilicalis* *Haliotis tuberculata*



Conus mediterraneus *Balanophyllia europaea*

Lineus geniculatus

Haliotis tuberculata Arbacia lixula *Paracentrotus lividus* *Sphaerechinus granularis*

Figure 3: Representative images of some species found on the boulder field.

Discussion

The molluscs were the most abundant phylum, especially the gastropods, which live mostly on the rocks and are able to attach to stones more easily. Their hard shell protects them from the power of the tides and waves, as well as from turning stones. Bivalves are filter feeders and do not prefer the oligotrophic water around Corsica with their low nutrient content (Hofrichter 2002). On the contrary, gastropods are mostly grazers or predators and find enough food on the vegetated rocks. There is a high abundance of overgrowing algae on these stones. Close to the station the frequency and intensity of destructed biota growing on the boulders is low.

Granite rock is a very hard substrate and consequently not accessible for lime drilling organisms, like some sponges or mollusks (Hofrichter 2002). Echinoderms in the area are either grazers (sea urgins), suspension feeders (some brittle stars, some sea cucumbers) deposit feeders (most sea cucumbers) or carnivors (sea stars). Through their bigger size they have larger territories and are not as abundant as the small mollusks. Also the number of counted species is noticeable lower compared to mollusks (Hofrichter 2002).

Crustaceans and annelids are for sure underrepresented in the catches, due to their high mobility or hidden live style. The same is true for the Cnidaria, though they are sessile forms living on the boulders. They are generally difficult to remove from the stone because they have adhesive discs and nematocytes for protection.

Comparison to the last years:

The determined phyla were more or less equally abundant compared to 2012. Compared with the years before some phyla were not collected, including Porifera, Echiurida and Tunicata. It has to be taken into account that the results heavily depend on the snorkeling skills of the collectors and the weather conditions.

Suggestion for improvement:

A connection between species richness and the stone size would be a possibility to restrict species to a smaller habitat inside the boulder field, including turnover rates of the stones and measurement of the flow rate inside the habitat. The biodiversity according to the habitat could be explored in more detail.

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Algae

Dana Michaelis, Isabella Hilti and Jascha Dehm

Introduction

Throughout this course, different types of macro-algae were collected to describe the diversity and the bathymetric patterns of the seaweed communities close to the STARESO Station in Calvi. Macro-algae are macroscopic, multi-celled and photosynthetic organisms, which are classified into red algae (Rhodophyta), green algae (Chlorophyta) and brown algae (Heterokontophyta). They are usually attached to hard substrates in coastal areas and are restricted to the photic zone (Goffredo and Dubinsky, 2014). Due to their size and their attachment to different substrates macro-algae are also named seaweed (Dawes, 1998). The three different groups of algae have different absorption characteristics due to their accessory photosynthetic pigments resulting in growth at various depths. Green algae are mainly located in the shallow water followed by brown algae, whereas red algae can still grow in deeper waters (Larkum *et al*., 2003). Macro-algae can be classified into different life-form categories due to the substratum they are attached to. During our study, the most frequent life-form was lithophytic/epilithic, which means that the algae are attached to stones, rocks or boulders. Macro-algae can also be divided into functional form groups like calcareous, corticated, foliose, filamentous or leathery (Short & Coles, 2001).

The biodiversity within the macro-algae is high. Currently, 1124 species of macro-algae are occurring in the Mediterranean Sea with about 20% of endemic species. This diversity is presently influenced by the introduction of invasive species. An invasion route is for example the Suez Canal, which connects the Red Sea with the Mediterranean Sea (Goffredo & Dubinsky, 2014).

Methods

Algal samples were collected from two locations (i.e. Cliffside exposed to sunlight, Cliffside not exposed to sunlight), as well as various depth profiles: 0-1m, 1-3m, >3m. This allowed for comparison of algal type with regard to habitat and exposure to sunlight and hence the light spectrum. Sampling and identification was carried out by all members of the Calvi 2014 excursion on various days between the 31st of August 2014 and the 2rd of September 2014 and was carried out via snorkelling whereby algae was plucked from the rocky surface by hand and transferred into allocated zip-lock bags. On land the samples were transferred to containers, buckets or petri-dishes to allow for species identification. Care was taken in order to avoid confusion or mixing of samples. Identification was done to the lowest possible scale and was conducted by consulting identification keys and literature.

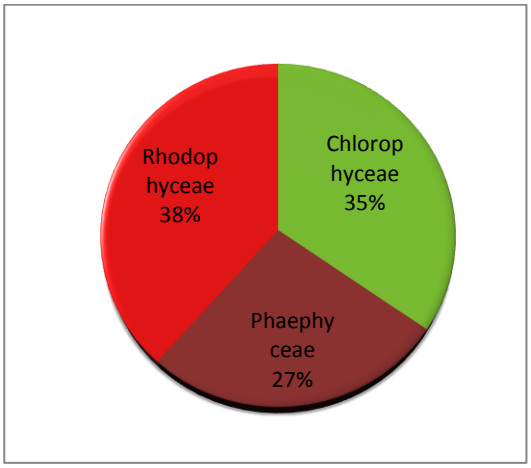

Figure 1: Percentage total of observed algae

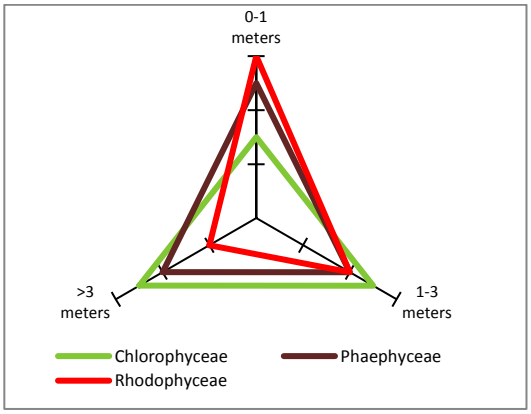

Figure 2: Comparison of algal distribution with regard to depth

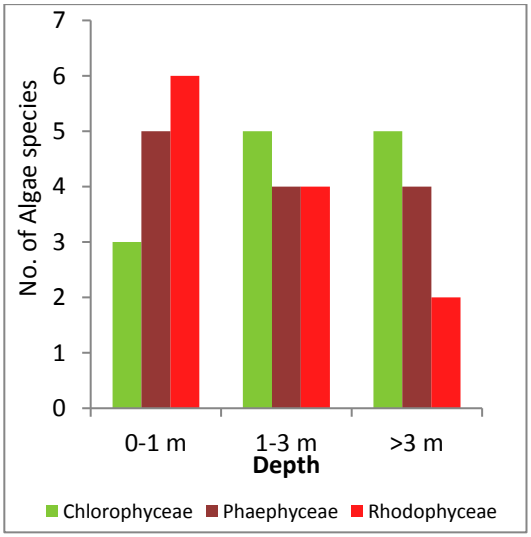

Figure 3: Algae distribution with regard to depth

Results

Overall 26 algae species belonging to 19 genera and 20 families were identified throughout the survey (refer to Table 1). Algae of the class Rhodophyceae recorded the greatest species number (10 species) making up 38% of the total species identified (Figure 1). The class Chorophyceae made up 35% (9 species) of the total species found and Phaephyceae accumulated to only 27% (7 species) (Figure 1).

Figure 2 and Figure 3 show that the different classes dominated different depth zones. Rhodophyceae dominated the surface water (0-1m) and decreased with increasing depth (Figure 2). At the surface 6 species were identified (Figure 3), namely (Table 1) *Amphiora rigida, Corallina elongate, Corallina officinalis, Peyssonnelia squamaria, Callithamnion corymbosum*, and *Laurencia obtuse*. At intermediate depth, species number decreased to 4 individuals namely (Figure 3), *Crouania attenuate, Amphiora fragilissima, Nemastoma dichotomum* and *Peyssonnelia squamaria* (Table 1). In deeper waters, only 2 Rhodophyceae were found (Figure 3); *Peyssonnelia squamaria* and *Peyssonnelia rubra* (Table 1). *P. Squamaria* was the only species of red algae found throughout the water column, whereas the other species were only found within the respective depth zones.

On the other hand, Chlorophyceae was found to be the least dominant at surface waters (0-1m) but the most dominant at intermediate (1-3m) and deep depths (>3m)(Figure 2). At the surface layer, only 3 species of Chlorophyceae were sampled (Figure 3), namely *Spongomorpha aeruginosa, Udotea petiolata* and *Udotea spinulos* (Table 1). Five species (Figure 3) were found at depths between 1-3m, i.e. *Halimeda tuna, Cladophora prolifera, Anadyomene stellata,*

Udotea petiolata and *Flabelia petiolata* (Table 1). The Chlorophyceae community in the deeper water (>3m) was also comprised of 5 species (Figure 3); *Codium bursa, Codium effusum, Halimeda tuna, Udotea spinulosa* and *Udotea petiolata* (Table 1). *Udotea petiolata* was the only green algae found throughout the different depth zones.

Phaeophyceae did not dominate a single depth zone (Figure 2), however in terms of species number this class was the least affected by changing depths (Figure 3) (i.e. species number was not significantly influenced by depth). In total 5 species were found at the surface layer of which only *Dictyota linearis, Dictyota linearis* and *Cystoseira sp.* were limited to this zone (Table 1). *Dictyota dichotoma* and *Padina pavonica* (Table 1) were found in all three depth zones. Apart from *D. Dichotoma* and *P. Pavonica* 2 other brown algae species were found at each of the depth zones below the surface zone, namely: *Dictyota implexa and Halopteris scoparia* (Table 1). Figure 4 describes the distribution of the algae classes with regard to exposure to sunlight. Rhodophyceae species was found to be of higher abundance on the cliff side less exposed to sunlight (i.e. shaded side), whereas Chlorophyceae and Phaeophyceae had higher species numbers on the side exposed to sunlight.

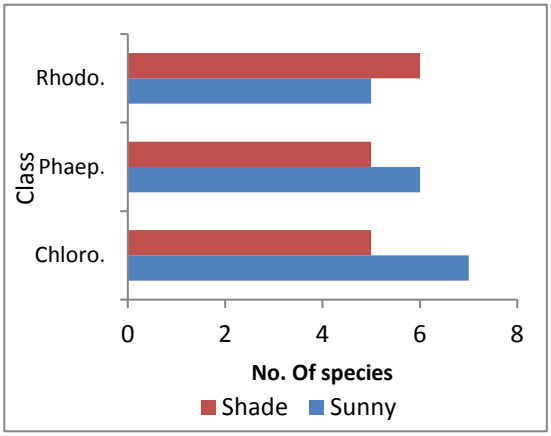

Figure 2: Algae distribution with regard to shade/sunny exposure

Table 1: Algal species list and collection zone

note: L --> licht, S--> schatten				
		Fundort		
Klasse-Ordnung-Familie	Art	0-1m	1-3m	3-5m
Chlorophyta (Grünalgen)				
Acrosiphoniales				
Acrosiphoniaceae	*Spongomorpha aeruginosa*	L		
Bryopsidales				
Codiaceae	*Codium bursa*			L
	Codium effusum			S
Halimedaceae	*Halimeda tuna*		S	S/L
Cladophorales				
Cladophoraceae	*Cladophora prolifera*		L	
Anadyomenaceae	*Anadyomene stellata*		L	
Ulvales				
Udoteaceaae	*Udotea petiolata*	S/L	S	S
	Udotea spinulosa	S/L		S
	Flabelia petiolata		S	
Phaeophyta (Braunalgen)				
Dictyotales				
Dictyotaceae	*Dictyota dichotoma*	S	S	S/L
	Dictyota implexa		L	L
	Dictyota linearis	S/L		
	Dictyopteris polypodioides	S/L		
	Padina pavonica	S/L	S	S/L
Ectocarpales				
Chordariaceae	*Cystoseira sp.*	L		
Stypocaulaceae	*Halopteris scoparia*		S	S
Rhodophyta (Rotalgen)				
Ceramiales				
Ceramiaceae	*Crouania attenuata*		L	
Corallinales				
Corallinaceae	*Amphiora fragilissima*		L	
	Amphiora rigida	S		
	Corallina elongata	L		
	Corallina officinalis	S		
Gigartinales				
Nemastomataceae	*Nemastoma dichotomum*		S	
Peyssonneliales				
Peyssonneliaceae	*Peyssonnelia squamaria*	S	S	S/L
	Peyssonnelia rubra			S
Rhodymeniales				
Ceramiaceae	*Callithamnion corymbosum*	S		
Rhodomelaceae	*Laurencia obtusa*	L		

Discussion

The structure of marine algae communities is influenced by various factors, of which the availability of sunlight (required for primary production) is probably the most crucial. The ability to harvest various light intensities is a key aspect of the community structure and vertical zonation of algae (Markager & Sand-Jensen, 1992; Wiencke & Bishof, 2012). Typically Chlorophyceae followed by Phaeophyceae have the shallowest depth range due to the dependency on the higher light frequencies. On the other hand, Rhodophyceae are able to utilize slightly lower light frequencies and therefore have the greatest depth range (Markager & Sand-Jensen, 1992).

The results of this survey show a slightly different pattern, that is, when looking at the vertical zonation Rhodophyceae are dominant closer towards the surface and species number decreases with depth. Phaphyceae also had slightly higher species numbers at the surface and did not change much with depth. The Chlorophyceae was found to have the least number of species present in the surface waters but increased with depth. This reverse zonation may be attributed to the fact that Chlorophyceae are generally more fleshy and less durable than Rhodophyceae. Hence growth at the surface is hindered due to exposure to tides or waves which would increase wear and tear, predation or dehydration of the plant. The Rhodophyceae and Phaphyceae are more robust and are therefore less at risk to predation, dehydration or wear and tear by waves.

The fact that Rhodophyceae are able to survive in deeper waters with reduced light intensity can be observed when comparing algal communities on the shaded side of the cliff with the side exposed to sunlight (Figure 4). More species of Rhodophyceae were found on the shaded side then the exposed side, whereas higher numbers of species of Chlorophyceae and Phaphyceae were found on the exposed side. This is likely due to Rhodophyceae being able to harvest lower-intensity-light then Chlorophyceae and Phaphyceae (Markager & Sand-Jensen, 1992).

The algal community plays an important role in the marine environment in the sense that it aids in primary production, construction, cementation, bio-erosion and that it forms microhabitats for various other organisms (Markager & Sand-Jensen, 1992; Wiencke & Bishof, 2012). Therefore an understanding and monitoring of the algal community structure and its zonation are crucial when studying/conserving or working with the marine environment (Wiencke & Bishof, 2012).

The number of species sampled and identified this year (26 species) was significantly less than in the 2012 and 2010 excursions (34 and 37 species, respectively). This rather large difference could have been due to sampling/identification error, however a possibility exists that the algal community at the sampling location is changing in terms of structure. This possible change in community structure could be attributed to alterations in grazing communities, outcompeting by more robust algae, changes in oceanographic and climatic factors and so on. Similar cases have been recorded before by Wiencke & Bishof (2012), and therefore further studies are recommended in order to confirm this speculation.

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Seagras

Verena Naschberger, Huajing Yan and Bianca Jansen

Introduction

Seagrass meadows in the Mediterranean Sea

Seagrass is a monocotyledonous plant and appears monotypic, which means that seagrass meadows consist of one species. In the Mediterranean Sea there are four common families: Posidoniaceae, Zosteraceae, Cymodoceaceae and Hydrocharitaceae. *Posidonia oceanica* is endemic in the Mediterranean Sea, all other species occur also in the Atlantic or in the Black Sea (see Table 1). All species show the same leaf morphology and grow at sedimentary substrate.

Table 1: Four common seagrass families in the Mediterranean Sea. M=Mediterranean Sea, AL= Atlantic, RS= Red Sea and BS= Black Sea.

Taxa	Distribution	Substrate	Depth [m]
Posidoniaceae			
Posidonia oceanica	M (endemic)	Coarse sand	3-40 (max. 50)
Zosteraceae			
Zostera marina	AL, M, BS	Mud, sand	Shallow, 6-10
Nanozostera noltii	AL, M, BS	Mud, sand	Shallow, 6-10
Cymodoceaceae			
Cymodocea nodosa	AL, M, BS pioneer species	Sand with finer material	Shallow, 1 – 35
Hydrocharitaceae			
Halophila stipulacea	RS, eastern M, Lessepsian immigrant	Mud, sand	10-15

Ecological importance of Posidonia oceanica

Posidonia oceanica plays an important role in stabilisation of sand, gravel and sludge. Seagrass acts as a so-called “sediment trap”, the water movement is stopped and nutrients of the fine sediment are accumulated. These nutrients are necessary for the fauna which inhabit the seagrass. Furthermore, a seagrass meadow increases the habitat structure. Each square metre is covered by approximately 1000 leaves with a length of up to 60 to 80 cm. Thus, a leaf area index (LAI) of 20 is reached, which means that per m^2 bottom area 20 m^2 leaf surface are built.

Moreover, the Mediterranean Sea is very nutrient poor and seagrass meadows are one of the most productive habitats and primary producers. *Posidonia oceanica* is responsible for a net carbon fixation of 3000 g C m^{-2} a^{-1} in decomposed and remineralized form, however a quarter is built by epiphytes. Per m^2 seagrass meadow 14 litres of oxygen are generated.

The increase of surface structure and 3D shape through the seagrass are important for the fauna. In this way, the seagrass offers protection, possibility to hide, spawning ground and food, although only few organisms feed on seagrass itself, but rather on the epiphytes.

Fauna of seagrass meadows

Per ha *Posidonia oceanica* up to 15 t of animal biomass may be measured. This consists of generalists as well as specialists. Seagrass meadows have a so-called “conveyer-growth” which means that photosynthetically active leaves are formed by the basal meristem. The main growth is in autumn and winter, thus sea grass leaves grow few cm per year. Co-evolution between plant and animals can be observed, due to coordinated life-cycles.

Endangered habitat

Destruction of seagrass meadows in general is caused by altered current conditions, due to construction of docks, piers and protective dams or ship´s propellers. Furthermore, the seagrass gets destroyed through mechanical damage, such as anchors or trawl nets. Reduction in seagrass meadows from nutrient overflow could be either due to direct impact from high nutrient concentrations or indirect impact from the increase in water turbidity. In consequence, the seagrass located at greater depths will die due to light limitation. Moreover, detergents or algal bloom can affect the seagrass meadows negatively, however this is pure speculation and not proven.

Methods

In order to examine animal life in a seagrass meadow of *Posidonia oceanica,* several samples were collected from different areas of the water in front of the STARESO station. These samples were collected at a depth of 6 meters. Some plants and parts of plants were collected into a plastic bag with lots of surrounding water, to catch as much of the organisms inhabiting the seagrass meadow as possible. The samples of *Posidonia* were kept to a minimum in order not to strain this protected species unnecessarily.

Back onshore the samples were placed into labelled buckets and later on brought to the laboratory, where they were searched for organisms using a microscope, classified taxonomically (using Riedl (1983) and internet [WoRMS and NBN]) and photographed (see Figure 4).

The samples were separated into

- Live plant with rhizome (L,R)
- Dead leaves (D)
- Free floating plant parts (W)

Results and Discussion

From all samples of *Posidonia* collected, a total of 48 species were found and identified, which were separated into 10 phyla. The largest phylum was Mollusca with 11 species identified, closely followed by Arthropoda with 10 species (see Table 2).

The species richness was the main focus of this survey rather than abundance. When looking at the specific location, the highest species richness was observed on the leaves of the *Posidonia*. A total of 21 species were found on the *Posidonia* leaves. In the dead *Posidonia*, there was also a high richness of species where 20 species were identified. Only 2 species were found on the rhizomes and 9 free floating species were found in the water.

The highest species diversity found on the *Posidonia* leaves belongs to the phylum Arthropoda, as shown in figure 1. Arthropod species richness amounted to almost half of the total species number found on the leaves. It is noteworthy that Cnidaria species were only found in dead *Posidonia* (see Figure 1-3) and occurred as the highest species richness in overall dead *Posidonia* (see Figure 2). Though live *Posidonia* had the largest amount of species found, no Cnidaria species were identified (see Figure 1).

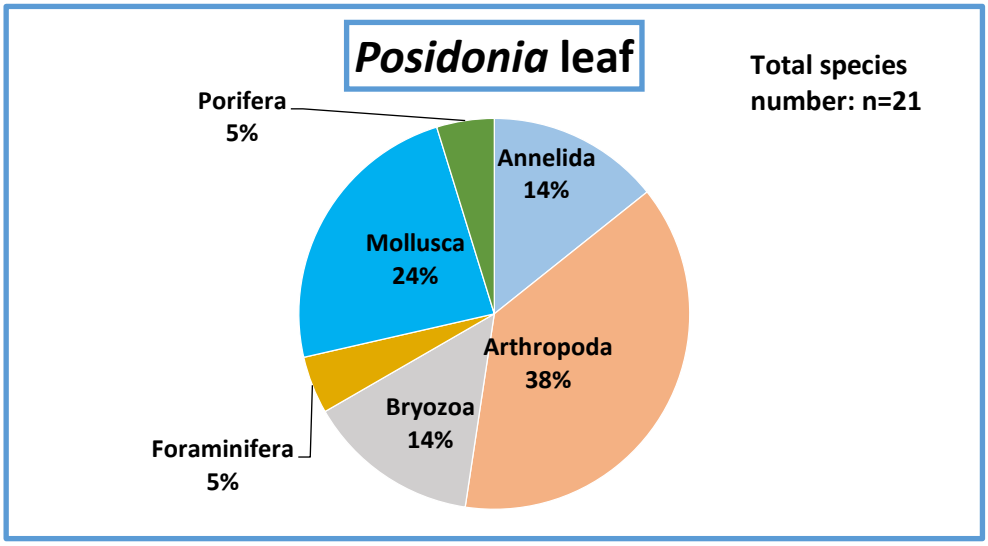

Figure 1: Percentage of each phylum found on live Posidonia

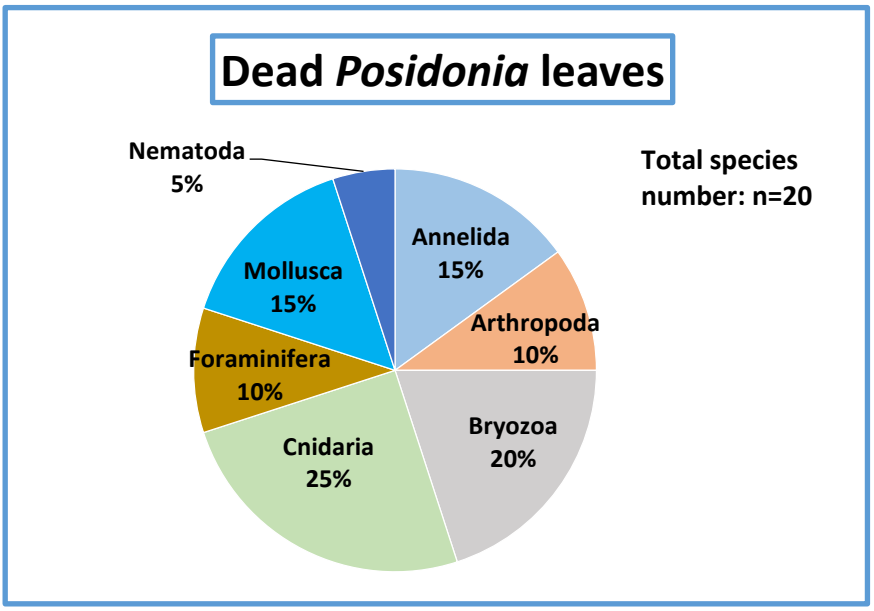

Figure 2: Percentage of each phylum found on dead Posidonia leaves.

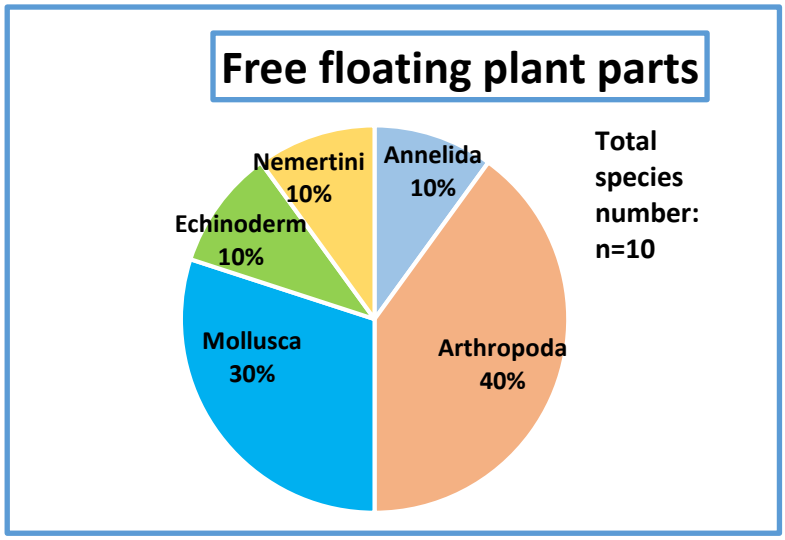

Figure 3: Percentage of each phylum found on free floating leaves.

Less species were found on free floating plant parts (see Figure 3), probably because species would need a place to anchor and use as shelter, feeding ground, or food source, which in this case would be on the *Posidonia*. Only half the number of species as on rooted *Posidonia* was found on the free floating plant parts with Mollusca and Arthropoda as the dominant phyla.

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Internet

- WoRMS: World Register of Marine Species http://www.marinespecies.org
- NBN: National Biodiversity Network http://nbn.org.uk/

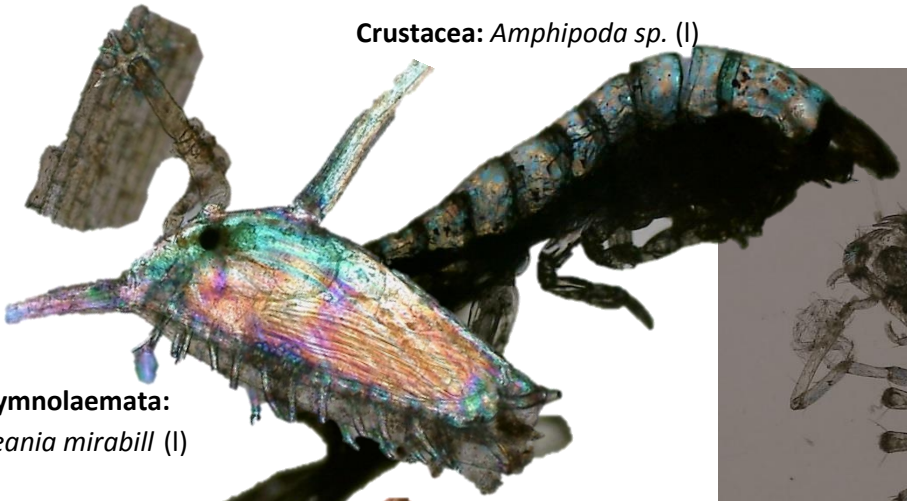
Table 2: Summary of all collected and classified organisms in sea grass meadow at a depth of 6 meter in front of the marine station STARESO in Calvi/Corsica. Samples were collected on 31st Aug. - 1st Sep. 2014. Species were identified using Riedl (1983) and WoRMS.

Phylum	Division	Class	Family	Species	Sample type			
					L	R	D	W
Bryozoa		Gymnolaemata	Aeteidae	*Aetea truncata*		X	X	
		Gymnolaemata	Beaniidae	*Beania mirabilis*	X			
		Gymnolaemata	Electridae	*Electra posidoniae*	X		X	
		Gymnolaemata	Farrellidae	*Farrella repens*			X	
		Gymnolaemata	Microporellidae	*Fenestrulina malusii*	X			
		Stenolaemata	Lichenoporidae	*Patinella radiata*	X			
		Stenolaemata	Tubuliporidae	*Tubulipora liliacea*			X	
				gen.sp.	X			
Nemertini	Hoplonemertea	Enopla	Oerstediidae	*Oerstedia dorsalis*				X
Annelida		Polychaeta	Eunicidae	*Eunice aphroditois*			X	
		Polychaeta	Nereididae	gen.sp.				X
		Polychaeta	Polynoideae	*Lepidonotus clava*	X			
		Polychaeta	Sabellidae	*Sabella pavonina*	X			
		Polychaeta	Serpulidae	*Janua pagenstecheri*			X	
		Polychaeta		gen.sp.	X		X	
Mollusca	Aculifera	Polyplacophora	Chitonidae	*Chiton olivaceus*	X			
	Conchifera	Gastropoda	Atlantoidea	gen.sp.				X
	Conchifera	Gastropoda	Cerithiidae	*Bittium latreillii*	X			
	Conchifera	Gastropoda	Creseidae	*Creseis sp.*				X
	Conchifera	Gastropoda	Littorinoidea	gen.sp.				X
	Conchifera	Gastropoda	Pyramidellidae	*Chrysallida sp.*			X	
	Conchifera	Gastropoda	Rissoidea	*Alvania lineata*	X	X		
	Conchifera	Gastropoda	Rissoidae	gen.sp.	X			
	Conchifera	Gastropoda	Vermetidae	*Vermetus triquetrus*			X	
Note: L = Leaf, R = Rhizome, D = Dead Seagrass, W = Water (Free floating)								

Phylum	Division	Class	Family	Species	Sample type			
					L	R	D	W
Mollusca	Conchifera	Gastropoda	Vermetidae	gen.sp.			X	
	Conchifera	Gastropoda		gen.sp.	X			
Nematoda				gen.sp.			X	
Arthropoda	Chelicerata	Acari	Halacaridae	gen.sp.	X			X
	Crustacea	Malacostraca	Alpheidae	*Athanas sp.*	X			
	Crustacea	Malacostraca	Amphipoda	gen. sp.				X
	Crustacea	Malacostraca	Caprellidae	*Caprella acanthifera*			X	
	Crustacea	Malacostraca	Idoteidae	*Idotea sp.*	X			
	Crustacea	Malacostraca	Ischyroceridae	*Ericthonius punctatus*	X			X
	Crustacea	Malacostraca	Maeridae	*Elasmopus rapax*	X			
	Crustacea	Maxillopoda	Ostracoda	gen.sp.	X			X
	Crustacea	Maxillopoda	Porcellidiidae	*Porcellidium sp.*	X			
	Crustacea	Maxillopoda	Tisbidae	*Tisbe furcata*	X		X	
	Pantopoda	Pycnogonida	Callipallenidae	*Callipallene emaciata*	X			
Echinodermata		Ophiuroidea	Amphiuridae	*Amphipholis sp.*				X
Note: L = Leaf, R = Rhizome, D = Dead Seagrass, W = Water (Free floating)								

Phylum	Division	Class	Family	*Species*	Sample type			
					L	R	D	W
Foraminifera		Globothalamea	Elphidiidae	*Elphidium crispum*			X	
		Globothalamea	Homotremidae	*Miniacina miniacea*			X	
				gen. sp.	X			
Porifera		Demospongiae	Suberitidae	*Suberites sp.*	X			
Cnidaria		Hydrozoa	Campanulariidae	*Campanularia hincksii*			X	
		Hydrozoa	Campanulariidae	*Clytia gracilis*			X	
		Hydrozoa	Campanulariidae	*Clytia hemisphaerica*			X	
		Hydrozoa	Eirenidae	*Campanopsis sp.*			X	
		Hydrozoa	Syntheciidae	*Synthecium evansi*			X	
Note: L = Leaf, R = Rhizome, D = Dead Seagrass, W = Water (Free floating plant parts)								

Crustacea: *Amphipoda sp.* (l)

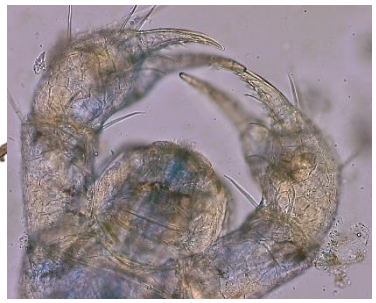


Gymnolaemata:
Beania mirabill (l)

Pantopoda: *Callipalene emaciata* (v)

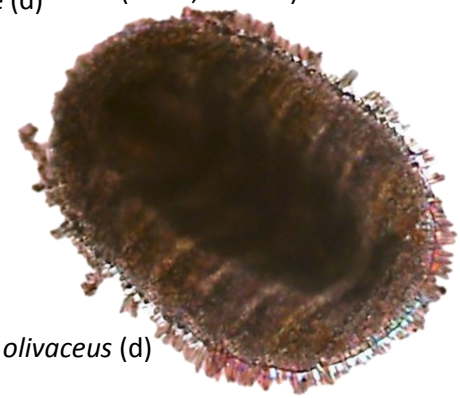
Gastropoda: *Creseis sp.* (l)

Acari: Halacaridiae (d)



Pantopoda: *Callipalene emaciata*
(Head, ventral)

Crustacea: *Tisbe furcata* (l)



Polyplacophora: *Chiton olivaceus* (d)

Crustacea: *Porcellidium sp.* (v)

Figure 4: Photographs taken with microscope camera from organisms found in Posidonia. d: dorsal view; l: lateral view; v: ventral view

Coralligène

Saskia Amann, Laura von Raffay,
and Raphael Strohmaier

Figure 1: Piece of rock overgrown by coralline algae, with shells and calcareous tubes of serpulid worms on it.

Introduction

For there is no clear definition of what exactly the Coralligène is, we used this term to describe marine secondary hard substrates mainly formed by coralline red algae. Other organisms contribute to the growth of this biogenous rock bottom, like bryozoans, serpulid polychaetes, cnidarians, molluscs, sponges, crustaceans and foraminifers. For the structure of this habitat mainly depends on the physical parameters and the biology of the major coralline algae, a wide variety of morphological appearances can be found. Growth of the Coralligène is limited by light abundance and physical factors like currents. Biological degradation of the habitat occurs mainly by boring bivalves like the *Lithophaga lithophaga* or sponges of the family Clionaidae, which can dissolve the calcium carbonate and find shelter in the massive structures of the Coralligène.

Generally, we can divide the habitat into two different types; The “Coralligène de trottoir” in the infralittoral zone and the “Coralligène de plateau” found in the circalittorale zone.

Whilst the Coralligène de trottoir appears on submerged vertical cliffs close to the surface, the Coralligène de plateau can be found on horizontal sea beds. The lower limit of growth is set by the penetration of light to the ocean floor.

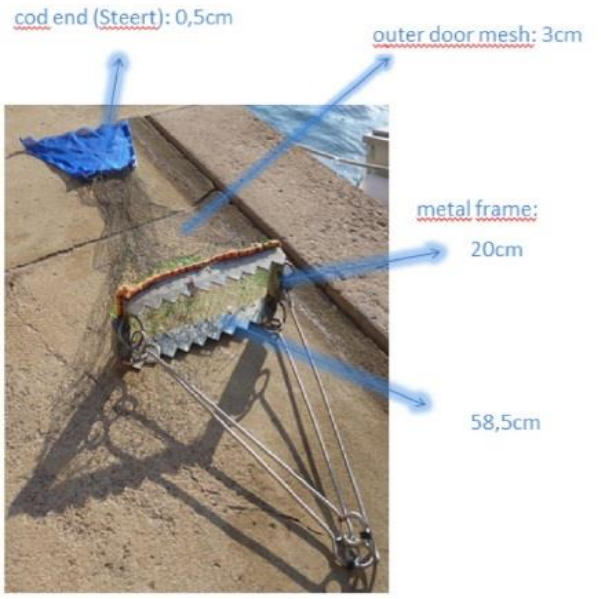


Figure 2: Illustration of our Dredge, with specific dimensions as used in the field.

Methods

For our studies, sampling has been conducted on the Coralligène de plateau in an average depth of 45 to 56 meters. In front of the Punta de l'Oscelluccia, it builds up on sandy sediments and is easy to be sampled via dredge.

Dredge:

- outer door mesh size: 3 cm
- Steert (cod end): 0.5 cm
- Frame opening: 20 cm X 58.5 cm

Our study site was right in front of STARESO, reached by the station motorboat in approx. 15 minutes. The dredge has been lowered to the ocean floor in 45 m depth and slowly towed horizontally through the secondary hard substrate for 10 minutes. By the time we stopped dredging, we reached an average depth of 56 meters. After dragging the sample out of the water, we returned to the station for further analyzes. Species identification was carried out after manually separating the whole dredge sample into smaller plastic boxes. When identification down to species level was complicated, we only defined the genus.

Table 1: List of all Identifications including Phylum and Family name. Some identification was only possible down to genus level.

	Coralligène	
	Date: 2nd September, 2014	
	Time: 14:53 – 15:03	
	Depth: 45m – 50m	
	Sampling Method: Dredge	
	Outerdoormesh: 3cm	
	Cod end: 0.5cm	
	Dredge opening: width 58.5cm / length 20cm	
	Identified: 2nd September, 2014	
Phylum	Family	Genus + Species
Annelida	Aphroditidae	*Aphrodite aculeata*
Annelida	Phyllococidae	*Eulalia sp.*
Annelida	Sabellidae	*Sabella paviona*
Annelida	Terebellidae	*Polycirrus aurantiacus*
Arthropoda	Diogenidae	*Dardanus arrosor*
Arthropoda	Inachidae	*Inachus sp.*

Arthropoda	Leucosiidae	*Ebalia edwardsi*
Arthropoda	Majidae	*Maya squinado*
Arthropoda	Paguridae	*Catapaguroides timidus*
Arthropoda	Parthenopidae	*Parthenope massena*
Chlorophyta	Caulerpaceae	*Caulerpa racemosa*
Chlorophyta	Codiaceae	*Codium bursa*
Chlorophyta	Halimedaceae	*Halimeda tuna*
Chlorophyta	Udoteaceae	*Udotea petiolata*
Chlorophyta	Valoniaceae	*Valonia utricularis*
Chlorophyta	Zosteraceae	*Zostera marina*
Chordata	Gobiesocidae	*Diplecogaster bimaculatus*
Chordata	Gobiesocidae	*Apletodon incognitus*
Chordata	Pyuridae	*Microcosmus sp.*
Cnidaria	Hormathiidae	*Calliactis parasitica*
Echinodermata	Amphiuridae	*Amphiura chiajei*
Echinodermata	Echinarachniidae	*Echinarachnius sp.*
Echinodermata	Echinasteridae	*Echinaster sepositus*
Echinodermata	Holothuriidae	*Holothuria polii*
Mollusca	Arcidae	*Arca noae*
Mollusca	Calliostomatidae	*Calliostoma laugieri*
Mollusca	Cardiidae	*Acanthocardia spinosa*
Mollusca	Cerithiidae	*Gourmya vulgata*
Mollusca	Lucinidae	*Lucinoma borealis*
Mollusca	Muricidae	*Bolinus brandaris*
Mollusca	Pectinidae	*Chlamys opercularis*
Mollusca	Pectinidae	*Chlamys varia*
Mollusca	Pectinidae	*Chlamys flexulosa*
Mollusca	Turritellidae	*Turritella turbona*
Nemertea	Cerebratulidae	*Cerebratulus fuscus*
Nemertea	Tetrastemmatidae	*Tetrastemma diadema*
Nemertea	Tubulanidae	*Tubularius superbus*
Rhodophyta	Corallinaceae	*Pseudolithophyllum expansum*
Rhodophyta	Corallinaceae	*Lithothamnium fruticulosum*
Rhodophyta	Corallinaceae	*Lithophyllum sp.*
Rhodophyta	Peyssonneliaceae	*Peyssonnelia squamaria*
Rhodophyta	Rhodomelaceae	*Vidalia volubilis*

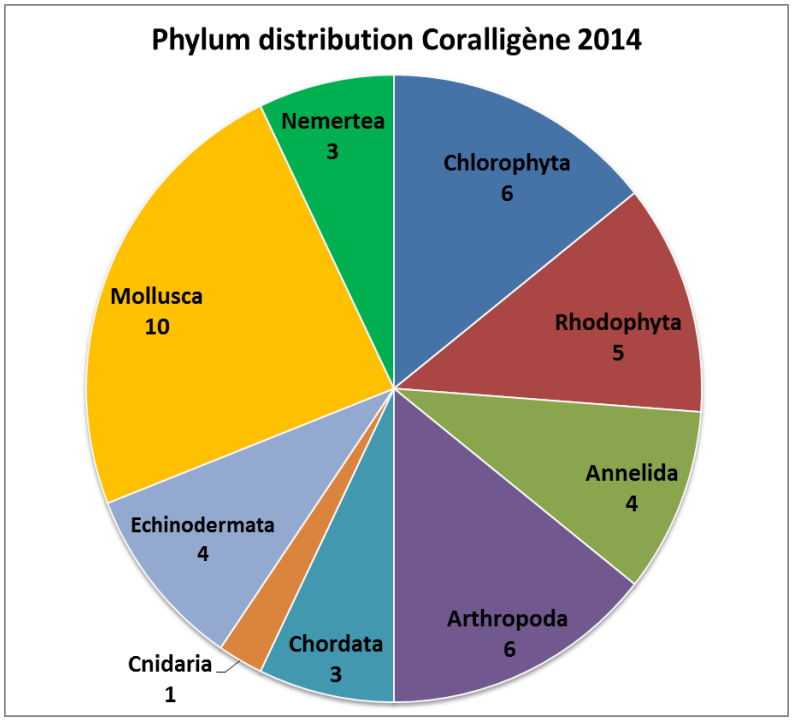


Figure 3: Proportion of identifications in the Coralligène sample by Phylum.

Results

With 10 mollusc species identified, this phylum dominated our dredge sample just before Arthropoda and Chlorophyta with 6 species identified each. Rhodophyta contributed 5 species, Annelida and Echinodermata 4. Nemertea and Chordata have been scarcer with only 3 species per phylum, and Cnidaria showed the least abundance in our sample with only one species identified (see Fig. 3). Most common families were the Pectinidae of the Mollusca phylum and the Corallinaceae belonging to the Rhodophyta. We could find 3 species in those families. The most common genus was *Chlamys* with 3 species inherent, belonging to the family of Pectinidae mentioned beforehand. A total of 42 identifications has been scored, with 37 species and 5 determinations down to genus level (Table 1).

Discussion

The first impression when retrieving the dredge to the surface was the high abundance of *Caulerpa racemosa*, an invasive green algae from the red sea. To avoid spreading this species, samples containing *C. racemosa* had been destroyed after processing. Molluscs seem to dominate the species composition of the Coralligène. Excluding the two groups of Algae, the molluscs contributed 32% of all animal species found in our samples. This might be due to the sandy substrate the Coralligène is located on in front of

STARESO. This sand is an important habitat and hiding ground for many molluscs. Over all, the results barely reflect the species numbers given by literature. The Dredge sampling method only gives a rough overview of benthic and bottom-dwelling organisms. Animals that are usually buried in the sediment can hardly be retrieved with this method.

Comparing the results to the dredge findings of the last years, we gathered a broad data set of not just Animalia but also several algae species. Species found in a second dredge sampling are not included in this list because the focus by then lay on mollusc species, therefore not reflecting the actual species diversity of Dredge samples.

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Plankton

Clara Hechenberger, Stefanie Kuen, and Raimund Schnegg

Introduction

In 1887, the term “plankton“ was coined by Viktor Hensen, at that time professor at the university of Kiel, Germany. Actually the term is derived from the Greek adjective *πλαγκτός - planktos*, meaning errant, and by extension "wanderer" or *drifter*. This term was used for the entire community of organisms in the open water column, which cover distances by being drifted rather than by their own abilty to swim. The highly diverse community of the marine plankton includes various unicellular and multicellular organisms, so large trophic-taxonomic differences can be found between the categories phytoplankton (unicellular photoautotrophic algae), zooplankton (heterotrophic protists and metazoans), and bacterio-plankton (prokaryotes), to which also the virio-plankton (viruses) belongs. Because of huge ranges of size in this group, size classification has been introduced. This classification begins with the extremely small size of viruses (0.2 m) and ends up with the size of large jellyfish and tunicates (2.0 m).

Life cycle

Species specificity is also given in differences of the life cycles. The group of the Holoplankton includes the pelagic species that spend all their life cycle as passively drifting plankton in the open water column. Meroplankton is called the larval stages and as an adult organism they are part of the Necton, which is a group that is partially able to move actively and change the swim direction independently of the water motion.

Trophic relationships

There are three basic ecological categories of organisms: producers, consumers and decomposers. The phytoplankton provides the photoautotrophic primary producers, which assemble their own food in deriving energy from the sunlight. Then there are herbivores or primary consumers, which directly graze on diatoms and dinoflagellates, and carnivores or secondary and tertiary consumers, which comprise the raptorial zooplankton feeding on animals. They are the basis of the food chain. And there are decomposers, e.g. bacteria and fungi, breaking down the organic matter of dead organisms into inorganic salts. The „marine snow“ is an effect of aggregated live and dead gelatinous planktonic organisms, that are stuck together by adhesive substances.

The majority of the marine vertebrates produce planktonic larvae. Different physiological mechanisms provide for a high degree of synchronisation of both sexes maturing and spawning in time and space. This results in successful and highly activated fertilisation. And for this reason, Meroplankton could dominate in the plankton community of coastal waters in certain seasons. Predation by planktonic predators (e.g. scyphomedusae, hydromedusae and siphonophores) and benthic predators (mostly sessile filter feeders) is the most significant possibility for the high mortality of the planktonic larvae. Of course fish are also important predators on invertebrate larvae, but also larval cannibalism is possible.

The microbial loop shows the different trophic levels in the water column. Zooplankton is dependent from Phytoplankton and so it is located in the photic zone (surface layer). Therefore most of the plankton is located in the upper 200 m of the sea. Bacterioplankton is able to remineralize organic material.

Plankton undergoes a vertical movement along the day. The particles move up when the sunlight appears and sink down afterwards. This movement influences strongly the food web, because predators depend on the abundance of Plankton and are reacting on the movement.

Material and Methods

Plankton was collected several times in a depth of about 6-8 m in the open sea. For the collection, a net was pulled for 15 minutes very slowly behind the boat. The low speed was chosen because of the importance not to harm the small and sometimes soft, gelatinous plankton particles. The length of the net was about 3 m (Figure 1). The diameter of the big opening at the beginning of the fishing tool was about 53 cm and at the end 9 cm. The first part collects the bigger particles, the finer substrates go to the thinner plastic cube. This collection container works as a filter and collects the very small Plankton particles. In the front the net has a diameter of 500 µm and in the end 250 µm. The collected plankton was afterwards put into a box with seawater. In the lab two different probes were taken for analysis. One pipette was taken from the middle of the box and one pipette full of plankton from the ground. The petri dishes with the collected plankton were then analysed with binoculars and microscopes and determined as good as possible with different identification literature. Some of the identified samples were photographed under the microscope and are listed with pictures below.

Figure 1: Plankton net used to collect the samples (left). It has a length of about 3 m. Filter unit for the small plankton particles for sampling bigger than 250 m (right).

Results

Adulta

Kingdom	Phylum	Class	Order	Family	Species
Chromista	Foraminifera	Globothalamea	Rotaliida	Globigerinidae	*Globigerinoides* sp.
Chromista	Myzozoa	Dinophyceae	Gonyaulacales	Ceratiaceae	*Ceratium contrarium*
Chromista	Myzozoa	Dinophyceae	Gonyaulacales	Ceratiaceae	*Ceratium massiliense*
Chromista	Ochrophyta	Bacillariophyceae	Chaetocerotanae incerta sedis	Chaetocerotaceae	*Chaetoceros densus*
Chromista	Radiozoa	n/n	n/n	n/n	Gen. spez.
Animalia	Platyhelminthes	Trematoda			
Animalia	Mollusca	Gastropoda	Thecosomata	Creseidae	*Creseis* sp.
Animalia	Mollusca	Gastropoda	Thecosomata	Creseidae	*Creseis acicula*
Animalia	Mollusca	Gastropoda	Thecosomata	Limacinidae	*Heliconoides* sp.
Animalia	Mollusca	Gastropoda	Thecosomata	Limacinidae	*Heliconoides inflatus*
Animalia	Arthropoda	Maxillopoda	Calanoida	Acartiidae	*Acartia* sp.
Animalia	Arthropoda	Maxillopoda	Calanoida	Calanidae	*Calanus* sp.
Animalia	Chordata	Appendicularia	Copelata	Oikopleuridae	*Oikopleura* sp.
Animalia	Chordata	Appendicularia	Copelata	Oikopleuridae	*Oikopleura dioika*
Animalia	Chaetognatha	Sagittoidea	Aphragmorpha	Sagittidae	*Sagitta* sp.

Larvae

Kingdom	Phylum	Class	Order	Family	Species
Animalia	Mollusca	Gastropoda	n/n	n/n	Veliger larva
Animalia	Sipuncula	Sipunculidae	n/n	n/n	Trochophora larva
Animalia	Phoronidae	n/n	n/n	n/n	Gen. spez.
Animalia	Arthropoda	Branchiopoda	Diplostraca	Podonidae	*Evadne* sp.
Animalia	Arthropoda	Branchiopoda	Diplostraca	Podonidae	*Evadne normanni*
Animalia	Arthropoda	Malacostraca	Decapoda	Crangonidae	*Crangon* sp.
Animalia	Arthropoda	Malacostraca	Decapoda	Protunidae	*Carcinus* sp.
Animalia	Arthropoda	Malacostraca	Decapoda	Upogebiidae	*Upogebia* sp.
Animalia	Echinodermata	n/n	n/n	n/n	Pluteus larva
Animalia	Chordata	Actinopterygii	n/n	n/n	Egg

The list contains a small part of all the organisms from all samples who are embossing the name plankton. The following pictures show some representatives of our plankton collection.

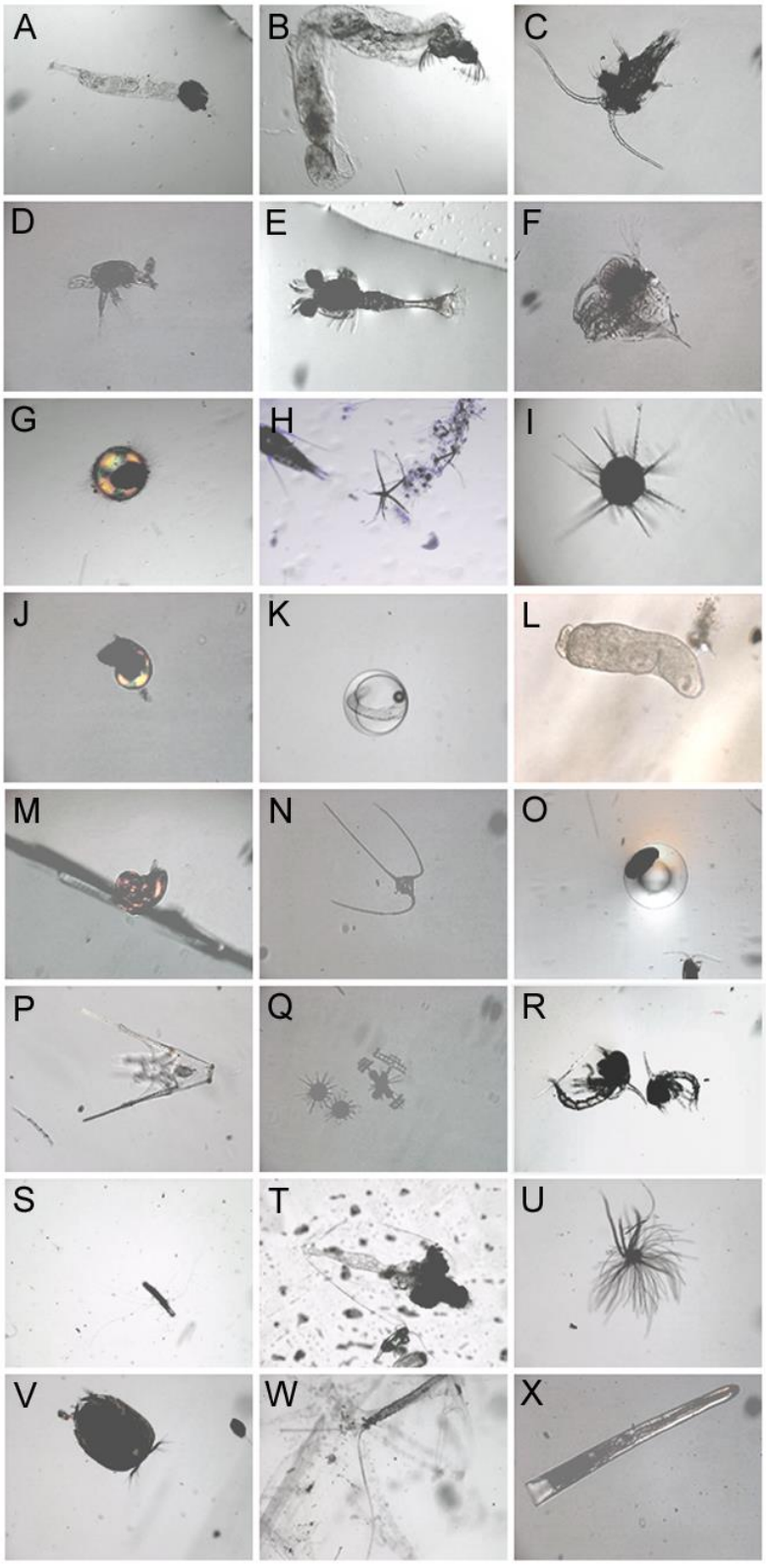

Figure 2: DIC contrast images of representative species. (A) Appendiculata spp., (B) Sagitta sp., (C) Acartia sp., (D) Crustacea Gen. spp., (E) Decapoda Gen. spp., (F) Evadne sp., (G) Foraminifera Gen. spp., (H) Phytoplankton Gen. spp., (I) Phyllostaurus siculus, (J) Gastropod embryo, (K) Teleostei egg, (L) Nematoda Gen. spp., (M) Veliger larva, (N) Cerratium sp., (O) Fish embryo, (P) Pluteus larvae, (Q) Radiolaria Gen. spp., (R) Zoea larvae, (S) Bacillariophycea sp., (T) Siphonophora larva, (U) Polly larva, (V) Porcellidium sp., (W) Thaliacea sp., (X) Thecosomata sp.

Discussion

During this excursion, students examined three plankton samples that were collected with the help of a plankton net pulled by boat. We have documented 7 single celled Chromista comprising 4 different phyla, namely Foraminifera, Myzozoa, Ochrophyta and Radiozoa as well as 23 species of multicellular animals. Among the adult animal species we have found representatives of at least 12 different phyla, including Platyhelminthes, Mollusca, Arthropoda, Chordata and Chaetognatha. Altogether the identified samples are very common for plankton and show a small part of the species richness of this habitat.

Remarkable is the high abundance of larval forms with 11 documented species of animal taxa comprising 6 phyla, namely Mollusca, Sipuncula, Phoronidae, Arthropoda, Echinodermata and Chordata. Larval forms may possibly use the current of the water to be distributed wider and inhabit new areas, while adult representatives are maybe less mobile, as for example Mollusca or other species. Despite being taxon and species rich, plankton has a high organismic density, but no thoroughly quantification was performed. To conclude, our results confirm that our plankton taken three times from one sampling place, is rich in species and individuals, so that still new species and taxa can be discovered during the following excursions.

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Makrofauna of the Sandy Beach

Werner Bader

Summary

On September 4th 2014 our group went to the “Mara Beach” to take samples of Macro- and Meiofauna. Therefore it was necessary to use different collection techniques. Macrofauna is the animal size of more than one mm. Animals of the Macrofauna live in different habitats like sea-grass, the stony and rocky floor of the sea as well as on sand grains and between the space of sand grains. These animals were caught with sieves, nets and by hand catching. The mesh size of the sieves were two to four mm.

The following species have been found on Mara Beach:

Phylum	Class	Ordo	Family	*Species*
Mollusca	Bivalvia	Heterodonta	Lucinidae	*Ctena decussata*
Mollusca	Bivalvia	Heterodonta	Chamoidea	*Chama sp.*
Mollusca	Bivalvia	Arcoida	Arcidae	*Barbatia barbata*
Mollusca	Bivalvia	Limoida	Limoidea	*Lima sp.*
Mollusca	Gastropoda	Patelloidea	Patelloida	*Patella sp.*
Mollusca	Bilvalvia	Pterioidea	Pinnidae	*Pinna nobilis*
Mollusca	Placophora	Chitonida	Chitonidea	*Chiton olivaceus*
Mollusca	Cephalopoda	Sepiida	Sepiidae	*Sepia officinalis*
Mollusca	Cephalopoda	Octopoda	Octopodidae	*Octopus vulgaris*
Mollusca	Bivalvia	Arcoida	Arcoidea	*Arca sp.*
Mollusca	Bivalvia	Veneroida	Chamidae	*Pseudochama gryphina*
Crustacea	Malacostraca	Decapoda	Paguroidea	*Pagurus sp.*
Echinodermata	Echinoidea	Camarodonta	Parechinidae	*Paracentrotus lividus*
Vertebrata	Actinopterygii	Perciformes	Labridae	*Symphodus cinereus*
Vertebrata	Actinopterygii	Perciformes	Labridae	*Symphodus rostratus*
Vertebrata	Actinapterygii	Pleuronectiformes	Bothidae	*Botus podas podas*
Vertebrata	Actinopterygii	Perciformes	Sparidae	*Lithognathus mormyrus*
Vertebrata	Actinopterygii	Atheriniformes	Atherinidae	*Atherina boyeri*
Vertebrata	Actinopterygii	Perciformes	Sphyraenidae	*Sphyraena sphyraena*
Vertebrata	Actinopterygii	Perciformes	Labridae	*Symphodus roissali*
Vertebrata	Actinopterygii	Perciformes	Sparidae	*Diplodus vulgaris*
Vertebrata	Actinopterygii	Perciformes	Serranidae	*Serranus scriba*
Vertebrata	Actinopterygii	Atheriniformes	Atherinidae	*Atherina hepsetus*

The species mentioned above were found in different habitats: the sandy and rocky ground near the beach, the open sea water near Mara Beach, the surrounding *Posidonia* as well as the surrounding rocky shore.

Specimen from the sandy ground of Mara Beach (caught by hand and sieves):

Phylum	Class	Ordo	Family	*Species*
Mollusca	Bivalvia	Heterodonta	Lucinidae	*Ctena decussata*
Mollusca	Bivalvia	Veneroida	Chamidae	*Pseudochama gryphina*
Mollusca	Bivalvia	Heterodonta	Chamoidea	*Chama sp.*
Mollusca	Bivalvia	Limoida	Limoidea	*Lima sp.*
Mollusca	Placophora	Chitonida	Chitonidea	*Chiton olivaceus*
Mollusca	Bivalvia	Arcoida	Arcoidea	*Arca sp.*

Specimen from the open sea water near to Mara Beach (caught by net):

Phylum	Class	Ordo	Family	*Species*
Vertebrata	Actinopterygii	Perciformes	Sparidae	*Diplodus vulgaris*
Vertebrata	Actinopterygii	Perciformes	Labridae	*Symphodus roissali*
Mollusca	Cephalopoda	Sepiida	Sepiidae	*Sepia officinalis*
Mollusca	Cephalopoda	Octopoda	Octopodidae	*Octopus vulgaris*
Vertebrata	Actinopterygii	Atheriniformes	Atherinidae	*Atherina boyeri*

Specimen from the surrounding rocky shore (caught by hand):

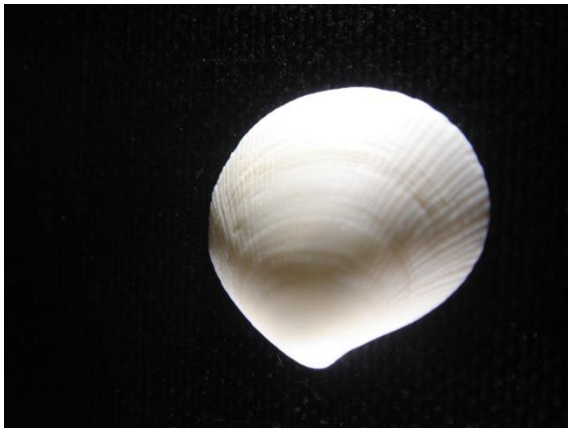
Phylum	Class	Ordo	Family	*Species*
Mollusca	Gastropoda	Patelloidea	Patelloida	*Patella sp.*
Mollusca	Bivalvia	Arcoida	Arcidae	*Barbatia barbata*
Mollusca	Bivalvia	Limoida	Limoidea	*Lima sp.*
Mollusca	Placophora	Chitonida	Chitonidea	*Chiton olivaceus*
Echinodermata	Echinoidea	Camarodonta	Parechinidae	*Paracentrotus lividus*

Specimen from the surrounding Posidonia (caught by hand and net):

Phylum	Class	Ordo	Family	*Species*
Mollusca	Bilvalvia	Pterioidea	Pinnidae	*Pinna nobilis*
Crustacea	Malacostraca	Decapoda	Paguroidea	*Pagurus sp.*
Vertebrata	Actinapterygii	Pleuronectiformes	Bothidae	*Botus podas podas*
Vertebrata	Actinopterygii	Perciformes	Sparidae	*Lithognathus mormyrus*
Vertebrata	Actinopterygii	Perciformes	Labridae	*Symphodus cinereus*
Vertebrata	Actinopterygii	Perciformes	Labridae	*Symphodus rostratus*
Vertebrata	Actinopterygii	Atheriniformes	Atherinidae	*Atherina hepsetus*

Image gallery ordered by Phylum:

Molluscs:



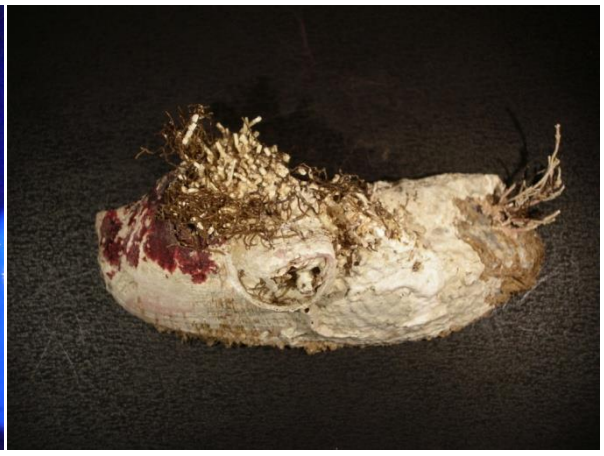
Ctena decussata

Chama sp.

Barbatia barbata

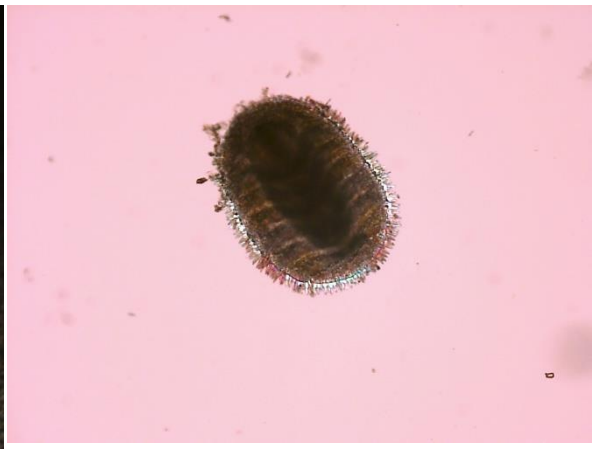
Lima sp.

Pinna nobilis

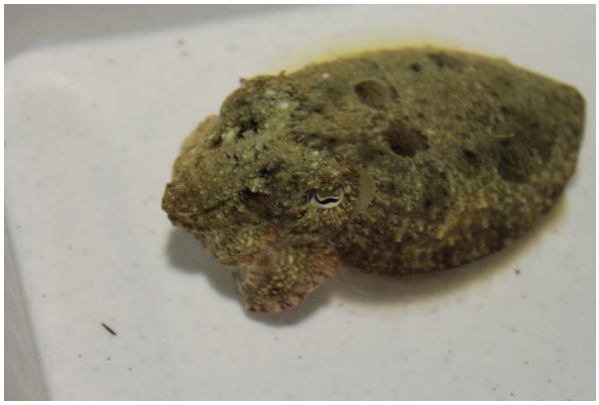


Arca sp.

Pseudochama gryphina



Chitonidea



Sepia officinalis

Octopus vulgaris

<u>Crustaceans:</u>

Pagurus sp.

<u>Echinoderms:</u>

Paracentrotus lividus

Vertebrates:

Symphodus cinereus

Symphodus rostratus

Diplodus vulgaris

Botus podas podas

Lithognathus mormyrus

Atherina boyeri

Serranus scriba

Symphodus roissali

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Meiofauna of the Sandy Beach

Nina Egger and Raimund Schnegg

Introduction

As the Meiofauna, zoobenthic species between 20µm-1mm sizes are denoted. The substrate, in which the Meiofauna is resident, is called the Mesopsammon. In this habitat, several niches can be used by the organisms. Since the species move in between the sand grains without perturbing them, the habitat can also be called interstitium, the inhabitants are therefore interstital.

Material and Methods

As already mentioned above, the size of these individuals is limited, so in the first step special equipment such as fine sifters are needed to sort out the organisms of the Mesopsammon. We used sifters of different mesh sizes to compare the different bio-communities within different grain sizes of the sediment. In the following we list a short protocol of how to conduct the extraction of meiofaunal species process.

Methods:
To gain the meiofaunal organisms several steps were carried out, this is explained in following steps:
Step 1: Prepare a 7.14% $MgCl_2$ solution.
Step2: Pour about 5 teaspoons of sand into an Erlenmeyer-flask.
Step 3: Mix a 1:1 $MgCl_2$-seawater solution.
Step 4: Pour this solution to the sample, shake it gently and incubate it for 10min.
Step 5: Shake the mixture again and then shifter it through one of the current shifters (Figure 1).

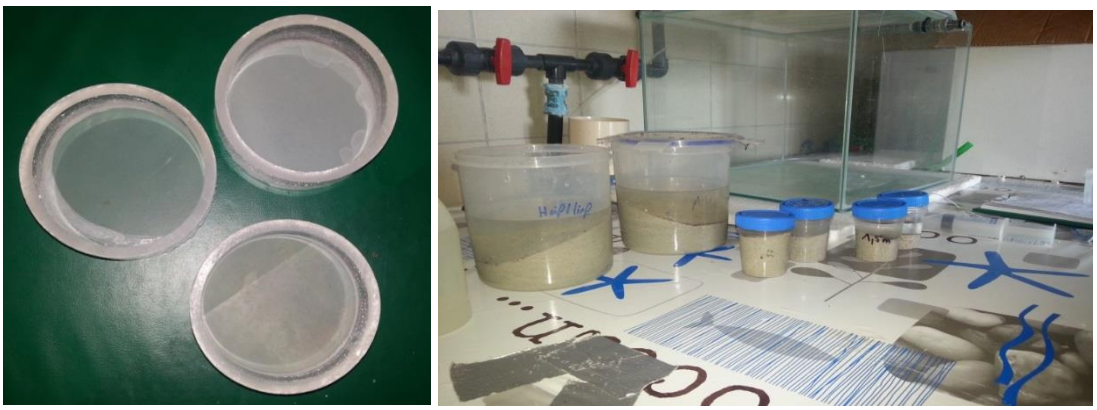

Figure 1: Filters of different mesh sizes and sand samples of the different sample depths

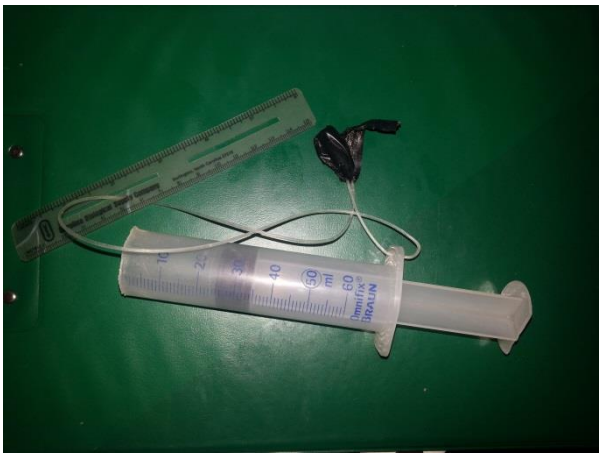

Figure 2: Core-sampler for consistent sample amounts

Measurement:

2 different depths were examined with three different sifter sizes (Figure 1).

15 µm	**40 µm**	**63 µm**
Hip-deep [1m]	Hip-deep [1m]	Hip-deep [1m]
Breast-deep [1.4m]	Breast-deep [1.4m]	Breast-deep [1.4m]

Results

The species found in the different mesh sizes are listed in Tables 1 – 3.

Table 1: Qualitative listing of the found exemplars within mesh size 15µm.

Phylum	***Class***	***Ordo***	***Family***	***Species***
Annelida	Polychaeta	n/n	n/n	*Gen. spez.*
Mollusca	Scaphopoda	n/n	n/n	*Gen. spez.*
Nemathoda	n/n	n/n	n/n	*Gen. spez.*
Acoelomorpha		Acoela	Sagittiferidae	*Symsagittifera corsicae*

Table 2: Qualitative listing of the found exemplars within mesh size 40µm.

Phylum	*Class*	*Ordo*	*Family*	*Species*
Ciliophora	Karyorelictea	Protostomatida	Trachelocercidae	*Tracheloraphis phoenicopterus*
	Litostomatea	Pleurostomatida	Litonotidae	*Litonotus cygnus*
Foraminifera	Globothalamea	Rotaliida	Asterigerinatidae	*Asterigerinata mamilla*
	Globothalamea	Rotaliida	Elphidiidae	*Cribroelphidium vadescens*
	Globothalamea	Rotaliida	Elphidiidae	*Elphidium crispum*
	Globothalamea	Rotaliida	Rosalinidae	*Neoconorbina terquemi*
	Globothalamea	Rotaliida	Rotaliidae	*Ammonia tepida*
	Tubothalamea	Miliolida	Spiroloculinidae	*Spiroloculina rostrata*
	incerta sedis	Lagenida	Vaginulinidae	*Lenticulina vortex*
	n/n	n/n	n/n	*Gen. spez.*
Arthropoda	Maxillipoda	Harpacticoida	Ectinosomatidae	*Microsetella norvegica*
				Gen. spez.
Annelida	Polychaeta	Phyllodocida	Hesionidae	*Hesione pantherina*
		Phyllodocida	Nereididae	*Nereis sp.*
Mollusca	Scaphopoda	Dentaliida	Dentaliidae	*Dentalium sp.*
				Gen. spez.
Nemathoda	n/n	n/n	n/n	*Gen. spez.*
Xenacoelomorpha		Acoela	Sagittiferidae	*Symsagittifera corsicae*

Table 3: Qualitative listing of the found exemplars within mesh size 63µm.

Phylum	*Class*	*Ordo*	*Family*	*Species*
Ciliophora	Karyorelictea	Protostomatida	Trachelocercidae	*Tracheloraphis phoenicopterus*
	Karyorelictea	Protostomatida	Trachelocercidae	*Tracheloraphis sp.*
	Litostomatea	Pleurostomatida	Litonotidae	*Litonotus cygnus*
Foraminifera	Globothalamea	Rotaliida	Cibicididae	*Lobatula sp.*
	Globothalamea	Rotaliida	Elphidiidae	*Elphidium crispum*
	Globothalamea	Rotaliida	Stainforthiidae	*Cassidorina sp.*

Arthropoda	Maxillipoda	Harpacticoida	Miraciidae	*Stenhelia inopinata*
Xenacoelomorpha		Acoela	Sagittiferidae	*Symsagittifera corsicae*
Chaetognatha	Sagittoidea	Phragmophora	Spadellidae	*Spadella sp.*

Representative bright field microscopic images:

Figure 3: Polychaeta Gen. spez.

Figure 4: Symsagitifera corsicae

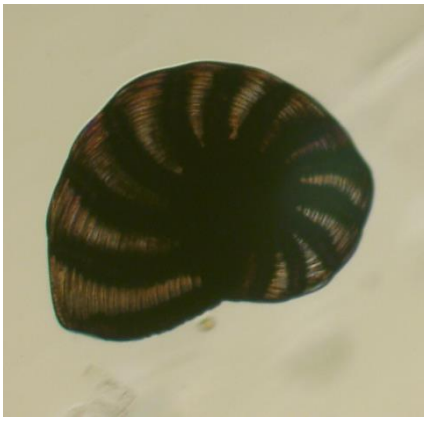

Figure 5: Foraminifera Gen. spez.

In the following, a short description of the various phyla found is given.

Ciliophora

The ciliophora are a heterogeneous group of heterotrophic, single-celled organisms. There are about 8.000 described species united by a common blueprint and a common way of reproduction. They possess multiple, short cilia, a specific structure of the cortex, a nuclear dimorphism as well as a special type of gamontogamy – the conjugation.

The cortex is composed by two structures – the pellicula and the root structures of the cilia – and reaches a thickness of 1 – 4 µm. The pellicula is made up by the cell membrane, which is sometimes coated by the so-called perilemma. Close to the cilia invaginations can be observed, the parasomal

pockets [sic?] where pinocytosis takes place. Underneath the plasmalemma lies a system of flattened vacuoles, the so-called alveoli. Ciliophora possess a distinctive mouth region, the cytostome, which defines the ventral side, as well as a cell after, the cytopyge, to eliminate larger food particles.

The most distinctive feature of the ciliophora is their nuclear dimorphism. They possess one to several somatic macronuclei and one to several micronuclei. The large macronucleus is responsible for the cell metabolism, while the assignment of the micronucleus lies solely in reproduction.

Larger, elongated ciliophora can be confused with microturbellaria. But they are easy to distinguish when observing their movement: ciliophora can move forward as well as backward, while microturbellaria, as all flatworms, can only move forward.

Foraminifera

Foraminifera are marine protozoa characterized by a shell (called test), reticulopodia and an alternation of generations. The test consists of ingested sand grains or self-made calcite. There are single-chambered tests (monothalme) or multi-chambered tests (polythalme). The chamber lumina in these polythalme tests are connected by openings, the eponymous foramina. The characteristic pseudopodia of the foraminifera extrude through small foramina of the test and form a fine network. This network of pseudopodia is called rediculopodia. The reticulopodia are characteristically streamed by cytoplasma, carrying along visible mitochondria, elliptical vesicles and food particles. Small (< 1 mm) foraminifera use bacteria and dinoflagellata as a food source, while larger species (up to several cm) live on photosynthesis products of their endosymbiontic, single-celled algae. Reproduction in foraminifera is connected with a heterophasic alternation of generations. A haploid sexual generation, the gamont, alternates with a diploid, asexual generation, the agamont. Besides their difference in chromosome number (haploid vs. diploid) do the two different generations differ in the number of nuclei – the gamont possesses always only one nucleus, while the agamont houses multiple nuclei.

Of the about 10.000 recent taxa most are marine and benthic, only a few are planktonic or live in fresh water. Foraminifera are known since the Cambrium since when they occurred in high abundance. Due to their good preservation and abundance they are often used as index fossils.

Harpacticoida, Maxillopoda, Arthropoda

Among the arthropoda, harpacticoid copepods are well known for their benthic lifestyle and can be found in meiofaunal samples in high abundance. Copepoda in general are a species rich taxon within the crustacean, with 12.000 species known so far. Copepoda play an essential role in the marine ecosystem as a food source for species of a higher trophic level due to their high abundance and individual density.

Free living copepods reach a size of 0.5 – 5 mm, only a few are larger. The body of free living copepods consists of a cephalothorax and 10 free body segments. The cephalothorax is composed of head and fused to a thoracomer. Thoracomer 2 – 6 each carry a pair of swimming legs. The 7th thoracomer is the genital segment, bearing the genital openings.

Phyllodocida, Polychaeta, Annelida

Polychaets comprise a group of annelida united by the character of having no clitellum. Therefore this group might by a paraphylum. As an attribute of the annelida they are segmented and, in contrast to the oligochaeta, bear multiple, in bundle organized bristles that insert in lateral appendages, the parapodia. There are about 9.000 described, mainly marine species.

Within the polychaete, the phyllodocida is the order with most of the motile polychaetes, the errantia, in contrast to the sedentary species, the sedentaria. Their segmentation is mostly homonomous and they are partly of great length and possess a high number of segments. The prostomium of phyllodocidae is well established with antennae, ventrally located palps and tentacle cirri. The phyllodocida are mainly benthic, but there are a number of holoplanktonic species as well.

Scaphopoda, Mollusca

The scaphopoda are a group of solely marine, 2 – 150 mm long, superficially bilateral symmetrical molluscs. They occur in seas all around the world from the eulitoral down to nearly 7000 m depth where they dwell in the sediment. There are about 600 species described so far. They bear a characteristic, elongated shell which is slightly curved, conical and open on both ends. The larger opening houses the majority of the mantle cavity and through this aperture foot and head appendages (so called captacula) can be protruded. The minor aperture serves to contact the sediment surface so that water can be forced through the mantle cavity.

Nematoda

Nematodes are considered to be the second largest animal phylum. Concerning their number of individuals they are by far the most abundant animals worldwide. As high the amount of species and individuals is, as divers are the habitats nematodes live in. They occur all around the world, some as cosmopolitans, where the habitat meets the requirement of the certain species. Many species are prominent parasites.

Their spindle shaped body makes them very suitable for the interstitial habitat. They move in-between the sand grains by undulating bending of their body axes. The bending is not a left-to-right bending as would be expected but a dorsal-ventral one as the animals lie on the left or right body side. Nematodes need a counterfort to move forward, without sand grains from which they can brace themselves against they remain stationary.

Acoela, Xenacoelomorpha

The Acoela are “flatworm-like” animals. This term is chosen because their systematic position is still highly debated. They form, together with their sister group Nemertodermatida, the taxon Acoelomorpha. For centuries this taxon was considered to be a basal group within the Platyhelminthes. In the beginning of the 21st century this few has changed by molecular systematics in the way that the Acoelomorpha were granted an even more basal position, as a sister group of all other bilaterians. Yet the most recent position within the animal tree of life is a sister group relation of the Acoelomorpha with the cryptic Xenoturbellaria. Together they form the taxon Xenacoelomorpha and are located within the deuterostomes as a sister group of the Ambulacralia.

The Acoelomorpha are characterized by a statocyst near the anterior end of the droplet-shaped body. Also located at the anterior part is the frontal organ, comprising gland- and sense-organs.

Another characteristic is the central digestive parenchyma which is organized as a syncytium. The mouth is located ventrally and no pharynx is developed. Some species possess symbiontic algae, which gives the Acoelomorph flatworms a colorful appearance as in the case of *Symsagittifera corsicae*, which we found in high abundance at the Sandy Beach.

Discussion

During the excursion we gained an insight of bio-communities in the interstitial habitat of the Sandy Beach. We detected representatives of a variety of taxa known to inhabit the interstitial. Most of the species were found in the range of 40 µm. This can be ascribed to the fact that most of the samples were examined from this fraction. In comparison only one sample of the 15 µm fraction was examined. Noteworthy is that the Chaetognath found in the 63 µm mesh size fraction does not belong to the Meiofauna, but might be protracted from the upper water column during sampling. Another alternative could be that it originates from the surface of the sand where it might forage. What could be expected was a higher abundance of Platyhelminthes, but we only observed Acoel flatworms. One cause could be that the acoel flatworms occur in such a high abundance, that they oust the Platyhelminthes. Another cause could be the spotted distribution of the interstitial fauna. We sampled only two closely located areas in 1 m and 1.4 m depth. This might be not representative and the locations are not significantly different from each other. One suggestion is, that at least one sample is taken from the shore, where it is known that for example Proseriata occur (RS personal communication with Robert Pjeta). Unfortunately the shore line of the Sandy beach is covered with dead and decomposing sea grass, which hampers sampling. But for future sampling it would be an option to search underneath the decomposing sea grass as well.

Excursion to the Beach of Girolata

Michaela Hittorf and Isabella Hilti

Introduction

Figure 1: The beach of Girolata

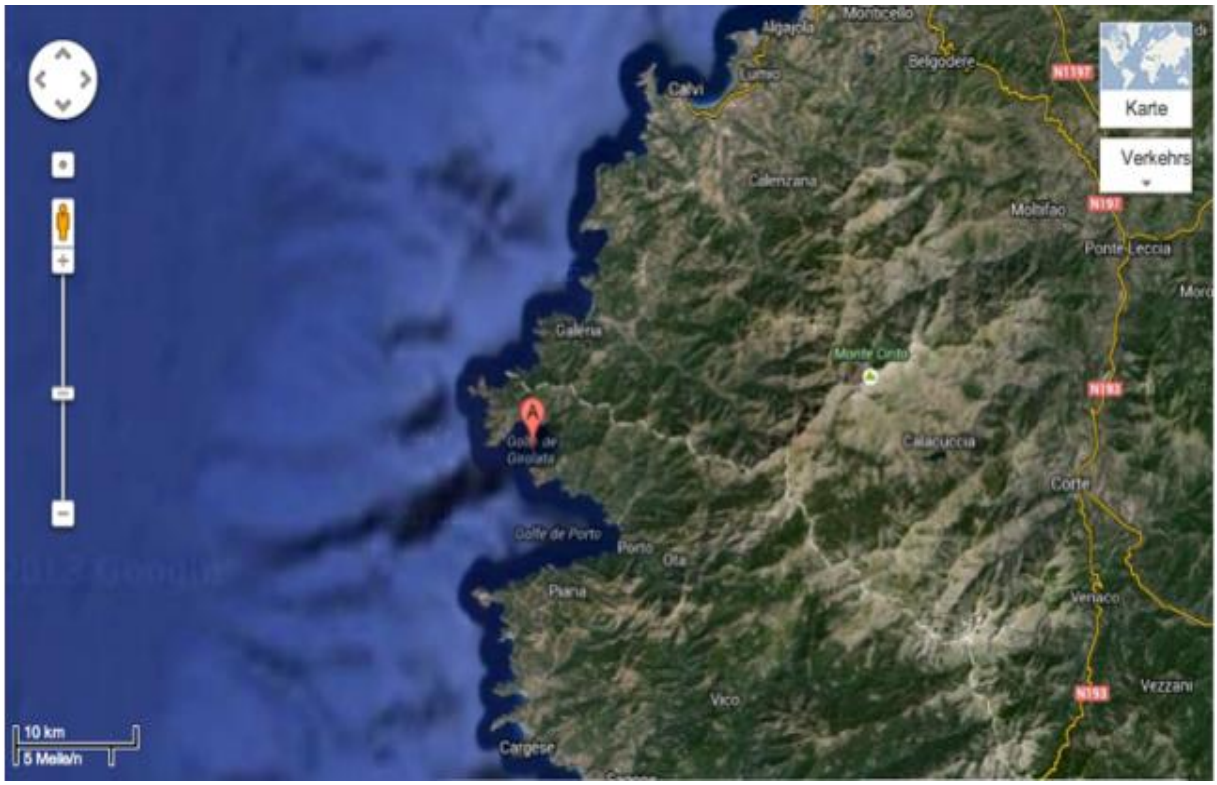

Figure 2: View to the Gulf of Girolata

The region Girolata has been a nature reserve since 1975 and lies on the north western coast of the island Corsica (Figures 1 and 2). It is also part of the Regional Natural Park of Corsica and was

added to the World Heritage List in 1983. The nature reserve extends over the Scandola peninsula, an imposing blood-red porphyritic rock mass. It was formed by volcanic activities in the Permian and comprises different geologically volcanic rocks such as porphyry, rhyolite and basalt. Beside beautiful beaches, the coastline also shows sheer and jagged red cliffs with many caves. Apart from the reddish rocks the vegetation is determined by scrubland. The Mediterranean Sea next to the Girolata beach is clear and has a lot of islets and caves, which includes a rich marine life. In the littoral zone of the sea in the nature reserve it is possible to find various species of marine algae which are typical for this part of the Mediterranean. Furthermore some species of red algae are found in this area, which only exist in this part of France. In addition to the algae, the marine environment shows a rich range of different littoral and sublittoral specimens due to clear water. Furthermore, it is important to allude that the Girolata beach is not a high energy beach like the Fango Delta.

Materials and Methods

Collecting was done with casher nets, by hand collecting and sieves. Moreover, open water observations were made. For further information about Girolata, species of the sandy beach and the ecological environment were observed and sampled. The specimens were determined by taxonomic literatures, for example Riedl. All living creatures, except the spotted weaver, were released after determination.

Results and Discussion

The total number of species found in Girolata was 34. Most of them were fish (20). Furthermore, five shells, three snails, two sea urchins, two insects, a cephalopod (Figure 4) and a maxillopod were found. The following table shows all specimens discovered by the students. Notably, this list contains only a fraction of specimens that can be found here.

Figure 3: Dasyatis pastinaca (Linnaeus, 1758)

Figure 4: Pinna nobilis (Linnaeus, 1758)

Figure 5: Octopus vulgaris (Lamarck, 1798)

Table 1: All species found in Girolata

Class	Family	Genus	Species	Synonym
Maxillopoda	Balanidae	-	-	Seepocken
Insecta	Cerambycidae	-	-	Bockkäfer
Insecta	Nymphalidae	*Caraxes*	*Caraxes jasius (*Linnaeus, 1766*)*	Erdbeerbaumfalter, Foxy Emperor
Bivalvia	Arcidae	*Barbatia*	*Barbatia barbata* (Linnaeus, 1758)	bärtige Archenmuschel
Bivalvia	Pinnidae	*Pinna*	*Pinna nobilis (*Linnaeus, 1758)	Steckmuschel, noble pen shell
Bivalvia	Chamidae	*Pseudochama*	*Pseudochama sp.*	
Bivalvia	Lucinidae	*Ctena*	*Ctena sp.*	
Bivalvia	Ostreidae	*Ostrea*	*Ostrea edulis* (Linnaeus, 1758)	Europäische Auster,
Gastropoda	Thaididae	*Thais*	*Thais haemastoma* (Linnaeus,1758)	Purpurschnecke, (*Stramonita haemastoma*)
Gastropoda	Patellidae	*Patella*	*Patella sp.*	
Gastropoda	Trochidae	*Monodonta*	*Monodonta turbinata* (Born, 1780)	Turbanschnecke (*Osilinus turbinatus*)
Cephalopoda	Octopodidae	*Octopus*	*Octopus vulgaris* (Lamarck, 1798)	common octopus
Echinoidea		*Paracentrotus*	*Paracentrotus lividus*(Lamarck, 1816)	Steinseeigel
Echinoidea	Arbaciidae	*Arbacia*	*Arbacia lixula*(Linnaeus, 1758)	Black Sea Urchin
Chondrichthyes	Dasyatidae	*Dasyatis*	*Dasyatis pastinaca* (Linnaeus, 1758)	gewöhnlicher Stechrochen, common stingray
Actinopterygii	Carangidae	*Seriola*	*Seriola sp.*	Amberjack
Actinopterygii	Carangidae	*Trachurus*	*Trachurus mediterraneus* (Linnaeus, 1758)	Mittlemeer-Bastardmakrele
Actinopterygii	Centracanthidae	*Spicara*	*Spicara smaris* (Linnaeus, 1758)	Picarel
Actinopterygii				Flatfish, Plattfisch; O:Pleuroneciformes
Actinopterygii	Sparidae	*Pagrus*	*Pagrus pagrus* (Linnaeus, 1758)	Common Seabream, Sackbrasse

Actinopterygii	Sparidae	*Lithognathus*	*Lithognathus mormyrus* (Linnaeus, 1758)	Marmor-Brasse, Striped Seabream
Actinopterygii	Sparidae	*Pagellus*	*Pagellus erythrinus* (Linnaeus, 1758)	Rotbrasse, common pandora
Actinopterygii	Sparidae	*Diplodus*	*Diplodus puntazzo* (Cetti, 1777)	Spitzbrasse, sharpsnout seabream
Actinopterygii	Sparidae	*Diplodus*	*Diplodus vulgaris* (Geoffroy Saint-Hilaire, 1817)	Zweibindenbrasse, common two-banded seabream
Actinopterygii	Sparidae	*Diplodus*	*Diplodus annularis* (Linnaeus, 1758)	Ringelbrasse
Actinopterygii	Sparidae	*Oblada*	*Oblada melanura* (Linnaeus, 1758)	Spiegelbrasse
Actinopterygii	Sparidae	*Sarpa*	*Sarpa salpa* (Linnaeus, 1758)	Goldstriemenbrasse, salema porgy
Actinopterygii	Labridae	*Coris*	*Coris julis* (Linnaeus, 1758)	Meerjunker, Mediterranean rainbow wrasse
Actinopterygii	Pomacentridae	*Chromis*	*Chromis chromis* (Linnaeus, 1758)	Mönchsfisch, Damselfish
Actinopterygii	Trachinidae	*Trachinus*	*Trachinus araneus* (Cuvier, 1829)	Spinnen-Petermännchen, spotted weever
Actinopterygii	Mullidae	*Mullus*	*Mullus surmuletus* (Linnaeus, 1758)	Streifenbarbe, striped red mullet
Actinopterygii	Serranidae	-	-	Grouper, Zackenbarsch (subfamily) Epinephelinae
Actinopterygii	Serranidae	*Serranus*	*Serranus scriba* (Linnaeus, 1758)	Schriftbarsch, painted comber
Actinopterygii	Atherinidae	*Atherina*	*Atherina presbyter* (Cuvier, 1829)	Ährenfisch,sand melt

As already mentioned, the list doesn't show the total species range of this habitat. Probably only the most common specimens living here were found during our short visit on the beach. Using improved sampling and observation methods, the number of different species can be increased. To carry out a comprehensive scientific analysis, a much longer period, maybe more than a month, would be needed. Additionally, abiotic parameters should be measured. Also the participants' prior knowledge had an influence on the results. This year was the first time that our course went on an

excursion to Girolata. In the past, students went to the Fango Delta and this is the reason why no comparison to previous courses has been made.

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Project Reports

Early Development of Sea Urchin Embryos

Werner Bader, Alexandra Grosbusch, Isabella Hilti,
Bianca Jansen, Marie Massmig, Verena Naschberger,
Iris Krainer, Jana Hobmayer, Bert Hobmayer

Sea urchin development on the example of *Sphaerechinus granularis*

Werner Bader

Introduction

Sea urchins or Echinoida belong to the Phylum of Echinodermata. In our marine excursion to Calvi, we work with three different species: *Arbacia lixula*, *Paracentrotus lividus* as well as *Sphaerechinus granularis* (Figure 1). As we have learned from the basic zoological course, there do exist two different groups of sea urchins: the Regularia and the Irregularia. Regularia have a major axis from the mouth to the anus and they exhibit a pentameric body shape, whereas the Irregularia show a secondary bilateral body axis. We collected our experimental animals, which all belong to the Regularia, in the harbor basin of the Stareso research Station at Revellata/Calvi. Typical habitats of the sea urchins we found are rocks, concrete walls in the harbor basin as well as on the near Sea floor in a depth of one to five meters. In former excursions, a mass of datasets have been collected especially for the two Species *Arbacia lixula* and *Paracentrotus lividus*. Therefore we aimed at completing a dataset for the third important species, *Sphaerechinus granularis*.

Figure 1: The three species of sea urchins found in Stareso:

Arbacia lixula	*Paracentrotus lividus*	*Sphaerechinus granularis*
Foto: by Raphael Strohmaier	Foto: by Sabine Gufler	Foto: by Werner Bader

Systematics of these species:

Phylum	Class	Order	Family	Species
Echinodermata	Echinoidea	Arbacioida	Arbaciidae	***Arbacia lixula***
Echinodermata	Echinoidea	Camarodonta	Parachiniidae	***Paracentrotus lividus***
Echinodermata	Echinoidea	Camarodonta	Toxopneustidae	***Sphaerechinus granularis***

Materials and Methods

The specimens were directly collected from the near harbor basin of Stareso and put into a bucket, which was filled with fresh sea water. This bucket was then put to the laboratory on the first floor on Stareso Marine Station. Then, further steps to get the gametes had to be done (Figure 2). We used a 0,5 M KCl solution which was placed in a syringe. The sea urchins were positioned into a measuring cup, filled with fresh sea water. This arrangement was placed in a larger plastic container, which was located below. The KCl was injected in the soft tissue in the mouth region lateral to the "Lantern of Aristoteles". Thereby the spawning was induced. Female sea urchins can be recognized on their orange to red color of their eggs and male sea urchins can be recognized on their white up to beige color of their sperms. If it was a male sea urchin, it was dissected and the gonads were cut out and put in a test tube to cool them down in a fridge in order to have more sperm stored for further fertilization experiments.

Figure 2: Spawning of a sea urchin by KCl injection and dissection of a male to get gonads.

Results

After controlled fertilization, the development could be observed under the microscope using DIC optics. The various developmental stages were documented (Figure 3), and the timing of the progression of development until the pluteus larval stage could be analyzed quantitatively.

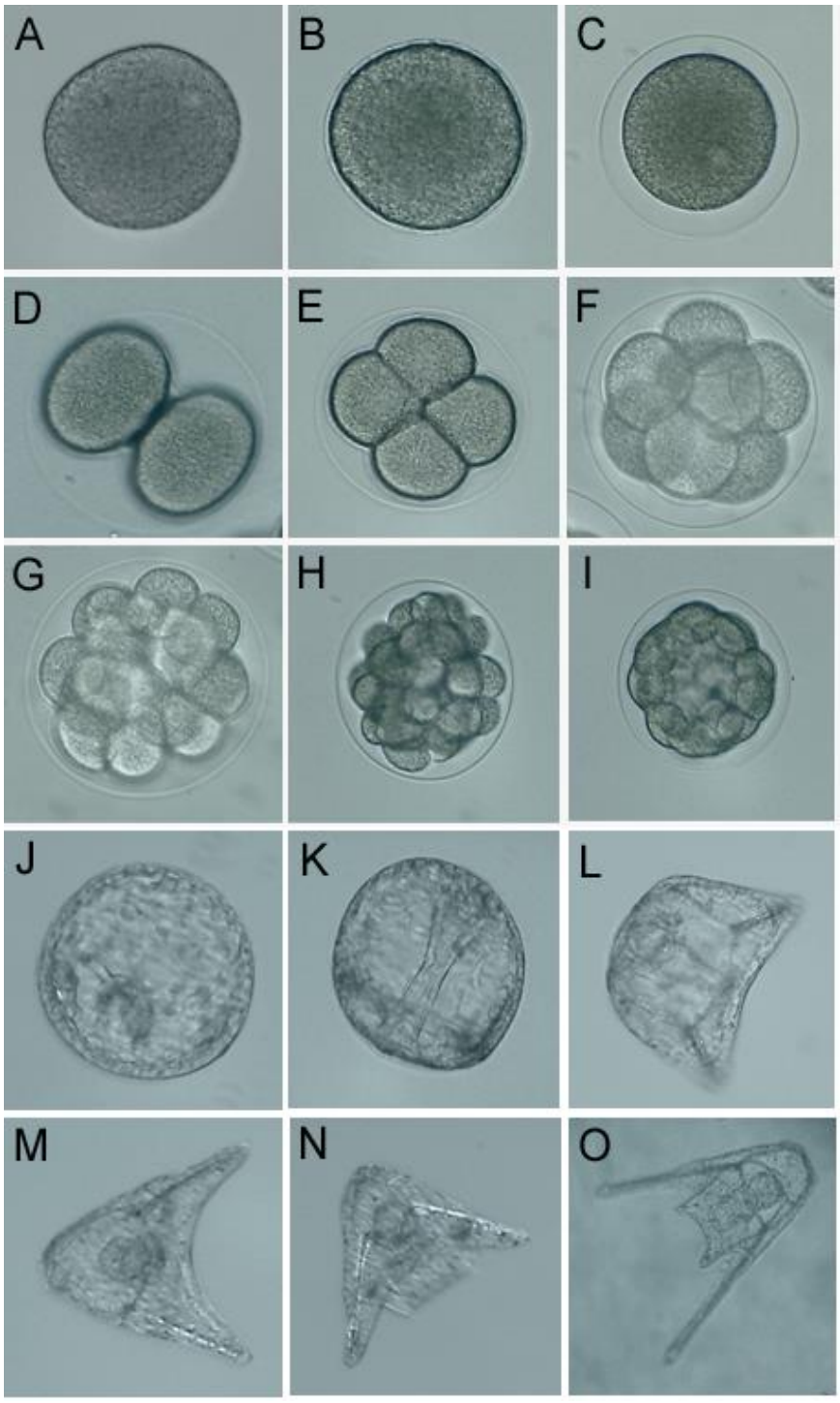

Figure 3: Developmental stages of **Sphaerechinus granularis*****. A. unfertilized egg, B. fertilized egg, C. fertilization membrane, D. 2-cell stage, E. 4-cell stage, F. 8-cell stage, G. 16-cell stage, H. 32-cell stage, I. blastula, J. early gastrula, K. gastrula, L. prism stage, M. late prism stage, N. early pluteus, O. pluteus.***

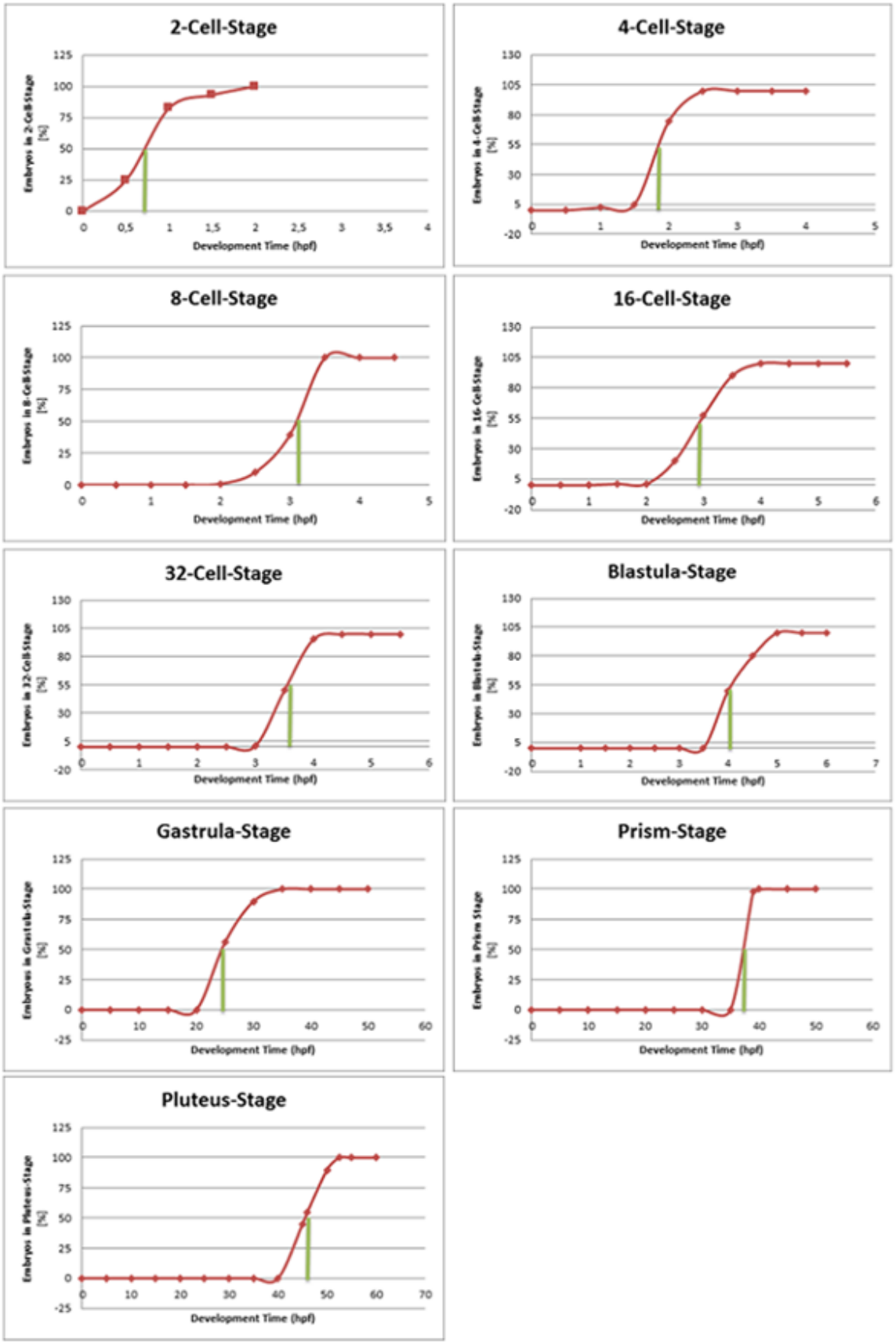

***Figure 4: Timing of developmental stages of* Sphaerechinus granularis**

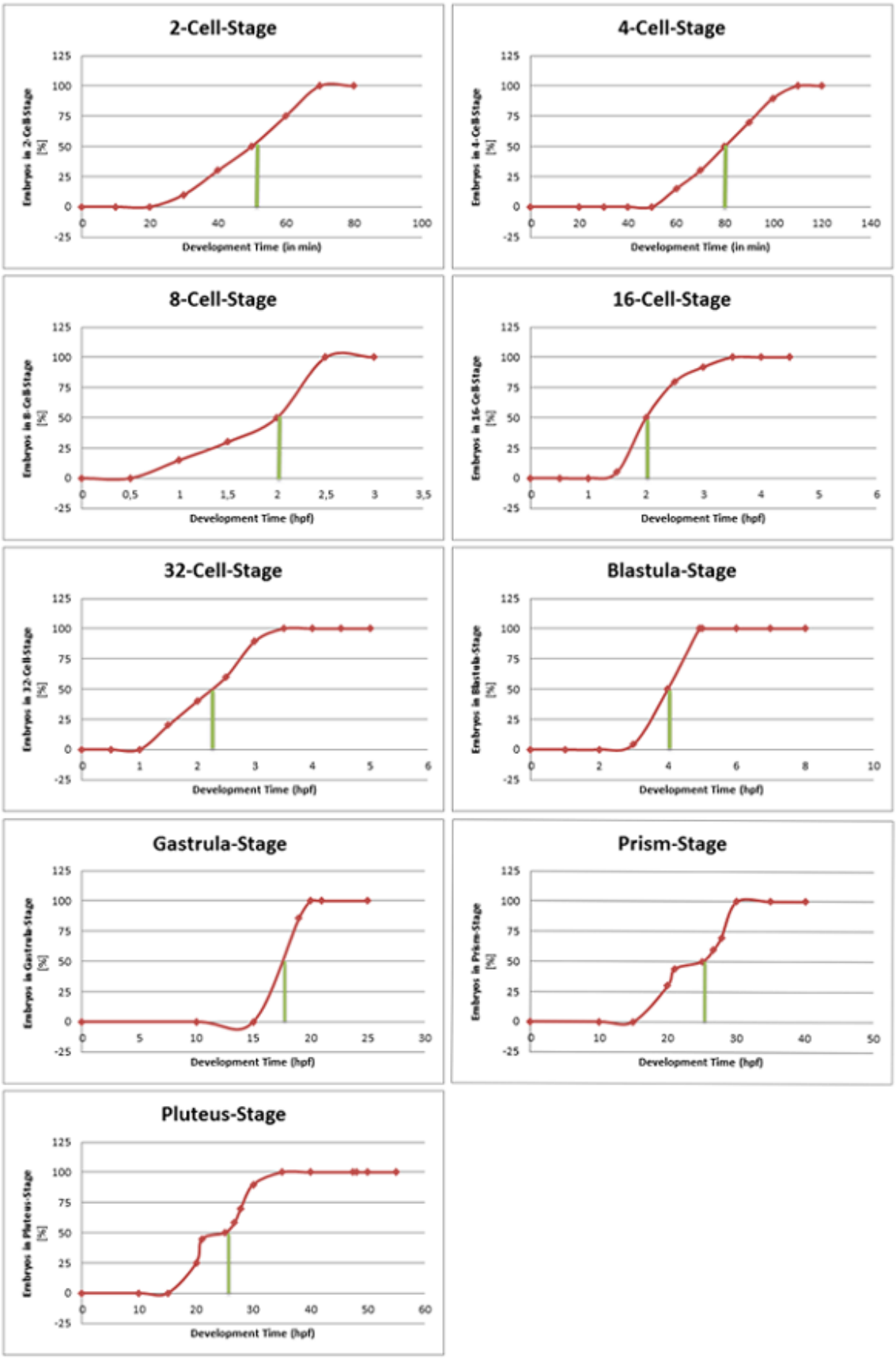

***Figure 5: Timing of developmental stages of* Arbacia lixula**

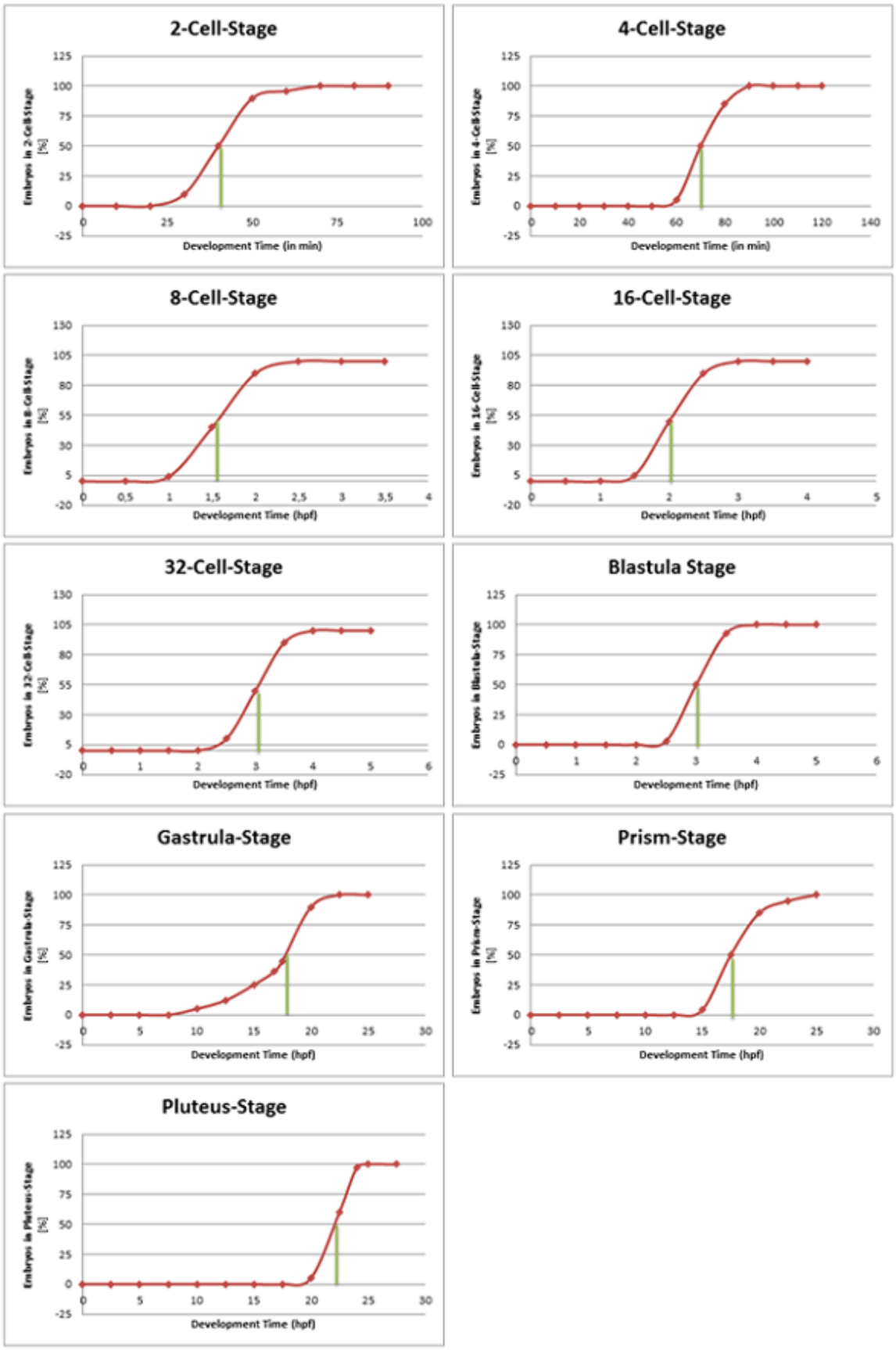

***Figure 6: Timing of developmental stages of* Paracentrotus lividus**

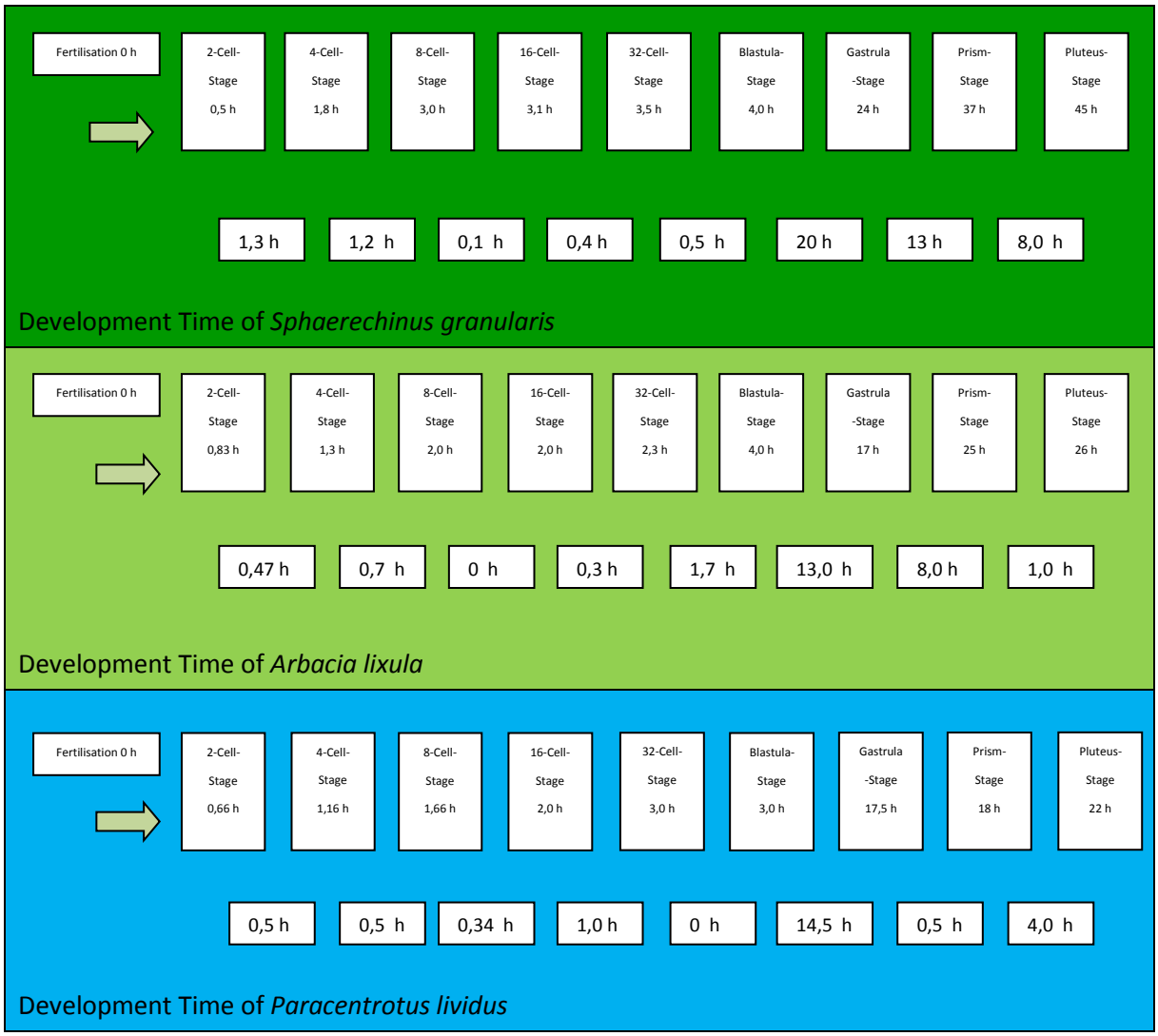

Figure 7: Comparative summary of the timing of development of the three species investigated.

Discussion

As we can recognize on the table above, the development time is significantly shorter in this year as compared with the data of the excursion to Calvi in 2012 and earlier. This is most likely due to the very good and very warm weather conditions up to 30° C all over the two weeks. They may be responsible for faster development and more rapidly occurring cleavages. Notably, also the fertilization rates were rather high during this course, even in the cross species fertilization experiments (see below).

Cross fertilization of sea urchins

Marie Massmig, Bianca Jansen

Introduction

Concerning the embryonic development of Bilateria, there still remain open questions. For instance the influence and origin of genetic control units in the early embryonic development. Sea urchins were chosen to be a model organism in the field of embryonic development for different reasons. First, they are easy to handle, their eggs are transparent and they have a fast embryonic development at least to the pluteus larvae. Furthermore, sea urchins, or echinoderms in general, are deuterostomes, so they are more closely related to vertebrates than other model organisms like *Drosophila* and *Caenorhabditis*. Therefore the scientific research can be transferred more simply (Wolpert, 2006).

Current studies are focused on maternal determinants, which polarize the egg cell before even a fertilization occurred. Maternal factors influence further development and fate of the fertilized egg. The hybridization of the three species, which can be found at the west coast of Corsica, *Arbacia lixula*, *Paracentrotus lividus* and *Sphaerechinus granularis* should give more information regarding fertilization and embryonic development. This study was focusing on fertilization rate, developmental variations and the phenotypic effects generated by the different genetic information of the egg (female; f) and the sperm (male; m).

Materials and Methods

Trypsin test

In nature the eggs are protected against a fertilization by foreign species and polyspermy with two blocking systems:

Fast blocking system: After the sperm is entering, the egg´s cell membrane depolarizes within 1 – 3 sec. and inhibits any other sperm to permeate. This blocking system lasts only a minute.

Slow blocking system: The second blocking system makes an impact one minute after the first sperm entered. The cortical granules located at the inner side of the cell membrane, start to fuse with membrane and vitelline envelope releasing several enzymes which results evolving a protective jelly layer, called *fertilization membrane* (Figure 1).

To avoid the slow blocking mechanism, there was a need to destroy the vitelline envelope (Figure 1) with trypsin, a proteolytic enzyme. To find the right concentration and time for the incubation in trypsin is important, because of its proteolytic impact on all structures of the egg and the

possibility of massive lowering the fertilizations rate. Thus the first part of the experiment has to be a series of tests with *Arabacia* and *Paracentrotus* eggs in different trypsin concentrations.

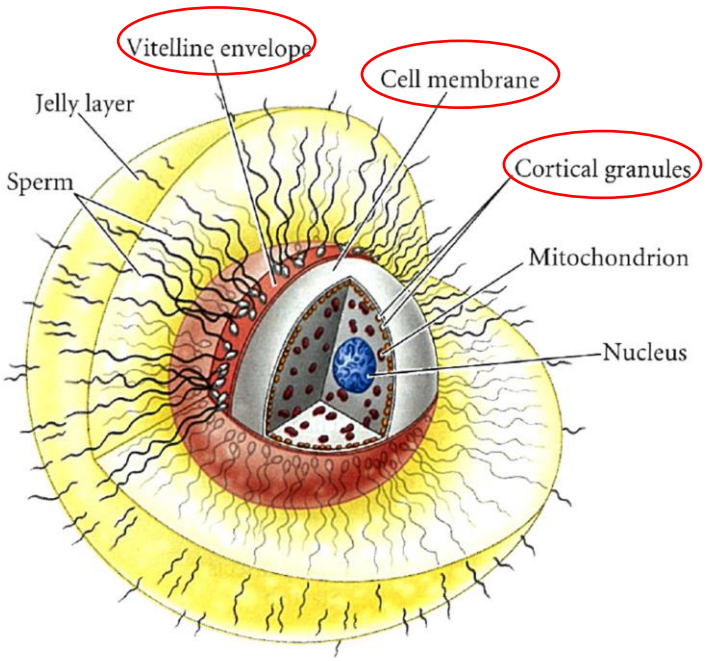

Figure 1: Structure of several layers surrounding a sea urchin egg with sperms penetrating it. (Modified from Gilbert, 2006)

One female and male each of *Arbacia* and *Paracentrotus* were forced to spawn by injecting 2 – 5ml of potassium chloride (KCl). The gonads of the males were removed and kept in the fridge to use them for several fertilizations. The eggs obtained were incubated for 15-30 minutes in a trypsin – seawater – solution (concentrations see Table 1). Seawater was filtered before using for all experiments to avoid foreign bodies.

Arbacia lixula	***Paracentrotus lividus***
Seawater (control)	Seawater (control)
0,4 g/l trypsin	2,0 g/l trypsin
0,8 g/l trypsin	4,0 g/l trypsin
1,2 g/l trypsin	
1,6 g/l trypsin	
2,0 g/l trypsin	

Table 1: Different concentrations of trypsin in filtered seawater for the trypsin test to find a good working concentration for hybridization of sea urchin.

After the incubation time, the eggs were washed three times in seawater and fertilized with the fitting sperm for each species. An hour later, the fertilized eggs were counted under the microscope and divided into fertilized/unfertilized and regular fertilized/irregular fertilized.

Hybridization

After determination of the appropriate incubation circumstances, fresh eggs of the three sea urchin species were treated with trypsin for 15 minutes in the following concentrations:

- *Arbacia*: 0,8 g/l and 1,2 g/l trypsin;
- *Paracentrotus* and *Sphaerechinus*: 1,5 g/l and 2,0 g/l trypsin

When incubation was finished, the eggs were washed in seawater three times and fertilized with foreign sperm:

- *Arbacia* (f) x *Paracentrotus* (m) -> **A(f)/P(m)**
- *Paracentrotus* (f) x *Arbacia* (m) -> **P(f)/A(m)**
- *Sphaerechinus* (f) x *Arbacia* (m) -> **S(f)/A(m)**
- *Sphaerechinus* (f) x *Arbacia* (m) -> **S(f)/P(m)**

One hour after fertilization (1hpf), the successfully fertilized eggs were counted under the microscope to detect the fertilization rate of hybrids. The gained hybrids were repeatedly moved to fresh seawater with the help of a microscope and pipette to remove dead and toxic embryos and to maintain a suitable surrounding. The developmental stages were documented with microscope and camera.

Results

Trypsin test with sperm of the same species

The test of a specific trypsin concentration was necessary, because the effectiveness of trypsin depends on the species and other influencing factors which remain to be discovered as for example the time of the year and the temperature. Because of a lag of time, the trypsin test was restricted to *Arbacia* and *Paracentrotus*. The trypsin tests with *Arbacia* showed a very high fertilization rate between 100% and 92 % up to a trypsin concentration of 1.2 g/l. In addition, the number of regular fertilized eggs was decreased to 35% by a trypsin concentration of 0.8 g/l, respectively to 4% with a trypsin concentration of 1.2 g/l (Figure 2a). The fertilization rate of the *Paracentrotus* eggs is decreasing constantly from 61 % in 2.0 g/l trypsin to 14% in 4.0 g/l trypsin. Whereas it has to be taken into account that in this experiments only two different concentrations where used. The percentage of regular

fertilized eggs is 3% in the treatment with 2.0 g/l trypsin and 100 % in 4.0 g/l trypsin, which can be explained by the, in general, very low fertilization rate in the second treatment (Figure 2b).

The tested trypsin treatments with an incubation time of 30 minutes have not been continued or taken into account for the further experiment, because the fertilization rate was too low.

Cross-species hybridization

Hybridization worked with all four species combinations in which different trypsin cconcentrations were used. The combination A(f)/P(m) reached the highest fertilization rate of 14 % in a trypsin concentration of 0.8 g/l, with 1.2 g/l of trypsin 9 % were fertilized and in the control 4 % were inseminated.

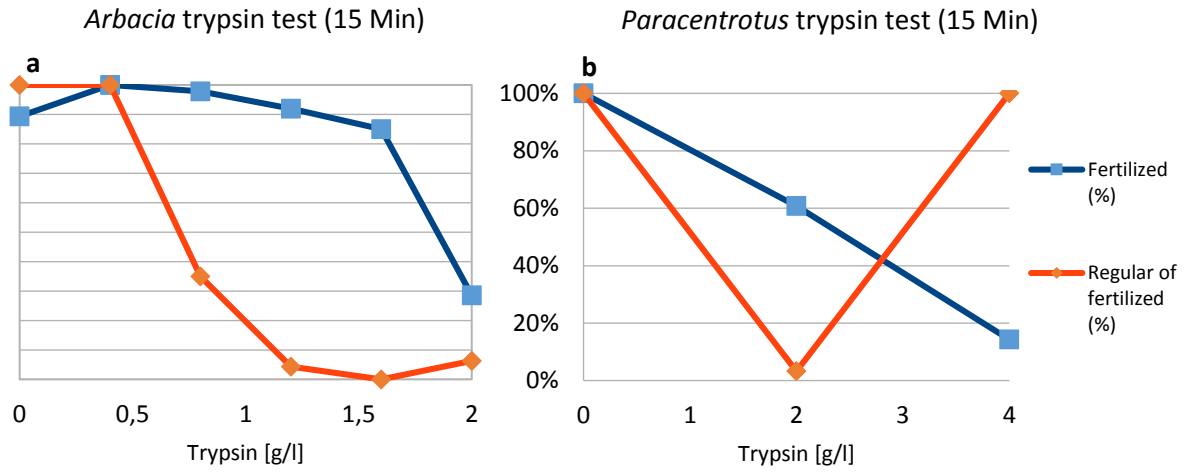

Figure 2: Trypsin test showing the percentage of fertilized and regular fertilized eggs depending on different trypsin concentrations with an incubation of 15 minutes. a) Arbacia fertilizations with a very low percentage of regular fertilized eggs and a high fertilization rate at a trypsin concentration between 0.8 and 1.2 g/l. b) Paracentrotus fertilizations with a very low percentage of regular fertilized eggs and a high fertilization rate at a trypsin concentration between 1.5 and 2.0 g/l.

Furthermore, the fertilization of *Paracentrotus* eggs with *Arbacia* sperm worked in 3% of the cases with a 1.5 g/l trypsin treatment and in 11 % of the cases with 2 g/l Trypsin. Additionally the combination S(f)/A(m) showed 14 % fertilized eggs with 1.5 g/l trypsin and a fertilization rate of 12 % in the treatment with 2.0 g/l trypsin. The last hybridization between *Sphaerechinus* eggs and *Paracentrotus* sperm reached the highest fertilization rate in the control (9 %) and 6 % with 2.0 g/l trypsin as well as 2 % with 1.5 g/l trypsin (Figure 3). In general it was noticeable that the eggs of *Paracentrotus* were more difficult to hybridize than the eggs of *Sphaerechinus* and in special more difficult than the eggs of *Arbacia*.

During the restricted time of the experiment, the highest stage of development differed between the species and treatments (Table 2). The highest developmental stage was reached with *Arbacia* eggs and *Paracentrotus* sperm which developed to a pluteus larvae. The combinations

including *Arbacia* sperm developed until the gastrula stage, whereas the combination of *Sphaerechinus* eggs with *Paracentrotus* sperm only reached blastula stage.

The hybrids are in general similar to a normal developed insemination. Additionally in the higher developed fertilizations as the pluteus larvae it can be seen that the outer shape of the larvae are more similar to the female species than to the male (Figure 4). Nevertheless during the experiments a lot of abnormal developments of the hybrids could be identified (Figure 5). It has to be assumed that these forms won´t be able to develop further.

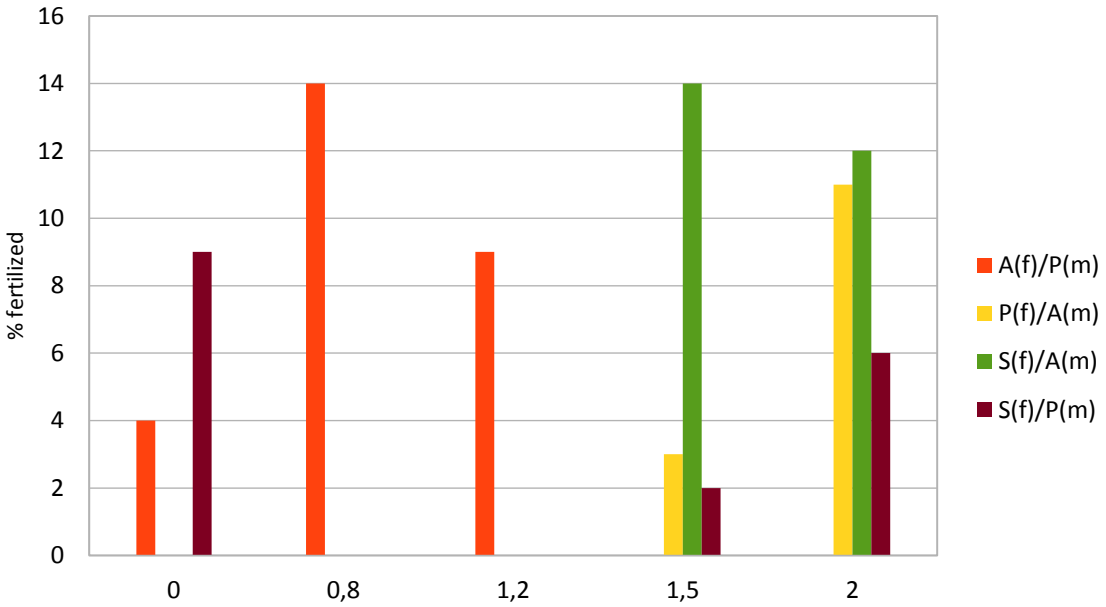

Figure 3: Percentage of successfully hybridized sea urchin eggs with various species combinations (1hpf).

Species	Trypsin [g/l]	Hpf	Developmental stage	Abundance
A(f)/P(m)	0	56	prism	common
	0,8		pluteus	rare
	1,2		pluteus	rare
P(f)/A(m)	0	42	unfertilized	common
	1,5		gastrula	rare
	2		gastrula	rare
S(f)/A(m)	0	68	gastrula	rare
	1,5		gastrula	rare
	2		gastrula	rare
S(f)/P(m)	0	68	blastula	rare
	1,5		blastula	rare
	2		blastula	rare

Table 2: Development stages of the different hybrids and their abundance at the latest hpf (hours past fertilization) observed.

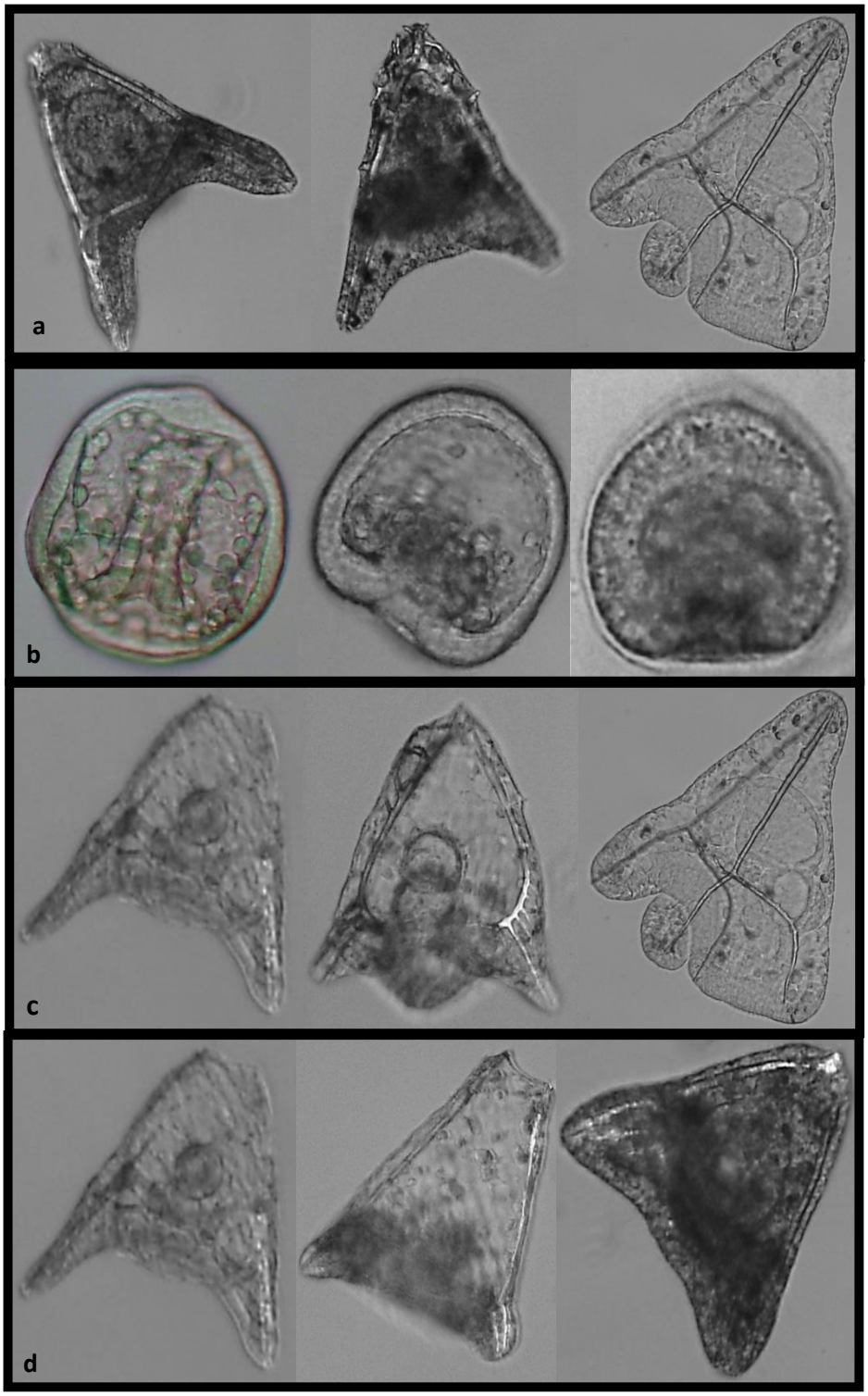

Figure 4: The latest observed developmental stages of the different hybrids in the middle and the corresponding female organisms at the left, respectively the male at the right. a) Pluteus larvae of Arbacia eggs and Paracentrotus sperm; 56 hpf; 1.2 g/l trypsin. b) Early gastrula of Paracentrotus eggs and Arbacia sperm; 42 hpf; 2.0 g/l trypsin. c) Pluteus larvae of Sphaerechinus eggs and Paracentrotus sperm; 68 hpf; 2.0 g/l trypsin. d) Pluteus larvae of Sphaerechinus eggs and Arbacia sperm; 68 hpf; 0.0 g/l trypsin (control).

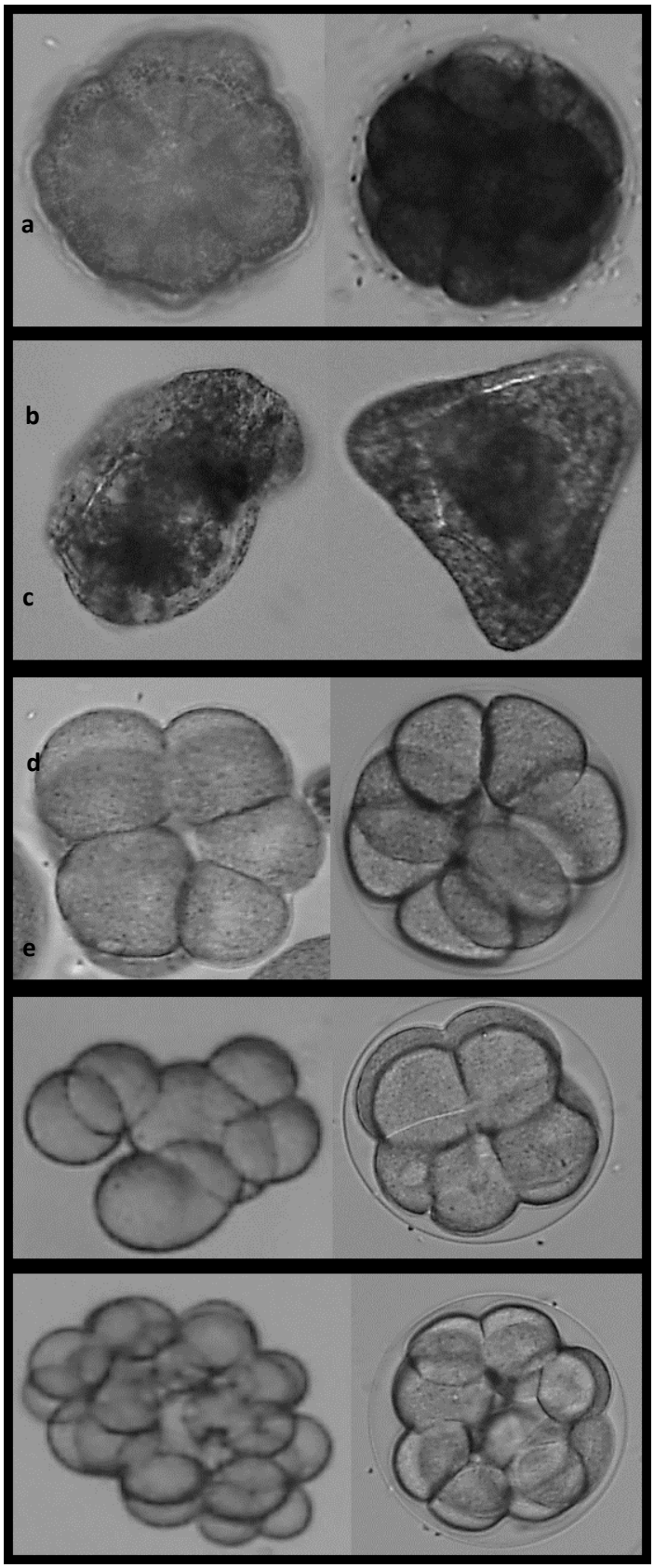

Figure 5: Different stages of irregular hybrids of sea urchin eggs. Left side deformed hybrids, right side normal fertilized egg of female species. Deformation occurs due to stabilization of the vitelline envelope which was digested by trypsin.
a) A(f)/P(m) 8-16 cell;
b) A(f)/P(m) prism;
c) P(f)/A(m) 8 cell;
d) P(f)/A(m) 16 – 32 cell;
e) P(f)/A(m) 8 cell

Discussion

First of all, it was remarkable that even without an incubation in trypsin the eggs were able to get fertilized by a different species (Figure 3). One possible explanation is the high abundance of sperm during the experiment, which was much higher than in nature. In contrast to the preliminary experiments in previous courses, all hybridizations have been possible this year. This could be influenced by the higher water temperature, the individual organisms and abiotic factors in the sampling area. In general the ability of hybridization could be supported by the narrow relationship and the high possibility of the different development stages to regenerate (Wolpert, 2006).

It was remarkable that the resulting hybrids were more similar to species which gave the eggs than to the male part of the hybridization. In addition to effects of the sex chromosome, the maternal determination can explain this observation. The sea urchin egg already has many determinants, which influence the development of the fertilized egg. For example the egg has an animal-vegetal asymmetry which predetermines the first two planes of cleavages (Wolpert, 2006). Furthermore, the maternal factors are responsible for the specification of micromeres (Wolpert, 2006).

To conclude, the experiment was successful and the fertilized egg even developed partly up to the pluteus larvae, the highest stage which can be reached in the laboratory until now. In future, the developmental timing should be observed so that a quantitative analysis is possible too. And if it is possible to cultivate sea urchins in a laboratory to the adult stage in the future, it has to be found out if hybrids are able to develop to an adult sea urchin.

Reaggregation experiments with sea urchin embryos

Verena Naschberger, Alexandra Grosbusch

Aim of this study

The aim of this project was to establish a protocol for re-aggregation of sea urchin embryos. Therefore, three different species were used, namely *Arbacia lixula*, *Paracentrotus lividus* and *Sphaerechinus granularis*. The main interest was to find out if the re-aggregated sea urchin embryos are able to rebuild the tissue structures and whether they can reconstruct the animal-vegetal axis.

Introduction

Echinoderms as model organisms

Sea urchins and sea stars belong to the phylum Echinodermata. Sea urchins are common models for developmental systems, because they are transparent and easy to handle. Furthermore, they are deuterostomes as the vertebrates, and thus are closely related. Sea urchins are very popular models for gastrulation and fertilisation studies (Wolpert et al., 2006).

Development of sea urchin embryos

After fertilisation, egg cleavages start, and the first two cleavages are along the animal-vegetal axis; thereby 2- (no illustration) and 4-cell stages are built. Further cleavages are equatorial and divide the embryo along the animal-vegetal axis. At the 16-cell stage, the vegetal pole consists of macromeres and four small micromeres. The 32-cell stage possesses four small and four large micromeres. At the 128-cell stage the blastula stage begins. Thereby, cells of the same size build a hollow sphere, which is surrounded by an epithelial sheet, which is composed of approximately 1000 ciliated cells. The ciliated blastula is rotating within the fertilisation membrane. At the gastrula stage, micromeres and macromeres located at the vegetal pole induce gastrulation (gut invagination). These micromeres are responsible for subsequent development of the larval skeleton and are the only cells whose fates are determined autonomously. Gut invagination then fuses with the mouth invagination (opposite site of embryo). The mouth finalizes the aboral-oral (A-O) axis in pluteus larvae (Wolpert et al., 2006; Gilbert, 2000).

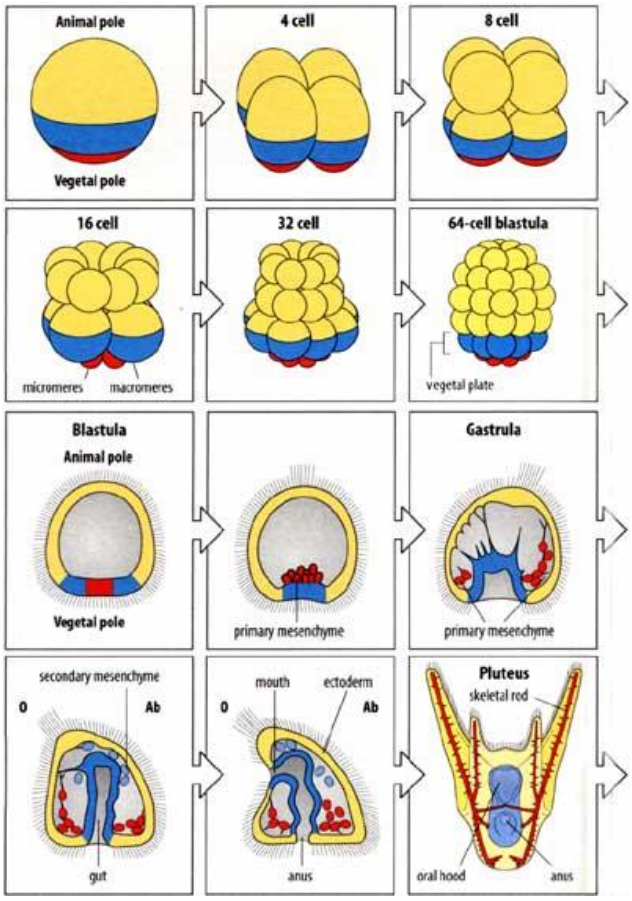

Figure 1: Embryonic development of the sea urchin from fertilised egg to the pluteus larvae.

Vegetal region and the organisation centre of sea urchin embryos

The major embryonic organisation centre possesses regulative capacities and is located at the vegetal pole. Moreover, this organisation centre is necessary for inducing complete body axis in sea urchin embryos. The micromeres of the vegetal pole induce macromeres to adopt endo-mesodermal fate. This phenomenon was confirmed by an experiment, in which micromeres implanted into the side of 32-cell stage induced a second gastrulation centre (Wolpert et al., 2006).
In Figure 2, the establishment of the organisation centre at the vegetal pole is summarized. The animal-vegetal axis is already established in the fertilised egg due to the localisation of maternal factors in the egg. As a result of wnt-pathway activation, β-catenin is accumulated in the nuclei of early blastomeres and acts with transcription factors which activate zygotic gene expression. Furthermore, β-catenin accumulation is very high in micromere nuclei and nearly absent in the animal pole, thus β-catenin accumulation is responsible for the establishment of the organisation centre at the vegetal pole. Moreover, micromeres induce the endo-mesoderm (Wolpert et al., 2006).

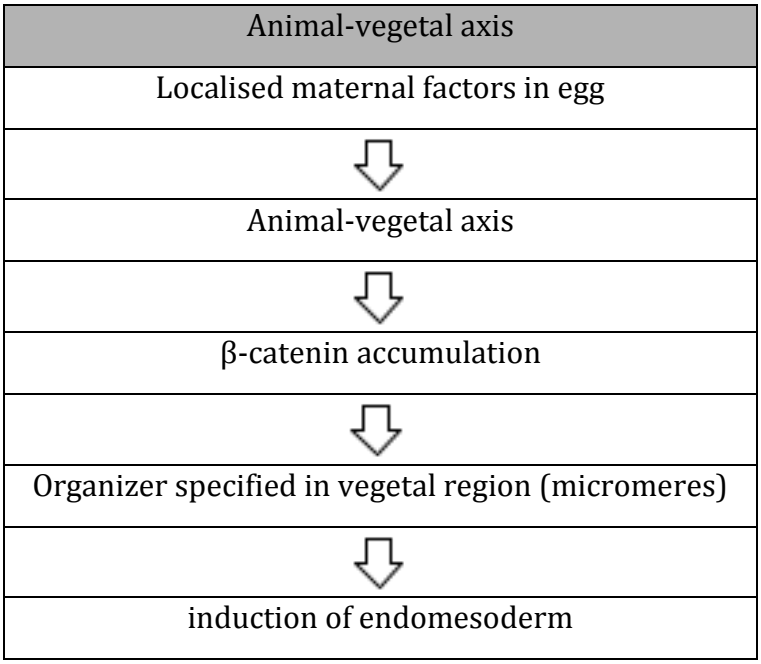

Figure 2: Establishment of the major organisation centre at the vegetal pole [modified according to Wolpert et al. (2006)].

Material and Methods

Experiments were conducted according to "Experimentelle Embryologie von marinen Evertebraten" (Banyuls, 2001), however with some modifications. We conducted four independent experiments with three different sea urchin species, *Arbacia lixula*, *Paracentrotus lividus* and *Sphaerechinus granularis*. In addition, different embryonic stages, such as 16-32 cell stage and blastula stage were used. In Table 3 the different experiments are illustrated. The first experiment was a pilot experiment to test the two different dissociation solutions (HEM and CF) and also the differences between the use of syringe and dissociation pipette.

Dissociation solutions: See Table 1 and Table2.

Dissociation

After fertilisation in petri dishes, the appropriate embryos, in this case free swimming blastulae, were incubated in calcium-free media for approximately half an hour. Then, the blastula stages were transferred into a 15 mL falcon tube and re-suspended with the above mentioned calcium-free sea water (CF) and hyaline extraction media (HEM). These calcium-free solutions are necessary for disrupting cell-cell contacts, the so called adherens junction. After incubation, the blastula stages were dissociated, due to mechanic shear of a syringe or dissociation pipette. For both techniques the cells were pipetted up and down five to ten times. The last step of dissociation was to check via microscope if single cells were available.

Table 1: Components of calcium-free sea water (CF).

calcium-free sea water	1000 mL
NaCl	26.5 g
KCl	0.7 g
$MgSO_4 7H_2O$	11.9 g
$NaHCO_3$	0.5 g
pH 8.2	
salinity 34-36 ppt	

Table 2: Components of hyaline extraction media (HEM).

hyaline extraction media	1000 mL
NaCl	17.5 g
KCl	0.75 g
$MgSO_4 7H_2O$	2.5 g
Glycine	22.5 g
TRIS	1.21g
EGTA	0.76g
pH 8.2	
salinity 34-36 ppt	

Centrifugation

After the dissociation step, single cells were centrifuged by hand centrifugation. Centrifugation time was as long as a pellet was built, this took approximately three minutes.

Reaggregation

The supernatant was removed and the pellet was layered with normal sea water to rebuild the cell-cell contact, due to the calcium in normal sea water. Therefore, the pellet was incubated for approximately half an hour. Afterwards the re-aggregated cell clump was cut into small pieces/aggregates.

Observation

The aggregates were observed at different times under the microscope and the medium was changed as often as possible. From the pilot experiment we concluded that HEM was more appropriate than CF. Moreover, the dissociation was better by syringe than by dissociation pipette, see figure 3. Therefore, we used HEM and syringe for further experiments. In experiment 2, the 16-32 cell stages were additionally incubated in 0.08 g and 0.12 g Trypsin for 15 minutes to digest the fertilisation membrane. Otherwise, the dissociation would not work because of the fertilisation membrane. After incubation the cells were washed three times with normal sea water to remove the trypsin traces. Further steps were conducted according to the pilot experiment.

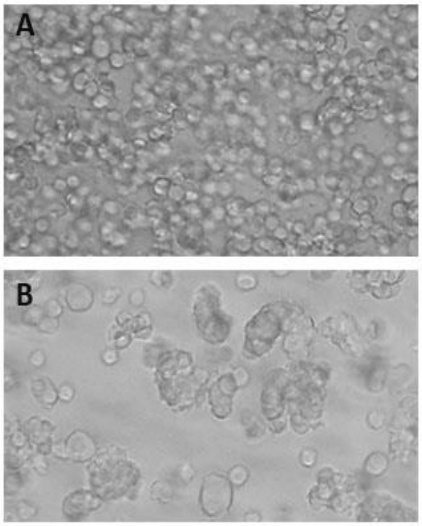

Figure 3: Comparison of syringe and dissociation pipette. In A the dissociation with syringe and in B dissociation within dissociation pipette in HEM are illustrated.

Results

The short overview in Table 3 summarizes the species of embryos, which were dissociated and re-aggregated in each experiment. Furthermore it indicates the stage of development at the time of dissociation and the resulting aggregate stages.

Table 3: Overwiew of all the experiments, summarizing species, dissociation stage and results.

	Species	***Developmental stages***	***Results***
Pilot Experiment	*Paracentrotus lividus*	Blastula stage	not viable
Experiment 2	*Arbacia lixula*	16-32 cell stage	only some pluteus stages observed
Experiment 3	*Paracentrotus lividus*	Blastula stage	a lot of pluteus stages observed
Experiment 4	*Sphaerechinus granularis*	Gastrula stage	not viable

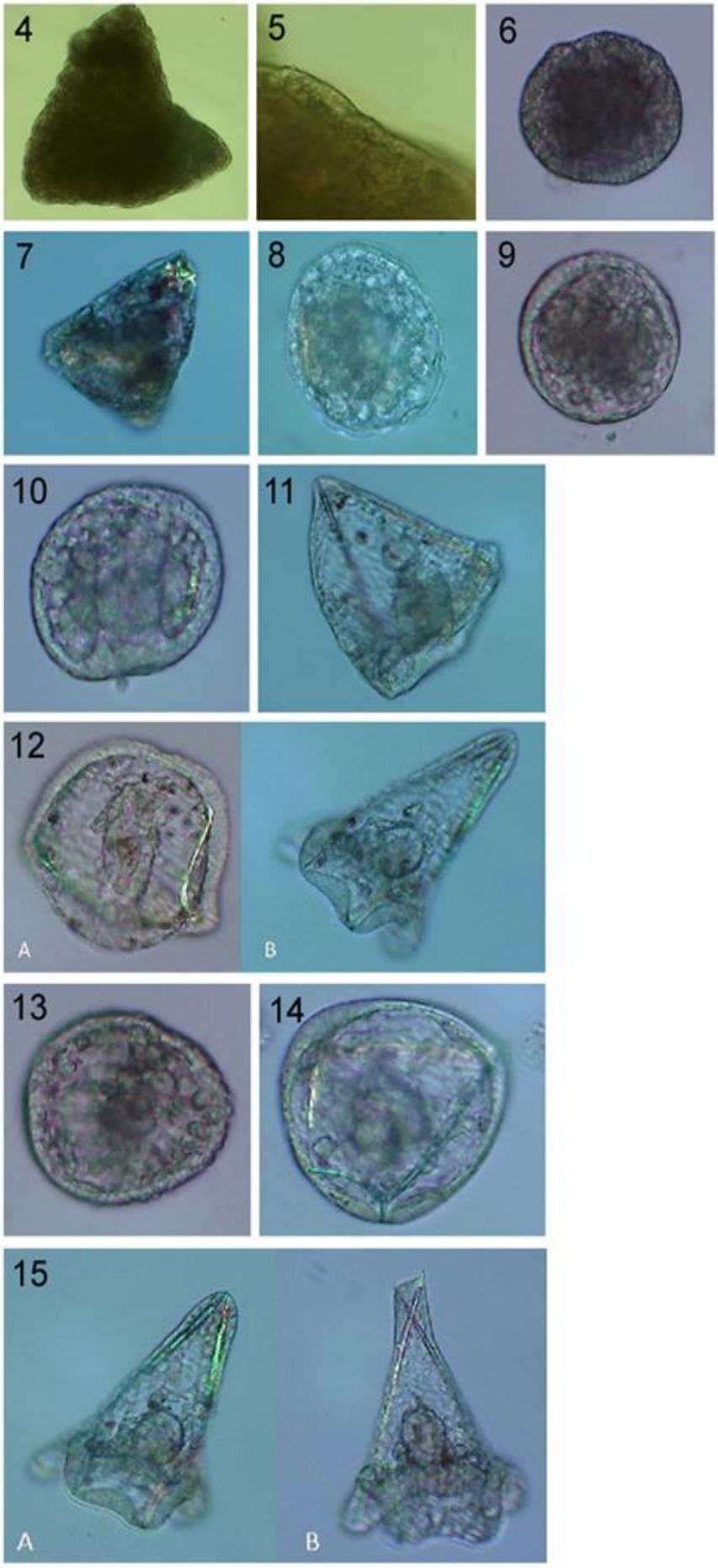
4
5
6
7
8
9
10
11
12
A
B
13
14
15
A
B

Figure 4: Reaggregated cells of Paracentrotus lividus 11,5h after reaggregation.
Figure 5: Monociliated cells forming epithelium.
Figure 6: Blastula stage of Arbacia lixula 20h after reaggregation.
Figure 7: Prism stage of Arbacia lixula 44h afterreaggregation.
Figure 8: Irregular gastrulation stage of Arbacia lixula 44h after reaggregation.
Figure 9: Blastula stage of Paracenrotus lividus 15h after reaggregation.
Figure 10: Late gastrula stage of Paracenrotus lividus 23h after reaggregation.
Figure 11: Prism stage of Paracenrotus lividus 39h after reaggregation.
Figure 12: Irregular pluteus stage of Paracenrotus lividus 39h after reaggregation. A: basal view, B: lateral view.
Figure 13: Irregular gastrula stage of Paracenrotus lividus 15h after reaggregation.
Figure 14: Irregular prism stage of Paracenrotus lividus 23h after reaggregation.
Figure 15: Comparison between a pluteus stage of Paracentrotus lividus 39h after reaggregation (A) and a pluteus stage of Paracenrotus lividus after 33h of normal development (B).

Pilot Experiment

As already said, the pilot experiment was done with larvae from *Paracentrotus lividus* which were dissociated at the stage of blastula. The aggregates of the dissociated cells were able to build a mono-ciliated epithelium, which is visible in Figure 5. So part of the cells in the aggregates were able to rearrange themselves, but because of the big size of the aggregates, a lot of the epithelial cells couldn't reach the surface and died. Thus the aggregates weren't viable and began to fell apart after one day. To increase the chance of survival of the aggregates in the further experiments the aggregates were cut in smaller pieces so that the size of the aggregates were similar to those of regular larvae.

***Experiment 2: 16-32-cell stage of* Arbacia lixula**

The next experiment was the only one done with embryos of *Arbacia lixula* and the only one dissociated and re-aggregated at the developmental stage of 16-32 cells. Unfortunately, only a few aggregates managed to regenerate. But among these aggregates some were able to develop into a regular prism stage. Figure 7 shows a regular prism stage, a triangular larva with spicules inside which built the larval skeleton. Even irregular stages were observed like in Figure 8, which shows an irregular gastrula stage. This larva shows an irregular development because mesoderm cells and spicula were present at the same time of development.

***Experiment 3: Blastulae of* Paracentrotus lividus**

Another experiment with embryos of *Paracentrotus lividus,* which were dissociated at the stage of blastula, was done. As can be seen in Figure 9, the aggregates were able to regenerate and build a regular blastula again. The greatest amount of regenerated blastula was observed during this assay. Furthermore, Figures 10 and 11 show a perfect gastrula stage with well-formed invagination

as well as a prism stage with spicules inside. Finally, rather regular pluteus larvae developed as shown in Figure 12.

***Experiment 4: Late blastulae of* Sphaerechinus granularis**

There aren't pictures of the aggregates of *Sphaerechinus granularis*, because all of them died after the first hours of re-aggregation. Obviously they weren't able to rearrange their cells so they died and the aggregates fell apart.

Discussion

After realisation of our experiments, we conclude that small aggregates tend to develop in a more normal way than larger aggregates. Gary Freeman (1988) observed the same during his experiments with echinoid embryos. So it is necessary, during the creation of aggregates, that their size is embryo-like. Only if the size is appropriate, the aggregates will be able to regenerate fully and develop into pluteus larvae.

Once the aggregates were cut into pieces of an appropriate size, a quick regeneration and reorientation of the cells can be observed. More precisely, after a few hours the outer layer of the aggregates is built of ciliated cells, leading to the conclusion that the cells were able to reorient after being mixed up. These aggregates were able to grow into later development stages, which indicates that the aggregates were able to reconstruct the animal-vegetal axis.

Moreover, there were some irregular stages like the irregular gastrula in Figure 13 or the irregular prism in Figure 14. The gastrula stage in Figure 13 shows mesoderm cells spread all over the larva. This could have been induced by micromeres, which were distributed all over the larva during the re-aggregation. If an excessive number of micromeres is spread all over the aggregate, they aren't able to rebuild the animal-vegetal axis. In regular gastrulae, the mesoderm cells were all on the same spot near the vegetal pole. From this spot the invagination is induced. At prism stage, the larval skeleton begins to develop, the first prismatic spicules appear. This can be seen in Figure 14, but the shape of the larva resembles more a gastrula than the characteristic triangular shape of a prism stage. In Figure 15, a pluteus stage of *Paracentrotus lividus* developed from our aggregates (A) and a normal developed pluteus stage of *Paracentrotus lividus* were compared. After considering the images, it can be concluded that they look more or less the same. According to our study, it could be said that re-aggregated larvae need more time to develop than normal larvae. Time of development between the stage of blastula and pluteus of *Paracentrotus lividus* takes in general 22 to 23h. Our aggregates needed 23h after re-aggregation to reach the stage of gastrula. But no meaningful conclusion could be drawn, because the time the aggregates needed to rebuild a blastula stage has not been recorded. In comparison, Nelson and McClay (1988) described their aggregates as smooth and ciliated after 6h of re-aggregation.

According to the study of Gary Freeman (1988), the presence of embryos, which were incompletely dissociated so that there were groups of un-dissociated cells mixed with single cells increases the percentage of aggregates that are able to develop into pluteus stage. Even if we

checked out whether the cells were completely dissociated, the possibility exists that incompletely dissociated cell groups were present. So it could be possible that in experiment 3 pieces of cell groups were present in our aggregates, which would explain why this experiment gave better results.

Otherwise the good results could also be related to the species and the development stage which were chosen for dissociation. Corresponding to our results *Paracentrotus lividus* is more suitable than *Arbacia lixula* or *Sphaerechinus granularis.* Furthermore embryos in the blastula stage are more able to regenerate than 16-32 cell stage or gastrula stage. However our results aren't very significant. For all experiments we used HEM, but according to Banyuls (2001) the use of HEM or CF depends on the species, which were analysed. We conclude that more data would be necessary to draw a significant conclusion, which we were not able to obtain because of a lack of time.

Manipulation of the animal-vegetal axis

Isabella Hilti, Werner Bader

Aims of this study

- To over-activate β-catenin in early embryos to investigate whether β-catenin is really responsible for axis formation.
- To find out whether pharmacological inhibitors of β-catenin signaling have a significant influence on the manipulation of the vegetal-animal axis.

Introduction

Development of sea urchin embryos starts with the fertilization of the egg. The fertilized egg then divides by radial cleavage. The first three cell clusters in the early stages of the embryo development are symmetric. The fourth proliferation is asymmetrically and the early embryo creates at one side the vegetal pole. The vegetal pole is the organizing center of the embryo. After some cleavages the vegetal pole develops four macromeres, which include a heap of cytoplasm and four tiny micromeres. Further cleavages follow and the proliferation results in a hollow spherical blastulae. The next step in the sea urchin development is the gastrulation (10 hpf – hours past fertilization). Gastrulation in the embryo initiates with the invagination on the vegetal pole. From then on, the primary body axis gets visible for the first time. The invaginated vegetal pole stretches right across the blastocoel and produces mouth, gut and anus. Nevertheless, it is remarkable that the development of the body axis comes into the picture much earlier in the development. The fourth cell division restrains unequal and conduces to a vegetal pole with micromeres in which maternal determents get distributed into the daughter cells. The unequal dispersion of these determents is important to specify animal as well as vegetal cell fates. Maternal determents are significant for intracellular signaling pathways, e.g. the canonical Wnt signaling pathway. In this signaling pathway ß-catenin plays an important role as maternal determent. The transcription factor ß-catenin accumulates in the nuclei of the blastomeres to activate zygotic gene expressions. Furthermore, ß-catenin occurs mainly in the micromere nuclei and is almost absent in the animal region. The maternal Dishevelled protein activates the canonical Wnt signaling pathway and thus ß-catenin gets stabilized.

ß-catenin is necessary to determine the vegetal fates. Usually, the transcription factor in the micromeres activates invagination of the cell at the vegetal pole. However, if the embryos are treated with certain pharmacological substances, such as Alp (Alsterpaullone), the GSK-3 kinase gets blocked. Consequently, more ß-catenin accumulates in cells. That leads to a vegetalization of the embryo. This results in an altered phenotype and embryo development.

Based on this knowledge, we performed experiments with two pharmacological substances to find out, what substances influence the intracellular Wnt signaling pathway on certain developmental steps.

Materials and Methods

Specimens:

- *Arbacia lixula*
- *Paracentrotus lividus*

Alp and BIO:

Both, Alsterpaullone (Alp) and BIO block the activation of the GSK3ß kinase and the CDKs (cyclin dependent kinases) in the canonical Wnt signalling pathway. GSK3ß kinase is responsible for the intracellular proteolysis of the multifunctional protein ß-catenin. Alp is as a competitive ATP-Inhibitor. It occupies the ATP-juncture at the GSK3ß kinase and prevents the transfer of phosphate from ATP on ß-catenin.

To harvesting eggs and sperm from *Arbacia lixula* and *Paracentrotus lividus*, it was necessary to treat them with potassium chloride. Afterwards, the eggs got fertilized with the sperm suspension. The development of the fertilized eggs proceeded normally until a certain development stage in untreated seawater. When the development of the embryos reached the 8- cell and 16-cell stage, the embryos were put into small petri dishes with 10 ml seawater. Furthermore, the both substances with different concentrations were pipetted into the petri dishes and got incubated for six hours in dark surrounding, in this case in a box. First, we treated the embryos of *Arbacia lixula* and *Paracentrotus lividus*, with two different concentrations of Alp. Afterwards, another experiment was started with the three concentrations of BIO. Thereby, it is essential to pipet the same volume of the concentrations. In our case, we pipetted always 10 µl of each concentration with a 20 µl pipette.

Finally, we had five chemically treated petri dishes of *Arbacia lixula* and five dishes of *Paracentrotus lividus*. It was also important to treat the embryos with control substance DMSO-stock 100%.

Table 1: Concentrations of substances.

Substances	Alp	BIO
	1,0 µM	0,5 µM
	5,0 µM	1,0 µM
		5,0 µM

Washing the embryos after treatment was very important. Thereby, the embryos were put in new small petri dishes containing 10 ml untreated seawater. This process was done three times

consecutively. Henceforth, later developmental stages of the embryos could be observed and documented.

Following this experiments, we started a third experimental implementation with BIO and *Paracentrotus lividus.* We repeated the experiment because in the first experiment, we incubated pharmacologically treated embryos for six hours in the dark and mistakenly in the fridge as well. The incubation in a cool surrounding was awry so the development of the embryos was delayed in time and some embryos died.

Results

In the developmental stages blastula, gastrula, prism stage and pluteus larva (early and late), the impact of the treatment with the pharmacological substances was clearly evident. Axis manipulation was clearly evident. We focused on three main criteria to consider the effect of the pharmacological substances on the ordinary development of the embryos. The first variable was premature death of the embryo, the second was irregular early development and the third were specific effects on axis formation.

Experiment 1: *Arbacia lixula*

In the first experiment, embryos of *A. lixula* were analyzed for an effect of Alp and BIO.

Treatment with Alp:

Table 2: Premature death of embryo; treatment with Alp.

	< 20%	20% - 80%	> 80%
DMSO	-	-	-
Alp 1,0 µM	x	-	-
Alp 5,0 µM	-	x	-

According to table 2, it is evident that the concentration Alp 5,0 µM had a great effect on the premature death of embryos. The concentration 1,0 µM was also toxic for sea urchin embryos. Nevertheless, the morality rate was fewer than 20% (Fig. 1). All embryos treated with the control substance DMSO survived.

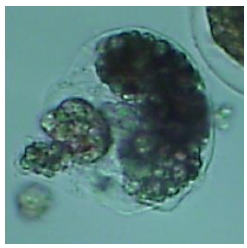

Figure 1: A. lixula, degenerated blastula, Alp 1,0 µM.

Furthermore, we examined whether Alp gives rise to irregular early development (2-cell stages to blastulae).

Table 3: Irregular early development; treatment with Alp.

	< 20%	20% - 80%	> 80%
DMSO	x	-	-
Alp 1,0 µM	x	-	-
Alp 5,0 µM	-	x	-

We ascertained that the best concentration for Alp was 5,0 µM for samples of *Arbacia lixula.* Moreover, a concentration of 1,0 µM caused only minor changes in the early development of embryos (Fig. 2). The control sample DMSO exhibited also few unexpected mutations. The possible reason why the control sample DMSO showed an irregularly early development was not clear. However, it might have resulted from the wrong incubation temperature, which was mentioned before.

Figure 2: A. lixula, irregular early development, Alp 1,0 µM.

Table 4: Specific effects on axis formation; treatment with Alp.

	Exogastrulae	Prism stage – animal flatted	Pluteus defect
DMSO	x	-	-
Alp 1,0 µM	-	-	-
Alp 5,0 µM	x	x	x

The effect of Alp 5,0 µM on axis formation is obvious in Table 4. The sample Alp 5,0 µM showed several modifications like exogastrulae (Fig. 3), animally flatted prism stages (Fig. 4) and pluteus defects. It was observed that also the DMSO sample presented exogastrulae but here we were no sure, whether they represented only degenerated blastulae. The sample Alp 1,0 µM showed no specific effects on the axis formation. In comparison with the control sample DMSO, the Alp-treated embryos showed a decelerated development and a slightly reduced animal pole.

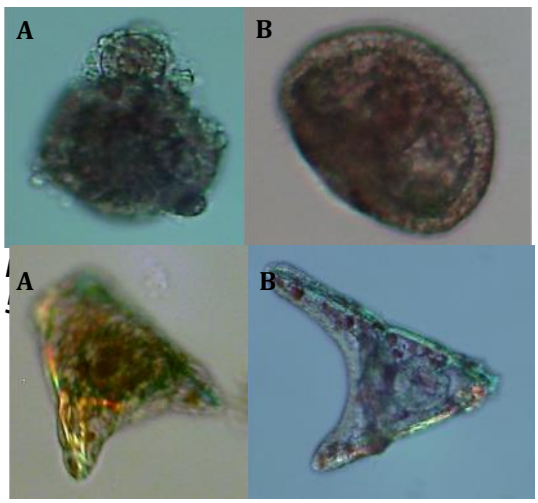

Figure 4: A. lixula: (A) Animally flatted prism stage , Alp 5,0 µM. (B) Wildtype prism stage.

Treatment with BIO:

	< 20%	20% - 80%	> 80%
DMSO	-	-	-
BIO 0,5 µM	x	-	-
BIO 1,0 µM	-	x	-
BIO 5,0 µM	-	-	x

Table 5: Premature death of embryos; treatment with BIO.

According to Table 5, BIO is more toxic than Alp on embryos of *A. lixula.* The lowest concentration with 0,5 µM already led to death. The higher the concentration of BIO, the higher was the percentage of the premature death of embryos. In the BIO 5,0 µM sample, most embryos died (Fig. 5).

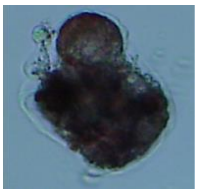

Figure 5: A. lixula, dead embryo, BIO 5,0 µM.

	< 20%	20% - 80%	> 80%
DMSO	-	-	-
BIO 0,5 µM	x	-	-
BIO 1,0 µM	x	-	-
BIO 5,0 µM	x	-	

Table 6: Irregular early development (2-cell stage to blastulae); treatment with BIO.

Due to the toxic effect of BIO and the high premature death of the embryos, minor irregular early development was expected. Only the embryos in the sample BIO 0,5 µM had the possibility to develop to a later stage.

Table 7: Specific effects on axis formation; treatment with BIO.

	Exogastrulae	Prism stage – animal flatted	Pluteus defect
DMSO	x	-	-
BIO 0,5 µM	x	-	-
BIO 1,0 µM	-	-	-
BIO 5,0 µM	-	-	-

As mentioned above, the observed exogastrulae in the DMSO sample could also be degenerated blastulae. Due to the high mortality rate in treatments with greater concentrations of BIO, only the low dosed sample (0,5 µM BIO) was able to generate exogastrulae (Fig. 6).

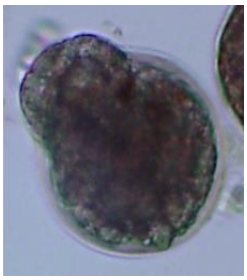

Figure 6: A. lixula, exogastrula, BIO 0,5 µM.

Experiment 2: *Paracentrotus lividus*

The second experiment displayed the effect of Alp and BIO on embryonic development of *Paracentrotus lividus.*

Treatment with Alp:

The treatment with the pharmacological substance Alp was conducted twice. Because of the wrong incubation implementation in the first stage, the experiment was repeated. Table 8 shows the toxic effect of Alp on embryonic development of *P. lividus.* Furthermore, this Table also indicates early irregular development.

Table 8: Premature death of embryos and early irregularly development (2-cell stage to blastulae); treatment with Alp.

	< 20%	20% - 80%	> 80%
DMSO	x	-	-
Alp 1,0 µM	x	-	-
Alp 5,0 µM	-	x	-

Most of the embryos of *P. lividus* survived the low dose treatment with Alp. Consequently, there were only few dead cells, before the embryos reached the blastulae stage. In the control sample, the possible reason for the early death was unknown. When the concentration was raised to 5,0 µM, the survival rate of the early embryos gradually declined to zero (Fig. 7).

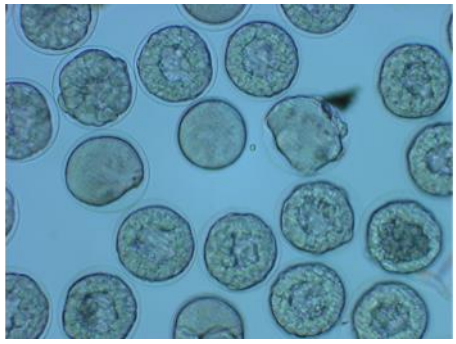

Figure 7: Paracentrotus lividus, premature block of development, Alp 5,0 µM.

In this case, it is possible to perceive that the low concentration caused a very small degree of irregular early embryonic stages. Increased concentration led to an unusual embryonic development and often resulted in premature death of the embryo.

The impact of Alp on axis formation:

Table 9: Specific effects on axis formation; treatment with Alp.

	Exogastrulae	Prism stage – animal flatted	Pluteus defect
DMSO	-	-	-
Alp 1,0 µM	-	-	-
Alp 5,0 µM	x	(x)	(x)

Exogastrulae were only obtained at a concentration of 5,0 µM Alp. Due to limited time, further observations could not be done. We assume that the embryos of *P. lividus* develop like the embryos of *A. lixula* and finally show animally flatted prism stages and faulty pluteus larvae.

Treatment with BIO:

To complete the experiments, *Paracentrotus lividus* was also treated with the pharmacological substance BIO.

	< 20%	20% - 80%	> 80%
DMSO	x	-	-
BIO 0,5 µM	x	-	-
BIO 1,0 µM	-	x	-
BIO 5,0 µM	-	-	x

Table 10: Premature death of embryos; treatment with BIO.

In these samples, the mortality rate increased with the concentration of BIO. Hence, BIO 5,0 µM was too high and led to complete mortality. A concentration of 1,0 µM was also high, but left some embryos alive. However, the living embryos were delayed in their embryonic development. Merely BIO 0,5 µM and DMSO allowed the advancement of the cells.

Table 11: Irregular early development (2-cell to blastulae); treatment with BIO.

	< 20%	20% - 80%	> 80%
DMSO	x	-	-
BIO 0,5 µM	x	-	-
BIO 1,0 µM	-	x	-
BIO 5,0 µM	-	-	x

Table 11 shows nearly the same result as Table 10. However, it had been observed that especially the sample BIO 1,0 µM led to irregular early development. Some blastulae with intact fertilization membranes and extended cells could be recognized (Fig. 8 and 9). Even though, most of the cells died in the sample BIO 1,0 µM and BIO 5,0 µM, not all cells died immediately after the treatment. Some cells survived but only developed until the 2-cell stage (Fig. 10).

Figure 8: P. lividus, blastula with fertilization membrane, BIO 1,0 µM.

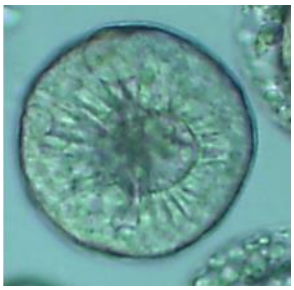

Figure 9: P. lividus, blastula with elongated cells, BIO 1,0 µM.

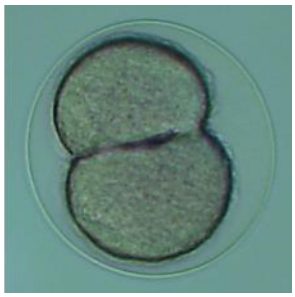

Figure 10: P. lividus, 2-cell stage, BIO 5,0 µM.

Table 12: Specific effect on axis formation; treatment with BIO.

	Exogastrulae	Prism stage – animal flatted	Pluteus defect
DMSO	-	-	-
BIO 0,5 µM	x	x	x
BIO 1,0 µM	-	-	-
BIO 5,0 µM	-	-	-

The concentration of BIO 0,5 µM induced a vegetalization during embryonic development, which led in an altered phenotype. In this case, the lowest concentration of BIO (0,5 µM) gave rise to exogastrulae, animally flatted prism stages and aberrant pluteus larvae (Figure 11 – 13).

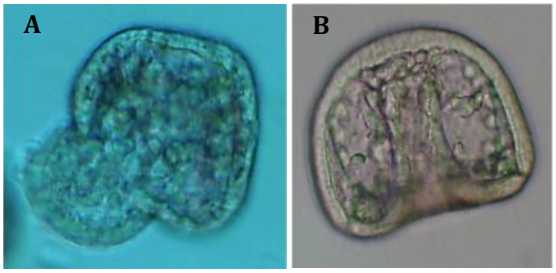

Figure 11: P. lividus: (A) Exogastrula, BIO 0,5 µM. (B) Wildtype gastrula.

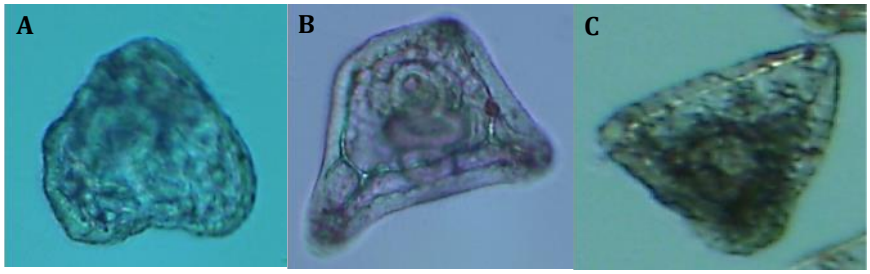

Figure 12: P. lividus: (A) Irregularly prism stage, BIO 0,5 µM. (B) Animalyl flatted prism stage, BIO 0,5 µM. (C) Wildtype prism stage.

Figure 13: P. lividus: (A) irregular early pluteus larvae, BIO 0,5 µM. (B) Animally flatted early pluteus , 0,5 µM. (C) Wildtype early pluteus larvae. (D) Wildtype late pluteus larvae.

Discussion

In our project week, we tested the pharmacological substance BIO, which is known to activate canonical Wnt signaling in deuterostomes by its inhibitory action on GSK3. We compared BIO with the known GSK3 inhibitor Alp. The project aimed to examine whether BIO and Alp have an impact on the canonical Wnt signaling pathway and on axis formation of sea urchin embryos. Unexpectedly, BIO reacts as a fairly strong toxic substance. A slight concentration of 0,5 µM suffices to manipulate the transcription factor β-catenin, which leads to an increased vegetal pole. Especially *Paracentrotus lividus* showed a strong vegetalization when treated with 0,5 µM BIO leading to an emergence of

exogastrulae, animally flatted prism stages and pluteus defects. By contrast, *Arabcia lixula* responded to BIO less strongly.

Alp-treated embryos showed the expected deceleration of development. In the case of Alp, embryos of *Arbacia lixula* increased their vegetal pole on cost of the animal pole more distinct than in *Paracentrotus lividus*. Thereby, β-catenin is stabilized in the vegetal area whereas in the animal area it gets phosphorylated by the GSK3β kinase. Alp-treated embryos presumably stabilized β-catenin in the vegetal pole and this led to an enlargement of the vegetal region and to the generation of exogastrulae. In the animal pole, β-catenin most likely got degraded. The best concentration to obtain such a manipulation of the animal-vegetal axis of sea urchin embryos is for samples of *A. lixula* 5,0 µM Alp. In contrast to the data of 2012, a concentration higher than Alp 1,0 µM, in our case Alp 5,0 µM, provides best results for axis formation. However, this high concentration also bring along a relatively high mortality rate (20 – 80%).

In comparison, the pharmacological substance BIO is an ideal alternative to Alp to manipulate the axis of sea urchin embryos, as it acted at much lower concentration. BIO is a superb inhibitor of the GSK3β kinase in the canonical Wnt signaling pathway. It is of great interest for both university and cancer research.

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Molluscs

Raphael Strohmaier, Nina Egger, Laura Von Raffay,
Clara Hechenberger, Michaela Hittorf, Stefanie Kuen,
Raimund Schnegg, Robert Gschwentner

Introduction

The term Mollusca (mollis = soft) was firstly described by Georges Cuvier in 1795 and the scientific study is called malacology. This specious taxon varies in body types and compose around 100 000 extant species, so they are, after the arthropods, the most diverse phylum. Concerning marine organisms, they are the largest phylum, comprising about 24 %. Most species live in marine habitats, especially in the benthic zone. Additionally they also live in freshwater and terrestrial habitats and they are highly diverse, concerning size, anatomical structure, behaviour and habitat. Cephalopoda (cuttlefish, octopuses and squid) are the most advanced species and some characteristics remind of vertebrates. The gastropods are the most numerous molluscs and account for approx. 80 % of the total.

Body plan

Even though there is a great range of anatomical diversity among molluscs, there are some characteristics that apply to all of them. Their body is bilaterally symmetrical, but some of them are also secondary asymmetric. It consists of the Cephalopodium (mouth, foot, sensory organs and nervous system) and the Visceropallium (inner organs). The body is structured in a threefold order: head, single muscular foot and visceral sac with stomach, heart, gonads, liver, kidney and gut form the torso. It has a single shell on the top which gets secreted by the glands of the Visceropallium. The mantle is lined with the epidermis. Its exact position varies between the groups. The whole soft body of the bivalves lies in an enlarged mantle cavity. The underside consists of a muscular foot, which has adapted to different purposes in different classes. It carries the statocysts, which can act as balance sensors.

Development

Sexual reproduction relies on external fertilization. They are producing eggs, which develop, by spiral cleavage, into trochophore larvae or more complex veliger larvae. Internal fertilization is also possible, but requires more complex reproductive systems. In contrast, the cephalopods differ in showing direct development, so the hatchling is a very small form of the adult.

Subclasses

Aculifera

The phylum of the molluscs consists of two subclasses: the Conchifera and the Aculifera. The characteristics of the Aculifera are the anterior-posterior axis and the shell of eight plates, denoted as the Polyplacophora, but the shell plates can also be absent as in the Aplacophora. The Aplacophora have no shell, but instead their cuticula has calcareous spiculae. The extant species of this group are called Solenogastres and Caudofoveata.

Conchifera

All higher molluscs are in the group of the Conchifera. Their habitats range from depths from the shore to up to 6 500 m depth. The highest diversity is given in the Gastropoda with 38 000 extant species. They are marine and fresh water living organisms, but as well terrestrial. Moreover they have a well-developed head and the Visceropallium shows a typical counter clockwise torsion of 180°. But sometimes they show a secondary re-torsion. Another group, the Scaphopoda, have a coned-shaped single shell, which is open at both ends. Normally they are buried in the sand and just by a small opening it is possible for them to let seawater flowing into the mantle cavity for oxygen supply. By a large radula, they are able to take up food, e.g. foraminifers, detritus and other sediment fauna. Cephalopoda are the most highly developed group of the molluscs and they just live in marine habitats, in the benthic as well as in the pelagic zone. Their most important feature is their highly developed nervous- and visual system. Bivalvia live in marine habitats, in brackish water and some of them also in freshwater. Their size ranges from 2 mm to 1 m and their maximum weight can be reached by *Tridacna gigas* with a weight of 400 kg. They can be found in shallow water as well as on deep sea floors. In comparison to other mollusc groups, they nearly lost all head structures, including the radula.
The weekly program

Sampling took place from September 7th to September 10th, 2012. Five habitats were chosen to find a range of species, which should be as varied as possible. On the first project day we sampled at the boulder field of the bay of Stareso. Then we collected some samples by pulling out of the Coralligène. Afterwards sampling was done at the bay of Revellata (Sandy Beach) and we collected at the Finger bay. On the last day before the presentation of our results, we focused on identification and archiving.

Materials and methods

For the collection of the molluscs several utensils were needed. Because different conditions reign in the diverse sampling sites, the corresponding molluscs have specific anatomic phenotypes. The aim of the course was to get as much information as possible about the composition of the mollusc fauna in this section of the Mediterranean Sea.

The six different sampling sites:

- **Sandy Beach** – 'Bay of Revellata'
- **Block field** – 'STARESO-bay'
- **Coralligéne**
- **Fingerbay**
- **Posidonia** – 'sea weed'
- **Girolata** – 'rocky beach'

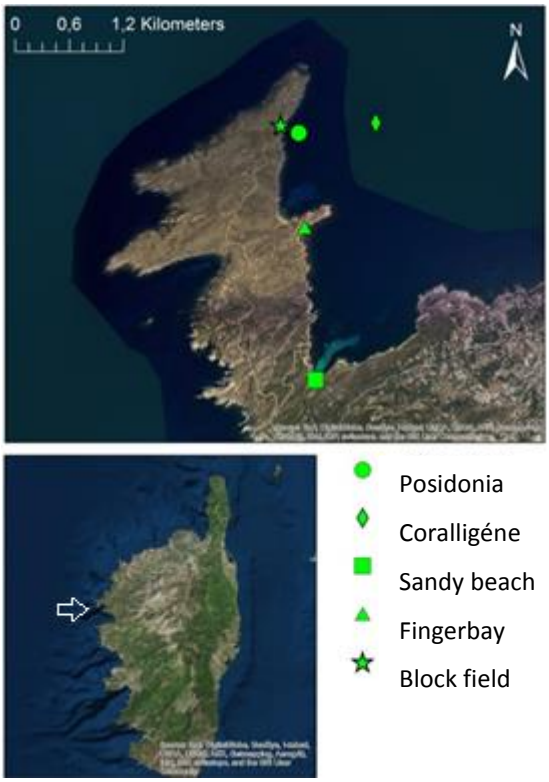


Figure 1: The six different sampling sites: Gulf of Revellata (upper legend); the Gulf of Girolata (bottom legend) shown by the white arrow.

Table 1: Overview of the materials used for each of the six different sampling areas.

Sandy beach	Boulder field	Coralligéne	Fingerbay	Posidonia	Girolata
zip-lock plastic bags	zip-lock plastic bags	boat	zip-lock plastic bags	zip-lock plastic bags	zip-lock plastic bags
sifters	microscope	dredge		bin bag	sifters
nets		big bucket		diver-knife	nets
		microscope		microscope	

Sandy Beach – 'Bay of Revellata'

Sandy habitats just as the Sandy Beach offer special habitats to molluscs. First to mention is the typical fine grained sand, which makes the inhabitants of this area different compared to those, which are adapted the environmental conditions of the other sample sites. The fine sand in combination to the continuous waves, cause a friction-effect. To resist this shear force, the molluscs need compact shells for protection. A marine slug for example wouldn't resist such conditions, because of its soft and therefor delicate skin.

For collection of the molluscs, which live in this sandy areas, sifters with a diameter of 20 cm, a depth of 6.5 cm, and a mesh size of 1 and 2 mm and also nets with a mesh size of 2 mm, were used. The other parts of the Sandy beach, which got examined, were the boulder fields on each side. Therefore nets and zip-lock plastic bags were used. An important fact, which should not be neglected is, that waves can transport nearly everything ashore. Of course molluscs are not t excluded here. So it cannot be claimed that all exemplars, which were found there, are actually inhabitants of this area. The caught molluscs were transferred into plastic pods filled with seawater and then immediately determined on site and written down.

Figure 2: Part of the Sandy beach with the boulder field on the left side.

Boulder field – 'STARESO-bay'

To the north side of the STARESO-bay, there is a pier to protect the STARESO-station against floods. To the south, the STARESO-bay is confined with cliffs. In this case, the rocks were the central sampling-areas. They got examined while snorkeling and picking up as many different species as possible. The found individuals got transferred into the zip-lock plastic bags and afterwards determined in the laboratory.

Figure 3: STARESO-bay sampling-site.

Coralligéne

Coralligène is also an important shelter for molluscs, because it offers heterogeneous structures and therefor constitutes a special kind of habitat for several marine species. The Coralligène samples were taken a few meters away from the STARESO bay. For this a motorboat was needed. For the sampling a dredge was led on the marine ground and dragged behind the boat, while it was slowly moving forward in about 50m depth. After about 10 minutes the full dredge got pulled up and the collected substance was put into a bottle of water. On the station the content of the bottle got sorted out and afterwards the molluscs were determined in the laboratory. *Caulerpa racemosa* (Fig.4) was not sent back to the ocean again, because it is an invasive species in the Mediterranean Sea.

Figure 4: Sorting out the sampled Coralligéne.

Figure 5: Parts of the Coralligéne incl. Caulerpa prolifera.

Fingerbay

The Fingerbay consists of sand, small stones and boulders and as mixture of different habitats, it provides a home for several different mollusc groups. The different individuals were picked up with zip-lock plastic bags and then immediately determined in the field.

Figure 6: Fingerbay in the South-East of the STARESO bay.

Posidonia – seagrass

The Posidonia sea grass grows in the middle area of the STARESO-bay. It's an important habitat for different animals to hide respectively to grow up – for fish and crustaceans as important as for molluscs. For the sampling, a part of the seaweeds got overlapped with a bin bag and then cut off with a diver-knife. In the bin bag the collected seaweed could be transferred to the laboratory to be examined. The other few sea weeds got convicted into the zip-lock plastic bags and also transferred to the laboratory. Because Posidonia weeds are getting rarer and are highly protected, the picking up of it had to be limited!

Figure 7: Bottom of STARESO-bay with Posidonia weed.

Girolata – 'rocky beach'

The Girolata bay is located at the north-west coast of Corsica. It is similar to the Fingerbay, consisting of sandy and stony areas, such as cliffs to the right and left side of the shore. For collecting the molluscs we used the zip-lock plastic bags and also nets, to then determine the exemplars instantly in the field.

Figure 8: Shore of Girolata - rocky beach.

Description of the sampling methods

For all sampling sites snorkeling with nets (Fig.11) and zip-locked plastic bags along the areas looking for molluscs was carried out except for the Coralligéne sampling. For this, as already mentioned, a boat was needed. The dredge, which scraped off the Coralligéne material (Fig.9) was slowly trailed behind the boat (Fig.10) for about 10 minutes to pick up enough material, which could be searched for molluscs.

Figure 9-11: Dredge, boat and nets for the sampling.

All molluscs, which were not identified in the field, were taken to the laboratory. Therefor they got transferred in zip-lock plastic bags or plastic pods, filled with seawater, if the individuals were still alive. In the laboratory they got sorted out in alive and not-alive individuals, respectively in gastropods and Bivalvia or other groups. Also to mention are the cephalopods, which were determined in the field and immediately got released back into the ocean afterwards. In the laboratory the alive as well as the non-alive individuals got photographed after determination. After this step, the animals, which were still alive, were put back into the sea again.

Results and Discussion

The Mediterranean Sea has an average depth of about 1 500m, which differentiates it from other seas. It`s mostly surrounded by land and the tides are not very strong. The dark blue colour is typical and it has a high biodiversity. More than 8500 macro-species are estimated and the number is increasing rapidly, especially because of the invasion through the Suez Canal. More than 500 alien species were detected until now. Particularly the molluscs are a very high abundant and divers group. They comprise up to 25% of the benthic macro-fauna, in number of species and in number of individuals. The course in 2010 collected and distinguished about 168 species and the last course in 2012 established 131 species. There are about 2300 different mollusc species occurring in the Mediterranean Sea (Coll et al., 2010), which means, that along the peninsula Revellata about 14% of all molluscs have been described within the courses.

Table 2: Collected species of the course in 2014 shown in an alphabetical order. Newly described species are marked (*).

class/family	**species**		**habitat**					
			Co	Bf	Fb	Po	Sa	Girolata
Bivalvia								
Anomiidae	*Anomia ephippium*	Linnaeus, 1758					S	
Arcidae	*Arca noae*	Linnaeus, 1758		S			S	
	Barbatia barbata	(Linnaeus, 1758)		S			S	?
Astartidae	*Astarte sulcata*	(da Costa,1778)	S					
Cardiidae	*Acanthocardia echinata*	(Linnaeus, 1758)	S					
	Acanthocardia spinosa	(Solander, 1786)	S					
	Cerastoderma glaucum	(Poiret, 1789)					S	
	Laevicardium crassum	(Gmelin, 1791)	S					
	Laevicardium oblongum (?)	(Gmelin, 1791)	S					
	Parvicardium exiguum	(Gmelin, 1791)					S	
	Parvicardium scabrum	(Philippi, 1844)	S, L					
	Parvicardium pinnulatum	(Conrad, 1831)	S				S	
	Plagiocardium papillosum	(Poli, 1795)					S	
Carditidae	*Cardita calyculata*	(Linnaeus, 1758)		S			S	
	Cardites antiquata	(Linnaeus, 1758)			S			
Chamidae	*Chama gryphoides*	Linnaeus, 1758		S			S	
	Pseudochama sp.	Odhner, 1917						?
	Pseudochama gryphina	(Lamarck, 1819)			S		S	
Donacidae	*Donax trunculus*	Linnaeus, 1758					S	
Glycymerididae	*Glycymeris glycymeris/insubrica*	da Costa, 1778	S					
Limidae	*Lima lima*	(Linnaeus, 1758)		L	?			
	Limaria inflata	Link, 1807					S	
Lucinidae	*Ctena decussata*	(O. G. Costa, 1829)					S	
	Ctena sp.	Mörch, 1861						?
	Loripes lucinalis	(Lamarck, 1818)					S	
Mactridae	*Mactra stultorum*	(Linnaeus, 1758)	S				S	
Mesodesmatidae	*Donacilla cornea*	(Poli, 1795)			L			
Mytilidae	*Modiolus barbatus*	(Linnaeus, 1758)			S			
	Musculus subpictus	(Cantraine, 1835)	S					
Noetiidae	*Striarca lactea*	(Linnaeus, 1758)					S	
Ostreidae	*Ostrea edulis*	Linnaeus, 1758		S,?				?

Pectinidae	*Chlamys flexuosa*	(Poli, 1795)	S					
	Chlamys hyalina	(Poli, 1795)	L					
	*Chlamys multistriata**	(Poli, 1795)	S					
	Chlamys varia	(Linnaeus, 1758)	S					
	*Hinnites distortus**	(da Costa,1778)	S					
	*Palliolum incomparabile**	(Risso, 1826)	S					
	Pecten jacobaeus	(Linnaeus, 1758)	S					
	Pseudamussium clavatum	(Poli, 1795)	S					
Pharidae	*Ensis ensis**	(Linnaeus, 1758)		?				
Pinnidae	*Pinna nobilis*	Linnaeus, 1758				L		?
Psammobiidae	*Gari depressa*	(Pennant,1777)	S					
Pteriidae	*Pinctada radiata*	(Leach, 1814)		S				
Tellinidae	*Tellina donacina*	Linnaeus, 1758	S				S	
Veneridae	*Dosinia exoleta*	(Linnaeus, 1758)					S	
	Gouldia minima	(Montagu, 1803)	S					
	Paphia aurea	(Gmelin, 1791)					S	
	*Timoclea ovata**	(Pennant,1777)	S					
	Venus verrucosa	Linnaeus, 1758			S			
Polyplacophora								
Acanthochitonidae	*Acanthochitona discrepans**	Brown, 1827		L				
Chitonidae	*Chiton olivaceus*	Spengler, 1797		L			L	
Lepidochitonidae	*Lepidochiton cancellatus**	Sowerby, 1840	L					
	Lepidopleurus cajetanus	Poli, 1791		L				
Cephalopoda								
Loliginidae	*Loligo vulgaris*			L				
Octopodidae	*Octopus vulgaris*	Lamarck, 1798		L				L
Sepiidae	*Sepia officinalis*	Linnaeus, 1758		L			L	
Gastropoda								
Aplysiidae	*Aplysia punctata**	(Cuvier, 1803)	?					
Atyidae	*Haminoea hydatis*	(Linnaeus, 1758)	S					
Buccinidae	*Buccinulum corneum*	(Linnaeus, 1758)		S				
	Cantharus dorbignyi	(Payraudeau, 1826)		S				
	*Chauvetia cf. brunnea**	(Donovan, 1804)	L					
Calliostomatidae	*Calliostoma granulatum*	(Born, 1778)					S	
	Calliostoma conulus	(Linnaeus, 1758)	L					
	Calliostoma zizyphinum	(Linnaeus, 1758)		S				
Calyptreidae	*Calyptraea chinensis*	(Linnaeus, 1758)	L					
	*Crepidula unguiformis**	Lamarck, 1822		S				
Capulidae	*Capulus ungaricus*	(Linnaeus, 1758)	S					
Cassidae	*Phalium undulatum*	(Gmelin, 1791)			S			
Cerithiidae	*Bittium latreillii*	(Payraudeau, 1826)					S	
	Bittium reticulatum	(da Costa, 1778)		?				
	Bittium scabrum	(Olivi, 1792)					S	
	Cerithium alucaster	(Brocchi, 1814)	L					
	Cerithium vulgatum	(Bruguière, 1792)	S	L		L	S	
	Cerithium rupestre (lividulum)*	Risso, 1826			L		S	
	Cerithium sp.	Bruguière, 1789		S				
Chromodoridae	*Chromodoris britoi* (Felimida britoi)*	(Ortea & Pérez, 1983)		L				
	Hypselodoris punctata (Felimare cantabrica*	(Bouchet & Ortea, 1980	?					
	*Hypselodoris tricolor**	(Cantraine, 1835)	S					
Clathurellidae	*Comarmondia gracilis**	(Montagu, 1803)	S					
Columbellidae	*Columbella rustica*	(Linnaeus, 1758)		L			S	
	Mitrella broderipi	(Sowerby, 1844)					S	
	Mitrella minor	(Scacchi, 1836)		L				
	Mitrella scripta	(Linnaeus, 1758)		L				
Conidae	*Conus mediterraneus?*	(Bruguière, 1792)			L		S	
	Conus ventricosus	Gmelin, 1791		L			L	
Costellariidae	*Vexillum savignyi*	(Payraudeau, 1826)					S	

Creseidae	*Creseis acicula*	(Rang, 1828)						
Cypraeidae	*Luria lurida*	(Linnaeus, 1758)		L				
Epitoniidae	*Epitonium clathrus*	(Linnaeus, 1758)	L					
Eulimidae	*Cythara albida**	Carpenter, 1864	S					
Fasciolariidae	*Fasciolaria lignaria*	(Linnaeus, 1758)		S				
	Fusinus labronicus	(Monterosato, 1884)	S					
Haliotidae	*Haliotis tuberculata lamellosa*	Lamarck, 1822		S, L				
	Haliotis tuberculata	Linnaeus, 1758			L		S	
Littorinidae	*Littorina neritoides*	(Linnaeus, 1758)			L			
Mangeliidae	*Bela nebula**	(Montagu, 1803)	S					
Muricidae	*Hexaplex trunculus*	(Linnaeus, 1758)		L				
	Muricopsis cristatus	(Brocchi, 1814)		L				
	Ocinebrina aciculata	(Lamarck, 1822)		S				
Nassariidae	*Cyclope donovania*	Risso, 1826					L	
	Cyclope neritea	(Linnaeus, 1758)					S	
Naticidae	*Naticarius dillwyni*	(Payraudeau, 1826)			S			
Patellidae	*Patella caerulea*	Linnaeus, 1758		S			S	
	Patella intermedia	Murray, 1857					S	
	Patella nigra	(da Costa, 1771)		S				
	Patella rustica	Linnaeus, 1758					S	
	Patella ulyssiponensis	Gmelin, 1791					S	
	Patella vulgata	Linnaeus, 1758		S				
	Patella sp.	Linnaeus, 1758						?
Phasianellidae	*Tricolia pullus*	(Linnaeus, 1758)					S	
	Tricolia tenuis	(Michaud, 1829)					L	
	Odostomia conoidea	(Brocchi, 1814)		S				
Plakobranchidae	*Thuridilla hopei*	(Vérany, 1853)		L				
Raphitomidae	*Raphitoma cordieri**	(Payraudeau, 1826)		L				
Rissoidae	*Alvania cimex*	(Linnaeus, 1758)		S			S	
	*Alvania reticulata**	(Montagu, 1803)		S				
	Rissoa splendida	Eichwald, 1830	S					
Strombidae	*Strombus decorus raybaudi*	Nicolay & Manoja, 1983	S					
Thaididae	*Thais haemastoma*	(Linnaeus, 1758)		L				
Triphoridae	*Triphora perversa*	(Linnaeus, 1758)	L					
Trividae	*Trivia arctica*	(Salander, 1797)	L					
Trochidae	*Clanculus jussieui*	(Payraudeau, 1826)		S, L				
	Gibbula divaricata	(Linnaeus, 1758)		S				
	*Gibbula drepanensis**	(Brugnone, 1873)					S	
	Gibbula richardi (Phorcus richardi)	(Payraudeau, 1826)			S			
	Gibbula umbilicalis	(da Costa, 1778)					S	
	Gibbula varia	(Linnaeus, 1767)		L			S	
	Jujubinus dispar	Curini-Galletti, 1982			?			
	Jujubinus exasperatus	(Pennant, 1777)	S	S			S, ?	
	Monodonta turbinata	(Born, 1780)		L			S	?
Turridae	*Raphitoma linearis (?)*	(Montagu, 1803)				L		
Turritellidae	*Turritella communis*	Risso, 1826	S					
Vermetidae	*Vermetus sp.*	Daudin, 1800			S, L			

Table 2 shows the list of the 133 molluscs found in Calvi during the course of 2014. This year we described about 20 new species, but there were less species collected than in the years before. To get an increased number of samples, more Coralligène should be collected, because of the high number of individuals in this area. Or the number of collectors should be higher, to get more samples and within a higher species richness. In total the number of different species which are identified is still becoming more, which means, that the biodiversity in Revellata and surrounding is still high. We cannot tell if the species richness is still given through the good ecological condition or if long term researches would show that the whole area is suffering under the high human pressure and the richness would decrease in a long term study. There is already an effect on the species living in the bays, especially through the high human impact and the global exchange of species with other oceans. But disregarding that point, the bay of Revellata is a very species rich area and offers a lot of different opportunities to collect a high amount of molluscs. Table 2 shows the individuals, the class/family and the localization where the animals were found. It is also shown if they were still alive (L) or dead (shell, S).

As shown in Table 2, some very common molluscs were found, as every year (*Arca noae, Thais haemastoma etc.).* In comparison to the other years (table 2 and figure 12), fewer Polyplacophora and Bivalvia were found. While the number of Bivalvia was just slightly lower than the years before (figure 14), the number of Polyplacophores was decreasing (figure 13). In this course there were no Scaphopods detected. According to the data of the last course, we would have expected them in the Coralligène, but despite intensive search there were no individuals found. Figure 12 points out, that the high number of species found in 2010 could not be reached in 2012 nor in 2014 but it was still higher than in 2004.

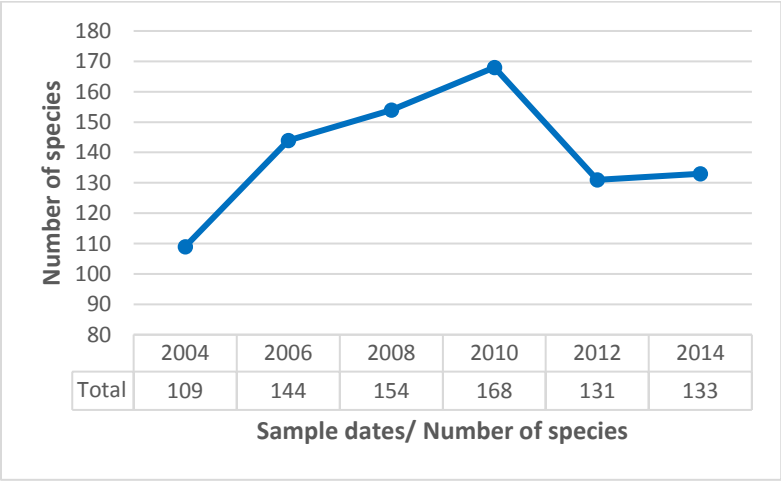


Figure 10: The numbers of collected species were increasing from 2004 to 2010 and decreasing drastically in 2012 and 2014.

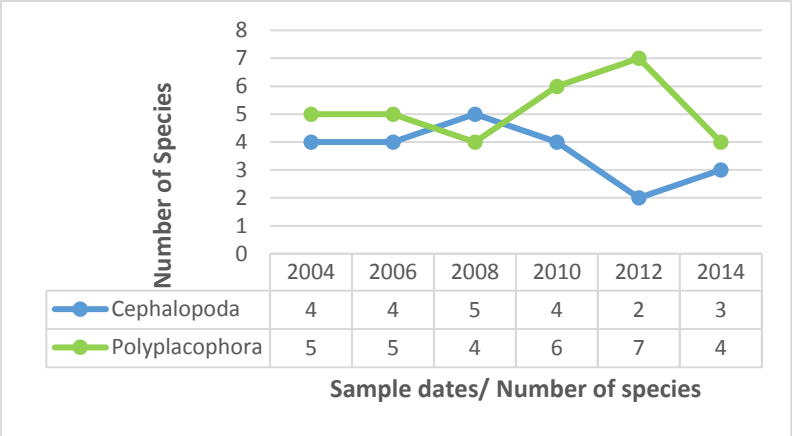


Figure 11: The numbers of Polyplacophora were increasing from 2008 to 2012 and decreasing until 2014, but there were more Cephalopoda found than in 2012.

Figure 13 reveals the slowly increasing number of Cephalopods found and the decreasing individuals of Polyplacophores. Compared to the years 2004, 2006 and 2008, the collected number was not that low, but 2010 and 2012 showed, that the number could be much higher.

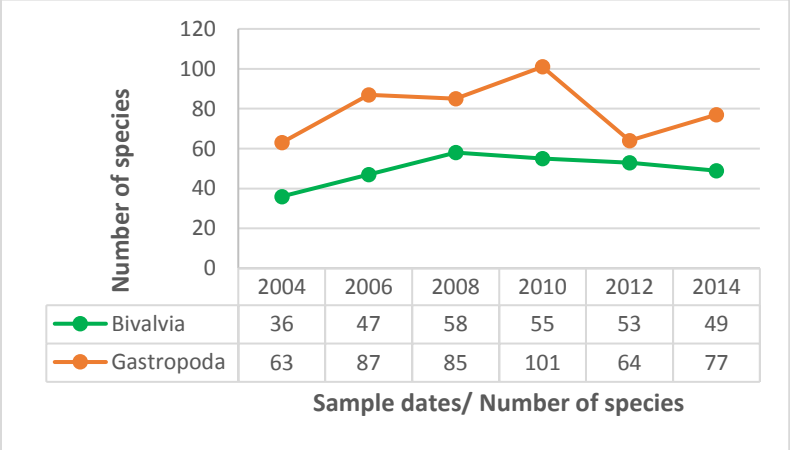


Figure 12: While the number of Gastropods was decreasing from 2010 to 2012 and then increases slightly, the number of Bivalvia is just increasing from 2004 to 2008 and then constantly decreasing to 2014.

Also the number of different Gastropod species, that were sampled, was higher in 2010. But still 2014 the amount was increasing compared to 2004 and therefore an improvement. The collected Bivalvia are nearly at the same level as from 2004 to 2012. Maybe all the results are depending on weather, temperature, samplers and also on the conditions in the sea of the whole area. While comparing the different sampling sites, some differences should be mentioned. For example, the most species were found in the boulder field (figure 15).

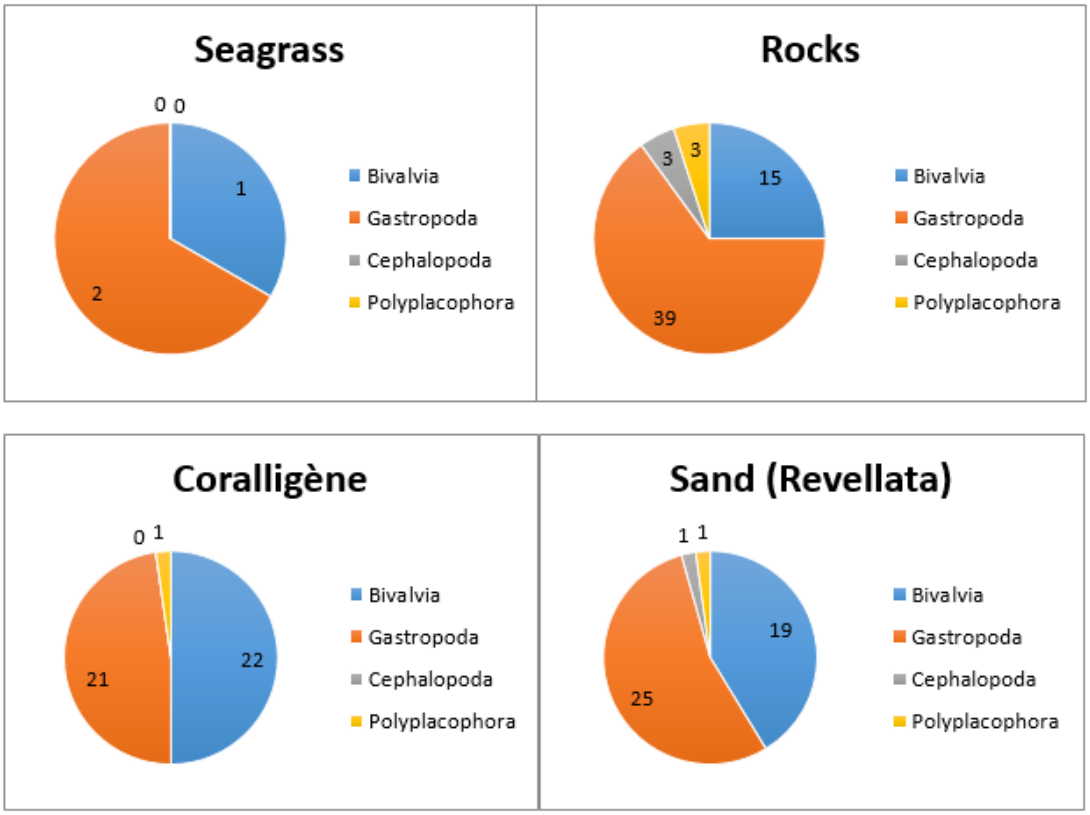


Figure 13: Species composition of the different habitats investigated.

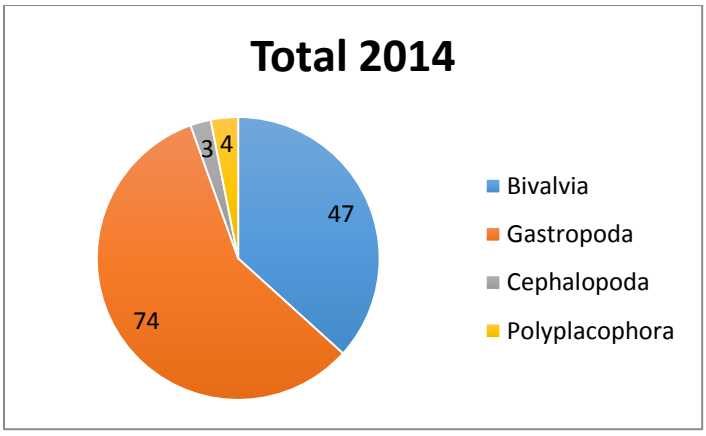


Figure 14: All species listed in a graph, comparing the different habitats.

In 2012, the habitat with the highest species richness was the Coralligène and also the Sandy beach was depicted with a lot of individuals. Our course could not collect as much individuals, especially in the Sandy beach, as the year before. Maybe more sampling trips would increase the number or a more extended sampling day.

In three habitats the Gastropods are the most common species, except of the Coralligène, where the Gastropods are nearly equal to the Bivalvia. They were collected in a high number and were also easy to collect. Gastropods are in the whole Mediterranean area highly abundant and contain a lot of different species. Also the Bivalvia were very abundant and were collected in a high number in

all areas. Because of their sessile life and the wide abundance of empty shells, those molluscs are easy to catch, especially with nets and sifters.

There were no Cephalopods found in the Coralligène. It was not surprising to us that they were not found there, because they are normally not occurring in this habitat. In the Posidonia meadows were not many species found, but due to the fact that this meadow is a protected area, invasive and numerous sampling was not possible. The Posidonia fields are also in a high depth, what makes the sampling difficult. But also in the open water column above the seagrass meadow were no Cephalopods seen, although they are likely found there.

Table 3: The number of species occurring in the three specific habitats. The Posidonia habitat has been neglected due to the few species detected there. The species of the Finger Bay have been included in the Rock habitat.

	Coralligène	Rock	Sand
Coralligène	38	1	3
Rock	1	45	13
Sand	3	13	26
In all three habitats	2	2	2
Total	44	61	44

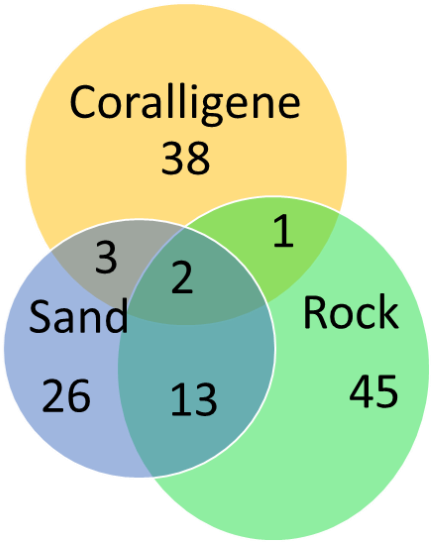


Figure 15: Venn-Diagram showing the overlapping occurrence of species in the specific habitats. The Posidonia habitat has been neglected due to the few species detected there. The species of the Finger Bay have been included in the Rock habitat.

As shown in Table 3 and the Venn-Diagram in Figure 17, we can conclude that some species occur in more than one habitat (the posidonia habitat has been neglected since only three species have been detected there, which we do not consider representative). For Example two species, namely

Cerithium vulgatum and *Jujubinus exasperatus* have been found in all three habitats (rock, sand, Coralligène). The highest overlap can be seen between rock and sand with 13 species found in both habitats. Yet this discovery could be biased by two compromises done for this investigation: firstly, the species found at the Finger Bay have been included into the rock habitat. It is true that the shoreline at the Finger Bay is mainly composed of rocks and stones, yet further into the sea the habitat changes to sand and posidonia. Secondly, some the species found at the Sandy Beach, designated as living in the sand habitat, are derived from different habitats and their shells are washed to the shore be wave action and current. Another fact is that the Sandy Beach is surrounded by rocky shore and also this area has been searched for molluscs that have been included as species of the Sandy Beach. Moreover the relative close distance between the Finger Bay and the Sandy Beach leaves space for all kinds of transitions between the habitats.

To summarize the course, the Gastropods were the most common species collected, as they were in the years before. They are a very divers and species rich group and occurring in the whole Mediterranean. Gastropods are typical representatives for bays and boulder fields, so it was not surprising to us to find them there in a high number. The habitat-characteristic and most common species are described in more detail in the following part.

Molluscs - common and special species - a picture catalog

I. Site/ habitat: **STARESO-bay** and "**Fingerbay**"; rocks
   - *Octopus vulgaris*
   - *Sepia officinalis*
   - *Monodonta turbinata*
   - *Thais haemastoma*
   - *Hexaplex trunculus*
   - *Patella sp.*
   - *Arca noae*
   - *Haliotis tuberculata lamellosa*

II. Site/habitat: **Posidonia meadow**/ seaweed
   - *Pinna nobilis*
   - *Chromodoris britoi*

III. Site/habitat: **Sandy beach- Revellata**; sand
   - *Pseudochama gryphina*
   - *Conus ventricosus*
   - *Ctena decussata*
   - *Loripes lucinalis*

IV. Site /habitat: **Coralligène**

- *Hypselodoris tricolor*
- *Hypselodoris villafranca*
- *Aplysia punctata*
- *Chlamys sp.*
- *Cerithium alucaster*

I. Site/ habitat: STARESO- bay and "Fingerbay"; Rocks

- ***Sepia officinalis***

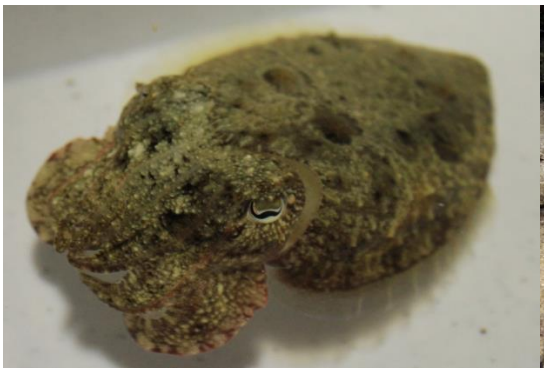
Sepia officinalis is a common Cephalopod of the Mediterranean Sea. The "Kalk-Schulp" of *Sepia officinalis* is used as grindstone for cage birds. It has a sac filled with ink consisting of concentrated melanin, which formerly was used to dye cloths of photo paper. Nowadays it is only used to dye pasta black.

- ***Octopus vulgaris***

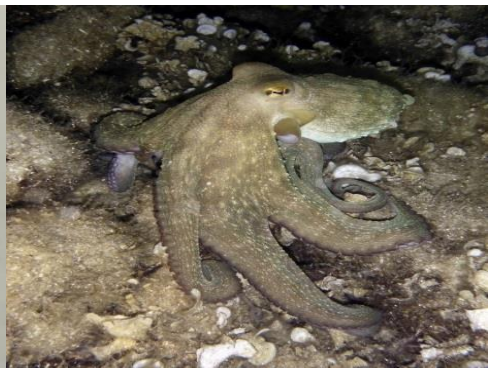
The *common octopus* is found in oceans worldwide. It is a predatory Cephalopod, very able to learn and capable of quick color changes, just as *Sepia officinalis* can do. During the day it hides in holes between rocks. Both of them belong to the foodstuff of humans.

- ***Loligo vulgaris***

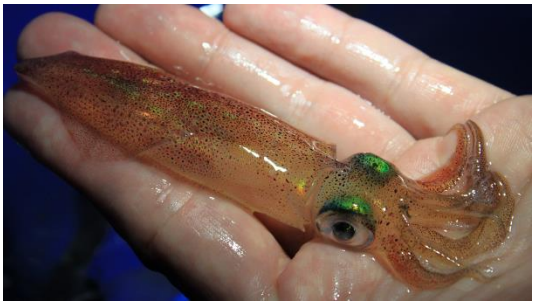
This species is widely abundant in the Mediterranean Sea. It can reach a maximum size of 75 cm and is a predator. Loligos prefer deep waters and rise up during the night. They are fast swimmers and are able to change their color. Although they are hunted very often, the species is not endangered.



Picture 1: Sepia officinalis



Picture 2: Octopus vulgaris



Picture 3: Loligo vulgaris

- ***Monodonta turbinata***

The *Turban shell* excels by a rounded and conical shell. The base of the spindle is marked by a strong tooth, an umbilicus is missing. The inside of the shell is overlaid with nacre and is very common on rocky shores.

Picture 4: Monodonta turbinata

- ***Thais haemastoma***

Thais haemastoma, also known as the violet sea snail, has a rounded or ovate shell. Often it is colored brown with white stripes and with an orange colored mouth. In times past, glands of violet snails were used to dye wool or cloths crimson red. It`s also known as the royal red, because it was often used for upper nobility.

Picture 5: Thais haemastoma

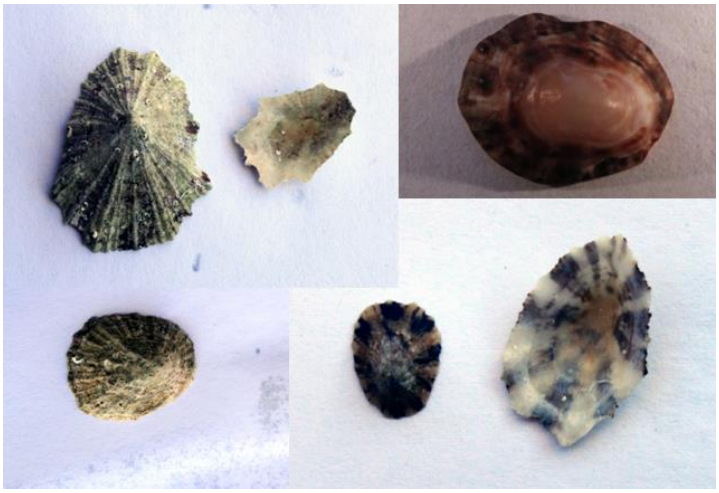
- ***Hexaplex trunculus***

Hexaplex trunculus, also known as banded dye-murex, is a common species in the Mediterranean Sea. Its shell is conical and banded white and brown. At risk its hypobranchial gland excretes a secretion, which changes color in action of light to indigo blue. In times past it was used to dye cloths.

Picture 6: Hexaplex trunculus

- ***Patella sp.***

The snail shell of patella looks like a small bowl without any coil. They are perfectly adapted to the rocky substrate. By excreting acid they are able to form the substrate, so that the bowl closes exactly up with the rock. The species are very variable and difficult to differentiate.



Picture 7: Patella sp.

- ***Arca noae***

Noah's ark shell is also a common species of the Mediterranean Sea, where it lives on all types of bottoms that contain hard substrates. Its shell is rounded and oblong, with a nearly straight upper rand.

Picture 8: Arca noae

- ***Haliotis tuberculata* form lamellosa**

The abalone resembles an external ear and its shell is rich in nacre.

II. Site/habitat: Eelgrass meadow/ seaweed

- ***Pinna nobilis***

Pinna nobilis, also known as the noble pen shell. It is a marine bivalve mollusk and large individuals can reach up to 120 cm of shell length. This mussel is a protected animal.

- ***Chromodoris britoi***

This species of colorful sea slug belongs to the Nudibranchia, which are marine gastropod molluscs.

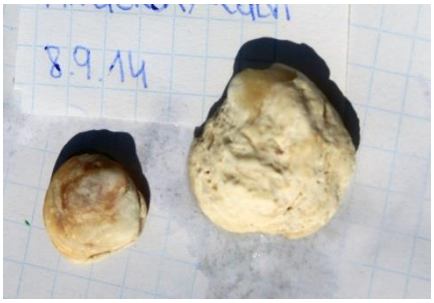
Picture 9: Pinna nobilis

Picture 10: Chromodoris britoi

III. Site/habitat: Sandy beach- Revellata; sand

- ***Pseudochama gryphina***

It inhabits rocky grounds at the upper sublittoral zone. But its shells can be found in high abundance at the sandy beach, where it is washed to by wave action.



Picture 11: Pseudochama gryphina

- ***Conus ventricosus***

The Mediterranean Cone shell is a sea snail of the family Conidae. Some representatives of this family are toxic.

Picture 12: Conus ventricosus

- ***Ctena decussata***

A primarily Mediterranean species, which is associated with seaweed in the sublittoral zone and shallow shelfs.

Picture 13: Ctena cecussata

Not to be confused with:

- ***Loripes lucinalis*,**

which is also a common species of sandy beaches of the Mediterranean Sea. The difference between *C. decussata* and *L. lucinalis* is the reticulate sculpture of the shell of *C. decussata,* while L. lucinalis possesses a smooth shell surface.

Picture 14: Loripes lucinalis

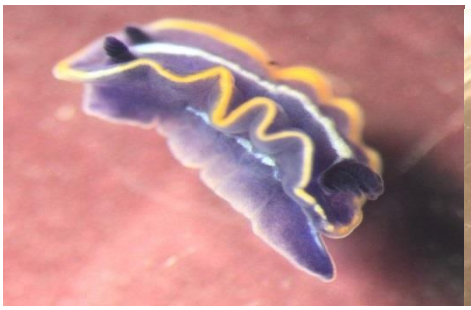
IV. Site /habitat: Coralligène

- ***Hypselodoris tricolor***

Also known as *Felimare tricolor*, it is a blue bodied Nudibranchia, with white-to-orange lines running longitudinally along its body.

- ***Hypselodoris villafranca***

Is also a Mediterranean species of the Nudibranchia. It is usually blue, often quite dark blue in color, with longitudinal yellow stripes.



Picture 15: Hypselodoris tricolo

Picture 16: Hypselodoris villafranca

- ***Aplysia punctata***

Aplysia punctata, of the family Aplysiidae, is also known as sea hare. Sea hares are marine Opisthobranchia.

Picture 17: Aplysia punctate

- ***Chlamys sp.***

Representatives of Chlamys species are small scallops of the family Pectinidae. They are very variable in color and size. In the Coralligène, they are a very abundant species, mostly colored red or brown.

Picture 18: Chlamys sp.

- ***Cerithium alucaster***

This is a marine mollusk of the family Cerithiidae. It has a slight shell with a lot of windings, which is why it has a pointed form.

Picture 19: Cerithium alucaster

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Fish Survey

Dana Michaelis, Henrike Wunderow, Huajing Yan, Ines Hrabie,
Lara Schmielau, Saskia Amann, Teresa Müllauer,
Jasha Dehm, Reinhold Hanel

Introduction

The Mediterranean littoral zones are dominated by rocky environments (Fasola, Canova, Foschi, Novelli, & Bressnan, 1997) and therefore constitute an optimal habitat for fish (Harmelin-Vivien, Harmelin, & Leboulleux, 1995). These environments are inhabited by various species simultaneously, especially in regions where marine management strategies have been put in place (Frau, Deudero, Cerdeno, & Alou, 2003).

The coexistence of various species in the same ecosystem would theoretically result in inter-species competition (Sala & Ballesteros, 1997). This scenario is avoided by resource partitioning, whereby species of fish in the same ecosystem are segregated in terms of food, habitat or time at which active (Sala & Ballesteros, 1997).

In the Mediterranean, there have been only few studies focusing on resource partitioning in fishes (Sala & Ballesteros, 1997), however since 1992 the University of Innsbruck (and later in cooperation with the Christian-Albrechs-Universitaet zu Kiel) have conducted surveys which involved the formulating of a fish species list as well as determining temporal changes with regard to substrate type and day/night periods. These surveys were conducted at coastlines along STARESO (Station de Recherches Sous-Marines et Océanographiques) research facility and adjacent bays. The line intercept transect and area transect methods are the general census methods used, due to these methods being cost effective (Gates, 1979), adequately precise and accurate, and cause close to no disruption/destruction (Halford & Thompson, 1994).

In 2014, the purpose of this survey was to: identify littoral fish species and add onto a pre-existing species list of the area, determine how the dominant species are involved in resource partitioning, and allow students to gain a better understanding through direct involvement with the methods.

To determine how species are segregated in terms of resource partitioning, the survey was divided into three parts. Firstly, the rhythmic changes in species composition throughout day and night periods were identified. This was conducted via a 24-hour line intercept transect (LIT) method.

Secondly, an area transect was conducted using the rapid visual technique, which allows for an estimation of species in a given area. This was conducted to allow for comparison of fish community structure between various habitats and locations. Lastly the feeding habits of selected fish species were determined via gut content analysis. In this survey, gut content analyses were conducted on the three sympatric species *Scorpaena porcus*, *Serranus scriba*, and *Labrus viridis*. Gut content analysis in this study used the volumetric method and allowed us a better understanding of food partitioning and niche separation in typical Mediterranean littoral fish community.

Materials and Methods

Total fish species list

The identification and subsequently, the formulation of the fish species list was conducted using various literature and fish guides (e.g. Riedl, 1983; Bergbauer & Humberg, 2009), which were used to identify fish sighted during snorkeling trips throughout the entire 13-day stay in Corsica. For fish species which were not identifiable at first glance, photos were taken for later idenification. These were then added and compared to a pre-existing list of previously sighted fish.

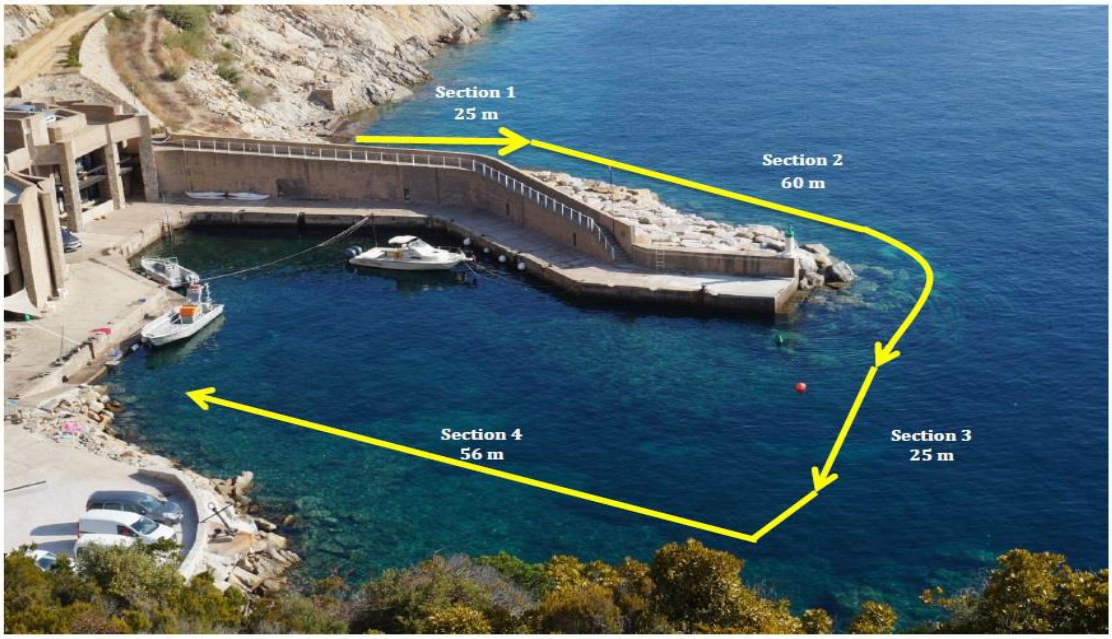

Figure 1: Showing the 24-hour transect line including sections.

24-hour transect

A 166 x 6 meter transect **(Figure 1)** was setup in such a way that various habitats were included in the transect, thus allowing for comparison between the communities. The transect line was then divided into four sections reflecting the various communities: Section 1: shallow water with rocky substrate and had a length of 25 m, Section 2: deep water with rocky substrate composed primarily of artificial tetrahedrons and had a length of 60 m **(Figure A).** Section 3: deep water with seagrass substrate and had a length of 25 m **(Figure B)**. Section 4: intermediate water depth with rocky substrate and a length of 56 m **(Figure C)**. A depth profile **(Figure 2)** was also conducted using a hand-held echosounder.

Fig. A: Artificial tetrahedrons in section 2.

Fig. B: Posidonia oceania covered the bottom of section 3.

Fig .C : Rocky substrate at section 4.

The transect was marked out with an orange line fixed to the substrate and stone markers **(Figure D)** were used to mark out the transect width. Buoys made of plastic bottles were used to mark out the sections **(Figure E)**.

Fig. D: Stone Marker to mark out width of transect.

Fig. E: Plastic Bottles used to mark Beginning/ end of sections.

The 24 hour transect was conducted at 2 hour intervals beginning at 12:00 noon on September 8th, 2014 and ended at the same time on September 9th, 2014. At each interval 2 snorkelers would swim along the transect at a steady pace stopping only between sections to record sightings. The abundances were recorded in categories namely; category A (1 individual), B (2-5 individuals), C (6-30 individuals), D (>30 individuals) and E (>100 individuals) and was entered into a Microsoft Excel Spreadsheet for data analysis.

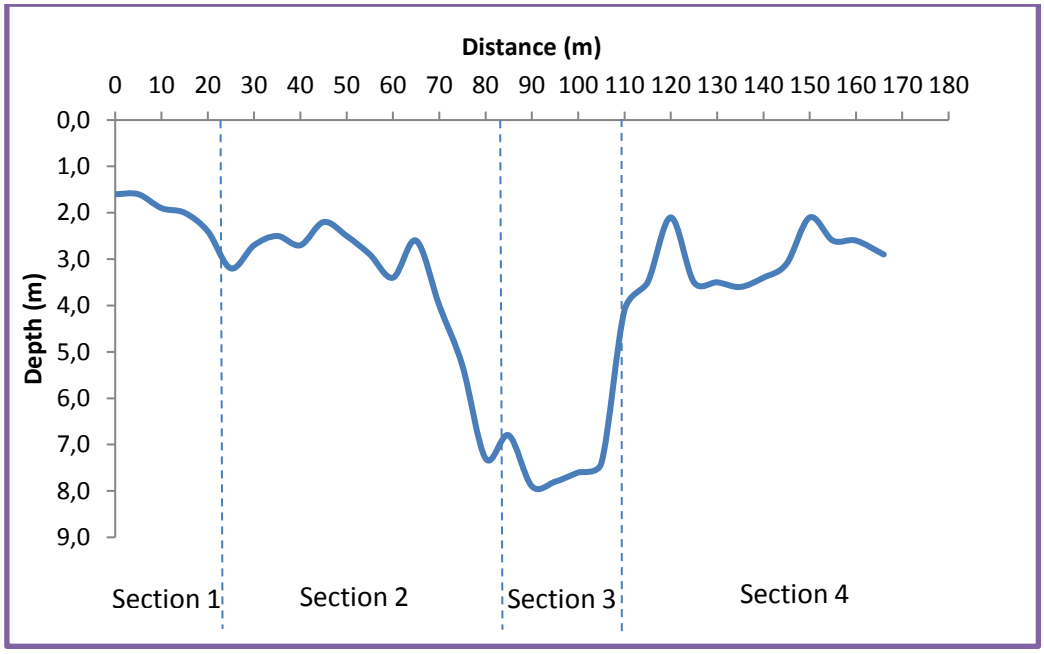

Figure 2: Depth profile of the 24-hour transect.

Area Transect

The rapid visual technique was carried out to conduct the area transect and is a method which involves randomly snorkelling for a defined time over a given substrate type, while recording all observed fish in the process.

In this case, the area transects were conducted over sandy, stony and seagrass substrates at two locations, namely Mar A beach (sandy beach) and a stony beach. Each of the sites was surveyed in duplicate by different pairs of snorkelers for ten minutes. Before being transferred into a Microsoft Excel spreadsheet for data analysis, observations were recorded on a "dive slate" following the same abundance categories as in the 24-hour transect.

Gut Analysis

Ten individuals of each species of interest (*Scorpaena porcus, Serranus scriba, Labrus viridis*) were captured from the vicinity around STARESO Research Station using hand nets and a harpoon. After capture, individuals were immediately frozen for later analysis.

Before analysis, all fish were defrosted, after which the weight, length, sex, and morph of each individual was noted, and a number was assigned. The fish was cut from the anus up towards the gill chamber and all intestine were taken out and placed onto a petri dish. The gut and intestine were cut open and all contents were taken out and washed with some tap water. The contents were then identified and separated into categories of fish, crustacean, mollusc, and others. The abundance of each category was estimated by eye as percentage of total contents.

The data was averaged within each species and the result represents the percent average value for each species. Fish individuals that had empty guts were excluded from the data average.

Results

Total fish species list

The Mediterranean is home to around 737 species of fish (Froese & Pauly, 2014), of which 67 species (9%) were sighted and identified this year, refer to Appendix 1. This comprises approximately 66% of the total species number (102 species) sighted by the previous Corsica excursions since 1992.

24 hour transect

To allow for simplified result analysis, fish observations from the 24 hour transect were divided into 3 parts (Sparidae, Labridae, Others) allowing comparison within the two dominant fish families, namely; Sparidae and Labridae.

1. Sparidae

The sea breams were observed throughout the entire 24 hour period, **(Figure 3)**, though slightly higher numbers were observed towards the late afternoon and early morning, suggesting possible crepuscular behaviour. Although sightings of sea breams remained relatively high throughout the night, it should be noted that the fish were inactive/motionless.

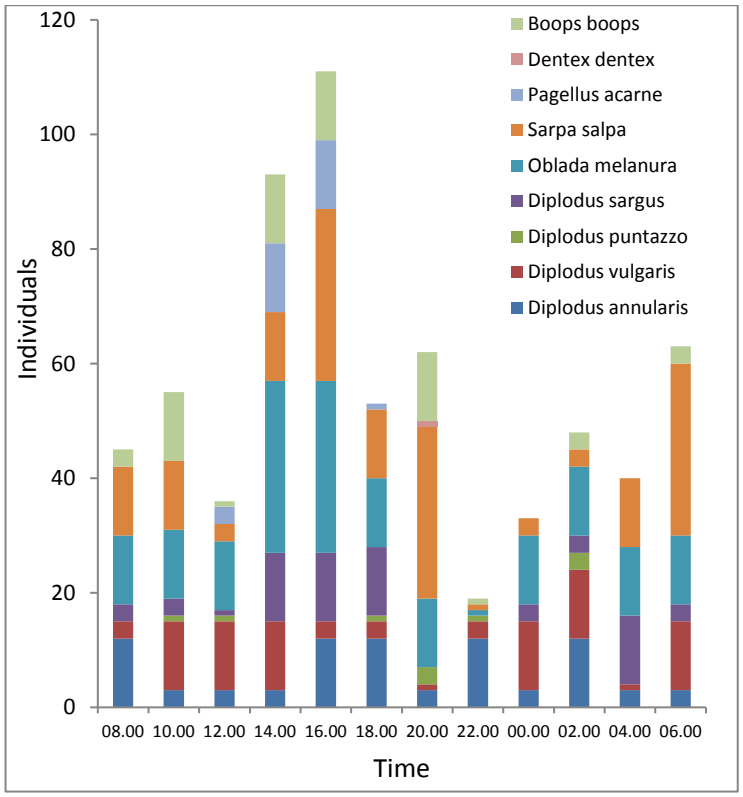

Figure 3: Sparidae species abundance over 24-hour period.

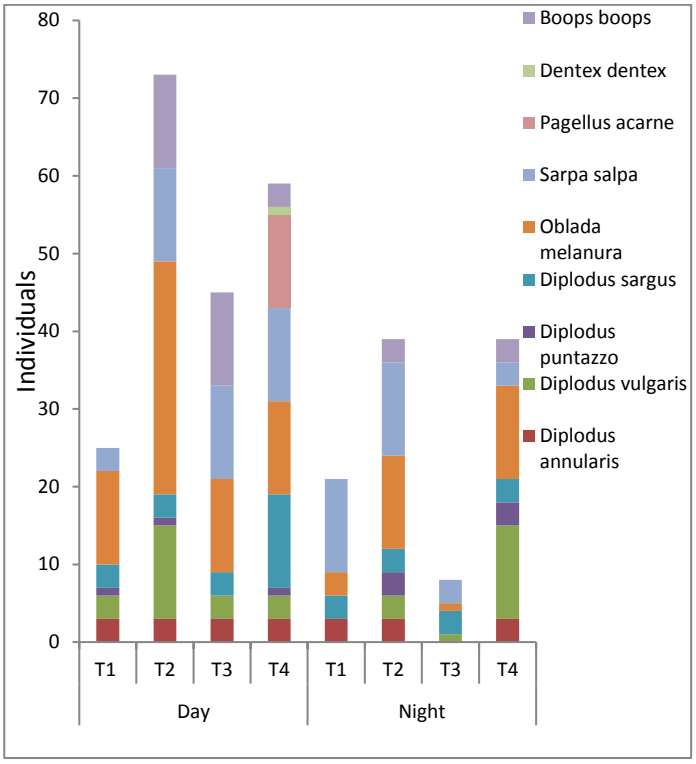

Figure 4: Sparidae distribution with regard to Transect Section.

Sarpa salpa and *Oblada melanura* were the most dominant species throughout the day and early morning hours **(Figure 3)**. Throughout the night, *S. salpa* and *O. melanura* observations reduced drastically, whereas the other sea bream observations remained relatively constant over time **(Figure 3)**.

Sparidae were found across all four sections, **(Figure 4)**, regardless of the substrate. However, a significantly higher amount could be observed over sections 2 and 4 during both day and night phases, suggesting preference to deeper water with rocky substrate over shallow water or seagrass substrate. All species were found at relatively equal proportions across all sections with the exception of *Boops boops* **(Figure 4)**, which was not observed over section 1 (shallow rocky), suggesting that the species prefers slightly deeper waters clearly below the intertidal zone.

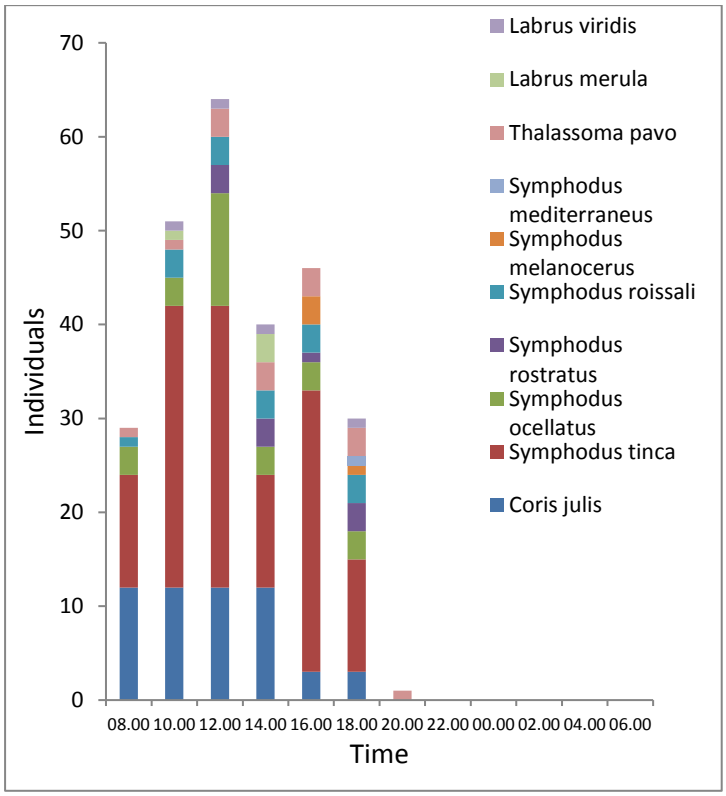

Figure 5: Labridae species abundances over 24-hour period.

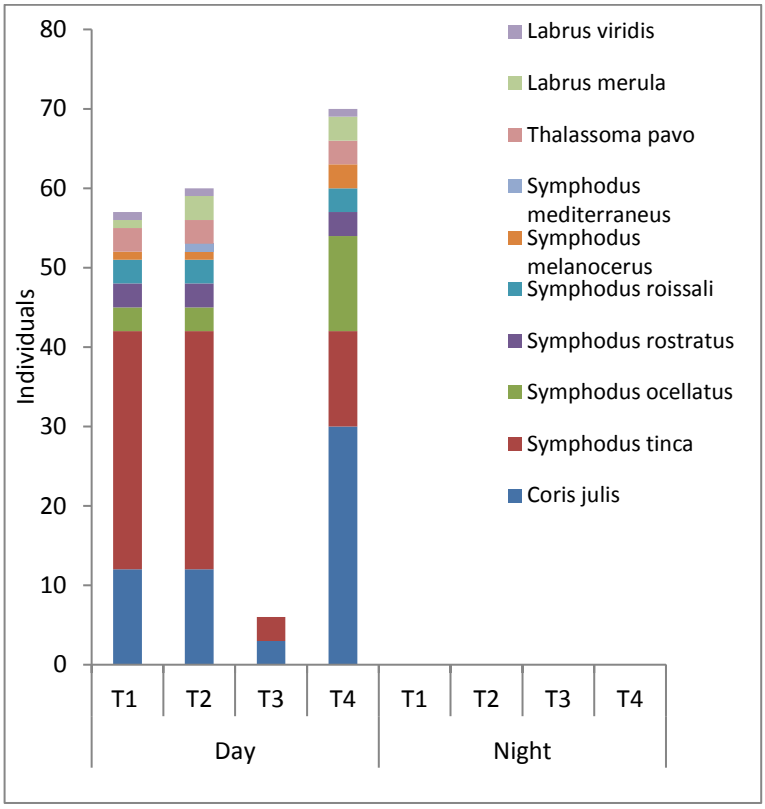

Figure 6: Labridae distribution with regard to Transect Section.

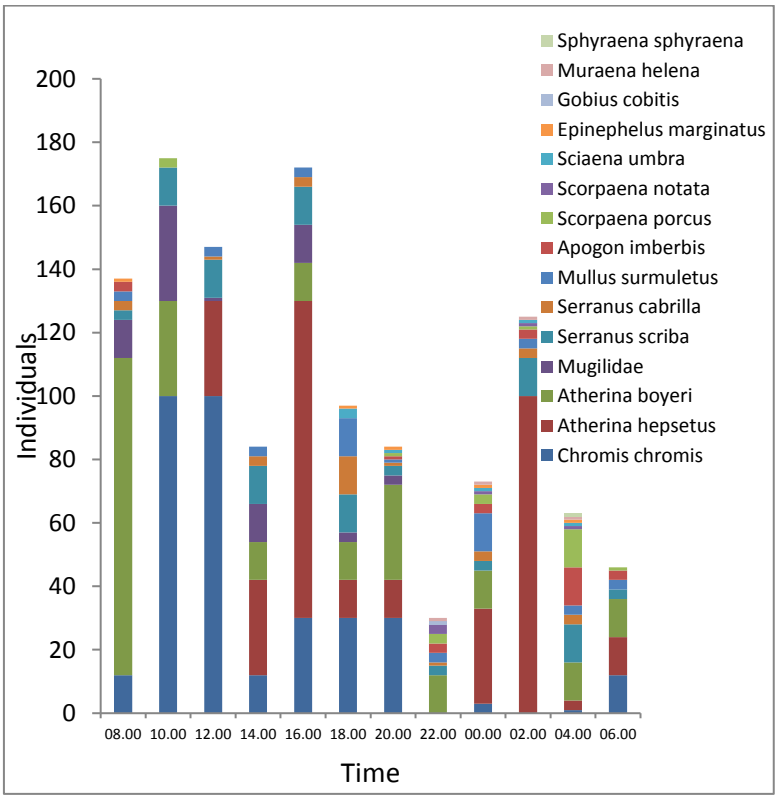

Figure 7: Other fish Species abundances over 24-hour period

2. Labridae

All wrasse species were only observed throughout the day **(Figure 5)**, thereafter no wrasse species were observed. Activity period began between 06:00 and 08:00 and ended between 18:00 and 20:00, with the peak abundance at noon **(Figure 5)**.

Symphodus tinca and *Coris julis* dominated the species counts with between 10 and 30 individuals recorded per daylight-time period **(Figure 5)**. The least recorded were both *Labrus viridis and Labrus merula* with 1-2 individuals per sighting.

Labridae were recorded across all 4 sections of the transect **(Figure 6)**, however section 3 (seagrass substrate) contained only *Symphodus tinca* and *Coris julis* at less than 5 individuals of each. In comparison, significantly higher numbers (>10) of *S. tinca* and *C. julis* were observed at the other sections. Section 4 recorded the highest abundance of wrasse species **(Figure 6)**.

3. Others Fish Species

Fish families with low species numbers were recorded and compared as "other fish species" **(Figure 7 & 8)**. This group was dominated throughout the day by *Atherina boyeri, Atherina hepsetus,* and *Chromis chromis,* which form swarms of high number of individuals **(Figure 7)**. Throughout the night, *C. chromis* observations sink to almost none,

suggesting that the species abandons its daytime open-water schooling behaviour to seek shelter for the night. *A. boyeri* and *A. hepsetus* numbers only slightly decrease due to swarm dissolution and spreading out of individuals throughout the water column. During the day all three species swarms were found in the water column below the surface, across all four sections of the transect **(Figure 8)**.

Serranus scriba, Serranus cabrilla and *Epinephalus marginatus* were found at all time points with 1 to 10 individuals, of whom *S. scriba* was the most frequent and *E. marginatus* the least frequent **(Figure 7)**. These species were also found at night, however they were mainly inactive atop the substrate. All three species were found relatively close to the rocky substratum in sections 1, 2 and 4 **(Figure 8)**.

Scorpaena notada, Scorpaena porcus and *Apogon imperbis* are mainly nocturnal fish species; hence an increase in abundance (3-5 individuals) was noted between the hours of 20:00 and 06:00 **(Figure 7)**. All three species were found mainly along sections 2 and 4 of the transect, where there was a rocky substrate and relatively deeper water **(Figure 8)**. Both *Scorpaena* species were found "sitting" on the rocky substrate, whereas *Apogon imperbis* was found swimming in the water column above rocky substrate.

Other notable species include *Sciena umbra* and *Muraena helena* were found to be active during dawn and dusk, with 1 - 4 individuals recorded for time periods between 04:00 and 08:00 and between 18:00 and 20:00 **(Figure 7)**. Both of these species were dominantly found in sections 2 and 3 near holes and crevices close to the substratum **(Figure 8)**.

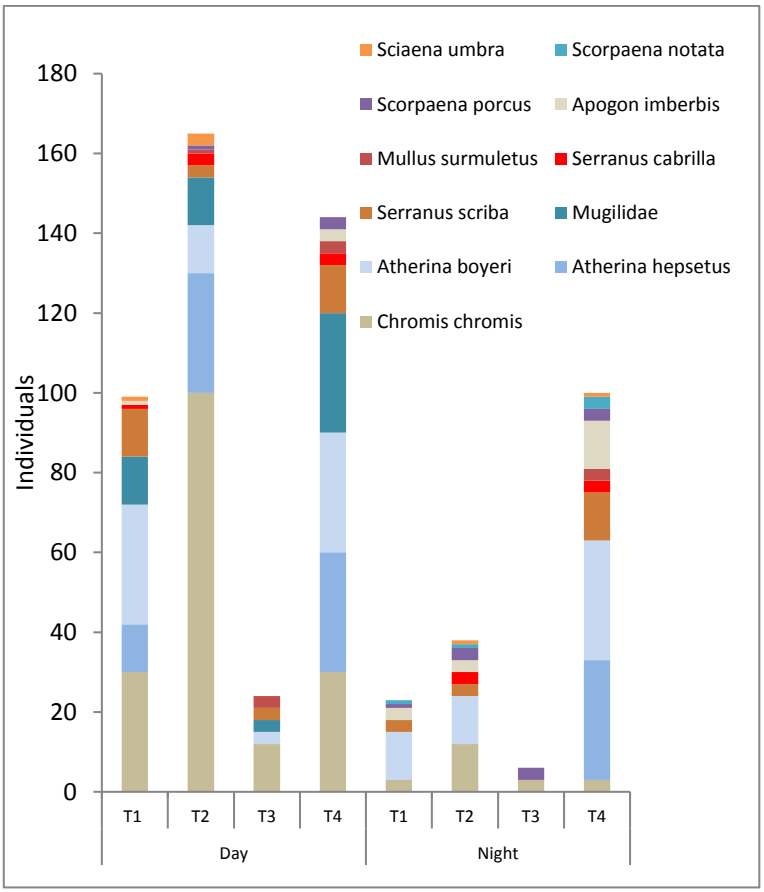

Figure 8: Other distribution with regard to Transect Section.

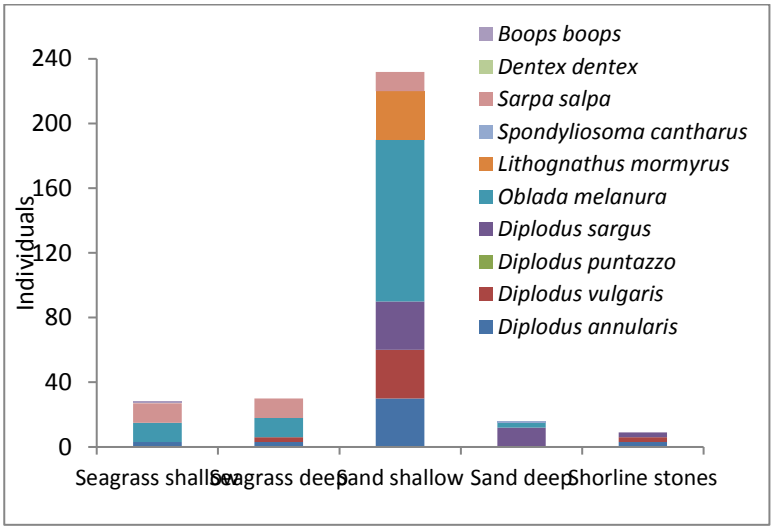

Figure 9: Sparidae abundance over different substrate types at Stony beach

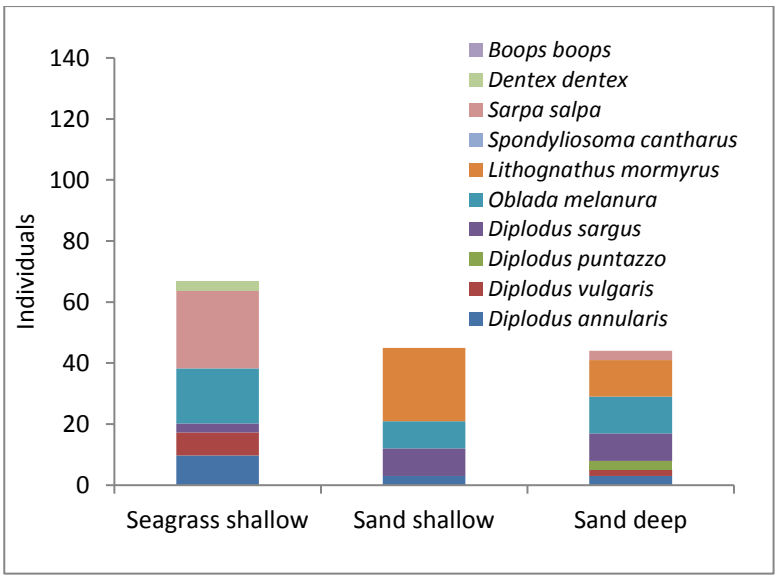

Figure 10: Sparidae abundance over different substrate types at Mara Beach.

Area transect

Overall, 26 fish species representing 12 families were visually censured with the area transect method at the stony beach. Out of these 26 fish species, 3 of them were only identified to family level. 24 fish species representing 13 families were visually censured at the sandy beach, while only 2 species were identified to family level.

The stony and sandy beach had most of the fish species in common. In all habitat types at the stony and sandy beach, the most abundant families were *Sparidae* and *Labridae* **(Figure 9, Figure 10, Figure 11 and Figure 12)**. The family of the *Labridae* was only absent over seagrass at the stony beach **(Figure 11)**.

The *Sparidae* preferred shallow sand areas at the stony beach, because highest abundances and the highest diversity of that family was observed there **(Figure 9)**.

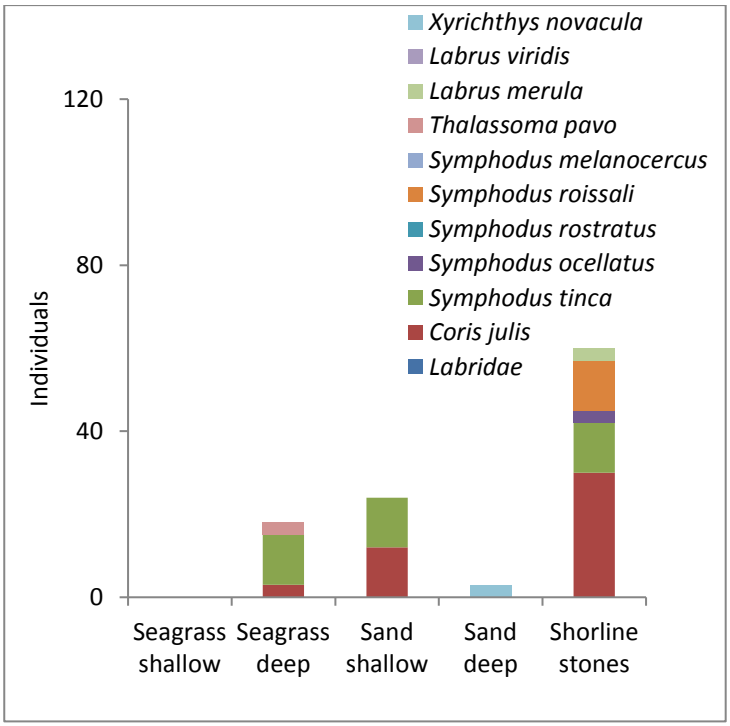

Figure 11: Labridae abundance over different substrate types at Stony beach.

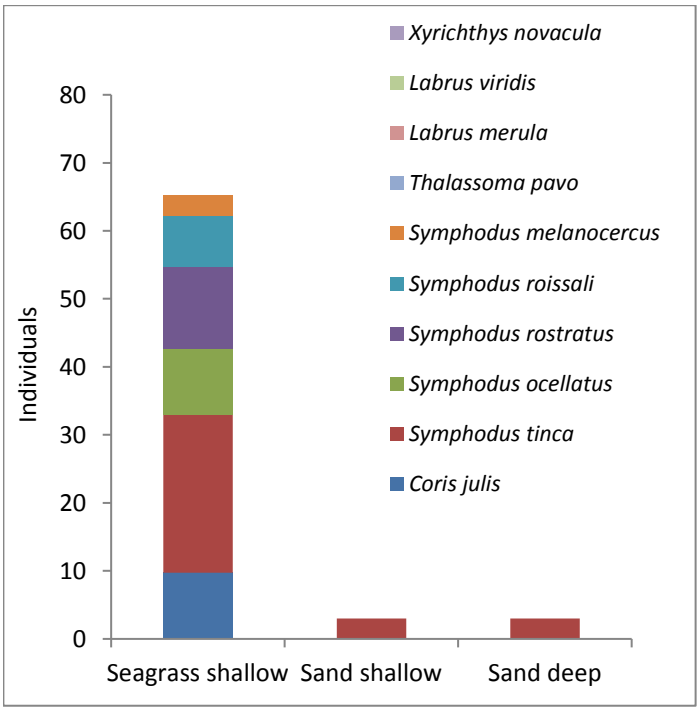

Figure 12: Labridae abundance over different substrate types at Mara beach.

At the sandy beach, the *Sparidae* were more evenly distributed over all habitat types **(Figure 10)**. The Sand steenbras (*Lithognathus mormyrus)* was exclusively observed over sandy substrate at both survey areas. Most of the species of Labridae were found along the shoreline at the stony beach **(Figure 11)**. The most abundant representative species was the Rainbow wrasse (*Coris julis)*. In contrast, the Labridae were almost exclusively observed over shallow seagrass at the sandy beach **(Figure 12)**. The East Atlantic peacock wrasse (*Symphodus tinca)* was the most abundant species and the only one, which occurred over all habitat types. Some species of

the Sparidae and Labridae family, for instance the Sharpsnout seabream (*Diplodus puntazzo)* and the Ornate wrasse (*Thalassoma pavo)* respectively, were only observed in one of the survey areas **(Figure 9, Figure 10, Figure 11 and Figure 12)**. The Spotted weever (*Trachinus araneus*), Greater weever (*Trachinus draco*) and the Common stingray (*Dasyatis pastinaca*) were primarily found at areas with a sandy substrate **(Figure 13 and Figure 14)**. The Garfish (*Belone belone)*, Yellow-mouth barracuda (*Sphyraena viridensis)* and the Wide-eyed flounder (*Bothus podas)* were sighted at various areas without specificity to substrate type **(Figure 13 and Figure 14)** though not necessarily linked to the substrate. *Chromis chromis* and species of the family Atherinidae were also sighted in high densities in both survey areas due to their swarming behaviour **(Figure 13 and Figure 14).**

The highest fish diversity at the sandy beach was found over shallow seagrass and at the stony beach over the stony shoreline. The abundances were also high over these substrates. Low abundances of fish species were especially found over deep sand.

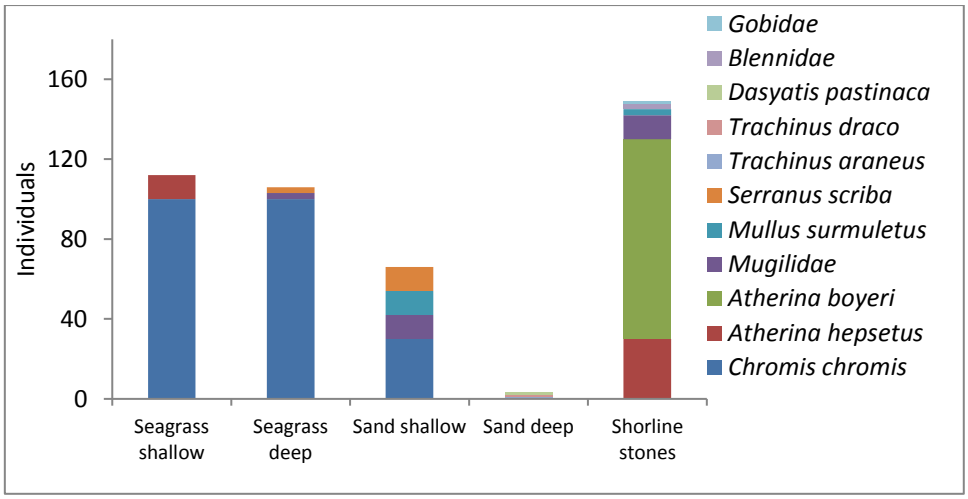

Figure 13: Other fish species abundance over different substrate types at stony beach.

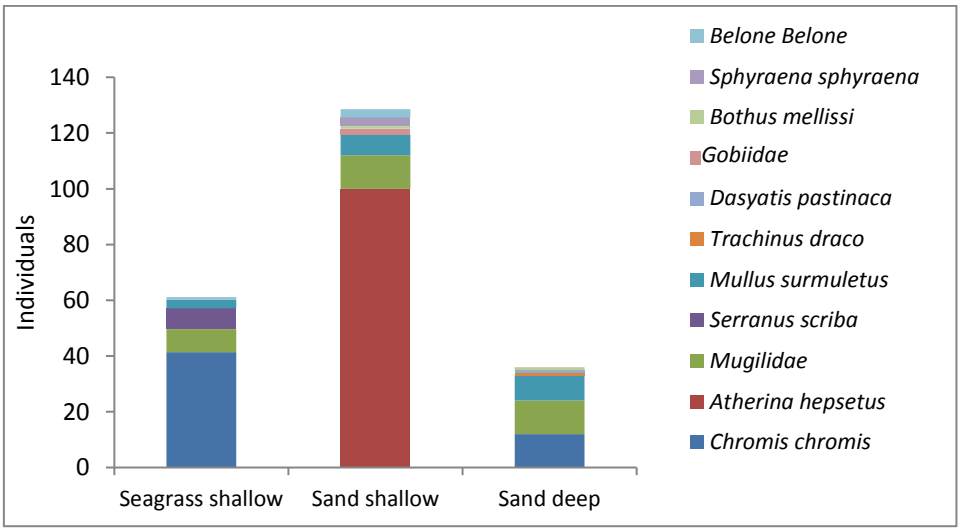

Figure 14: Other fish species abundance over different substrate types at Mara beach.

Gut Analysis

Scorpaena porcus gut analysis showed that this species mainly consumed crustacean as 75% of their diet while also consuming a small amount of fish at 23%. *Serranus scriba* consumes approximately half crustacean and slightly less fish. However, it is interesting to note that in one specific individual, only an ascidian was found taking up the entire gut content, which when shown in a pie chart with other data, reflect a large amount of 'others' that might not fully represent the rest of the individuals. *Labrus viridis* consumes almost completely fish at 98.6% and only a very small amount of crustacean of 1.4%.

Table 1: Table Showing number of guts analyzed and whether it is filled or empty.

Species	Total guts analyzed	Guts full	Guts empty
Scorpaena porcus	10	10	0
Serranus scriba	10	9	1
Labrus viridis	10	7	3

When looking into the amount of empty guts compared across the species, it is noticed that *Labrus viridis* had the most amount of empty guts and *Scorpaena porcus* had no empty guts. This could suggest that a difference in feeding behaviour where *Scorpaena porcus* would generally feed constantly whereas *Labrus viridis* most likely would wait for the ideal prey.

Apart from the variablitiy in the prey type, a difference in feeding time also can be inferred when looking at activity periods of the species **(Figure 18)**. *Serrnus scriba* and *Labrus viridis*, which feed on crustaceans and fish, respectively, are active throughout the day whereas Scorpaena porcus, which feeds on crustaceans is primarily nocturnal.

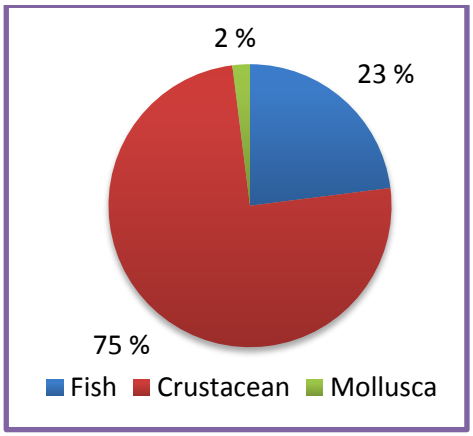

Figure 15: Percentage gut content of Scorpaena porca

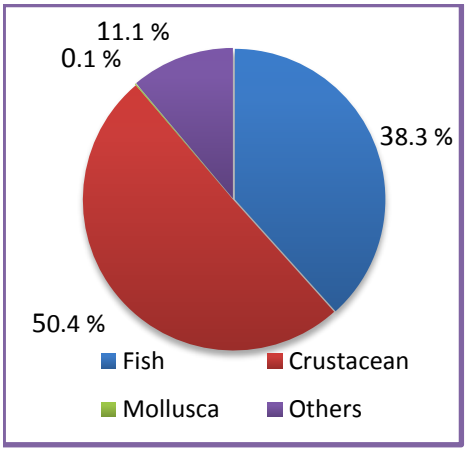

Figure 16: Percentage gut content of Serranus scriba.

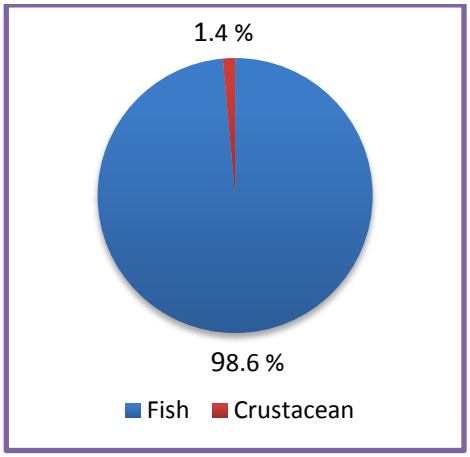

Figure 17: Percentage gut content of Labrus viridis.

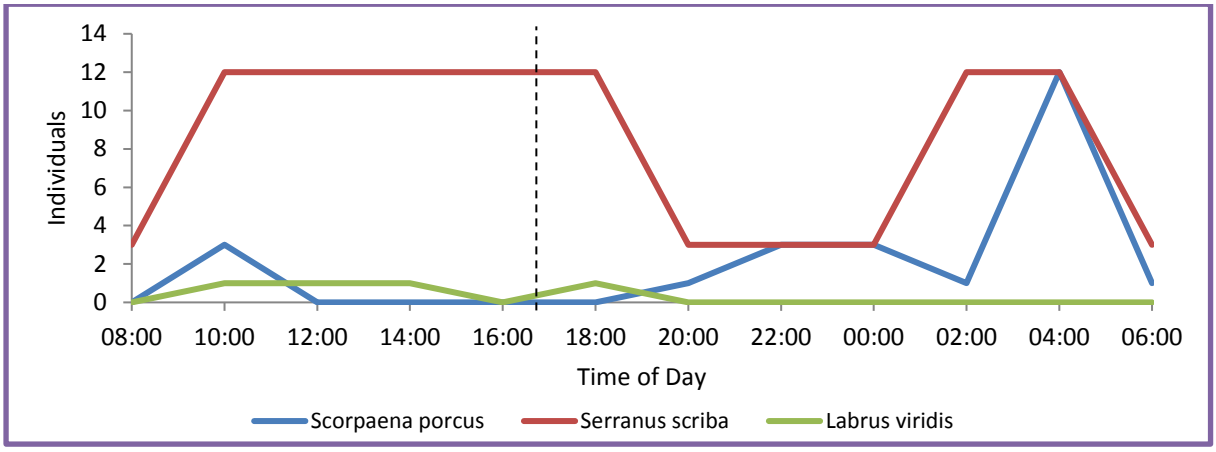

Figure 18: Activity Patterns of Scorpaena porcus, Serranus scriba and Labrus viridis.

Discussion

This study shows that species richness and abundance clearly vary with substrate types. Spatial variation in fish assemblages is influenced by factors like competition, predation, recruitment and composition of benthos (Letourneur *et al.,* 2003) as well as abiotic factors like habitat complexity (Gratwicke & Speight, 2005). Nonetheless some species were revealed to be generalists as they were sighted over all substrates.

The highest species richness and abundance was found over seagrass and over the rocky shoreline. Similar patterns were observed by Giakoumi & Kokkoris (2012). They studied fish communities in relation to habitat complexity in the North-eastern Mediterranean (Cyclades Archipelageo) and could show that substrate types largely determine the fish communities. Habitats with high heterogeneity, for example hard bottom with seagrass patches, have high species richness and abundances. In contrast, sandy areas showed lower abundances and species numbers due to lower habitat complexity. These habitats differ also in their community concerning their exclusive species.

Overall, the stony beach was more complex in its habitat structure and depth with smaller sand areas, which could be the reason more fish was observed.

The 24-hour transect line was also used to determine species distribution with regard to water depth and substrate type. The majority of the species was found in sections 2 and 4 of the transect. This was where there was intermediate to deep-water depth and the substrate composed of larger boulders and rocks. A deeper water column allows for swarming fish such as *C. Chromis* or *A. boyeri* to have access to pelagic plankton required for nutrition, the large boulders or rocks form holes and crevices which are ideal hiding/resting places for various fish. This unique habitat is possibly the key aspect, explaining the highest species number in sections 2 and 4.

Only little is known about the food preferences of *Serranus scriba* (Zolezzi, 1939; Arculeo et al. 1993) and *Scorpaena porcus* (Harmelin-Vivien et al. 1989). Our study confirms the importance of fish and decapod crustaceans in both fish diets. We can presume that both species fall into the feeding category of macrophagic carnivores as defined by Bell and Harmelin-Vivien (1983) and that both predators arranged themselves to feed on nearly the same species by their activities during day for *S.*

scriba and night for *S. porcus.* According to our study, *Labrus viridis* seem to feed mainly on fish (98.6%), which was inconsistent with Harmelin-Vivien et al. (1983) where *L. viridis* consumed only 43.2 % of fish. The rest consisted of crustaceans and polychaetes, which couldn´t be observed in our study, even though some gut content components showed high digestive states and could not be unambiguously assigned to distinct prey items, a crustacean or polychaete origin seemed unlikely due to the absence of exoskeleton or chetae remains.

The 24-hour transect shows that the structure of the fish community around the STARESO research facility varies according to daily cycles. Within a day (24-hour period) three recognisable communities (crepuscular, diurnal and nocturnal) are noticeable. Hobson (1974) describes that fish activity patterns change throughout a day to best suit and reduce competitions in feeding activities. The change in diel and nocturnal communities can be best described by the complete disappearance of the wrasses (Labridae, **Figure 5**), inactivity of the sea breams (Sparidae, **Figure 3**), the dissolution of swarms (**Figure 7**) of *Chromis chromis*, *Atherina boyeri* and *Atherina hepsetus*, and the appearance of nocturnal (**Figure 7**) carnivorous fish such as *Scorpaena notada* , *Scorpaena porcus* and *Apogon imperbis.*

The crepescular community was slightly more difficult to observe and were represented mainly by *Sciena umbra* and *Muraena helena* (**Figure 7**), which were observed mainly at early morning and late evening hours. Although larger abundances of *Serranus scriba* were observed at early hours of the day, the species was active throughout the day, suggesting passive crepescular behaviour. Possible reasons that no clearer image of crepescular communities were observed could be due to the time interval between conducting the survey, i.e. at 06:00 the sun was just coming up and at 08:00 it was already up and a similar scenario played out during sunset. The 2 hour gap between the survey times could have been enough to miss the majority of the crepescular community.

It needs to be considered that these data have to be viewed critically as a result of a lack of replication, i.e. only two replicates for the area transect and none for the 24hour transect. Another key aspect could be a bias in fish observations, even though training was carried out insecurities still existed with few species, this coupled with the lack of nocturnal fish identification-training could have resulted in misidentifying certain species. Similar problems with identification existed when analyzing gut content. The digestion stage varied between the fish and in some cases was so far on that identification was limited.

In this study, visual census of activity time was coupled with gut content analysis and with species distribution due to habitat type. The collective data provided convincing argument that fish communities are influenced by resource partitioning. Even though each method can be conducted independently, the combination of the various methods provides for more convincing data and consequently a more detailed insight into fish community structure.

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List of fish species observed throughout the course

Familie	Art	deut. Name	2014	2012	2010	2008	2006	2004	2002	2000	1998	1996
Torpedinidae Zitterrochen	*Torpedo marmorata*	Marmorzitterrochen	-	-	x	-	-	-	-	-	-	-
Dasyatidae Stachelrochen	*Dasyatis pastinaca*	Gewöhnlicher Stachelrochen	x	x	x	x	x	x	-	-	-	-
	Dasyatis violacea	Pelagischer Stechrochen	-	-	-	-	x	-	-	-	-	-
Myliobatidae Adlerrochen	*Myliobatis aquila*	Adlerrochen	-	-	-	-	x	x	x	-	-	-
Congridae Meeraale, Conger	*Conger conger*	Meeraal	x	-	-	x	-	x	-	x	x	-
Anguillidae Aale	*Anguilla anguilla*	Aal	-	-	-	-	x	x	-	x	x	-
Murenidae Muränen	*Muraena helena*	Mittelmeermuräne	x	x	x	x	x	x	x	x	x	-
Engraulidae Sardellen	*Engraulis encrasiculus*	Sardelle	x	-	-	-	x	-	-	-	-	-
Gadidae Dorsche	*Phycis phycis*	Gabeldorsch	x	x	-	-	x	-	x	-	-	-
Belonidae Hornhechte	*Belone belone*	Europäischer Hornhecht	x	x	x	x	x	-	x	-	-	-
Atherinidae Ährenfische	*Atherina boyeri*	Kleiner Ährenfisch	x	x	x	x	x	x	x	x	x	-
	Atherina hepsetus	Großer Ährenfisch	x	x	x	x	x	x	-	x	-	-
Mugilidae Meeräsche	*Liza aurata*	Goldmeeräsche	x	x	x	x	x	x	x	x	x	-
	Mugil cephalus	Kurzkopf-Meeräsche	-	-	-	-	x	x	-	-	-	-
	Oedalechios labeo	Kastenmaul-Meeräsche	x	x	x	x	x	x	x	-	-	-
Syngnathidae Seenadeln	*Hippocampus guttulatus*	Langschnäuziges Seepferdchen	-	-	-	-	-	-	-	-	x	-
	Syngnathus typhle	Grasnadel	-	-	-	-	-	-	x	-	-	-
Scorpaenidae Skorpionsfische	*Scorpaena notata*	Roter Drachenkopf	x	-	x	x	x	x	x	x	x	-
	Scorpaena porcus	Kleiner Drachenkopf/Brauner Drachenkopf	x	x	x	x	x	x	x	x	-	-
	Scorpaena scrofa	Großer Drachenkopf/Meersau	x	-	x	x	x	-	x	-	x	-
Triglidae Knurrhähne	*Chelidonichthys sp.*	Knurrhahn	-	-	-	-	-	x	-	-	-	-
Serranidae Zackenbarsche	*Anthias anthias*	Roter Fahnenbarsch	-	x	-	-	-	-	x	-	-	-
	Epinephelus marginatus	Brauner Zackenbarsch	x	x	x	x	x	x	x	x	x	-
	Serranus cabrilla	Sägebarsch	x	x	x	x	x	x	x	x	x	-
	Serranus scriba	Schriftbarsch	x	x	x	x	x	x	x	x	x	-
Dicentrarchidae Wolfsbarsche	*Dicentrarchus labrax*	Wolfsbarsch	-	x	x	-	-	x	-	-	-	-
Apogonidae Kardinalbarsche	*Apogon imberbis*	Meerbarbenkönig	x	x	x	x	x	x	x	x	x	-
Carangidae Stachelmakrelen	*Seriola carpenteri*	Mittelmeer-Bernsteinmakrele	-	x	x	x	x	-	-	-	-	-
	Seriola dumerili	Große Bernsteinmakrele	x	-	-	-	x	x	-	-	-	-
	Trachinotus ovatus	Ostatlantische Gabelmakrele	-	-	-	-	x	-	-	-	-	-
	Trachurus mediterraneus	Mittelmeerstöcker	x	-	-	-	-	x	-	-	-	-
Centracanthidae Pikarellen	*Spicara maena*	Gefleckte Pikarelle	-	x	-	x	x	-	x	x	-	-
	Spicara smaris	Schnauzen-Pikarelle	x	-	x	-	-	-	-	-	-	-
Sparidae Brassen	*Boops boops*	Gelbstrieme	x	x	x	x	x	x	x	-	-	-
	Dentex dentex	Zahnbrasse	x	x	x	x	x	x	x	-	x	-
	Diplodus annularis	Ringelbrasse	x	x	x	x	x	x	x	x	x	x
	Diplodus puntazzo	Spitzbrasse	x	x	x	x	x	x	x	x	x	x
	Diplodus sargus sargus	Geißbrasse	x	x	x	x	x	x	x	x	x	x
	Diplodus vulgaris	Zweibindenbrasse	x	x	x	x	x	x	x	x	x	x
	Lithognathus mormyrus	Marmorbrasse	x	x	x	x	x	x	x	-	x	-
	Oblada melanura	Bandbrasse	x	x	x	x	x	x	x	x	x	x
	Pagellus acarne	Achselfleckbrasse	x	x	-	x	-	x	x	-	-	-
	Pagellus erythrinus	Rotbrasse	x	-	-	x	-	-	-	x	x	-
	Pagrus pagrus	Sackbrasse	-	-	-	-	-	-	x	-	-	-
	Sarpa salpa	Goldstrieme	x	x	x	x	x	x	x	x	x	x
	Sparus aurata	Goldbrasse	x	x	x	x	x	x	x	x	x	x
	Spondyliosoma cantharus	Streifenbrasse	x	x	x	x	x	x	x	x	-	x
Haemulidae Grunzer	*Pomadasys incisus*	Bastard-Grunzer	-	-	x	-	-	-	-	-	-	-
Sciaenidae Umberfische	*Sciaena umbra*	Meerrabe	x	x	x	-	x	x	x	-	-	-
Mullidae Meerbarben	*Mullus barbatus*	Rote Meerbarbe	-	-	-	-	x	x	-	-	-	-
	Mullus surmuletus	Streifenbarbe	x	x	x	x	x	x	x	x	x	-
Pomacentridae Riffbarsche	*Chromis chromis*	Mönchsfisch	x	x	x	x	x	x	x	x	x	-
Labridae Lippfische	*Coris julis*	Meerjunker	x	x	x	x	x	x	x	x	x	x
	Labrus merula	Amsellippfisch	x	x	x	x	x	x	x	x	x	x
	Labrus viridis	Grüner Lippfisch	x	x	x	x	x	x	x	-	x	x

	Symphodus cinereus	Grauer Lippfsich	x	x	x	x	x	x	x	-	x	x
	Symphodus doderleini	Doderleins Lippfisch	-	-	-	-	-	x	-	-	-	-
	Symphodus mediterraneus	Mittelmeer-Lippfisch	x	x	x	x	x	x	x	x	-	-
	Symphodus melanocercus	Schwarzschwanz-Lippfisch	x	x	x	x	x	x	x	x	x	x
	Symphodus ocellatus	Augenlippfisch	x	x	x	x	x	x	x	x	x	x
	Symphodus roissali	Fünffleckiger Lippfisch	x	x	x	x	x	x	x	x	x	x
	Symphodus rostratus	Schnauzenlippfisch	x	x	x	x	x	x	x	x	x	x
	Symphodus tinca	Pfauen-Lippfisch	x	x	x	x	x	x	x	x	x	x
	Thalassoma pavo	Meerpfau	x	x	x	x	x	x	x	x	x	-
	Xyrichthys novacula	Mittelmeer-Schermesserfisch	x									
Trachinidae Petermännchen	*Trachinus araneus*	Spinnenqueise	x	-	-	-	-	-	-	-	x	-
	Trachinus draco	Gewöhnliches Petermännchen	x	x	x	x	x	-	-	-	-	-
	Trachinus radiatus	Strahlen-Petermännchen	x	-	-	-	-	-	-	-	-	-
Uranoscopidae Himmelsgucker	*Uranoscopus scaber*	Himmelsgucker	-	-	-	x	x	-	-	-	-	-
Tripterygiidae Dreiflosser	*Tripterygion tripteronotus*	Roter Spitzkopf-Schleimfisch	x	x	x	x	x	x	x	-	x	-
Blenniidae Schleimfische	*Adiablennius sphynx*	Sphinx-Schleimfisch	x	x	x	x	x	-	x	-	x	-
	Coryphoblennius galerita	Amphibischer Schleimfisch	x	-	x	x	-	-	-	-	-	-
	Lipophrys fluviatilis	Flußschleimfisch	x	x	x	x	x	x	-	-	-	-
	Lipophrys nigriceps	Schwarzkopfschleimfisch	x	-	x	x	x	x	-	-	-	-
	Lipophrys trigloides	Grauer Schleimfisch	-	-	-	-	x	x	-	-	-	-
	Parablennius gattorugine	Gestreifter Schleimfisch	x	x	x	x	x	x	x	-	x	-
	Parablennius pilicornis	Gelber Schleimfisch	-	-	x	x	-	-	-	-	-	-
	Parablennius rouxi	Langstriemenschleimfisch	x	x	x	x	-	x	x	-	x	-
	Parablennius sanguinolentus	Blutstriemen Schleimfisch	x	x	x	x	x	x	x	-	x	-
	Parablennius zvonimiri	Hirsch-Schleimfisch	x	x	x	x	-	x	x	-	x	-
	Salaria pavo	Pfauenschleimfisch	-	-	-	-	-	x	x	-	x	-
Clinidae Klippfische	*Clinitrachus argentatus*	Silbriger Schleimfisch	-	-	-	-	-	x	-	-	-	-
Gobiidae Grundeln	*Gobius bucchichi*	Anemonengrundel	-	-	-	-	x	x	x	-	x	-
	Gobius cobitis	Riesengrundel	x	-	-	x	x	x	x	-	-	-
	Gobius geniporus	Schlankgrundel	-	-	-	-	-	x	-	-	-	-
	Gobius paganellus	Felsengrundel	-	-	-	-	x	-	-	-	-	-
	Pomatoschistus minutus	Sandgrundel	-	-	x	x	x	-	-	-	-	-
Gobiesocidae Schildfische	*Diplecogaster bimaculata*	Zweifleck-Schildbauch	x	-	x	-	-	-	-	-	-	-
	Gouania wildenowi	Stumpfschnäuziger Schildbauch	-	-	x	-	-	-	-	-	-	-
	Lepadogaster candolii	Rotflecken Ansauger/Rotfleck-Schildbauch	-	-	-	x	x	x	x	-	x	-
	Lepadogaster lepadogaster	Blaufleckiger Ansauger	-	-	x	-	-	-	-	-	-	-
	Opeatogenys gracilis	Seegras-Schildbauch	-	x	-	-	x	x	-	-	-	-
Callionymidae Leierfische	*Callionymus pusillus*	Kleiner Leierfisch	-	x	x	x	-	-	-	-	-	-
Sphyraenidae Barrakudas	*Sphyraena viridensis*	Mittelmeer-Barrakuda	x	x	x	x	x	x	-	-	-	-
Bothidae Butte	*Arnoglossus kessleri*	Kessler-Butt	-	-	-		-	-	-	-	x	-
	Bothus podas	Breitaugenbutt	x	x	-	x	x	-	-	-	-	-
	Zeugopterus regius	Zwergbutt	-	-	x	x	-	x	x	-	-	-
Soleidae Seezungen	*Solea lascaris*	Warzen-Seezunge	-	-	-	x	x	-	-	-	-	-
Balistidae Drückerfische	*Ballistes carolinensis*	Mittelmeer-Drückerfisch	-	-	-	x	-	-	-	-	x	-
		99	64	55	63	64	68	65	55	33	46	17
		Gesamt	2014	2012	2010	2008	2006	2004	2002	2000	1998	1996