

The Effect of Substratum Surface on the Biodiversity of Marine Fouling Community

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ABSTRACT

Fouling is when unwanted materials developed on a solid surface and flourish. In this experiment, two sets of ceramic plates with a rough and smooth surface on each side were deployed in a fish farm located in the South China Sea for three months in different periods of the year at different depth. The biofouling communities developed on the surfaces of ceramic plates were counted and the alpha and beta diversity were calculated. A total of 18 biofouling species and species associated with the biofouling community was observed. The results suggested that surface smoothness is a key factor shaping the biofouling community structure. In additional, biofouling communities developed at different seasons also showed remarkable differences. Overall, my finding suggested that biofouling is an important ecological event and is an often-ignored ecosystem with an unexpectedly vivid biodiversity.

Introduction

Fouling is caused by the accumulation of non-living deposits and the attachment and growth of marine sessile benthic organisms. It can be classified into two broad categories: microfouling and macrofouling. Macrofouling is caused by matter of either inorganic or biological origin, such as animals and plants, which are also referring to marine biofouling. Biofouling organisms settle on any submerged surfaces and grow quickly, forming a layer that covers the surface, attracting and trapping more biofouling organisms to settle on top of the existing biofouling organisms. Biofouling species are generally fast-growing, short-lived, and tightly tied to the environment. But in general, biofouling organisms quickly flourish, aggregate, and eventually form a community with physical 3D structures, in which a variety of organisms from different taxa living inside. On the other hand, biofouling organisms such as bryozoans, sponge, bivalves and polychaetes are mostly filter feeders that may also help to improve the water's clarity and provide nursery habitat for marine invertebrates at different trophic level, such as herbivorous snails, deposit feeding sea urchin and polychaetes and predatory fish and crustaceans. Hence, it is important to understand that biofouling community is also an important ecological system in the marine environment.

Previous studies suggested the extent and severity of fouling are dependent on variables such as temperature, rainfall, pollution, nutrient availability, water salinity, and fouling materials. In this study, I studied if the surface feature is an important factor in determining the fouling species composition. I studied the fouling community developed on a ceramic brick plate, with a smooth ceramic side and a rough side. I also studied if differences in season and depth can affect the fouling species composition.

Methods and Materials

Development of the fouling community

The sample of fouling was collected from a fish farm located in the DaPeng Peninsula, the South China Sea, (GPS: 22.564800, 114.535782); two ceramic brick plates of the same size (10cm x 20cm) were placed in the fishfarm by hanging the plate at about 1 meter below sea level at 3rd of July and were acquired three months later during the China National day Golden Week holiday in the 3rd of October. Subsequently, a set of nine ceramic brick plates were placed in the fishfarm by hanging the plate at 1 meter, 2 meters and 3 meters below sea level at the 3rd of October and retrieved at the 23th of December (Xmas).

Photographing of fouled plates and animal specimens

The brick was taken out of its container and placed under a white background with a scale right beside it. A camera was then fixed 15 cm away from the brick plate to have a top-down view. The rough and smooth sides of each fouled plate was photographed, with labels to illustrate the replicate number, surface type (rough or smooth) and the depth of each plate (see Figure 1 & 2). After the picture was taken, each of the bricks was placed into a new container with fresh seawater taken from the fish farm inside. Half of the yellowish seawater was then poured out of the old container into the sink. The organism specimens that had fallen off from the two bricks were carefully transferred into separate Petri dishes by using a pair of tweezers. Each specimen was then placed into a smaller petri dish to take pictures with a scale beside the animal specimen using a camera mounted on a microscope. This step was then repeated for the other animal specimens that had fallen of the other piece of brick. The numbers of specimens of each species were then counted and recorded with the species name and type.

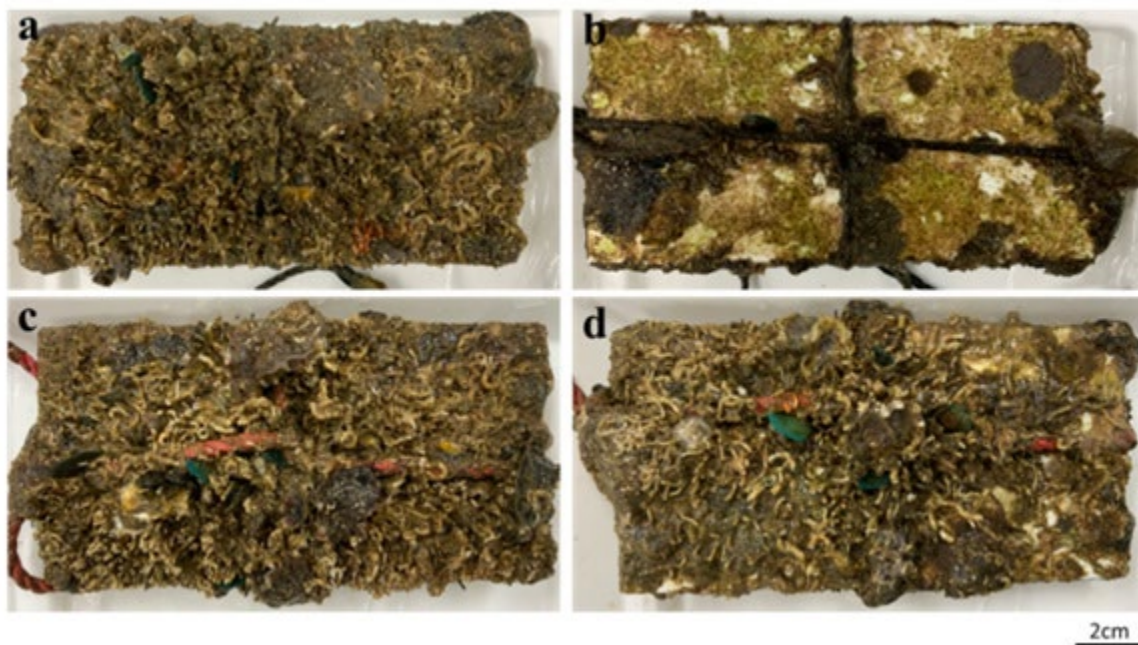


Figure 1.

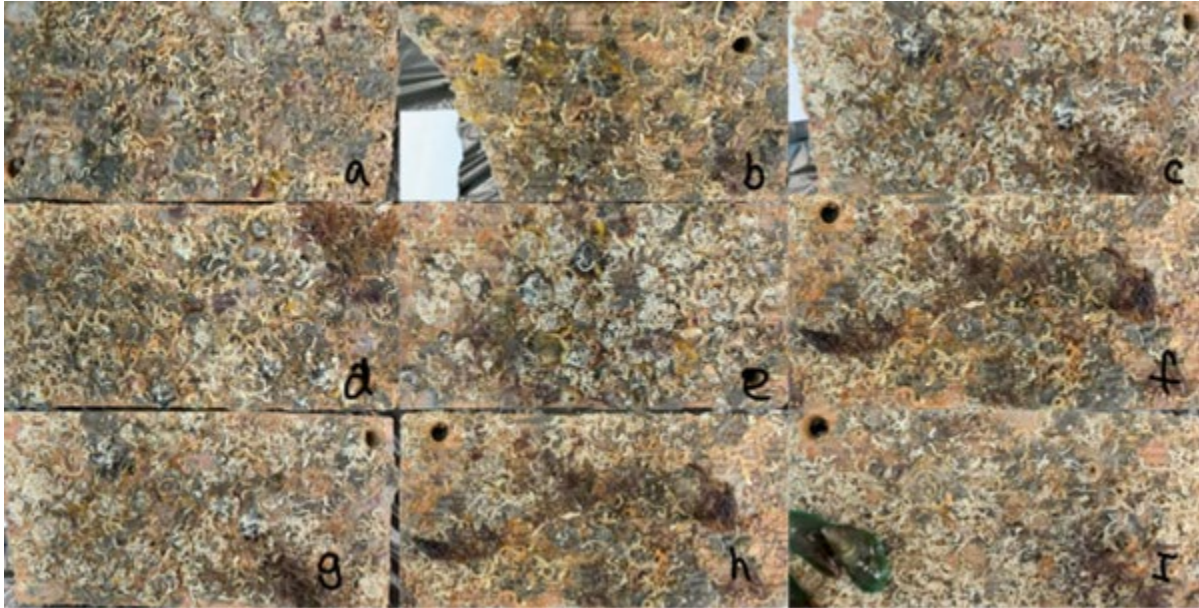


Figure 2.

Results

Fouling in different seasons and the fouling community biodiversity

Figure 1 shows two fouling plates replicates developed during the summer, from 2nd of July to 3rd of October 2022 at 1m depth (Fig. 1). Figure 2 shows nine fouled plates developed during the autumn, from 3rd of October to 24th of December 2022. The plates were placed in 3 depths: 1m, 2m and 3m, each depth with three replicates. Fouling on the smooth and rough side of the ceramic plates was documented (Fig. 2 & 3).



Figure 3.

The diversity of sessile organisms

The fouling animals found during the two experiment periods include green mussels (*Perna viridis*, Mollusca), five species of polychaetes (Annelida), one species of oyster (Mollusca), one species of clam (*Bivalvia*, Mollusca), colonial ascidians, three species of bryozoan (Bryozoa), one species of sponge (Porifera), and one species of barnacle (Cirripedia, Arthropoda). The photos record of these sessile fouling organisms is shown in Figure 4 and their characteristics features are described in the following.

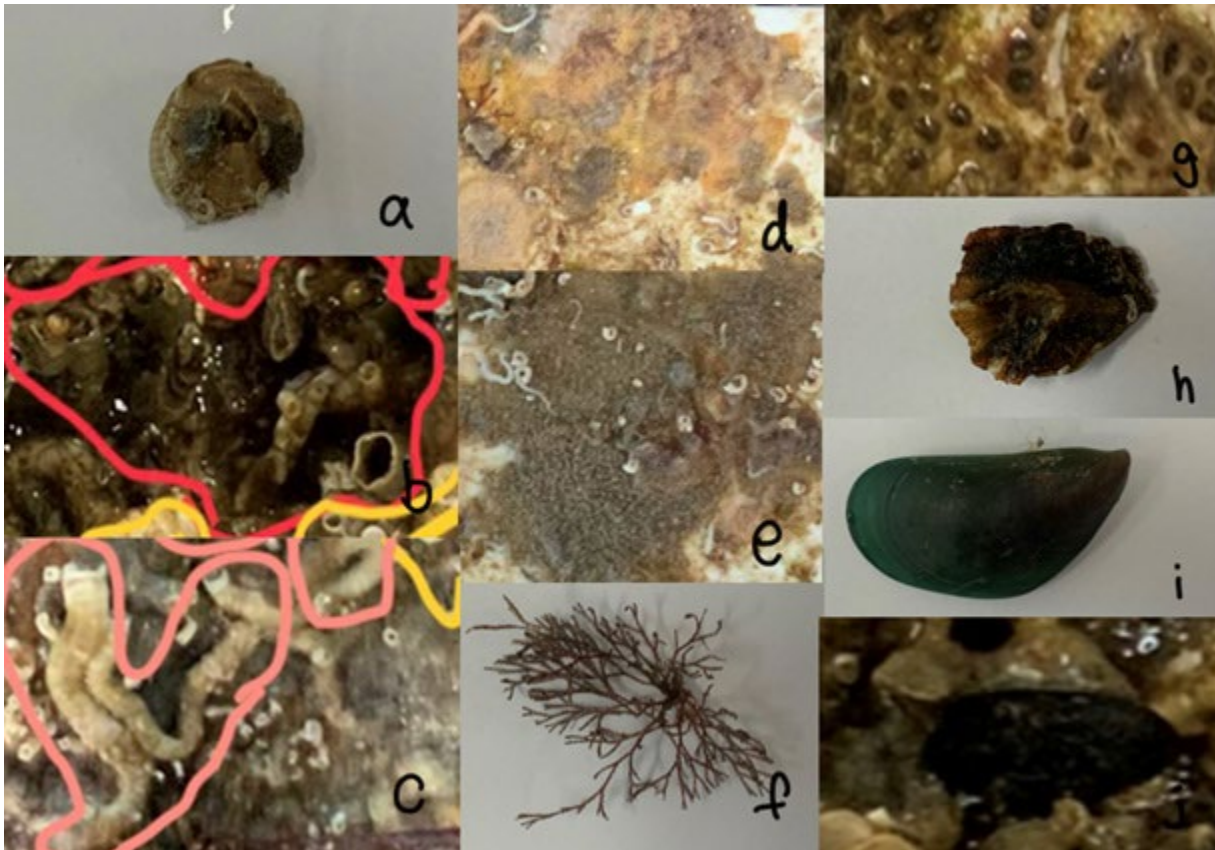


Figure 4.

Arthropoda:: Crustacea::Thecostraca (Barnacles) (Fig. 4a)

Barnacles are sessile animals; they have no need for the walking legs that many of their crustacean relatives possess. Their legs have adapted over time to a different use: to sweep tiny food particles from the water column and pass them to their mouth parts inside their protective plates. There are more than 1,400 species of barnacles found in the world's waterways [10]. The specimen observed in the fouling plate was an acorn barnacle. The shell was in general resembling the shape of a volcano and was white in color with multiple vertical stripes on each shell plate.

Annelida::Polychaete (Phylum::class) (Fig. 4b, 4c, 5e)

Polychaete sp. 3, which the tube was shown in Fig. 4b and the body was shown in Fig. 5e, has multiple thread like tubular structure extending from the head part of the tubeworm. They have a flower-like head. The tubes are yellowish white and have colored bands for filter feeding and gas exchange. We suspect it could be *Polydora cornuta* which is a complex of species of tube-building estuarine and marine polychaetes. It occurs both in fouling communities and on muddy sea floors but is unable to bore into calcareous substrata and eat things like phytoplankton, benthic microalgae, and detritus. Adults of the *Polydora cornuta* species complex have up to 90 segments, can reach a length of 12-32 mm, and are light tan in color [7]. Most specimens have four eyes in a trapezoidal arrangement on the

prostomium, with a small, triangular occipital tentacle located posteriorly to them. A ciliated groove runs on either side of the caruncle. The prostomium is flanked by a pair of long, prehensile palps, which stretch back to chaetigers 15–35. The palps each have a ciliated groove along their lengths, bordered by a fine black line but lacking conspicuous pigment (Fig. 5e).

Polychaete sp. 5, as shown in Fig. 4c, is a serpulid tube worm called *Hydroides elegans*. It secretes a calcareous tube on hard surfaces like rocks, coral, and mangroves, just like other serpulid worms [11]. It is a common fouling organism that settles on marine infrastructure and may compete with other fouling species in many warm-water harbors. In some areas of its introduced range, it interferes with oyster settlement and causes extensive mortality. Serpulids have a feathery crown of modified prostomial palps, called radioles. The radioactive tubes can be folded and withdrawn into the tube. One of the radioles is modified to form an operculum, which acts as a stopper when the animal contracts. *Hydroides elegans* has a white tube, about 1.3 to 2.5 mm in diameter. The tubes are brittle and inconsistent, occasionally possessing longitudinal and transverse ridges, but they are typically smooth and feed on phytoplankton.

Bryozoa:: Stenolaemata (Bryozoan) (Fig. 4d, e & f)

The phylum bryozoans are a diverse group of aquatic invertebrates with zooids. Although some species' colonies can measure more than 0.5 meters in diameter, individual zooids are often little longer than one millimetre. A lophophore is a strange ring of ciliated tentacles that bryozoans have similar to brachiopods and phoronids for gathering food particles suspended in the water. There are roughly 5,000 living bryozoan species classified into three classes of: the freshwater dwelling Phylactolaemata, the marine Stenolaemata, and the Gymnolaemata (mostly marine).

Three bryozoan species were observed during the experimental period. The first bryozoan species (sp. 1) (Figure 4d) is an encrusting species with greyish color colonies. The colonies could be found on both the smooth and rough side of the plate and flourish in the plate fouling community developed during summer and autumn. This species usually attaches to the tube of the polychaete tubeworm species *Hydroides elegans*. The second bryozoan species (sp. 2) (Figure 4e) is a species in the genus *Watersipora*, which is an encrusting species with colony zooids in yellowish-white color. The species was mainly found on the surface of smooth side of the brick plate. The final bryozoan species is *Bugula neritina*, a arborescent bryozoan (sp. 3) (Figure 4f) with purple color that grows mainly on the rough side of the brick.

Chordata :: Ascidiacea :: Chordata::colonial Ascidian (Tunicate) (Fig. 4g)

Sea squirt, also called ascidian, are members of the invertebrate class Ascidiacea (subphylum Urochordata, commonly known as Tunicata), marine creatures having some rudimentary vertebrate traits, is referred to as a sea squirt or ascidian. Sessile (permanently attached to a surface), potato-shaped creatures known as sea squirts may be found in all oceans, from the intertidal zone to the deepest parts. They frequently live on ship hulls, rocks, huge seashells, and the backs of enormous crabs. Some species live alone, while others exist in communities or colonies. The tunic, which serves as the body's outer protective layer, is made of a material that resembles cellulose. Sea squirts have two sizable pores: the mouth or branchial aperture, which directs water into the body cavity, and the exit pore. Pharyngeal cilia push water through the animal. The pharynx has gill openings that let water to move through while capturing food and oxygen from the water current. Despite the fact that all adults are simultaneous hermaphrodites (having both male and female reproductive organs that function), eggs shed into the water are fertilized by sperm from other individuals. The notochord, a flexible rod-like structure shared by all vertebrates, and the nerve cord are in the muscular tail. The larva uses a sucker on the front of its body to adhere to a suitable location for metamorphosis. Later, the tail vanishes along with its notochord and nerve cord. Most sea squirts are sessile as adults, but others can move by latching onto one part of the body and letting go of another. By budding, reproduction also takes place. A lobe or protrusion with a cluster or chain of zooids breaks off around the base of the adult sea squirt; the zooids move to a new location to form new individuals. While "complex" colonial ascidians develop a colony where the zooids are embedded in a shared tunic, colonial sea squirts are more autonomous. Several zooids may share a single, common cloacal aperture, but each zooid has its own branchial aperture [13].

Mollusca::Bivalvia

Subclass: Autobranchia ::Pteriomorphia (Oyster) (Fig. 4h)

Oyster, any bivalve mollusc found in temperate and warm coastal waters of all seas that belongs to the family Ostreidae. The oyster shell's two valves, which are different in shape, have rough surfaces that are frequently a filthy grey color. Convexity describes the upper valve as being higher in the centre than at the edges. The lower valve, which is bigger, flatter, and fastened to the bottom or another surface, has smoother edges. Both valves have white, smooth surfaces on the inside. An elastic ligament holds the valves together at their thin ends. The valve is closed by a sizable central muscle that resists the ligament's strain. The microscopic hair-like projections (cilia) on the gills suck water inside by wavelike movements while the valves are kept slightly open. In one hour, two to three litres could travel through the oyster. Food is provided by tiny organic particles that are filtered out of the water. Although hermaphroditism does exist, most oysters are either males or females, like other bivalves. *Ostrea edulis* demonstrates a condition known as sequential hermaphroditism, in which an individual switches sexes in response to seasonal or water temperature variations [14].

Subclass: Mytilida::Mussel (Fig. 4i)

Bivalve molluscs known as mussels that are members of the marine family Mytilidae and are widely distributed, frequent in chilly waters [15]. Marine mussels often are wedge- or pear-shaped shapes, between 5 and 15 centimetres in size. They may be smooth or ribbed and often have a hairy covering. Many types of shells have dark blue or dark greenish brown exteriors with pearly interiors. Byssus threads, which are protein-based threads used by mussels to attach themselves to solid objects or to one another, are frequently seen in dense clusters.

Unidentified Bivalve sp. (Fig. 4j)

Bivalve, class Bivalvia, have more than 15,000 species of clams, oysters, mussels, scallops, and other Mollusca that have a shell that is separated into left and right valves from the front to the rear are considered bivalve (class Bivalvia) [16]. At a hinge, the valves are attached to one another. Primitive bivalves consume sediment, but the respiratory gills of most species have evolved into filtering structures known as ctenidia. Bivalves have lost the head and the radular rasping apparatus typical of most molluscs, in line with a primarily sedentary and deposit-feeding or suspension-feeding lifestyle. The length of a vacuole can range from roughly one millimetre to more than 137 centimetres. Classification is based on hinge structure and shell shape. The majority of species that burrow on the surface have tiny, round or oval shells with equal left and right valves. The shells of animals that burrow deeper are laterally compressed, allowing for faster movement through the sediments. Surface-burrowing animals might develop a sculptured exterior shell with radial ribs, concentric lines, and projections to reinforce the shell against damage and predation. Some bivalves have been able to colonize hard surfaces on wave battered coasts because to their triangular morphology, ventral flattening, and stable attachment to firm substrates via byssal threads. Some bivalve groups maintain the byssus in their adult forms, which is a characteristic of the larval stage.

The diversity of the fouling associated community

We also recorded and counted the benthic organisms associated with the sessile fouling community developed during the summer. From the two replicate plates, we recorded the presence of one species of brittle stars (Echinoderm), one species of jellyfish (Cnidaria), one species of amphipods (Arthropoda), one species of crab (Arthropoda), one species of flatworm, one species of nudibranch (Mollusca) and one species of polychaete. The photos of these benthic species are shown in Figure 5 and their characteristic features are described in the following.

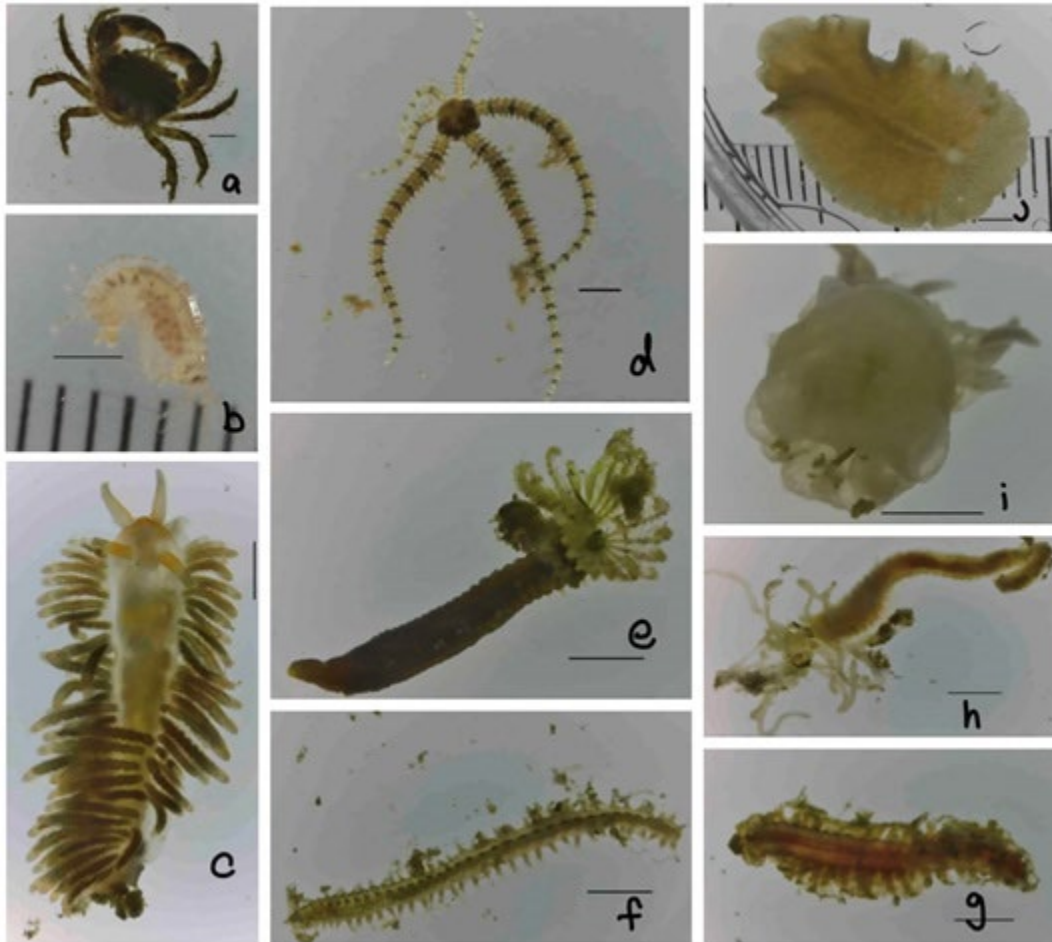


Figure 5.

Arthropoda :: Crustaceans :: Malacostraca::Decapodas (Crabs) (Fig.5a)

Crabs belongs to the crustacean family Decapoda (phylum Arthropoda). They are frequently omnivorous and scavengers, like many other crustaceans, but many are predatory, and some are vegetarian [1]. The crabs found were all about one centimetre in width, brownish in colour, with hairy legs and a pair of nearly identical clamps.

Arthropoda:: Crustacea:: Malacostraca:: Eumalacostraca::Amphipoda (Fig. 5b)

Any member of the invertebrate order Amphipoda (class Crustacea) inhabiting all parts of the sea, lakes, rivers etc. About 6,000 species have been described and are often mistaken for tiny shrimp [2]. They are important food for many fishes and invertebrates and important as scavengers of carrion. The amphipod species observed in this study is associated with the fouling community. Their body length varies from 1 to 140 mm and are active swimmers and strong jumpers [3]. The body colour is a yellowish-white colour, almost transparent (Figure 4b).

Mollusca:: Gastropoda ::Heterobranchia::Nudibranch (Fig. 5c)

There are more than 2,000 known species of nudibranch [4] Although they are present in all of the world's seas, shallow, tropical areas are where they are most common. They can reach lengths of up to 12 inches or as little as 0.25 inches [5]. They are carnivores, and the tops of their heads are home to two rhinophores, which are extremely sensitive tentacles used to detect prey. The food that nudibranchs eat gives them color, which aids in camouflage. Some even store the repulsive toxins of their prey and secrete them to ward off predators. The nudibranch species observed in this study has yellowish-green colour, with two rhinophores on the top of the head, and two lateral tentacle-like structures running along the body.

Echinodermata:: Ophiuroidea (Brittle star) (Fig. 5d)

Brittle stars have 2,100 known species. Their long, thin arms are usually five in number and spiny; they regularly break off and are regenerated. Brittle stars typically hide under rocks or in crevices during the day and emerge at night to feed [6]. The brittle star species observed in this study is yellow in color with brown strips. The specimen shown in the figure (Fig. 5d) shown has three new grown arm which is likely that it just broken off and started to generate.

Annelida:: Polychaete (Phylum::class)

There were four observed polychaete species.

Unidentified polychaete sp. 2 (Fig. 5e)

Polychaete species 2 (sp. 2) are tubeworm that was identified by their mud tubes built on plate surfaces and has a flower-like head. The tubes are yellowish white and have colored bangs for filter feeding and gas exchange. We suspect the species could be *Polydora cornuta* which reportedly occurs in both fouling communities and on muddy sea floors but is unable to bore into calcareous substrata and eat things like phytoplankton, benthic microalgae, and detritus [7].

Unidentified polychaete sp. 1 (Fig. 5f)

Polychaete species 1 (sp. 1) has a toothlike structure, which means that they are carnivorous. It has triangular-shaped parapodia.

Unidentified polychaete sp. 4 (Fig. 5g)

Polychaete species 4 (sp. 4) cannot be identified since the head is injured and nowhere to be seen (figure 4g).

Unidentified polychaete sp. 3 (Fig. 5h)

Polychaete species 3 (sp. 3) has tentacles on its head segment and I suspect it is a deposit feeder (likes to eat mud) (Figure 4h).

Cnidaria:: Scyphozoa (Jellyfish) (Fig. 5i)

Jellyfish are members of the class Scyphozoa (phylum Cnidaria), a group of invertebrate animals composed of about 200 described species [8]. Scyphozoan jellyfish can be divided into two types: those that are free-swimming medusae and those that are sessile (those that are attached to seaweed and other objects by a stalk). The jellyfish specimens observed in this study are yellowish white in color and have bell-shaped bodies with small round legs.

Platyhelminthes (Flatworm) (Fig. 5j)

Flatworm, also called platyhelminth, are free-living and about 80 percent of all flatworms are parasites living on or in another organism and securing nourishment from it. They are bilaterally symmetrical and lack specialized respiratory, skeletal, and circulatory systems. They have a head region that contains concentrated sense organs and nervous tissue (the brain) [9]. The flatworm specimens observed in this study is of a yellowish-brown color, and is flat, with an oval-shaped body.

Overall pattern of fouling species abundance

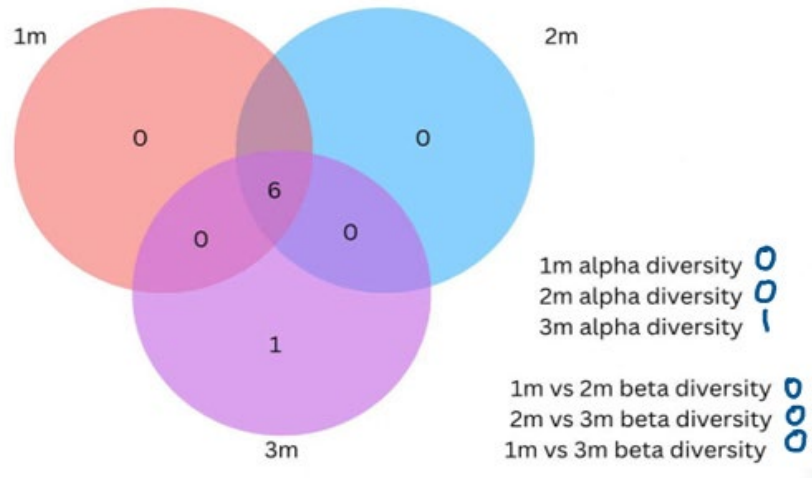
Fouling species on the smooth side of replicate plate 1 include 8 oysters, 4 green mussels, 5 barnacles, 1 jellyfish, and polychaete were the fouling species immersed to the surface on the smooth side of the bricks. Fouling species on the smooth side of the replicate plate 2 include 1 oyster, 1 green mussel, 5 colonial ascidians, 1 encrusting bryozoan, 1 barnacle, and 5 polychaete. Fouling species on the rough side of the red string include 7 oysters, 5 green mussels, 4 bivalves, and two species of polychaetes (sp. 3 and 5). Fouling species on the rough side of the black string include 3 oysters, 3 green mussels, 1 bivalve, 2 sponges, and two species of polychaetes (sp. 3 and 5).

Comparison between the rough and smooth sides (1 meter), from summer to golden week

From summer to golden week, the bricks were placed 1 meter below sea level. Oysters, green mussels, colonial ascidians, encrusting bryozoans, barnacles, jellyfish, and Polychaete were the fouling species immersed to the surface

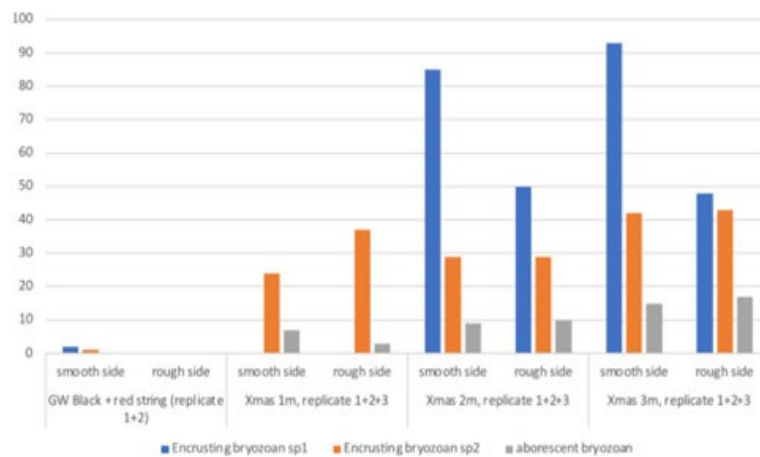
Comparison between the fouling communities developed on plates hanged in 1-, 2-, and 3-meter depth from October to December (Xmas)

Oysters, encrusting bryozoan sp1, sp2, sp3, barnacles and polychaete sp2 and sp5 was commonly found on all three depths during Xmas. Green mussels were only found 3 meters under sea level.



Change in abundance in bryozoans, the major group of sessile fouling organisms

Bryozoans sp. 1 appeared on the smooth side of the brick during from summer to golden week at 1 meter below sea level, but it did not appear at 1 meter below sea level from golden week to Xmas, it only appeared at 2 and 3 meters below sea level. Another point that ought not to be overlooked is that it is most abundant on the smooth side of the bricks as shown on the graph. Bryozoans sp. 2 only appeared on the smooth side during summer to golden week but appeared on each side and on all three meters below sea level for all the trail during golden week to Xmas. Bryozoans sp. 3 did not appear from summer to golden week, however, it appeared on both sides.



Discussion

Oysters, green mussels, colonial ascidians, encrusting bryozoans, barnacles, jellyfish, and Polychaete were the fouling species immersed to the surface on the smooth side of the bricks. Oysters, green mussels, bivalve, polychaete, colonial ascidians, and sponges were the fouling species immersed onto the surface of the rough side of the brick. The results show that bivalves and sponges are only immersed on the rough side of the brick, while barnacles and encrusting bryozoans are unique to the smooth side of the brick. Most bivalves cultivate on ropes, which have a surface comparable to the rough brick sides that are wave-shaped like ropes. Sponges normally live on the uneven sea floor or on ropes, as seen in the fish farm, which is similar to the rough side of the brick. Barnacles and encrusting bryozoan reside on sturdy objects like rocks, pilings, or boats, which are similar to the smooth side of the bricks, which implies the reason why some fouling only cultivates on the smooth or rough side of the brick. This result indicates that the coating or paint does not act as an antifouling material on the smooth side of the brick. Some possible reasons for the differences and variations observed in the fouling communities are seasonal variations in when the bricks are placed in the South China Sea. Since climate is an important factor in the survival of species, this is since the temperature of the sea varies in different time periods, which is why some species may not be able to sustain that environment or the food source for that species may not be available. Data shows that encrusting bryozoan sp1 did not appear at all from golden week (October) to Xmas (December) at 1 meter below sea level, for either for the former reasons given above. Similar observations have shown in the species arborescent bryozoan, from summer to golden week (October), arborescent bryozoan was not found on either side of the plates but appeared on all of the bricks from golden week (October) to Xmas (December). This suggests that some species will not survive and emerge because of climate change. As for encrusting bryozoan sp1 and sp2 they both appeared on the smooth side of the plates from summer to golden week (October), however, it did not appear on the rough side. Even if they appear on the smooth side, the number of encrusting bryozoans on the bricks is not abundant compared with those found on the bricks from golden week (October) to Xmas (December). This shows that the climate for bryozoan from summer to October is less suitable for them than that of October to December.

Polychaete is more abundant at 3 meters below sea level from golden week (October) to Xmas (December) even though polychaete can still be found at the depth of 1 and 2 meters below sea level, however polychaete is most abundant from summer to golden week (October), data shows that six species of polychaete are found during that time on the tow brick placed at 1 meter below sea level. Contrary to that, only two species were found from golden week (October) to Xmas (December).

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