

Matilde Costa

**BENTHIC COMMUNITIES IN SHIPWRECKS ALONG THE
PORTUGUESE CONTINENTAL COAST**



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**BENTHIC COMMUNITIES IN SHIPWRECKS ALONG THE
PORTUGUESE CONTINENTAL COAST**

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Trabalho efectuado sob a orientação de:

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2016

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(Matilde Costa Correia)

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*maggie and milly and molly and may
went down to the beach (to play one day)*

*and maggie discovered a shell that sang
so sweetly she couldn't remember her troubles, and*

*milly befriended a stranded star
whose rays five languid fingers were;*

*and molly was chased by a horrible thing
which raced sideways while blowing bubbles: and*

*may came home with a smooth round stone
as small as a world and as large as alone.*

*For whatever we lose (like a you or a me)
it's always ourselves we find in the sea.*

E. E. Cummings

Abstract

Shipwrecks are the most conspicuous type of artificial reefs. Besides their impact on local biodiversity, they have a major impact on the diving industry, being some of the preferred diving sites worldwide. Artificial reefs have been used for a long time with various purposes across different regions. There are multiple studies about their effect on fish and benthic communities, but there is little research focusing on shipwrecks specifically. This thesis is the first survey of shipwrecks on the Portuguese coastline, comparing their benthic communities with adjacent natural reef communities, and analysing the spatial distribution patterns within each shipwreck. Four shipwrecks and four nearby natural reefs between Sesimbra and Tavira were sampled with video-transects. The data were divided into two datasets: one dataset included densities of organisms that could be individually counted and the other included percent cover of morphological groups that could not be individually assessed (e.g., algae and incrusting fauna). The density analysis revealed similar communities between shipwrecks along the coastline despite their different ages. Surprisingly, in most cases shipwrecks had different communities from natural rocky reefs, with 40 to 70% of biodiversity not coming from the local reefs. Also unexpectedly, there were almost no differences between vertical and horizontal surfaces or between the exposed and protected sides of the wrecks, but intact (vs fragmented) parts of the shipwrecks harbour more biodiversity. Even though shipwreck age is likely to influence the observed colonization stage, it is hypothesized that a maximum of biodiversity is quickly reached, and thereafter a dynamic equilibrium is established, with the number of species remaining approximately constant, despite species compositional changes over time. Age also reduces the differences between artificial and natural reefs, making them more similar in terms of biodiversity. Overall, if remaining structurally in a good condition, shipwrecks can increase the local macrobenthic biodiversity.

Key-words: Artificial reefs; natural reefs; macrobenthic biodiversity; video-transects

Resumo

Os naufrágios representam provavelmente o tipo de recife artificial mais conspícuo. Além do impacto nos ecossistemas e biodiversidade locais, constituem sítios de mergulho muito populares por todo o mundo. O impacto que naufrágios podem ter na indústria do mergulho é imenso. No geral, recifes artificiais são estruturas feitas pelo Homem que podem assumir várias formas, desde barcos a cubos de betão feitos propositadamente para serem afundados no mar. Em diferentes áreas, recifes artificiais têm funções diferentes, entre as quais gestão das pescas, prevenir a pesca de arrasto, compensação de perda de habitat e proteger habitats costeiros. Os recifes artificiais podem ter impactos positivos ou negativos nos ecossistemas. Consequências entre as quais mudanças no hidrodinamismo da área, alterações nas distribuições de sedimento e nas comunidades biológicas podem ser positivas ou não.

Esta tese representa o primeiro registo das comunidades bentónicas dos naufrágios na costa Portuguesa. Escolheram-se quatro naufrágios, um na costa oeste (Sesimbra) e três na costa sul (em Sagres, Portimão e Tavira): River Gurara, Torvore, Oliveira e Carmo e Titan. Por cada naufrágio amostraram-se também recifes rochosos locais, de modo a comparar as comunidades em cada tipo de recife: natural e artificial. Os objectivos principais deste projecto foram registar pela primeira vez as comunidades que habitam os naufrágios e criar uma base de dados disponível online, acessível a qualquer pessoa, que possa ser consultada e actualizada em estudos futuros sobre colonização de recifes artificiais. As hipóteses a testar foram: 1) Os naufrágios têm comunidades bentónicas diferentes uns dos outros e estas diferenças deverão estar relacionadas com os diferentes estados de colonização dos naufrágios devido às diferentes idades; 2) A biodiversidade deve aumentar com a idade do naufrágio, devido aos estados de colonização mais avançados; 3) Naufrágios com mais de oito anos (idade baseada num estudo prévio) devem apresentar comunidades semelhantes às dos recifes rochosos locais; 4) As partes intactas dos naufrágios devem possuir mais biodiversidade que as partes fragmentadas; 5) Os lados dos naufrágios mais expostos às correntes devem possuir mais biodiversidade que os lados mais protegidos; 6) Superfícies verticais nos naufrágios devem possuir mais biodiversidade que as superfícies horizontais, devido à taxa de sedimentação mais elevada nas superfícies horizontais que acaba por sufocar organismos filtradores.

O registo das comunidades bentónicas foi feito usando video-transectos em mergulho. Um estudo preliminar feito num dos naufrágios indicou que era necessário realizar no mínimo três transectos para amostrar a diversidade dos recifes, embora tenham sido feitos mais transectos em alguns locais de estudo. No total foram feitos 16 transectos nos quatro naufrágios, seis num, quatro noutra e três nos dois restantes. Cada recife natural foi amostrado com três transectos. Os dados foram divididos em duas bases de dados: uma com a densidade de organismos (número de indivíduos por metro quadrado) e a outra com percentagem de cobertura de animais incrustantes e algas. As hipóteses foram testadas através de análises de variâncias multivariadas (PERMANOVA). Os resultados significativos foram analisados recorrendo a NMDS e análises SIMPER.

No geral, as análises de densidades mostraram resultados mais claros do que as análises de percentagem de cobertura. Os naufrágios da costa portuguesa aqui analisados têm na maioria dos casos comunidades bentónicas semelhantes. Os únicos significativamente diferentes foram o River Gurara e o Oliveira e Carmo. Estas diferenças estão provavelmente relacionadas com as diferentes idades dos naufrágios. Naufrágios podem ser habitats semelhantes a ilhas, em que a biodiversidade aumenta durante os primeiros anos até ao máximo possível dado o ambiente local. A partir deste ponto a colonização de novas espécies continua a acontecer, mas através de um equilíbrio dinâmico no qual algumas espécies se extinguem enquanto novas espécies aparecem. Factores como distúrbios físicos, relacionados com pesca de arrasto por exemplo, também podem contribuir para uma atenuação das diferenças entre o resto dos barcos.

Em alguns dos casos, os naufrágios têm comunidades semelhantes às dos recifes naturais da área. No geral, 40% da biodiversidade dos naufrágios não está presente nos recifes naturais locais, o que permite explorar a hipótese de que naufrágios são habitats disponíveis para serem colonizados por espécies que não ocorrem nos recifes naturais locais, levando a um aumento de biodiversidade local. Naufrágios mais antigos têm comunidades mais semelhantes às naturais. Comparativamente, o naufrágio mais antigo (99 anos) tem 44% de espécies em comum com o recife rochoso amostrado, enquanto o naufrágio mais recente (4 anos) só tem 20% em comum com o recife natural. Na maioria dos casos, os testes mostraram diferenças significativas entre naufrágios e recifes rochosos.

Na comparação entre as comunidades bentónicas presentes em estruturas verticais e horizontais dos naufrágios, houve algumas diferenças, principalmente nas percentagens de

cobertura mas sem um padrão de segregação biológica aparente. Esta semelhança estará provavelmente relacionada com o facto de as estruturas verticais e horizontais não terem uma extensão suficientemente grande para permitir o desenvolvimento de grupos de organismos diferentes. Na comparação entre os lados expostos e protegidos das correntes, a ausência de diferenças entre lados expostos e protegidos pode dever-se a uma colonização homogénea em todo o naufrágio sem uma zonação provocada pelas correntes, ou a uma suposição errada de haver um lado do navio mais exposto que outro.

As estruturas intactas dos naufrágios albergam muito mais biodiversidade que as partes fragmentadas dos barcos, o que pode ser um indicador de que naufrágios intactos e em boas condições albergam comunidades mais diversas e consequentemente mais saudáveis.

Uma das conclusões mais importantes desta tese foi a criação de um “catálogo” das espécies que se podem encontrar nos naufrágios da costa portuguesa. Este catálogo estará disponível online para que possa ser consultado pelo público em geral e referenciado pela comunidade científica em futuros estudos sobre colonização em recifes artificiais. Além da informação já recolhida, deu-se início ao desenvolvimento de um projecto (Project Baseline) com base em mergulhadores recreativos voluntários que se disponibilizarão a desenvolver um registo fotográfico contínuo destes naufrágios ao longo do tempo, permitindo um estudo sobre colonização a longo prazo e também com o objectivo de chamar a atenção da comunidade não científica para a importância destes naufrágios.

Palavras Chave: Recifes Artificiais; recifes naturais; biodiversidade macrobentónica; video-transectos

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1. Introduction

1.1 Overview – Shipwrecks and Artificial Reefs

Shipwrecks represent the most charismatic and valued type of artificial reef structures for SCUBA divers (Blout, 1981; Stolk et al. 2005), which frequently associate artificial reefs specifically with shipwrecks. A general definition for artificial reefs is that they are man-made structures placed on the sea bottom, including a variety of structures that goes from sunken vessels to specially designed structures of limestone, steel, and concrete, built to become long lasting artificial reefs. Most SCUBA divers are aware that artificial reefs are structures that provide habitat for marine populations (Stolk et al. 2005), and some are known to sustain even more density and variety of biota than natural reefs in the area (Clark and Edwards, 1999; Diamant et al. 1986; Wilhelmsson et al. 1998).

Recreational SCUBA diving is usually considered a marine ecotouristic activity, representing just a small part of the greater anthropogenic impact on the marine environment, and at a certain point it was possibly the fastest growing recreation activity in the world (Dignam, 1990; Orams, 1999; Tabata, 1992), with a constant growth in the number of operators and participants. There is some evidence that points out the inappropriate divers behaviour and the high volume of dives as a stress factor for marine flora and fauna (Davis and Tisdell, 1995; Harriott et al. 1997; Hawkins and Roberts, 1993; Medio et al., 1997). Artificial reefs come into place here as an alternative diving site, removing the pressure from the natural reefs (Polak and Shashar, 2012).

In Europe, artificial reefs started being placed in the bottom of the sea during the second half of the 1900s and since then multiple types of structures have been sunken by several countries, most of which in the Mediterranean Sea (Jensen, 2002). Across different countries, artificial reefs have the purpose of contributing to fisheries management, prevent trawling, protect coastal habitats (Baine, 2001) and develop touristic activities related with the diving industry (Stolk et al. 2009). By 2011, Europe had 3422 artificial reefs deployed (Fabi et al. 2011).

In Portugal the largest artificial reef project was responsibility of IPMA (Instituto Português do Mar e da Atmosfera). They developed an artificial reef complex with an estimated area of influence of 70 km², implemented in southern Portugal in 1990. Two pilot artificial reefs were installed to assess the environmental impact and fishing yields of the reef as well as its usefulness for managing fish stocks and increasing coastal resources. After this pilot project,

artificial reefs were recognized as a tool for the integrated management of coastal areas in Portugal (Santos 1997). Five larger artificial reefs were deployed in the same area between 1998 and 2010, making it one of the largest reef system in Europe (Fabi et al. 2011). Between 2012 and 2013 there was a private initiative to sink four warships in the south coast to create new diving sites. The area is named Ocean Revival Park and the warships rest on the bottom of the sea in the shores of Portimão, south Portugal. Other than these sunken concrete blocks and vessels, there are multiple structures along the whole Portuguese shore that have created artificial reef habitats (vessels, cranes and bombers). Lists and descriptions of these artificial reefs can be found in diving centers websites (Subnauta, Haliotis and Submania), as they are usually used as diving sites.

1.2 Impacts of Artificial Reefs

The impact that an artificial reef has on its surrounding ecosystem depends, amongst others, on its durability. The durability of artificial structures depends on their overall condition: not becoming buried by sediment in the seabed and/or not being severely damaged by hydrodynamic forces and corrosion over time are key to maintain structures in good condition. Naturally, the type of material used for the construction of artificial reef structures also influences its durability. However, in the particular case of shipwrecks, their “lifespan” depends substantially on the hydrodynamic forces of the region and on the assemblages that live on it. For example, fouling organisms prevent direct exposure of steel to seawater, therefore enhancing even further the durability of the structure (Gabriele et al., 1999; Sun et al., 2003).

Other than the durability of the structure, the amount of time it takes for an artificial reef to reach the same level of biodiversity as the natural habitats in that area should also be taken into consideration (Dunford et al. 2004). Even though artificial reefs have been frequently used as a compensatory tool for marine habitat loss (Seaman 2007), previous research showed that young reefs cannot actually provide biodiversity levels and assemblage structures similar to the ones from natural reefs (Walker et al. 2007, Simon et al. 2013). Other studies indicate that the period of time until biodiversity levels on the artificial reef became similar to the ones on the natural reef can be between eight and 25 years (Burt et al. 2011; Perkol-Finkel et al. 2006). Comparing artificial with natural reefs is important in order to assess the performance of the artificial reefs, but this comparative analysis is complicated due to differences in reef size, heterogeneity, age and degree of isolation (Svane and Petersen, 2001). The lack of research comparing biodiversity between older artificial and natural

habitats makes it very hard to predict the effectiveness of this frequent compensatory measure for habitat loss.

Several case studies point to a positive effect of artificial reefs in biodiversity increase and conservation (e.g., Marzialetti et al. 2002; Ponti et al. 2002), but that is not always the case. Artificial reef deployment without proper planning may have negative impacts. These impacts include changes in hydrodynamic forces and consequent alteration in the distribution of sediment and biological communities, as well as changes of benthic settlement patterns in the area (Gonçalves et al. 2007; Danovaro et al. 2002). The presence of artificial reefs or even just the proximity to one may impact the soft-bottom sediment benthic assemblages living in that area. It also can modify the distribution and composition of the available food sources and even the biological interactions between different parts of the food web (Danovaro et al. 2002). Negative impacts may also include some types of pollution, such as ecosystem exposure to chemical contaminants that resulted from leeching of the artificial reef structures, especially due to lack of planning such as in the case of a shipwreck, or the degradation and fragmentation of the structure itself, due to lack of robustness, causing damage to sensitive ecosystems (OSPAR Commission 2008).

1.3 Colonization of Artificial Reefs

Colonization of new marine habitats generally happens through the dispersal and settlement of larvae and spores (Roberts et al. 1997). It depends on various factors such as the structure age (Nicoletti et al. 2007; Relini et al. 1994), depth and light availability (Relini et al. 1994), slope (Knott et al. 2004; Boaventura et al. 2006), rates of sedimentation and orientation of the reef in relation to prevailing currents (Baynes and Szmant 1989). Overall, barnacles, polychaetes, bivalves, hydroids and bryozoans are known to be pioneer species, quickly occupying the majority of the available area of a new habitat (Ardizzone et al. 1989; Bailey-Brock 1989; Relini et al. 1994; Hatcher 1998; Boaventura et al. 2006; Moura et al. 2006). Authors have shown changes in pioneering species abundance over time, given the colonization of more competitive species. For example, barnacle abundance is expected to decrease due to the colonization of sponges that are generally slower to recruit to new habitats (Thanner et al. 2006). These more competitive species are usually slow-growing and long-lived, appearing frequently during the later stages of community succession (Bailey-Brock 1989; Boaventura et al. 2006). However, once established (after at least 5 years), they can dominate artificial reefs (Thanner et al. 2006).

Predicting long-term colonization patterns on artificial reef structures is challenging due to the influence and interaction of many environmental and biological variables. Yet, some studies point to differences between young shipwreck communities, when compared with local natural reef communities. For example, solitary species such as barnacles and bivalves has been found to dominate in recently sunken shipwrecks (Walker et al. 2007). Over the long-term, one study found that the development of the benthic community in the new (artificial) substratum was composed by five distinctive phases (Nicoletti et al. 2007): 1) recruitment of Pioneer species (during the first month); 2) mussel (*Mytilus galloprovincialis*) dominance in the first two years; 3) regression of mussels on the third and fourth years; 4) absence of mussels in ten years' time, and 5) dominance of Bryozoans bioconstruction twenty years after the artificial reef deployment.

1.4 Exposure to Currents

Currents and wave action are known to influence the colonization of artificial habitats (Grove and Sonu, 1985), because currents carry nutrients, organic matter and planktonic larvae across artificial reefs, and remove sediment from the structures (McAllister 1981; Baynes and Szmant 1989) enabling further settlement of spores and larvae. It has been observed that artificial reefs that are perpendicular to prevailing currents are more productive, and that currents influence growth forms, abundance and diversity of corals (Mathews, 1981; Roberts et al. 1981; Done, 1983). Even zones with strong currents and high turbulence allow the growth of healthy communities of corals (Roberts et al. 1997). A previous study (Baynes and Szmant 1989) suggested that to increase benthic growth on an artificial habitat, a maximization of the surface area exposed to laminar current flow and of the vertical surfaces would be necessary.

1.5 Surface Inclination

Surface inclination can influence the development of benthic communities because of its impact on sedimentation rates and on light exposure (Sebens 1986; Glasby 2001; Irving & Connell 2002). The accumulation of sediments is usually higher on horizontal surfaces, increasing the possibility of smothering sessile organisms (Baynes & Szmant 1989) and preventing effective settlement of recruits. Horizontal surfaces are also more exposed to light than vertical surfaces (Brakel 1929). Light can influence epibenthic organism abundance in different ways: 1) many sessile invertebrate larvae display photonegative behavior immediately before settlement, showing a preference to settle on shaded substrate (Dybernm 1962; Thorsonm 1962; Olson, 1983); 2) different light intensities might indirectly harm an

organism by benefiting its competitors; 3) low light intensities can be stressful for photosynthetic organisms.

1.6 Objectives

There have been studies along the Portuguese coastline regarding natural reefs (Gonçalves et al. 2008; Boavida et al. 2016 a, b) and artificial reefs (Santos et al. 2005; Moura et al. 2006), but only one on one of the shipwrecks (Coelho et a. 2012). Through the use of videos to sample biodiversity, this thesis was the first survey to characterize the macrobenthic community on Portuguese shipwrecks along the continental coastline between Sesimbra and Tavira. Rocky reefs located close to the shipwrecks were sampled to assess if wrecks and natural reefs support similar assemblages and to try to ascertain the source populations responsible for the colonization of the shipwrecks.

The main purpose of this study was to create a biodiversity “baseline” database which could be used as a reference for studies on the colonization of artificial habitats, and to gather knowledge which could allow for proper planning of deployment and management of artificial structure projects. Four shipwrecks were sampled along the Portuguese west and south coasts and the expectations were that: 1) Macrobenthic communities should differ between shipwrecks, with differences related to their different ages and locations; 2) Biodiversity should increase with the shipwrecks age due to the more advanced state of succession in the colonization process; 3) Shipwrecks older than eight years should have communities similar to the ones on natural rocky reefs; 4) The intact parts of a shipwreck should harbour more biodiversity than the degraded parts; 5) The shipwreck side more exposed to the sea currents should have more biodiversity; 6) Vertical surfaces in the shipwrecks should have more biodiversity than the horizontal surfaces, due to smothering of filtering organisms by sediment deposition on the horizontal surfaces.

2. Materials and Methods

2.1 Sampling Locations

There are two distinct sampling regions, both belonging to the Portuguese continental coast. One on the West coast, in Sesimbra, while the other sampling region is the South coast, Algarve (Figure 1 locates the four sampling locations in the coastline). The list of sampled shipwrecks, natural rocky reefs and respective information can be consulted in Table 1.

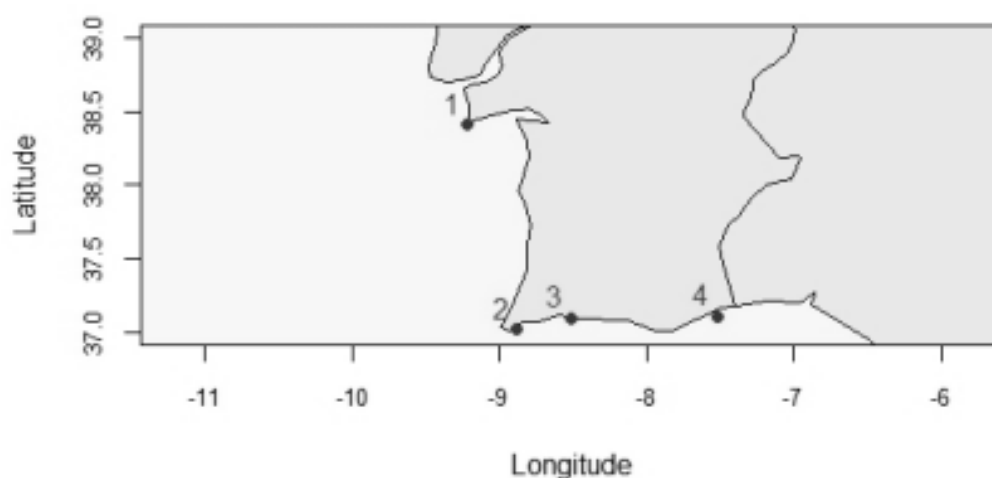


Figure 1 - Map of the sampling sites, 1 – River Gurara and Paredes do Cabo; 2 – Torvore and Falésia; 3 – Oliveira e Carmo and Ponta da Piedade; 4 – Titan and Pedra da Cacela.

Table 1 - List of sampling sites, with the date of the sinking, the age of the shipwrecks, the maximum depth of each site, their location and coordinates.

| Sampling Sites | Sunk in | Age (years) | Max. Depth (m) | Length (m) | Location | Type | Coordinates |
|------------------|---------|-------------|----------------|------------|----------|------------|-----------------------------------|
| Oliveira e Carmo | 2012 | 4 | 33 | 85 | Portimão | Shipwreck | N 37°05'816'' W 8° 35'114' |
| Titan | 2002 | 14 | 28 | 30 | Tavira | Shipwreck | N 37°05'899'' W 7°31'321'' |
| River Gurara | 1989 | 27 | 25 | 90 | Sesimbra | Shipwreck | N38°24'39.85'', W9°13'12.65'' |
| Torvore | 1917 | 99 | 32 | 97 | Sagres | Shipwreck | N37° 01'07.2'' W8° 52'54.8'' |
| Ponta da Piedade | - | - | 7 | . | Lagos | Rocky Reef | N37°04'45.69'' W8°40'1.40'' |
| Pedra da Cacela | - | - | 12 | . | Tavira | Rocky Reef | N37°08'35.1'' W7°32'15.5'' |
| Paredes do Cabo | - | - | 18 | . | Sesimbra | Rocky Reef | N38°24'39.85'', W9°13'12.65'' |
| Falésia | - | - | 18 | . | Sagres | Rocky Reef | N 37° 0'19.70'' W 8°55'39.51'' |

2.1.1 Sesimbra

River Gurara and Paredes do Cabo are both inside the Prof. Luiz Saldanha Marine Park (in Parque Natural da Arrábida). This area has some unique features: it is located near the northern limit of one of the main upwelling systems in the North Atlantic (Wooster et al., 1976), and it is a biogeographic and oceanographic transition zone between warm and cold temperate waters, being protected from the North and Northwest winds by the Arrábida's ridge (e.g. Gonçalves et al., 2002). Subtidal rocky reefs in this area are dominated by boulders originated from the erosion of the cliffs and by a bedrock with fissures and crevice, creating a complex diversity of macro and microhabitats (Horta e Costa et al. 2013).

The two sampling sites in Sesimbra were the stern of the shipwreck River Gurara and the rocky reef right next to it (0.6 km away), known as Paredes do Cabo. River Gurara was a Nigerian cargo motor vessel, 175 m long, which sunk and broke in half due to a storm in 1989, near Cape Espichel. It is partially destroyed, but it is still possible to navigate on the wreck and recognize the different parts of the ship. Paredes do Cabo, the name of the diving spot, refers to the rocky wall area of the cliff, with an average depth of 15 m.

2.1.1 Algarve

The continental shelf of the Portuguese South coastline encompasses an oceanographic transition zone due to the combined influence of the North Atlantic and Mediterranean currents, reason why the benthic fauna in this area is classified as Atlantic-Mediterranean. Some areas (i.e. Sagres) are known to be highly hydrodynamic, and this force can actually have an ecosystem structuring influence up to 10 m of depth (Dolbeth et al. 2007).

The sampling sites in the South Coast are Sagres, Portimão, Lagos and Tavira. Sagres is the western most part of Algarve, with a complex substrate, comprised mainly of boulders that are an extension from the land cliffs into the ocean, ending in a sandy seabed. Portimão and Lagos, between Sagres and Tavira, have much gentler slopes, with rocky reefs that present a low relief and are interspersed with patches of sediment (Boavida et al. 2016a) that varies between sandy and muddy. Tavira's coastline is composed of sandy beaches, with a very gentle slope, low relief rocky reefs and a sandy seabed.

The two sampling sites in Sagres were Falésia, the rocky reef next to the cliff, and Torvore, a Norwegian steam ship that sunk during the First World War due to explosive charges, which is 4.34 km away from Falésia. Although very damaged, many parts of the structure are still easily recognizable. The shipwreck is at a depth between 28 and 32 m, set on a sandy and

muddy bottom. The rocky reef, Falésia, is a habitat relatively protected from waves, right next to the harbor, composed primarily by boulders, with a maximum depth around 18 m.

The two sampling sites in Portimão and Lagos were the Corvette Oliveira e Carmo (Portimão) and the rocky reef in Ponta da Piedade (Lagos), 9 km away. Oliveira e Carmo is an 85 m long ship that sunk in 2012, as part of the Ocean Revival Park (www.oceanrevival.org) and now rests in a 33 m deep muddy/sandy bottom with its full structure almost completely intact. Ponta da Piedade is a shallow had a maximum depth of 7 m.

The two sampling sites in Tavira were Titan and the rocky reef Lage da Cacela, 5.5 km away. Titan sunk in 2002, while placing concrete blocks of artificial reefs. It is still completely intact, 30 m long and 18 m wide and rests on a sandy bottom at 28 meters deep. Lage da Cacela has a maximum depth of 12 m, and is composed by several rocks completely surrounded by sand, with a main rock being very long and with a rift.

2.2 Data collection

The surveys were done using video-transects while SCUBA diving. Each transect was 30 m long and took on average 4 minutes to record. Even though the pilot study (described in section 2.4) allowed an estimation of the minimum number of transects needed to sample the species richness on each type of reef, the sampling ended up being opportunistic: if it was possible to do more than the minimum number of transects (three), depending on the amount of gas left or pn being able to go back to the site a second time, than more transects were made. River Gurara ended up being sampled with six transects, Oliveira e Carmo with four, Titan and Torvore with three. The rocky reefs were all sampled with three video-transects each. The system used for filming included cameras GoPro Hero 3 and 4, two lasers separated by 7.4 cm (projecting a light in the video which was used as scale) and a video light with 3600 lumens of intensity.

As it has been explained before, the same diver was responsible for the videos through all the dives, but the diving teams varied between two and three people along the sampling. At least one more diver was required to be present to ensure safety. The second (and third diver when present) were positioned behind the diver, so no sediment was lifted in a way which could affect the image of the video and the impact in benthic fishes and crustaceans was reduced (not “scaring” them off). During the dives of the pilot study, the support diver was responsible for laying a line, using a reel, so the transects were precisely 30 m. After several

dives, the swimming speed of the diver recording the videos became constant (7.5 meters per minute), and there was no need of laying the transect line from that point on. This was a big advantage because the task of laying the transect line underwater was very time consuming. Due to the lack of natural light at the shipwrecks depth, a lantern was used so all the videos were lit by an external light, which was positioned on an angle which allowed for the lighting of the whole vision field of the camera. The structures being filmed were not always leveled, so keeping a perfectly parallel positioning of the camera and the correct angle of the lantern required constant adjustments along the dive. When it was necessary to capture more details, the diver with the camera would stop and even go back if necessary.

2.3 Data Analysis

The underwater videos were viewed in Windows Media Players, without image editing. The data collected was divided in two distinctive databases. One database included taxa in which organisms or colonies could be individually counted, like gorgonians or sea stars. The other database included incrusting organisms like sponges and the algae morphological groups, which for most part could not be individually counted. Algae were not identified in taxa but in morphological groups (Steneck and Dethier, 1994) due to the difficulty of identifying in the videos smaller species or algae that occurred in large patches.

The data used for density analysis was counted directly from the videos into the database. Densities (number of individuals per m²) required an estimation of the sampled area, which was made using the laser lights. As it has been explained before, the lasers projected two light points into the video, which had a constant distance of 7.4 cm between each other. Using the same screenshots used for percent cover analysis (explained in the next paragraph), the average width of the area filmed was calculated using the software Coral Point Count with Excel extensions, and multiplied by the total length sampled in that site (number of transects x 30 m).

Percent cover was analyzed through a subsample of the videos. Systematic snapshots of the video-transects every 10 seconds, using GOM Player software. Only snapshots that corresponded to images completely parallel to the object (part of the wreck or rock) were used, therefore not all sites were sampled with the same number of snapshots (Table 2). The snapshots were then analyzed with CPCe (Coral Point Count with Excel extensions) to estimate the taxa percent cover.

Table 2 - List of the number of snapshots used for percent cover analysis per site

| Sampling Sites | Number of Snapshots |
|------------------|---------------------|
| Oliveira e Carmo | 32 |
| Titan | 31 |
| River Gurara | 95 |
| Torvore | 31 |
| Ponta da Piedade | 34 |
| Pedra da Cacela | 29 |
| Paredes do Cabo | 24 |
| Falésia | 30 |

2.3.1 Comparing shipwrecks communities

Diversity indexes were performed to allow a comparison between communities. The following were computed using BiodiversityR package in R: Shannon-Wiener, Simpson and the taxonomic distinctness index (Kindt et al. 2005). The taxonomic distinctness index, besides focusing on the distribution of abundances amongst species, also focuses on the taxonomic relatedness of species in the samples (Clark & Warwick 2001), unlike the Shannon-Wiener and Simpson indexes, which only consider the number of species present and their abundance (Clarke & Warwick, 2001). The taxonomic distinctness index is regarded as one of the most accurate indicators in biodiversity analyses (Clarke & Warwick, 2001).

Shannon-Wiener (H') - It is based on the species abundances ratio, taking into account the evenness and the species richness, and p_i is the ratio of individuals of the species i

$$H' = - \sum p_i \log(p_i)$$

Simpson index (D) – considers that biodiversity is inversely related with the probability of randomly finding two individuals of the same species. p_i is the ratio of individuals of the species i :

$$D = \frac{1}{\sum_{i=1}^s p_i^2}$$

Data was previously transformed with square-roots and converted to a Bray–Curtis similarity matrix. Data transformation is used frequently when the measured variable does not have a normal distribution or when there are different standard deviations in the different groups of samples. The square-root transformation is usually applied in situations involving counts.

Bray-Curtis index is generally used with abundance or count data (Greenacre and Primicerio, 2013). To test differences in benthic assemblages, a permutation multivariate ANOVA was performed (PERMANOVA with 9,999 permutations and α set at 0.008; Anderson, 2001), with pairwise comparison. The site was the only factor, which was fixed and had four levels. The α is set at 0.008 and not at 0.05 as it is commonly seen in ecology statistics due to the Bonferroni correction, in which the p-values are divided by the number of comparisons (Wright, 1992). For every PERMANOVA test with significant differences, an analysis of dispersion (PERMDISP) was made.

PERMANOVA significant results were further analyzed by NMDS (Nonmetric multidimensional scaling) and SIMPER (Similarity Percentages analysis), to determine which species have the biggest role in the dissimilarities between sites.

PERMANOVA is a non-parametric method for multivariate analysis of variance, widely used in ecology, which tests differences in the composition and relative abundances of species in samples. PERMANOVA is used in situations in which the simultaneous responses of potentially non-independent variables (usually abundances of species in an assemblage) have been measured (Anderson, 2001). NMDS is a flexible ordination technique which can be applied in multiple scenarios and it aims to represent the position of communities in multidimensional space, as way of easing the interpretation of the data. The SIMPER analysis assays the contribution of each species to the similarities or dissimilarities between groups. PERMANOVA, PERMDISP, NMDS and SIMPER were computed in PRIMER 6 with PERMANOVA+ (Anderson et al., 2008). Tables of all PERMDISP and SIMPER analysis can be found in the Appendixes.

The workflow was constant across all hypothesis testing: data transformations, similarity matrixes, multivariate testing and analysis of significant results were performed the same way as in the shipwrecks community comparisons.

2.3.2 Comparing shipwrecks with natural rocky reefs

The data was treated in a similar way as in the previous chapter (divided in two sets, one with densities and other with percent covers). Data was previously transformed with square-roots, and converted to a Bray–Curtis similarity matrix. To test differences between assemblages in shipwrecks and rocky reefs, several PERMANOVA were performed (9,999 permutations and α set at 0.05), one per shipwreck-reef pair. The reef type was the only factor, which was fixed

and had 2 levels (shipwreck or natural reef). PERMANOVA significant results were further analyzed by NMDS and SIMPER

2.3.3 Zonation within shipwrecks

The zonation (here defined as community differentiation between habitats with distinct characteristics) within the shipwrecks was tested through three fixed factors: integrity, protection and orientation. Integrity focused on the good condition of the wreck and had two levels, intact or fragmented. Protection was related with the exposure of the wreck to the sea currents, and its two levels were exposed or protected. Orientation was related with the structures of the wrecks, and its two levels were vertical and horizontal.

The data was treated in a similar way as in the two previous chapters. Data was previously transformed with square-roots, and converted to a Bray–Curtis similarity matrix. To test differences in benthic assemblages on the different zones within the same shipwreck (integrity, exposure and orientation), a permutation multivariate ANOVA was used (PERMANOVA with 9,999 permutations and a set at 0.05). Significant results were further analyzed by NMDS and SIMPER

2.4 Pilot Study

River Gurara and Paredes do Cabo were the first sites to be sampled so they could be used for a pilot study. The choice of sampling these sites first was opportunistic: they were the easiest to get to. The main purpose of this pilot study was to assess the appropriate sampling effort: the minimum number of video-transects needed to estimate species richness. It also had the purpose of refining some aspects of the method use to sample the macrobenthic assemblages (video-transects), like the adequate distance between the camera and the bottom while filming, the most appropriate swimming speed and the best position for the lantern. The third purpose was to attempt to do a preliminary assessment of the factors with a possible impact in structuring the communities.

This preliminary analysis lead to a few species accumulation curves (Figure 2 and 3). These accumulation curves show the number of new taxa sampled per minute of video. Overall, three transects (12 minutes of video in total) were enough to sample the species richness in the shipwreck, considering that each transect had a length of 30 meters and took an average of 4 minutes to record. There was a necessity of a higher sampling effort on the port side of River, likely related with two factors: the position of the shipwreck, which exposes more the

port side to the sea currents, and the difference of integrity between sides (the port side has many more structures still completely intact than the starboard side). In this case and for the rest of the shipwrecks in the study, we considered that the side of the shipwreck facing the coastline was more protected from the sea currents, while the side facing the open ocean was more exposed. The species accumulation curves for the port and starboard sides indicate that 8 minutes (two transects) were enough to sample most of the biodiversity on the port side, and four minutes (one transect) was enough to sample the starboard side. For Paredes, the natural reef, the minimum of minutes needed to sample most of the species richness is not clear, but 12 minutes (three transects) were estimated to be enough (Figure 3).

Something to consider is that the average of minutes per transect depends on the diver performing the video-transects: different divers will have different speeds. All the dives in this project were done with the same diver being responsible for recording the video-transects, to ensure their comparability. If this is not the case, then the species accumulation curves should relate the number of new taxa per transect and not per minute of video.

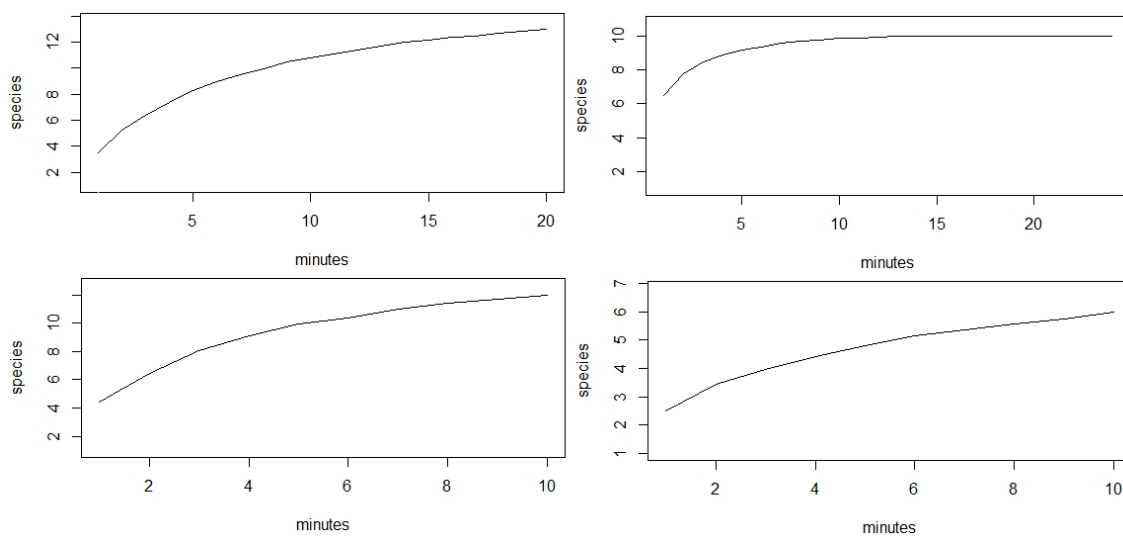


Figure 2 - Species accumulation curves for River Gurara showing the number of new species per minute of video. Top left: individually counted organisms; Top right: incrusting organisms and algae; Bottom right: individually counted organisms in the port side; Bottom right: individually counted organisms in the starboard side.

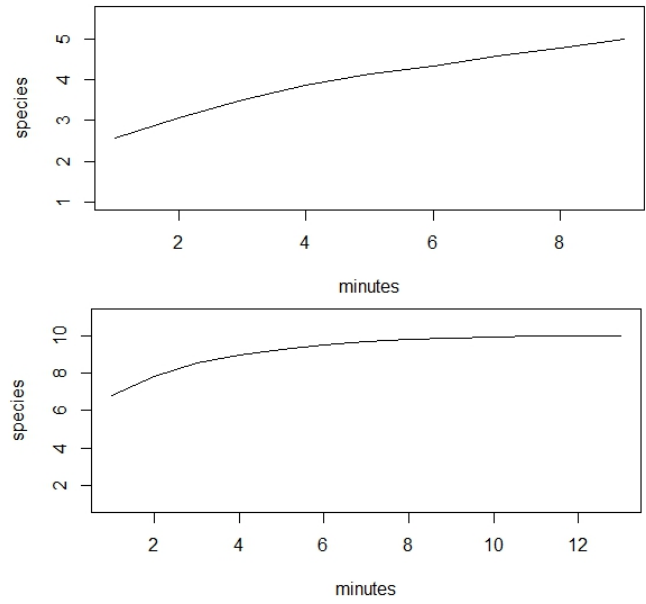


Figure 3 - Species accumulation curves for Paredes do Cabo showing the number of new species per minute of video. Top: non-colonial organisms; Bottom: incrusting organisms and algae.

3. Results

3.1 Comparison between shipwrecks

3.1.1 Comparing shipwrecks communities

The survey of the four shipwrecks recorded a total of 30 taxa (Table 3 does not include the taxa analyzed through percent cover, *Bryozoa*, *Cnidaria*, *Porifera* and *Balanidae*) and six morphological groups of algae (corticated foliose, corticated macrophytes, crustose, filamentous, foliose algae, leathery macrophytes), during approximately 68 minutes of video and 126 snapshots. The shipwrecks had very similar values for alpha diversity (a variation from 11 to nine taxa), and the highest number of algae morphological functional groups was recorded in River Gurara, followed by Titan, Torvore and Oliveira e Carmo. The best represented taxa groups were *Aiptasia mutabilis*, *Actinothoe sphyrodeta*, *Gorgoniidae*, *Holothuria sp.*, *Mytilidae* and *Scorpaena sp.* (Figure 14).

The various diversity indexes estimated do not present consistent results. The taxonomic distinctness index ($\Delta+$) indicated that all samples are within the 95% confidence interval (Figure 3). The highest taxonomic distinctness was observed in Oliveira e Carmo (92.84), followed by River Gurara (90.38), Torvore (90.32) and Titan (77.09). Other indexes (Shannon and Simpson) indicate that River has the most diverse community (1.76 and 0.79), followed by Torvore (1.14 and 0.59), Oliveira (0.35 and 0.17) and in the end, Titan (0.22 and 0.07).

Table 3 - List of the all the taxa mean densities (individual per m²) sampled in the shipwrecks. *Gorgoniidae* and *Holothuria sp.* are in bold because the densities were estimated at this level, but in some cases it was possible to identify at specie and genus level, therefore they are listed bellow but without a density value.

| Taxa | Filo | Torvore | River Gurara | Titan | Oliveira e Carmo |
|--|----------------------|---------|--------------|-------|------------------|
| <i>Actinopterygii</i> spp. 1 | <i>Chordata</i> | 0 | 0 | 0 | 0.01 |
| <i>Actinothoe sphyrodeta</i> | <i>Cnidaria</i> | 0 | 0.34 | 59.57 | 2.91 |
| <i>Aiptasia mutabilis</i> | <i>Cnidaria</i> | 0 | 0.22 | 0.53 | 0 |
| <i>Alcyonium</i> sp. | <i>Cnidaria</i> | 0 | 0.02 | 0 | 0 |
| <i>Alicia mirabilis</i> | <i>Cnidaria</i> | 0 | 0.02 | 0.02 | 0.01 |
| <i>Asciacea</i> sp. | <i>Chordata</i> | 0 | 0.07 | 0.09 | 0 |
| <i>Cancer pagurus</i> | <i>Arthropoda</i> | 0 | 0 | 0.02 | 0 |
| <i>Cerianthus membranaceus</i> | <i>Cnidaria</i> | 0.11 | 0 | 0 | 0 |
| <i>Conger conger</i> | <i>Chordata</i> | 0.07 | 0 | 0.02 | 0 |
| <i>Diaphorodoris luteocincta</i> | <i>Mollusca</i> | 0 | 0.01 | 0 | 0 |
| <i>Echinaster (Echinaster) sepositus</i> | <i>Echinodermata</i> | 0.23 | 0 | 0 | 0 |
| <i>Felimare</i> sp. | <i>Mollusca</i> | 0 | 0.01 | 0 | 0 |
| <i>Gobius</i> spp. 1 | <i>Chordata</i> | 0 | 0.02 | 0 | 0 |
| <i>Gorgoniidae</i> | <i>Cnidaria</i> | 1.75 | 0.31 | 1.19 | 0.03 |
| <i>Eunicella</i> spp. 1 | - | - | - | - | - |
| <i>Eunicella</i> spp. 2 | - | - | - | - | - |
| <i>Leptogorgia Sarmentosa</i> | - | - | - | - | - |
| <i>Holothuria</i> sp. | <i>Echinodermata</i> | 1.47 | 0.30 | 0 | 0 |
| <i>Holothuria arguinensis</i> | - | - | - | - | - |
| <i>Holothuria forskali</i> | - | - | - | - | - |
| <i>Holothuria mamata</i> | - | - | - | - | - |
| <i>Marthasterias glacialis</i> | <i>Echinodermata</i> | 0.08 | 0 | 0 | 0.01 |
| <i>Mytilidae</i> | <i>Mollusca</i> | 0 | 0 | 0 | 31.52 |
| <i>Palinurus elephas</i> | <i>Arthropoda</i> | 0.06 | 0 | 0 | 0 |
| <i>Parablennius gattorugine</i> | <i>Chordata</i> | 0 | 0 | 0 | 0.01 |
| <i>Parablennius pilicornis</i> | <i>Chordata</i> | 0 | 0 | 0.02 | 0 |
| <i>Parablennius</i> sp. | <i>Chordata</i> | 0 | 0 | 0.04 | 0.06 |
| <i>Parablennius</i> spp. 1 | - | - | - | - | - |
| <i>Parablennius</i> spp. 2 | - | - | - | - | - |
| <i>Paracentrotus lividus</i> | <i>Echinodermata</i> | 0 | 0 | 0 | 0.03 |
| <i>Paramuricea clavata</i> | <i>Cnidaria</i> | 0.06 | 0 | 0 | 0 |
| <i>Scorpaena notata</i> | <i>Chordata</i> | 0 | 0 | 0.02 | 0 |
| <i>Scorpaena</i> sp. | <i>Chordata</i> | 0.09 | 0 | 0.57 | 0.16 |
| <i>Octopus vulgaris</i> | <i>Mollusca</i> | 0 | 0.01 | 0 | 0 |

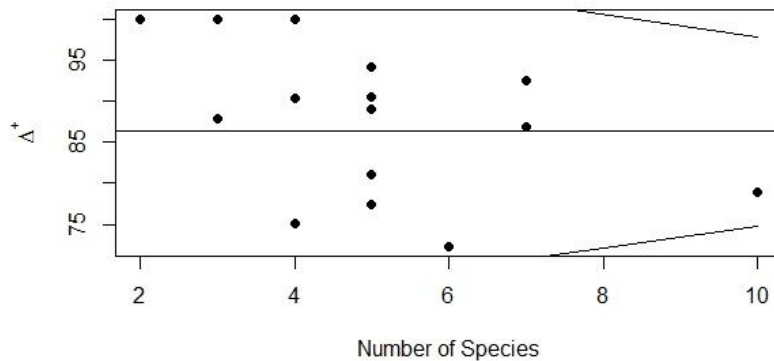


Figure 4 - Index of taxonomic distinctness. Lines indicate average and 95% confidence interval.

Only two shipwrecks had densities significantly different from each other: River Gurara and Oliveira e Carmo ($P = 0.006$). The remaining shipwrecks had no significant differences between them ($P > 0.008$; Table 4). The NMDS showed an Oliveira community very distant from the rest: transects are isolated from all the others, showing a distinctive community. River Gurara transects are by far the most disperse in the figure, showing a higher diversity than in any other wreck (Figure 3). The SIMPER analysis showed that the taxa most responsible for differences between the two wrecks were *Mytilidae*, *Actinothoe shyrodeta*, *Holothuria sp.* and *Gorgoniidae*. The first two were more abundant in Oliveira e Carmo, and the second two on River Gurara. Tables for all SIMPER analyses are found in the Appendixes.

The four shipwrecks had percent covers significantly different from each other ($P = 0.001$) (Table 5). In this case and in multiple situations across all the results, PERMADISP suggested that the differences found by PERMANOVA were caused by a dispersion effect in the samples (when $P > 0.05$) (Appendixes Tables 24 to 40). However, in all of those cases, PERMANOVA should be robust to such an effect (Anderson, 2001; Anderson et al., 2008). The NMDS here suggests that Oliveira transects are slightly less isolated, but a differentiation between transects in all the shipwrecks is still obvious (Figure 5). The SIMPER analysis shows that hydroids and algae assemblages, crustose algae and *Balanidae* are consistently the taxa responsible for the differentiation between shipwrecks: the same assemblages are recorded across all shipwrecks, but their abundance varies considerably, setting the communities apart.

Table 4 - PERMANOVA pairwise test based on a Bray–Curtis similarity matrix of square-root transformed benthic organisms densities.

| Pairwise test | PERMANOVA | | |
|---------------|-----------|---------|--------------|
| | t | P(perm) | Unique perms |
| TO, RG | 1.6813 | 0.035 | 84 |
| TO, TI | 2.9331 | 0.119 | 10 |
| TO, OC | 5.6453 | 0.031 | 34 |
| RG, TI | 2.1046 | 0.027 | 84 |
| RG, OC | 3.1356 | 0.006 | 209 |
| TI, OC | 3.9177 | 0.031 | 35 |

Note – TO torvore; RG River Gurara; TI Titan; OC Oliveira e Carmo

Table 5 - PERMANOVA pairwise test based on a Bray–Curtis similarity matrix of square-root transformed benthic percent covers.

| Pairwise test | PERMANOVA | | |
|---------------|-----------|---------|--------------|
| | t | P(perm) | Unique perms |
| OC, RG | 8.984 | 0.001 | 999 |
| OC, TI | 8.7072 | 0.001 | 997 |
| OC, TO | 9.4426 | 0.001 | 998 |
| RG, TI | 5.2794 | 0.001 | 999 |
| RG, TO | 2.0971 | 0.001 | 999 |
| TI, TO | 5.5216 | 0.001 | 999 |

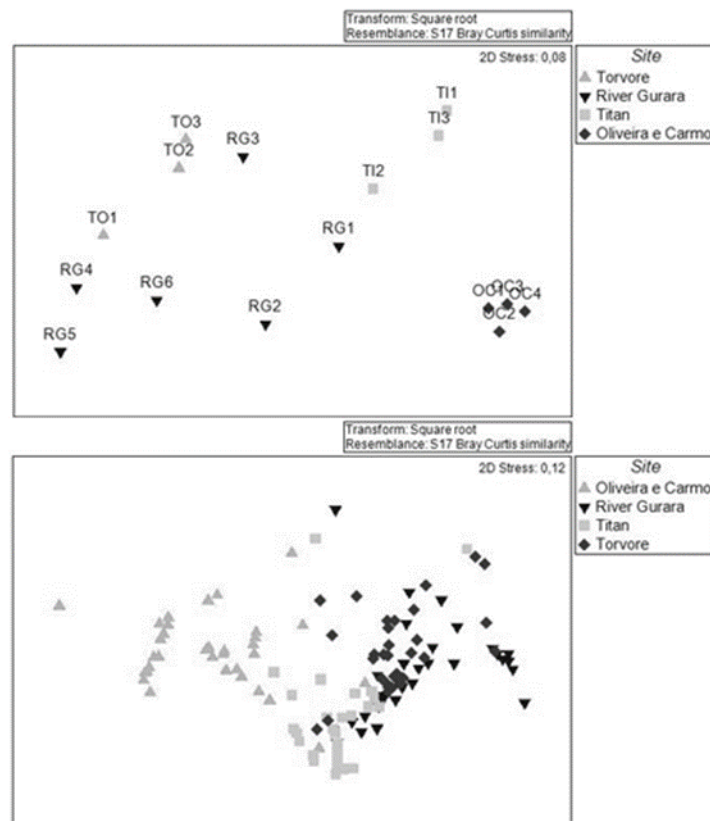


Figure 5 - NMDS on the Bray–Curtis square-root-transformed data of benthic organisms in all of the Shipwrecks. On top: densities; On the bottom: percent cover.

3.1.2 Comparing shipwrecks with natural rocky reefs

The survey of the four natural rocky reefs recorded a total of 24 taxa (Table 6) and seven morphological groups of algae, during approximately 55 minutes of video and 114 snapshots. From the 30 taxa sampled in the shipwrecks, 43% (13) were found in common with the rocky reefs.

The densities of benthic assemblages differed significantly between the oldest and youngest shipwrecks (River Gurara and Oliveira e Carmo) and their adjacent rocky reefs ($P = 0.036$; $P = 0.024$) (Table 7). The NMDS with data from all the wrecks and rocky reefs shows two clearly differentiated groups, with all wrecks assemblages being clearly separated from the rocky reefs (Figure 6). The NMDS of Oliveira e Carmo (the youngest wreck) and its nearby rocky reef shows a stronger differentiation between wreck and rocky reef than that of River Gurara (the oldest; Figure 7). The SIMPER analysis shows that *Paracentrotus lividus*, *Actinothoe sphyrodeta*, *Gorgoniidae* and *Holothuria sp.* are the taxa most responsible for the differences between River Gurara and its adjacent rocky reef. *P. lividus* is only present on the rocky reef. *A. sphyrodeta*, *Gorgoniidae* and *Holothuria sp.* were more abundant on the shipwreck. In Oliveira e Carmo, *Mytilidae*, *Anemonia viridis*, *A. sphyrodeta* and *P. lividus* were the main responsible for the differences between wreck and rocky reef. *Mytilidae* and *A. sphyrodeta* were much more abundant on the wreck, while *P. lividus* and *A. viridis* were more abundant on the rocky reef.

The percent cover of benthic assemblages differed significantly between all of the shipwrecks and the adjacent rocky reefs (PERMANOVA $P = 0.001$ for all) (Table 8). All NMDS suggest a clear differentiation between shipwrecks and rocky reefs (Figure 8). The SIMPER analysis shows that hydroids, algae assemblages and crustose algae are consistently the taxa responsible for most of the differences between shipwrecks and natural reefs, with hydroids and algae assemblages being constantly more abundant on the rocky reefs. The crustose algae are sometimes more abundant on the wrecks and other times more abundant in the rocky reefs.

Table 6 - List of the all the taxa mean densities (individual per m²) sampled in the rocky reefs.

| Taxa | Paredes do Cabo | Pedra da Cacela | Falésia | Ponta da Piedade |
|----------------------------------|-----------------|-----------------|---------|------------------|
| <i>Actinothoe sphyrodeta</i> | 0 | 2.38 | 0 | 0 |
| <i>Aiptasia mutabilis</i> | 0 | 3.57 | 4.10 | 0 |
| <i>Anemonia viridis</i> | 0 | 2.55 | 0.17 | 4.84 |
| <i>Asciidae sp.</i> | 0 | 0 | 0.02 | 0 |
| <i>Blenniidae</i> | 0 | 0.02 | 0 | 0 |
| <i>Diaphorodoris luteocincta</i> | 0.03 | 0 | 0 | 0 |
| <i>Felimare sp.</i> | 0 | 0 | 0.02 | 0 |
| <i>Gobius auratus</i> | 0 | 0.02 | 0 | 0 |
| <i>Gobius xanthocephalus</i> | 0 | 0 | 0.02 | 0 |
| <i>Gorgoniidae</i> | 0 | 4.71 | 1.83 | 0 |
| <i>Gymnangium montagui</i> | 0 | 0 | 0.02 | 0 |
| <i>Holothuria sp.</i> | 0.20 | 0.02 | 0.53 | 0.67 |
| <i>Palinurus elephas</i> | 0 | 0 | 0.02 | 0 |
| <i>Parablennius pilicornis</i> | 0 | 0 | 0.02 | 0 |
| <i>Parablennius sp.</i> | 0 | 0.057 | 0 | 0.02 |
| <i>Paracentrotus lividus</i> | 1.90 | 2.52 | 0.075 | 1.23 |
| <i>Sabella spallanzanii</i> | 0 | 0.02 | 0 | 0 |
| <i>Sabellida</i> | 0 | 0.06 | 0 | 0 |
| <i>Serpula sp.</i> | 0 | 0.02 | 0 | 0 |
| <i>Scorpaena maderensis</i> | 0 | 0.02 | 0 | 0.02 |
| <i>Scorpaena porcus</i> | 0 | 0.15 | 0 | 0 |
| <i>Scorpaena sp.</i> | 0 | 0.08 | 0 | 0 |
| <i>Sphaerechinus granularis</i> | 0 | 2.12 | 0.21 | 0 |
| <i>Octopus vulgaris</i> | 0 | 0.02 | 0 | 0 |

Table 7 - PERMANOVA tests based on a Bray–Curtis similarity matrix of square-root transformed benthic organisms densities in shipwrecks and in the adjacent rocky reefs.

| Source | PERMANOVA | | | | | |
|---|-----------|--------|--------|----------|---------|--------------|
| | df | SS | MS | Pseudo-F | P(perm) | Unique perms |
| Torvore vs Falésia | | | | | | |
| Type | 1 | 5826.7 | 5826.7 | 6.1567 | 0.098 | 10 |
| Residuals | 4 | 3785.6 | 946.4 | | | |
| Total | 5 | 9612.3 | | | | |
| River Gurara vs Paredes do Cabo | | | | | | |
| Type | 1 | 6760.6 | 5826.7 | 3.3882 | 0.036 | 28 |
| Residuals | 6 | 11972 | 946.4 | | | |
| Total | 7 | 18733 | | | | |
| Titan vs Pedra da Cacela | | | | | | |
| Type | 1 | 5978.1 | 5978.1 | 6.1021 | 0.098 | 10 |
| Residuals | 4 | 3918.7 | 989.68 | | | |
| Total | 5 | 9869.8 | | | | |
| Oliveira e Carmo vs Ponta da Piedade | | | | | | |
| Type | 1 | 15176 | 15176 | 31.946 | 0.024 | 35 |
| Residuals | 5 | 2375.3 | 475.07 | | | |
| Total | 6 | 17552 | | | | |

Table 8 - PERMANOVA tests based on a Bray–Curtis similarity matrix of square-root transformed percent cover organisms densities in shipwrecks and in the adjacent rocky reefs.

| Source | PERMANOVA | | | | | |
|---|-----------|-----------------------------|-----------------------|----------|---------|--------------|
| | df | SS | MS | Pseudo-F | P(perm) | Unique perms |
| Torvore vs Falésia | | | | | | |
| Type | 1 | 10705 | 10705 | 13.743 | 0.001 | 998 |
| Residuals | 59 | 45959 | 778.97 | | | |
| Total | 60 | 56664 | | | | |
| River Gurara vs Paredes do Cabo | | | | | | |
| Type | 1 | 22721 | 22721 | 19.669 | 0.001 | 998 |
| Residuals | 54 | 62381 | 1155.2 | | | |
| Total | 55 | 85102 | | | | |
| Titan vs Pedra da Cacela | | | | | | |
| Type | 1 | 51449 | 51449 | 53.546 | 0.001 | 999 |
| Residuals | 58 | 55728 | 960.83 | | | |
| Total | 59 | 1.078x10 ⁵ | | | | |
| Oliveira e Carmo vs Ponta da Piedade | | | | | | |
| Type | 1 | 1.139x10 ⁵ | 1.139x10 ⁵ | 139.68 | 0.001 | 998 |
| Residuals | 72 | 587261.7265x10 ⁵ | 815.64 | | | |
| Total | 73 | | | | | |

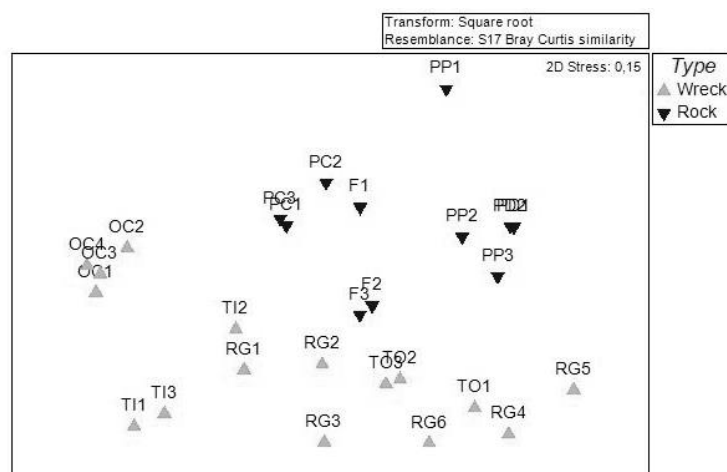


Figure 6 - NMDS on the Bray–Curtis square-root-transformed densities of benthic organisms in all the shipwrecks and adjacent rocky reefs.

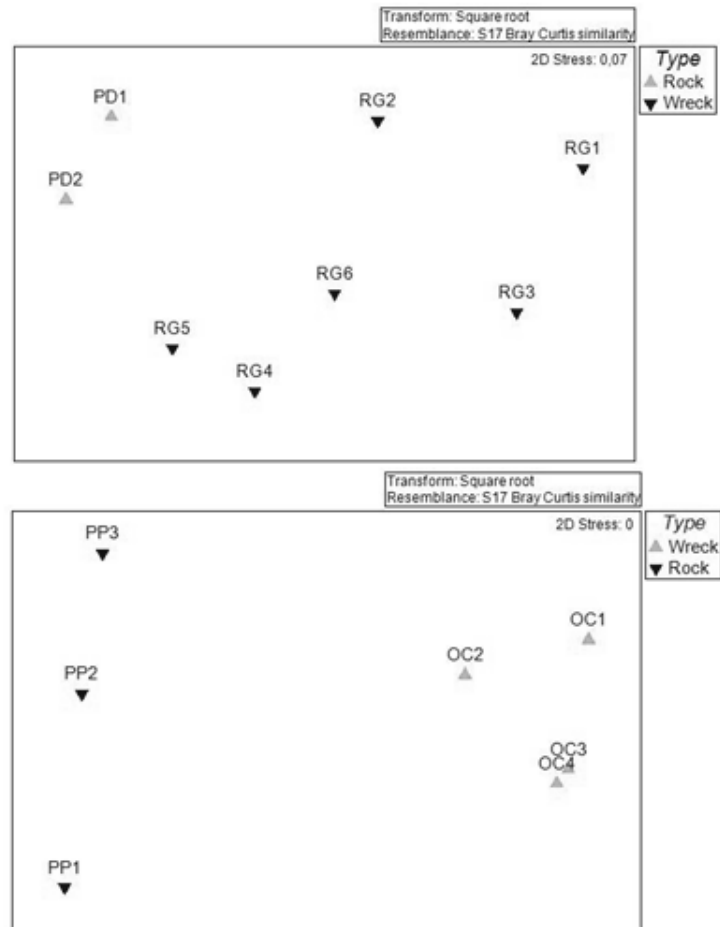


Figure 7 - On top: NMDS on the Bray–Curtis square-root-transformed densities of benthic organisms in River Gurara and Paredes do Cabo; On the bottom: NMDS on the Bray–Curtis square-root-transformed densities of benthic organisms in Oliveira e Carmo and Ponta da Piedade.

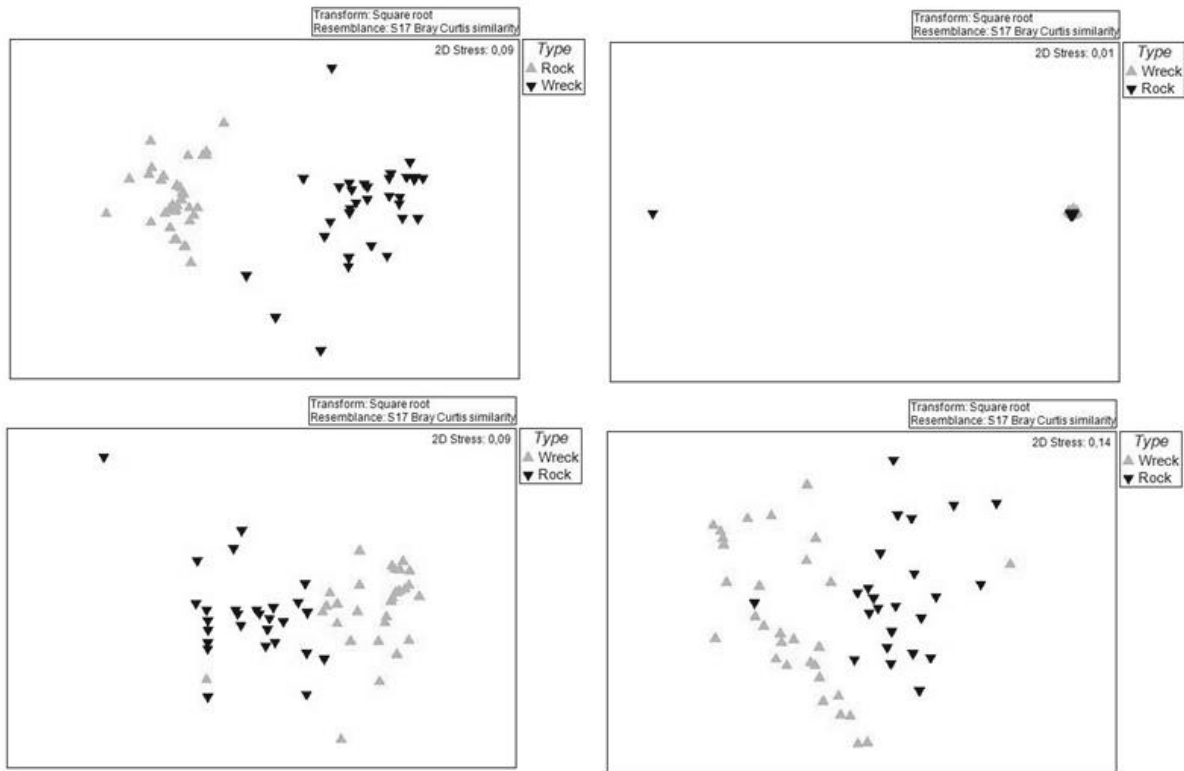


Figure 8 - On the top left: NMDS on the Bray–Curtis square-root-transformed percent covers of benthic organisms in Oliveira e Carmo and Ponta da Piedade; On the top right: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms in Torvore and Falésia (the figure is unclear about the number of samples because they are overlapped; Torvore was sampled with 31 snapshots and Falésia with 29); On the bottom left: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms in Titan and Pedra da Cabela; On the bottom right: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms in River Gurara and Paredes do Cabo.

3.1.3 Zonation within shipwrecks

Integrity

Benthic assemblage densities differed significantly between fragmented and intact structures in one of the shipwrecks (River Gurara, $P = 0.048$) (Table 9). The NMDS shows the transects of the fragmented structures with very little differentiation, constituting just a small part of the total diversity that exists on the intact structures (Figure 9). The SIMPER analysis shows that *Holothuria sp.*, *Actinothoe sphyrodeta*, *Asciidiacea sp.* and *Gorgoniidae* are responsible for most of the differences between intact and fragmented structures in River Gurara. *Holothuria sp.* and *Asciidiacea sp.* were more abundant on the fragmented parts of the shipwreck, while *Actinothoe sphyrodeta* and *Gorgoniidae* were more abundant on the intact parts.

The percent cover data of benthic assemblages were significantly different between intact and fragmented structures in both cases (River Gurara $P = 0.001$, Torvore $P = 0.004$) (Table 10). Both NMDS show some differentiation between fragmented and intact structures assemblages, but in Torvore the transects of the fragmented parts show a higher diversity than in Titan. Also, in both cases, the transects that sampled the fragmented parts of the wrecks seem to only have a fragment of the total diversity that can be found in the intact parts of the structure (Figure 10). The SIMPER analysis shows that Hydroids and algae assemblages and crustose algae still are the taxa responsible for most of the differences, and this will be constant along the protection and orientation analyses.

Table 9 - PERMANOVA tests based on a Bray–Curtis similarity matrix of square-root transformed benthic organisms densities in intact and fragmented structures in River Gurara and Torvore.

| Source | PERMANOVA | | | | | |
|---------------------|-----------|--------|--------|----------|---------|--------------|
| | df | SS | MS | Pseudo-F | P(perm) | Unique perms |
| River Gurara | | | | | | |
| Integrity | 1 | 6831.7 | 6831.7 | 1.9809 | 0.048 | 114 |
| Residuals | 10 | 34487 | 3448.7 | | | |
| Total | 11 | 41319 | | | | |
| Torvore | | | | | | |
| Integrity | 1 | 703 | 703.77 | 0.73424 | 0.68 | 10 |
| Residuals | 4 | 3834 | 958.5 | | | |
| Total | 5 | 4537.8 | | | | |

Table 10 - PERMANOVA tests based on a Bray–Curtis similarity matrix of square-root transformed benthic organisms percent cover in intact and fragmented structures in River Gurara and Torvore.

| Source | PERMANOVA | | | | | |
|---------------------|-----------|--------|--------|----------|---------|--------------|
| | df | SS | MS | Pseudo-F | P(perm) | Unique perms |
| River Gurara | | | | | | |
| Integrity | 1 | 12633 | 12633 | 12.988 | 0.001 | 998 |
| Residuals | 29 | 28206 | 972.63 | | | |
| Total | 30 | 40839 | | | | |
| Torvore | | | | | | |
| Integrity | 1 | 5118.4 | 5118.4 | 6.3074 | 0.004 | 999 |
| Residuals | 29 | 23533 | 811.48 | | | |
| Total | 30 | 28651 | | | | |

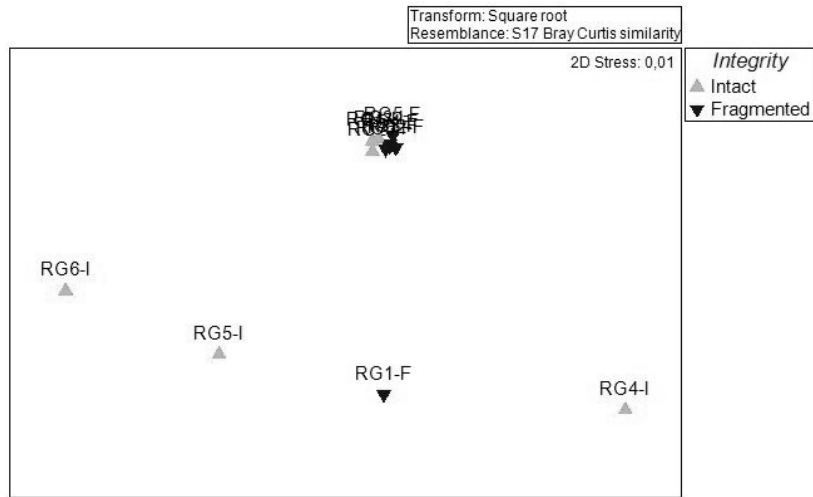


Figure 9 - NMDS on the Bray–Curtis square-root-transformed densities of benthic organisms on intact and fragmented structures in River Gurara.

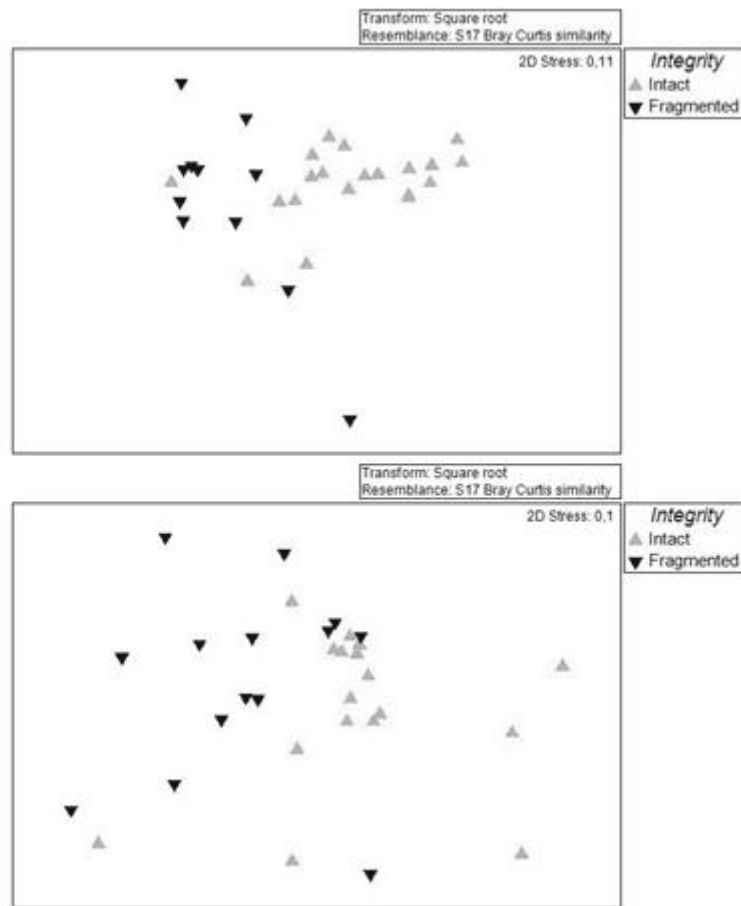


Figure 10 - On top: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms on intact and fragmented structures in River Gurara. On the bottom: NMDS on the Bray–Curtis square-root-transformed percent cover benthic organisms on intact and fragmented structures in Torvore.

Protection

Benthic assemblages densities did not differ significantly between exposed and protected structures in any of the shipwrecks, but the percent cover differed significantly in two of the shipwrecks ($P = 0.002$ in River Gurara; $P = 0.006$ in Titan) (Table 11). The NMDS actually shows that exposed and protected transects do not have much differentiation, but with some observations being clearly apart from the rest of the group (Figure 11).

Table 11 - PERMANOVA tests based on a Bray–Curtis similarity matrix of square-root transformed percent cover of benthic organisms in the exposed and protected sides of the shipwrecks.

| Source | PERMANOVA | | | | | |
|-------------------------|-----------|--------|--------|----------|---------|--------------|
| | df | SS | MS | Pseudo-F | P(perm) | Unique perms |
| Torvore | | | | | | |
| Protection | 1 | 1759.3 | 1759.3 | 2.083 | 0.172 | 999 |
| Residuals | 28 | 23649 | 844.62 | | | |
| Total | 29 | 25409 | | | | |
| River Gurara | | | | | | |
| Protection | 1 | 11346 | 11346 | 11.47 | 0.002 | 999 |
| Residuals | 30 | 29677 | 989.22 | | | |
| Total | 31 | 41022 | | | | |
| Titan | | | | | | |
| Protection | 1 | 3348.1 | 2248.1 | 4.299 | 0.006 | 999 |
| Residuals | 29 | 22585 | 778.81 | | | |
| Total | 30 | 25934 | | | | |
| Oliveira e Carmo | | | | | | |
| Protection | 1 | 67057 | 670.57 | 0.62344 | 0.597 | 998 |
| Residuals | 37 | 39797 | 1975.6 | | | |
| Total | 38 | 40468 | | | | |

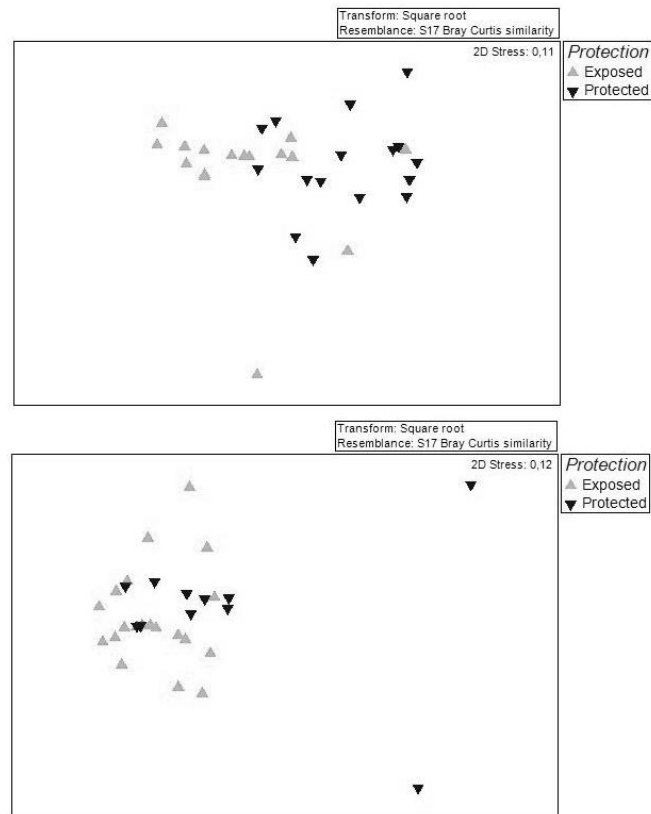


Figure 11 - On top: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms on the exposed and protected sides of River Gurara. On the bottom: NMDS on the Bray–Curtis square-root-transformed percent cover benthic organisms on the exposed and protected sides of Titan.

Structure Orientation

Benthic assemblages densities only differed significantly between horizontal and vertical surfaces in one of the shipwrecks (Oliveira e Carmo, $P = 0.026$). The remaining three shipwrecks did not have significant differences ($P > 0.05$) (Table 12). The NMDS plot (Figure 12) suggests that there is a lot of differentiation between the horizontal and vertical surfaces in Oliveira e Carmo, and the transects of the horizontal surfaces show more diversity than the ones from the vertical surfaces. The SIMPER analysis shows that *Mytilidae*, *Actinothoe sphyrodeta*, *Scorpaena sp.* and *Gorgoniidae* are responsible for most of the differences between horizontal and vertical surfaces in Oliveira e Carmo. *Mytilidae* was much more abundant on vertical surfaces, while the other three were more abundant on horizontal surfaces.

Benthic assemblages analysed as percent covers were significantly different between horizontal and vertical structures in three of the shipwrecks ($P = 0.001$ for River Gurara; $P = 0.031$ in Titan; $P = 0.018$ in Torvore). In Oliveira e Carmo no significant differences were

found ($P = 0.446$) (Table 13). The NMDS plots (Figure 12 and 13) show a clear differentiation between samples of the horizontal and vertical surfaces.

Table 12 - PERMANOVA tests based on a Bray–Curtis similarity matrix of square-root transformed benthic organisms densities in vertical and horizontal surfaces in each shipwreck.

| Source | PERMANOVA | | | | | |
|-------------------------|-----------|--------|--------|----------|---------|--------------|
| | df | SS | MS | Pseudo-F | P(perm) | Unique perms |
| Torvore | | | | | | |
| Orientation | 1 | 2652.9 | 2652.9 | 1.1046 | 0.408 | 10 |
| Residuals | 4 | 9606.6 | 2401.6 | | | |
| Total | 5 | 12260 | | | | |
| River Gurara | | | | | | |
| Orientation | 1 | 4706.3 | 4706.3 | 1.3391 | 0.244 | 312 |
| Residuals | 9 | 31632 | 3514.6 | | | |
| Total | 10 | 36338 | | | | |
| Titan | | | | | | |
| Orientation | 1 | 1364.4 | 1364.4 | 1.0339 | 0.483 | 10 |
| Residuals | 4 | 5278.5 | 1319.6 | | | |
| Total | 5 | 6642.9 | | | | |
| Oliveira e Carmo | | | | | | |
| Orientation | 1 | 4778.4 | 4778.4 | 14.227 | 0.026 | 35 |
| Residuals | 6 | 2105.1 | 335.86 | | | |
| Total | 7 | 6793.5 | | | | |

Table 13 - PERMANOVA tests based on a Bray–Curtis similarity matrix of square-root transformed benthic organisms percent cover in vertical and horizontal surfaces in each shipwreck.

| Source | PERMANOVA | | | | | |
|-------------------------|-----------|--------|--------|----------|---------|--------------|
| | df | SS | MS | Pseudo-F | P(perm) | Unique perms |
| Torvore | | | | | | |
| Orientation | 1 | 3751.7 | 3751.7 | 4.3695 | 0.018 | 999 |
| Residuals | 29 | 24900 | 858.61 | | | |
| Total | 30 | 28651 | | | | |
| River Gurara | | | | | | |
| Orientation | 1 | 17290 | 17290 | 0.92274 | 0.001 | 998 |
| Residuals | 30 | 23733 | 791.09 | | | |
| Total | 31 | 41022 | | | | |
| Titan | | | | | | |
| Orientation | 1 | 3291.6 | 3291.6 | 4.2159 | 0.031 | 985 |
| Residuals | 29 | 22642 | 780.76 | | | |
| Total | 30 | 25934 | | | | |
| Oliveira e Carmo | | | | | | |
| Orientation | 1 | 962.32 | 962.32 | 0.92274 | 0.466 | 998 |
| Residuals | 38 | 39630 | 1042.9 | | | |
| Total | 39 | 40592 | | | | |

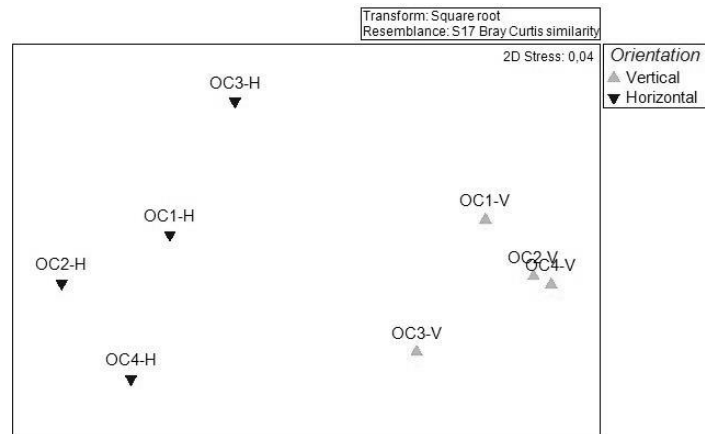


Figure 12 - NMDS on the Bray–Curtis square-root-transformed densities of benthic organisms in horizontal and vertical structures of Oliveira e Carmo.

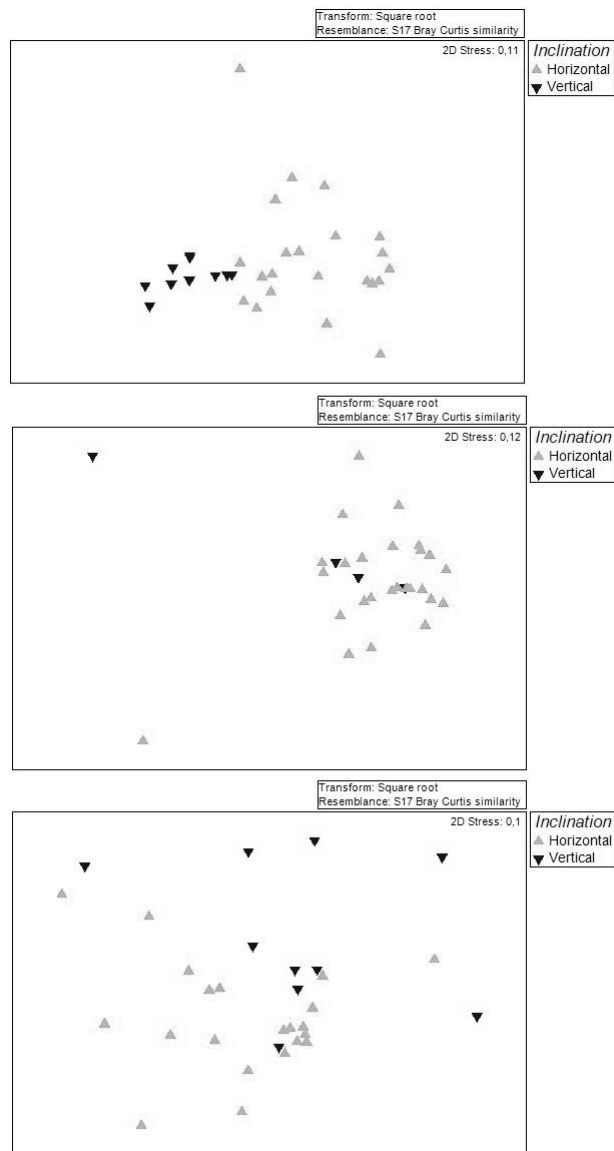


Figure 13 - On top: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms vertical and horizontal surfaces of River Gurara; In the middle: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms vertical and horizontal surfaces of Titan; On the bottom: NMDS on the Bray–Curtis square-root-transformed percent cover of benthic organisms in vertical and horizontal surfaces in Torvore.

4. Discussion

4.1 Comparing shipwreck communities

Hypothesis: *Macrobenthic communities should differ between shipwrecks, with differences related to their different ages and location*

Density data showed that only two shipwrecks had significantly different benthic assemblages: River Gurara (27 years old wreck, 90 m long in the west coast) and Oliveira e Carmo (4 years old, 85 m long in the south coast). Surprisingly, this implies that the rest of the shipwrecks, despite being very distant from each other (up to 300 km approximately), located in different areas subjected to distinct hydrodynamics (García-Lafuente et al. 2006), with different structures and ages, have similar macrobenthic assemblages. These are best represented by bivalves (Mytilidae), fish (*Scorpaena* sp.), echinoderms (*Holothuria* sp.), anemones (*Aiptasia mutabilis* and *Actinothoe sphyrodeta*) and gorgonian corals (Figure 14).

Even though it is possible to explore some explanations for the most relevant community structuring factors, it is not possible to state for sure which factor is the cause of the differences found. At first sight, the fact that these two wrecks, River Gurara and Oliveira e Carmo have significant differences between each other could be related to their distance and location, because one of the wrecks, River Gurara, is on the west coast and the other, Oliveira e Carmo, on the south coast. Yet, River Gurara is not significantly different from the other shipwrecks located on the south coast. Another possible explanation is that the age of the shipwrecks has a structuring effect in the community, since younger wrecks should be at an earlier stage in succession, dominated by species that settle and establish quickly, while older wrecks should be found at a mature successional stage with longer-lived species and higher diversity (Perkol-Finkel et al. 2006; Walker et al. 2007; Burt et al. 2011). Oliveira is only four years old while River Gurara is 27, and from all the shipwrecks on the south coast, River is only different from the youngest one. The reason the rest of the shipwrecks in the south coast are not different from Oliveira can be because they are much more geographically closer and all are influenced by the Portuguese–Canary eastern boundary current (García-Lafuente et al. 2006). But it is also important to consider that until recently, Torvore (southwest coast) and Titan (southeast coast) were in areas where trawl fishing was allowed. This extremely destructive fishing method (Eastwood et al. 2007, Foden et al. 2011) may have caused several setbacks on the normal successions of the colonization process of these

boats, and this may explain the absence of significant differences of the wrecks' assemblages with the very young assemblages of Oliveira e Carmo. There are previous studies that show how physical disturbances can work as an obstacle for normal colonization and succession (e.g. Cummings 1994).



Figure 14 - The best represented taxonomical groups in the shipwrecks (from top to bottom, left to right): Mytilidae, *Scorpaena* sp., *Holothuria* sp., *Aiptasia mutabilis*, Gorgoniidae and *Actinothoe sphyrodeta*.

The hypothesis that shipwrecks would have different communities from each other was not supported. This can be due to a low resolution in the identification of some of the taxa (mainly gorgonias, sea cucumbers, mussels and sponges). Sometimes incorrect lighting and an inappropriate filming angle caused difficulties in the identification of the species in the

image. The sampling method used, video-transects, works perfectly with conspicuous species (corals, polychaets, echinoderms), but it has some limitations with smaller or incrusting species (algae or sponges). Overall, the limitations are related with the level of detail required for the identification of a species. These limitations can be increased with poor image quality of the videos (usually related with the camera itself) and with the diver inexperience in filming underwater. Also, comparing different artificial reefs is hard due to the multiple factors that vary simultaneously (for example: age of the reef, location, habitat complexity). An ideal scenario would involve a large number of replicated reefs (Svane and Petersen 2001), in which only one factor would vary at a time, but this is not a possibility when working with real situations.

Hypothesis: Biodiversity should increase with the shipwrecks age due to the more advanced state of succession in the colonization process

Focusing on the age effect, it was expected that older shipwrecks would have a community more diverse (Ardizzone et al. 1989), but the taxonomic distinctness index and the alpha diversity had very similar values for all the shipwrecks except for Titan (smallest, southeast coast), which has a significantly lower taxonomic distinctness index. This sets aside the hypothesis that with age, the biodiversity of artificial reefs increases due to a more advanced state of succession in the colonization process.

It is important to take into consideration that, within a certain range (distance, location, age), the type of reef may have a stronger impact on biodiversity than anything else. Artificial reefs are “island-habitats” (Svane and Petersen 2001); the theory of island biogeography (MacArthur & Wilson, 1967) has been used before to test succession and recruitment in artificial reefs (Cummings, 1994). This theory states that in an isolated site the number of species present should be a result of emigration and extinction rates and so it explains the rates and mechanisms of colonization of new habitats. The first step in the colonization process is influenced by the reef size and by distance to the reefs of the source populations. As species accumulate on the new reef, the colonization rates decrease and some species start become extinct until a dynamic equilibrium is reached. This is probably why older shipwrecks did not have more biodiversity: after a certain period of time (the time needed for the dynamic equilibrium to be reached), age does not impact biodiversity anymore. The reef size impact was not analyzed in this study due to the complexity of assessing the area of influence of the

shipwrecks. The influence of the distance to the reefs of the source populations is analyzed on the following chapter of this discussion.

All shipwrecks had significantly different benthic assemblages, but with an Oliveira e Carmo assemblage less distant than in the density analysis. All shipwrecks had present the same algae morphological groups and encrusting fauna but with different abundances, and those different abundances were probably what caused the differences. These groups of organisms did not show any clear pattern of abundance or distribution other than having a higher number of algae morphological groups on the shallower wrecks (River Gurara and Titan). The analysis with percent cover data had results that did not allow for any major conclusions. The low resolution on the identification of the taxa in these groups of organisms may have been a setback on the analysis. An identification to species or genus level could have led to more conclusive results.

4.2 Comparing shipwrecks with natural rocky reefs

Hypothesis: *Shipwrecks older than eight years should have communities similar to the ones on natural rocky reefs*

There is a general lack of research comparing the biodiversity of artificial and natural reefs. This is worrisome because the use of artificial reefs is a frequent compensatory measure for habitat loss, coastal protection and recreational activities (Seaman Jr. & Jensen 2000).

Most of the species found exclusively in the shipwrecks were small benthic fishes (Table 14). This can be related with the type of structure. Vertical structures in artificial reefs are known to be more attractive for the settlement and recruitment of fish larvae than the moderately sloped bottoms usually found on natural reefs (Rilov and Benayahu, 2000). The high rugosity of artificial reefs increases the availability of refuge from predators (Sherman et al., 2002), increasing the survival rates of juveniles (Simon et al. 2011).

Submerged artificial structures are commonly colonized by nonindigenous species (Glasby et al. 2007). These invasions can threaten the native marine biodiversity, as nonindigenous species are ranked as the second cause for habitat loss (USGS, 2008). Fortunately, according to the Global Invasive Species Database (www.iucngisd.org), none of these species here recorded as being present exclusively on the shipwrecks is an invasive species.

Table 14 - Density of the taxa found exclusively in shipwrecks

| Taxa | Torvore | River Gurara | Titan | Oliveira e Carmo |
|--|---------|--------------|-------|------------------|
| Actinopterigi spp. 1 | 0 | 0 | 0 | 0.01 |
| <i>Actinothoe sphyrodeta</i> | 0 | 0.34 | 59.57 | 2.91 |
| Alcyonium sp. | 0 | 0.02 | 0 | 0 |
| <i>Alicia mirabilis</i> | 0 | 0.02 | 0.02 | 0.01 |
| <i>Cancer pagurus</i> | 0 | 0 | 0.02 | 0 |
| <i>Cerianthus membranaceus</i> | 0.11 | 0 | 0 | 0 |
| <i>Conger conger</i> | 0.07 | 0 | 0.02 | 0 |
| <i>Diaphorodoris luteocincta</i> | 0 | 0.01 | 0 | 0 |
| <i>Echinaster (Echinaster) sepositus</i> | 0.23 | 0 | 0 | 0 |
| Gobius spp. 1 | 0 | 0.02 | 0 | 0 |
| Mytilidae | 0 | 0 | 0 | 31.52 |
| <i>Parablennius gattorugine</i> | 0 | 0 | 0 | 0.01 |
| <i>Parablennius pilicornis</i> | 0 | 0 | 0.02 | 0 |
| Parablennius sp. | 0 | 0 | 0.04 | 0.06 |
| <i>Paramuricea clavata</i> | 0.06 | 0 | 0 | 0 |
| <i>Scorpaena notata</i> | 0 | 0 | 0.02 | 0 |

Based on previous studies (Burt et al. 2011; Perkol-Finkel et al. 2006) it was expected that shipwrecks older than eight years would have communities similar to the rocky reef ones. The percent cover analysis indicated that all shipwrecks were significantly different from the nearby rocky reefs, while density points to only two shipwrecks (River and Oliveira) bearing assemblages different from the reefs. The consistent differences found for percent cover data are likely related to the type of organisms that this analysis includes (incrusting macrofauna and a majority of algae). Algae growth is strongly influenced by light intensity, and some natural reefs sampled were significantly shallower than the wrecks (15 m shallower on average), having a higher light intensity, which should have an impact on the algae community. The density analysis showed that Torvore had a similar community to the nearby rocky reefs, as Titan did too. As the study previously mentioned predicted, artificial reefs with ages from 8 to 25 years should have communities that are similar to the nearby rocky reefs. Torvore is 99 years old, Titan is 14 and Oliveira e Carmo, which had different assemblages from its nearby rocky reef, is 4. For these cases, the results are in agreement with the previous research (shipwrecks older than eight years have communities similar to the

ones on the natural reefs, and shipwrecks under eight years do not). River Gurara seems to be the exception. This 27 years old wreck was the closest to the respective sampled rocky reef and still the assemblages were different. A possible explanation is the influence of the hydrodynamic forces in this particular area. The shallower reef had a much lower biodiversity than the shipwreck, which being deeper is more protected from the waves impact. A previous study on the south coast of Portugal discovered a gradient of decreasing hydrodynamic forces with increasing depth (from 1.2 to 32 m depth), with a reverse gradient happening with biodiversity: increasing diversity with increasing depths (Dolbeth et al. 2007). This may explain the differences with River Gurara, even though the wreck is not on the south but on the west coast, hydrodynamics should have a structuring role with communities in that area too.

Forty percent of the taxa of the shipwrecks do not come from the nearby rocky reefs. When comparing each shipwreck with its nearby natural reef individually, it seems like the older the shipwreck the more in common it has with its surrounding biodiversity. Oliveira e Carmo (four years) only has 20% of its diversity present in the natural reefs, while Titan (14 years) has 41%, River Gurara (27 years) has 25% and Torvore (99 years) has 44%. The next attempt was to compare the samples of the shipwrecks with available online databases of biodiversity for the south coast areas of Sagres, Lagos and Portimão (Boavida et al. 2016 a,b). For one of these studies, the samples were recorded between 40 and 70 m depth. The studied shipwrecks had 60% of taxa in common with the deep rocky reefs, therefore having the same 40% of wrecks' taxa that were not present in local deep reefs. For the other study, the samples were recorded between 60 and 100 m, and only 30% of the taxa found in the shipwrecks were present in the deep rocky reefs.

Considering data from shallow and deep reefs combined, there is apparently 40 to 70% of the shipwrecks biodiversity that cannot be found in local natural reefs. This may be related with a depth gradient, but it is not possible to test this hypothesis because so far there is no record of sampled natural reefs in these areas at closer depths to the shipwrecks. Another possibility is that artificial structures just do not have similar communities to the ones found in natural reefs. Even though outdated, there is plenty of research suggesting that recruitment in subtidal assemblages is localized within sites (Stoner, 1992; Petersen and Svane, 1995; Osman and Whitlatch, 1998). Local recruitment patterns in these studies suggest that subtidal assemblages may be viewed as closed or semi-closed systems.

This thesis shows unexpected results, because a large percentage of the shipwrecks' biodiversity does not come from local biodiversity pools, but when compared to the few studies that compare artificial with natural biodiversity, there is no record of an artificial reef that has a similar community as other local natural reefs (Clark and Edwards, 1994; Glasby 1997; Glasby and Connel, 1999, Carvalho et al. 2013).

Based on this thesis results and on previous published research, even though artificial reefs cannot mimic natural biodiversity, they may still have a positive impact in the ecosystem by actually increasing local biodiversity, because at least 40% of the shipwrecks diversity does not come from a local pool. Some studies raise the hypothesis that artificial reef do not actually increase biodiversity, they just relocate and agglomerate local populations in them (Grossman et al. 1997; Brickhill et al. 2005), but if part of the community does not come from a local pool, then apparently this is not the case. The increase in the general biodiversity for the region was also not related to colonization of the wrecks by invasive species.

4.3 Zonation within shipwrecks

The few studies done about shipwrecks do not mention how well preserved the wreck is. This was the first time that the impact of the ship's structures integrity on biodiversity was analysed.

Titan and Oliveira e Carmo, the youngest shipwrecks, were still almost completely intact (Oliveira e Carmo has lost a third of its structure, but the part that remains is in perfect condition). The two older shipwrecks, Torvore and River Gurara, had part of their structures completely collapsed, while other parts were still completely intact. Therefore, only the oldest wrecks were used to assess the impact of structures integrity on biodiversity. Overall, there is a difference in the type of assemblages found in the fragmented and intact parts of the shipwrecks, except for density of Torvore. As mentioned previously, for many years Torvore was in a fishing area. It was common for divers to find this wreck full of fishing nets, so fishing probably had a very damaging effect on the biodiversity of this wreck for many years. This damage may be the reason why there is no major difference between assemblages of intact and fragmented parts in this wreck: both were damaged for many years. Only recently fishing stopped being allowed in this area, so it would be relevant to monitor the succession of the colonization of this wreck during the following years.

Overall, the intact parts of the wrecks harbour more biodiversity than the fragmented parts. The diversity found in the fragmented parts of the wrecks actually seems to represent only a

small fraction of the total diversity present on the intact structures. *Actinothoe sphyrodeta* and *Gorgoniidae* were more abundant on the intact parts of the wrecks, while *Holothuria sp.* and *Ascidiacea sp.* were more abundant on the fragmented parts. The intact parts of the wreck are usually big and tall structures that may change currents and hydrodynamics in a convenient way for passive filter feeders such as gorgonians and anemone. The fragmented parts of the wrecks are usually a habitat with more rugosity and more horizontal, leading to a higher sedimentation rate which is convenient for organisms like *Holothurias* which feed on sediment.

The comparison between exposed and protected sides (in relation to sea currents) did not show any clear patterns, even though there were differences in a minority of the cases. While performing the dives, there were some differences between exposed and protected sides in some of the shipwrecks which were seen, but the overall assemblages did not change significantly between sides of the wrecks. It is possible that the level of protection/exposure to sea currents does not change a lot between sides, at least in these cases, and the colonization process happens more or less homogeneously.

In half of the cases, there were no differences between assemblages in horizontal and vertical surfaces on the shipwrecks. According to previous studies (Glasby 2000), it was expected that vertical surfaces would harbour more biodiversity than the horizontal surfaces due to a higher sedimentation rate on horizontal surfaces which leads to smothering of filter feeders and prevents settlement of young recruits. That was apparently not the case here, and actually in Oliveira e Carmo (which was the only wreck with significantly different densities), the abundance of *Mytilidae* (filter feeders) was significantly higher on horizontal surfaces.

The studies focusing on these comparisons were made in concrete cubs, which have a clear differentiation between their horizontal and vertical structures. This is not necessarily true for shipwrecks. The differentiation between horizontal and vertical structures is not always so obvious, because many times the structures are partially collapsed, so both vertical and horizontal surfaces have high rugosity and a lot of niches, probably having more in common as an habitat than different.

4.4 Conclusion

This study was the first survey of the shipwrecks and consequently the first comparison between shipwrecks and natural communities in Portugal. It was also the first simultaneous survey of multiple artificial reefs at such a large geographical scale. It is extremely important

that this type of data are made available to scientists, managers and the general public, easily accessible to everyone so they can be continuously used for research. One of the major outcomes of this thesis was a biodiversity “baseline” database, which can quickly be improved through citizen science projects. This is a pioneering initiative that is expected to allow the research in this thesis to be used widely and to continue in the future as a citizen initiative. One of the major goals was to establish a volunteer project in these shipwreck that are already some of the most popular diving sites in the country. Using an online platform where volunteers can upload their pictures, the point is to create a continuous record of the communities changes during different seasons along the years, eventually allowing for a study about long term colonization processes on shipwrecks. This project is already being developed, using the Project Baseline platform. Project Baseline is a global, aquatic conservation initiative that intends to establish environmental underwater baselines through divers volunteer work, using images. The goal of Project Baseline is to increase public awareness about the state of water now and in the future (more information in www.projectbaseline.org).

Also, extending this project to a broader geographical scale would be relevant to assess the impacts that shipwrecks have on local biodiversity in different types of locations (from the Mediterranean Sea to the Baltic Sea for example). Artificial reefs are used with different purposes in different places, but the studies usually focus on concrete cubs and similar kinds of structures. Focusing on shipwrecks that have a higher importance for the diving community across the world, therefore have a bigger economical impact would be even more important than focusing on the concrete units, because shipwrecks associate their economical impact with a biodiversity conservation impact. An overall update of research in this field is needed, because it was very hard to find bibliography focusing on artificial reefs which was not extremely outdated.

From the hypotheses previously mentioned, there were some surprising results. Overall, the macro benthic communities of the shipwrecks along the Portuguese coastline are similar between each other, but not always similar to the natural communities. The age of the shipwreck most probably has an impact on biodiversity, but only until a certain point. This pattern might indicate that after the maximum of biodiversity is reached, the community develops a dynamic equilibrium in which while some species become extinct new species colonize the habitat.

Shipwreck communities have a big fraction that does not seem to come from local natural reefs. This can be related with insufficient sampling, but it is possible that shipwrecks lead to an increase in local biodiversity due to their availability to be colonized by propagules from populations coming from distinct sources other than the local reefs.

Zonation within shipwrecks did not show many clear patterns other than the comparison between intact and fragmented structures. The intact parts of the shipwrecks are able to harbour more biodiversity. It was evident from the data that a good conservation of the shipwreck condition leads to healthier communities.

Overall, these shipwrecks are extremely important sites in the diving industry, and even have the advantage of relieving pressure from the natural reefs which are also used as diving sites. The fact that they are frequently visited by divers does not seem to have a destructive impact on their communities, and it is even possible that shipwrecks can actually increase local biodiversity. Therefore, there should be a bigger focus on shipwreck biodiversity (worldwide, not only in Portugal), with the purpose of trying to explore and increase their economical and conservational value as much as possible.

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Appendixes

SIMPER Tables

Table 1 Simper analysis on organisms densities in River Gurara and Oliveira e Carmo, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | RG | OC | RG-OC |
|-----------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| <i>Mytilidae</i> | 0.00 | 5.58 | 55.58% |
| <i>Actinothoe shyrodeta</i> | 0.41 | 1.68 | 12.92% |
| <i>Holothuria sp.</i> | 0.47 | 0.00 | 4.83% |
| <i>Gorgoniidae</i> | 0.32 | 0.12 | 3.48% |
| <i>Scorpaena sp.</i> | 0.00 | 0.28 | 2.90% |
| <i>Aiptasia mutabilis</i> | 0.26 | 0.00 | 2.36% |
| <i>Asciacea sp.</i> | 0.20 | 0.00 | 2.00% |

Table 2 Simper analysis on organisms percentage covers in River Gurara and Oliveira e Carmo, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | RG | OC | RG-OC |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Crustose Algae | 4.52 | 0.26 | 22.11% |
| <i>Balanidae</i> | 0.04 | 4.21 | 21.19% |
| Hydroids + Algae assemblages | 3.89 | 2.01 | 16.62% |
| <i>Porifera</i> | 0.00 | 2.59 | 13.07% |
| <i>Cnidaria</i> | 0.00 | 0.86 | 4.30% |

Table 3 Simper analysis on organisms percentage covers in Titan Oliveira e Carmo, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | OC | TI | OC-TI |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| <i>Balanidae</i> | 4.21 | 0.63 | 21.74% |
| Hydroids + Algae assemblages | 2.01 | 6.40 | 26.46% |
| <i>Porifera</i> | 2.59 | 0.00 | 14.71% |
| <i>Cnidaria</i> | 0.86 | 0.00 | 48.84% |

Table 4 Simper analysis on organisms percentage covers in Torvore and Oliveira e Carmo, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | OC | TO | OC-TO |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Crustose Algae | 0.26 | 3.83 | 19.46% |
| <i>Balanidae</i> | 4.21 | 0.04 | 22.96% |
| Hydroids + Algae assemblages | 2.01 | 3.41 | 14.00% |
| <i>Porifera</i> | 2.59 | 0.00 | 14.17% |
| <i>Bryozoa</i> | 0.00 | 1.10 | 5.89% |

Table 5 Simper analysis on organisms percentage covers in River Gurara and Titan, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | RG | TI | RG-TI |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Crustose Algae | 4.56 | 0.68 | 22.84% |
| <i>Balanidae</i> | 0.04 | 0.63 | 3.28% |
| Hydroids + Algae assemblages | 3.89 | 6.40 | 20.75% |
| Corticated Foliose Algae | 0.45 | 0.14 | 3.21% |
| Filamentous Algae | 0.40 | 0.00 | 2.05% |

Table 6 Simper analysis on organisms percentage covers in River Gurara and Torvore, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | RG | TO | RG-TO |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Crustose Algae | 4.86 | 3.83 | 14.18% |
| Hydroids + Algae assemblages | 3.89 | 3.41 | 15.85% |
| Corticated Foiose Algae | 0.45 | 0.00 | 2.66% |
| Filamentous Algae | 0.40 | 0.00 | 1.98% |
| <i>Bryozoa</i> | 0.21 | 1.10 | 6.18% |

Table 7 Simper analysis on organisms percentage covers in Titan and Torvore, with the average abundance on each one and the percentage of dissimilarities.

| | TI | TO | TI-TO |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Crustose Algae | 0.68 | 3.83 | 20.02% |
| <i>Balanidae</i> | 0.63 | 0.04 | 2.60% |
| Hydroids + Algae assemblages | 6.40 | 3.41 | 21.48% |
| <i>Bryozoa</i> | 0.00 | 1.10 | 6.64% |

Table 8 Simper analysis on organisms densities in River Gurara and Paredes do Cabo, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Rock | Shipwreck | RG-PC |
|--------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| <i>Paracentrotus lividus</i> | 1.38 | 0.00 | 37.41% |
| <i>Actinothoe sphyrodeta</i> | 0.00 | 0.41 | 8.79% |
| <i>Gorgoniidae</i> | 0.00 | 0.32 | 7.08% |
| <i>Holothuria sp.</i> | 0.44 | 0.47 | 6.31% |
| <i>Ascidacea sp.</i> | 0.00 | 0.20 | 5.40% |
| <i>Aiptasia mutabilis</i> | 0.00 | 0.26 | 5.24% |
| <i>Diaphodoris luteocincta</i> | 0.12 | 0.04 | 3.00% |

Table 9 Simper analysis on organisms densities in Oliveira e Carmo and Ponta da Piedade, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Rock | Shipwreck | OC-PP |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| <i>Paracentrotus lividus</i> | 5.58 | 0.00 | 7.97% |
| <i>Actinothoe sphyrodeta</i> | 1.68 | 1.09 | 14.14% |
| <i>Holothuria sp.</i> | 0.08 | 0.00 | 5.91% |
| <i>Mytilidae</i> | 5.58 | 1.05 | 47.02% |
| <i>Anemonia viridis</i> | 0.00 | 5.91 | 15.45% |

Table 10 Simper analysis on organisms percentage cover in River Gurara and Paredes do Cabo, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Rock | Shipwreck | OC-PP |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids + Algae assemblages | 3.89 | 3.63 | 13.93% |
| Crustose Algae | 4.56 | 2.34 | 13.61% |
| Leathery macrophytes | 0.34 | 2.83 | 13.00% |
| Corticated Foliose | 0.45 | 3.02 | 12.74% |

Table 11 Simper analysis on organisms percentage cover in Titan and Pedra da Cacela, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Rock | Shipwreck | TI-PC |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids + Algae assemblages | 6.40 | 1.68 | 35.53% |
| Crustose Algae | 0.68 | 3.66 | 22.72% |
| <i>Balanidae</i> | 0.63 | 0.12 | 4.41% |

Table 12 Simper analysis on organisms percentage cover in Torvore and Falésia, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Rock | Shipwreck | TO-FA |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids + Algae assemblages | 3.41 | 1.91 | 14.74% |
| Crustose Algae | 3.83 | 5.68 | 16.21% |
| <i>Bryozoa</i> | 1.10 | 0.10 | 6.82% |

Table 13 Simper analysis on organisms percentage cover in Oliveira e Carmo and Ponta da Piedade, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Rock | Shipwreck | OC-PP |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids + Algae assemblages | 2.47 | 2.01 | 9.62% |
| Crustose Algae | 3.34 | 0.26 | 12.53% |
| Corticated Macrophytes | 6.65 | 0.00 | 28.59% |
| Balanidae | 0.00 | 4.21 | 17.90% |
| Porifera | 0.43 | 2.59 | 10.18% |

Table 14 Simper analysis on organisms densities in intact and fragmented structures in River Gurara, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Intact | Fragmented | RG |
|--------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| <i>Holothuria sp.</i> | 0.7 | 2.19 | 30.30% |
| <i>Actinothoe sphyrodeta</i> | 1.51 | 0.29 | 14.14% |
| <i>Asciacea sp.</i> | 0.33 | 0.83 | 11.34% |
| <i>Gorgoniidae</i> | 1.35 | 0.00 | 9.70% |
| <i>Gobius cruentatus</i> | 0.17 | 0.33 | 7.01% |
| <i>Aiptasia mutabilis</i> | 0.41 | 0.17 | 4.72% |
| <i>Diaphodoris luteocincta</i> | 0.00 | 0.17 | 2.84% |

Table 15 Simper analysis on organisms percentage cover in intact and fragmented structures in Torvore, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Intact | Fragmented | TO |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Crustose algae | 3.09 | 4.73 | 14.85% |
| Hydroids + algae assemblages | 3.85 | 2.87 | 12.37% |
| <i>Bryozoa</i> | 0.48 | 1.85 | 10.78% |

Table 16 Simper analysis on organisms percentage cover in intact and fragmented structures in River Gurara, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Intact | Fragmented | RG |
|--------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids and algae assemblages | 5.43 | 0.88 | 24.39% |
| Crustose algae | 3.87 | 5.86 | 16.01% |
| Corticated foliose | 0.31 | 0.73 | 4.91% |
| Leathery macrophytes | 0.00 | 1.00 | 4.28% |
| Filamentous algae | 0.33 | 0.57 | 3.62% |

Table 17 Simper analysis on organisms percentage cover in the exposed and protected sides in Titan, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Exposed | Protected | TI |
|--------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids and algae assemblages | 6.82 | 5.64 | 19.94% |
| Crustose algae | 0.28 | 1.42 | 9.15% |
| <i>Balanidae</i> | 0.80 | 0.34 | 5.70% |

Table 19 Simper analysis on organisms percentage cover in the exposed and protected sides in River Gurara, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Exposed | Protected | RG |
|--------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids and algae assemblages | 5.57 | 2.21 | 20.74% |
| Crustose algae | 3.22 | 5.91 | 16.67% |
| Corticated foliose algae | 0.43 | 0.47 | 4.19% |
| Filamentous algae | 0.08 | 0.72 | 3.57% |
| Leathery Macrophytes | 0.00 | 0.69 | 3% |

Table 20 Simper analysis on organisms densities in vertical and horizontal structures in Oliveira e Carmo, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Vertical | Horizontal | OC |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| <i>Mytilidae</i> | 5.21 | 1.95 | 32.86% |
| <i>Actinothoe sphyrodeta</i> | 0.49 | 1.30 | 11.89% |
| <i>Scorpaena sp.</i> | 0.06 | 0.27 | 2.62% |
| <i>Gorgoniidae</i> | 0.12 | 1.25 | 0.95% |

Table 21 Simper analysis on organisms percentage cover in vertical and horizontal structures in Torvore, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Vertical | Horizontal | TO |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids + Algae assemblages | 3.42 | 3.37 | 12.37 |
| Crustose algae | 4.59 | 2.24 | 17.97 |
| <i>Bryozoa</i> | 1.11 | 1.07 | 9.40 |

Table 22 Simper analysis on organisms percentage cover in vertical and horizontal structures in River Gurara, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Horizontal | Vertical | RG |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids + Algae assemblages | 2.42 | 7.12 | 44.40% |
| Crustose algae | 5.63 | 2.22 | 34.55% |
| Corticated foliose algae | 0.65 | 0.00 | 6.54% |
| Filamentous algae | 0.50 | 0.00 | 4.92% |

Table 23 Simper analysis on organisms percentage cover in vertical and horizontal structures in Titan, with the average abundance of the taxa that are responsible for differences in the communities and the percentage of dissimilarities that each taxa causes.

| | Horizontal | Vertical | TI |
|------------------------------|-------------------|-------------------|-----------------|
| Species | Average Abundance | Average Abundance | Dissimilarities |
| Hydroids + Algae assemblages | 6.62 | 4.48 | 56.96% |
| Crustose algae | 0.55 | 1.61 | 26.12% |
| Balanidae | 0.73 | 0.00 | 10.53% |

PERMDISP Tables

Table 24 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms densities in the shipwrecks

| Deviations from Centroid | | | |
|---------------------------|-------|---------|------|
| F= 20,147 | df1=3 | df2=12 | |
| P(perm)=0,002 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Torvore | 3 | 23,17 | 5,62 |
| River Gurara | 6 | 44,11 | 2,84 |
| Titan | 3 | 26,91 | 4,70 |
| Oliveira e Carmo | 4 | 10,46 | 0,57 |

Table 25 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percentage cover in shipwrecks

| | | | |
|---------------------------|------|---------------|------|
| Deviations from Centroid | | | |
| F= 1,3755 | | df1=3 df2=130 | |
| P(perm)=0,331 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Oliveira e Carmo | 40 | 28,10 | 2,40 |
| River Gurara | 32 | 32,07 | 2,87 |
| Titan | 31 | 24,44 | 2,83 |
| Torvore | 31 | 26,22 | 2,81 |

Table 26 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms densities in River Gurara and Paredes do Cabo.

| | | | |
|---------------------------|------|-------------|-------|
| Deviations from Centroid | | | |
| F= 56,297 | | df1=1 df2=6 | |
| P(perm)=0,043 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Wreck | 2 | 5,2154 | 0 |
| Rock | 6 | 44,114 | 2,837 |

Table 27 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms densities in Oliveira e Carmo and Ponta da Piedade.

| | | | |
|---------------------------|------|-------------|---------|
| Deviations from Centroid | | | |
| F= 5,9541 | | df1=1 df2=5 | |
| P(perm)=0,036 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Wreck | 4 | 10,46 | 0,57046 |
| Rock | 3 | 23,701 | 6,4355 |

Table 28 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percentage cover in River Gurara and Paredes do Cabo.

| | | | |
|---------------------------|------|--------------|------|
| Deviations from Centroid | | | |
| F= 1,8168 | | df1=1 df2=54 | |
| P(perm)=0,228 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Wreck | 32 | 32,069 | 2,87 |
| Rock | 24 | 26,464 | 2,89 |

Table 29 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percentage cover in Titan and Pedra da Cacela.

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 0,61672 | | df1=1 | df2=58 |
| P(perm)=0,528 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Wreck | 31 | 24,427 | 2,825 |
| Rock | 29 | 27,696 | 3,0492 |

Table 30 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percentage cover in Torvore and Falésia.

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F=3,7808 | | df1=1 | df2=59 |
| P(perm)=0,1 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Wreck | 31 | 26,223 | 2,808 |
| Rock | 30 | 18,429 | 2,8606 |

Table 31 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percentage cover in Oliveira e Carmo and Ponta da Piedade.

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F=5,2956 | | df1=1 | df2=72 |
| P(perm)=0,1 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Wreck | 34 | 21,215 | 1,5886 |
| Rock | 40 | 28,099 | 2,4033 |

Table 31 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms densities in River Gurara intact and fragmented structures

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 2,501 | | df1=1 | df2=16 |
| P(perm)=0,265 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Intact | 9 | 55,538 | 5,1413 |
| Fragmented | 9 | 43,781 | 5,366 |

Table 33 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in River Gurara intact and fragmented structures

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 0,96415 | | df1=1 | df2=29 |
| P(perm)=0,434 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Intact | 20 | 23,32 | 3,4785 |
| Fragmented | 11 | 29,401 | 5,4734 |

Table 34 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in Torvore intact and fragmented structures

| | | | |
|----------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 4,0727x10 ⁻² | | df1=1 | df2=29 |
| P(perm)=0,844 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Intact | 17 | 22,657 | 3,9525 |
| Fragmented | 14 | 23,791 | 3,9117 |

Table 35 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in Oliveira e Carmo horizontal and vertical surfaces

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 0,269 | | df1=1 | df2=6 |
| P(perm)=0,265 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Vertical | 4 | 12,413 | 2,2996 |
| Horizontal | 4 | 17,114 | 3,6941 |

Table 36 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in Titan horizontal and vertical surfaces

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 0,53892 | | df1=1 | df2=29 |
| P(perm)=0,611 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Vertical | 27 | 22,592 | 2,3022 |
| Horizontal | 4 | 28,149 | 13,125 |

Table 37 PERMDIP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in River Gurara horizontal and vertical surfaces

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 16,804 | | df1=1 | df2=30 |
| P(perm)=0,001 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Vertical | 22 | 29,136 | 2,6685 |
| Horizontal | 10 | 11,92 | 1,9665 |

Table 38 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in Torvore horizontal and vertical surfaces

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 1,5026 | | df1=1 | df2=29 |
| P(perm)=0,205 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Vertical | 21 | 22,467 | 2,9761 |
| Horizontal | 10 | 29,134 | 4,8215 |

Table 39 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in Titan exposed and protected sides.

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 0,91045 | | df1=1 | df2=29 |
| P(perm)=0,395 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Exposure | 20 | 20,783 | 2,011 |
| Protected | 11 | 26,121 | 6,6776 |

Table 40 PERMDISP – based on a Bray–Curtis similarity matrix of square-root transformed organisms percent cover in River Gurara exposed and protected sides.

| | | | |
|---------------------------|------|---------|--------|
| Deviations from Centroid | | | |
| F= 5,8051x10-2 | | df1=1 | df2=30 |
| P(perm)=0,865 | | | |
| Means and standard errors | | | |
| Group | Size | Average | SE |
| Exposure | 16 | 25,808 | 5,1036 |
| Protected | 16 | 27,133 | 2,0508 |

Figures

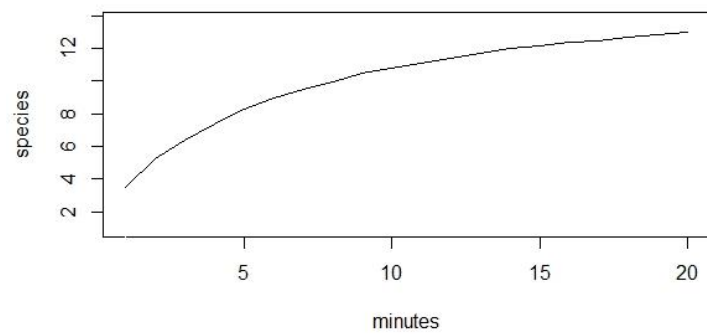


Figure 1 Species accumulation curve for the taxa of River Gurara, considering the number of new taxa per minute of video. After 10 minutes of video, the number of new species appearing per minute is smaller.

Species Catalog

Actinopterygii i spp. 1



Actinothoe sphyrodeta



Aiptasia mutabilis



Alcyonium sp.



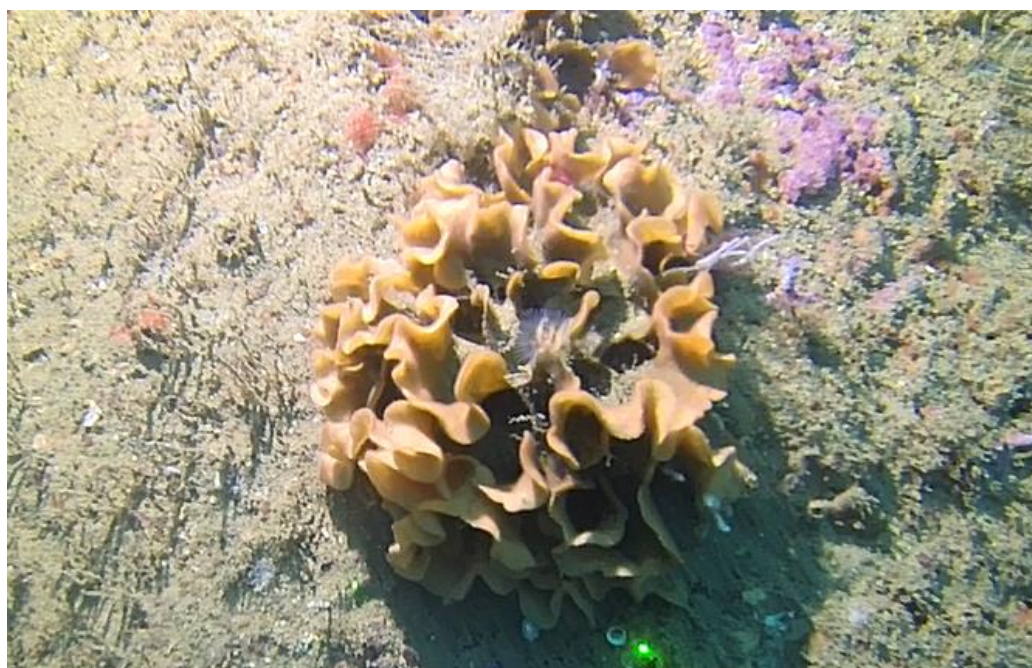
Alicia mirabilis



Asciacea sp.



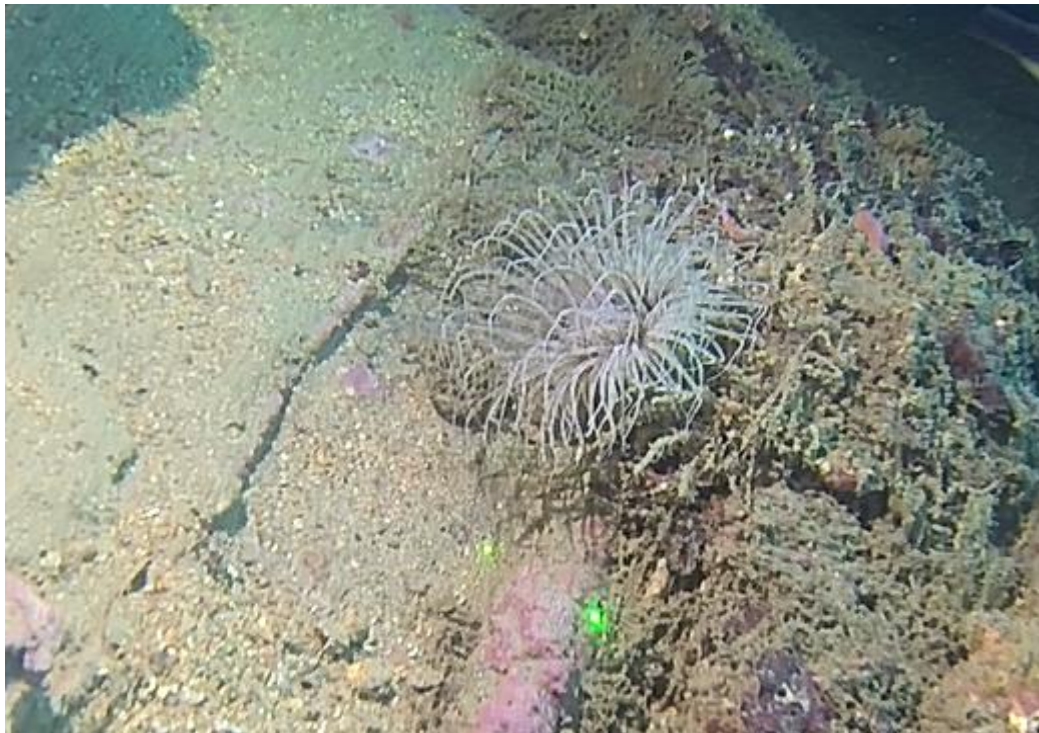
Bryozoa



Cancer pagurus



Cerianthus membranaceus



Conger conger



Diaphorodoris sp.



Echinaster (Echinaster) sepositus



Felimare sp.



Gobius sp.



Eunicela spp. 1 and *Eunicella spp. 2*



Leptogorgia Sarmentosa



Holothuria arguinensis



Holothuria forskali (left and middle) and *Holothuria mamata* (on the right)



Marthasterias glacialis



Mytilidae



Palinurus elephas



Parablennius gattorugine



Parablennius pilicornis



Parablennius spp. 1



Parablennius spp. 2



Porifera spp. 1



Porifera spp. 2



Porifera spp. 3



Porifera spp.4



Porifera spp. 5



Porifera spp. 6



Octopus vulgaris



Paracentrotus lividus



Savalia savaglia



Scorpaena notata



Scorpaena sp.



Submitted application form for the Project Baseline

New Project Application

Thank you for your interest in joining this grassroots, Global Underwater Explorers aquatic conservation initiative. Project Baseline is made possible through the ongoing commitment of our driven volunteers around the world, program sponsors and donors. By contributing to Project Baseline, you are joining a global team of concerned and invested citizens who share underwater observations, often hidden and out of sight, with divers and non-divers alike. Without the dedication, support and involvement of hundreds of Project Baseline volunteers, we would not be able to work towards the fulfillment of our mission. Please complete this online application. Once submitted, the Program manager will review your application and reply to your request within 1 - 2 weeks. Please contact the Program manager with any questions during the application process. vanessa@gue.com Note: by completing this application, you are expressing your intent to participate in Project Baseline. No information provided here will be automatically uploaded to the internet or to the Project Baseline database.

*Required

Are you a certified scuba diver? *

Yes ▾

If you are a certified diver, which agency issued your current certification(s)?

*

Your certification affiliation does not influence your application standing.

GUE / SSI

Please indicate your central focus or involvement in the scuba industry: *

Your level of involvement will not influence your application standing. Check multiple boxes as needed.

- Expedition/Exploration Diver
- Technical Diver
- Recreational Diver
- Scientific Diver
- Dive Shop Owner or Employee
- Eco or adventure tourism operator
- Cave Diver
- Other:

Project Name

This is generally a short, one to three word title that indicates the general geographic extent your team wishes to monitor and can appeal to both divers and non-divers alike. You are not bound to this Project name as it may be subject to change before your Project is accepted.

Shipwrecks of the Portug

Project Manager Name *

Please include your first and sur names.

Matilde Costa

Project Manager Email *

Due to the international nature of Project Baseline, email is our most common form of communication.

:ildecostaa@hotmail.com

Project Manager Hometown or City *

Faro

Project Manager Home Country *

Portugal

Project Structure

For more information about the Project, Site, and Station structure of a typical Project Baseline Project, please visit this resource:

http://projectbaseline.org/sites/default/files/PB_Station_Setup_2015.pdf

Project Structure Overview



Project Structure Overview



Briefly Describe Your Project *

Many teams begin Projects because they are interested the long-term documentation of an aquatic area that is frequented on a regular basis for training, exploration or recreational purposes. Some Projects are born from a desire to explore and document real and/or perceived change in environmental conditions that have been observed by divers over a period of time. Please describe your fundamental motivation for initiating a new Project. (Examples of Project descriptions are available on most Project webpages under this menu option: <http://projectbaseline.org/projects>)

Shipwrecks in the Portuguese coastline are very popular diving sites. Some of them are visited weekly or even more frequently during summer. Even though they are visited so frequently and most of them are easily accessible, there was no information about the biodiversity that this wrecks harboured until I did my master thesis exactly about the wrecks benthic biodiversity. One of the major problems I had with my research was that there was no long term studies

documentation of an aquatic area that is frequented on a regular basis for training, exploration or recreational purposes. Some Projects are born from a desire to explore and document real and/or perceived change in environmental conditions that have been observed by divers over a period of time. Please describe your fundamental motivation for initiating a new Project. (Examples of Project descriptions are available on most Project webpages under this menu option: <http://projectbaseline.org/projects>)

easily accessible, there was no information about the biodiversity that this wrecks harboured until I did my master thesis exactly about the wrecks benthic biodiversity. One of the major problems I had with my research was that there was no long term studies about colonization of this types of artificial reefs. The goal with this Project Baseline is to have a long term collection of data which eventually can allow for an evaluation of changes in this habitats over time.

Site Name(s) *

List one or more Sites your team intends to monitor. A Project must have one Site assigned. Please list by common name. Geographic coordinates (lat/lon) are not required for Sites.

Wreck River Gurara
Wreck Torvore
Wreck Oliveira e Carmo

Stations *

Stations are the exact locations within a Project area where underwater

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We're here to help you get started!

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Wreck Torvora
Wreck Oliveira e Carmo

Stations *

Stations are the exact locations within a Project area where underwater observations are collected. Each Site must have at least one Station assigned. Please provide the names of each Station, the Site that each Station belongs to and the geographic coordinates (lat/lon) of each proposed Station. These Stations may be subject to change as your Project undergoes initiation.

As a start, each wreck should only have one station. All the sites have a cable that connects a surface buoy to the bottom of the sea, in an area very close to the wreck. The stations in each site should be the closest vertical structure seen from the descending point (cable).

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Local Environmental Contacts

Project Baseline encourages all teams to engage with their communities to foster public awareness of local, underwater environmental conditions. Please identify and list three aquatic environmental management entities or organizations (academic, governmental, non-governmental) that will benefit from your work associated with Project Baseline.

Haliotis; Mar Ilimitado; Subnauta

This three names correspond to some of the divecenters in each location. Projects Baseline should encourage the local diving communities to become more active and participatory in local conservation projects.

Membership Notice *

Project Baseline is the conservation initiative of Global Underwater Explorers and relies heavily on donations and membership dues to continue our work. We appreciate our volunteer's membership support. To learn more about becoming a GUE member, please visit this link: <http://www.globalunderwaterexplorers.org/membership/levels>

I have read this statement.

Do you represent a formal or informal existing dive group/team/club or organization? *

Your group affiliation does not influence your application standing.

Yes

No

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If yes, please indicate the name of your group and website, or social media URL if applicable.

How did you hear about Project Baseline? *

Facebook

Google

Twitter

YouTube

Vimeo

Word of Mouth

Diving Group

Other:

More Project information

Still have a few minutes and want to learn more about how to establish a Project in your community? Visit this page - <http://projectbaseline.org/project-structure> - to look over our resources. Available as PDFs for download.

Submit

Never submit passwords through Google Forms.

Shipwrecks Species Database