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Wrecks as artificial reefs: benthic biota associated with scuttled wrecks in Malta and implications for conservation

MASTER'S THESIS

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Declaration

I hereby declare that I have done this thesis entitled “Wrecks as artificial reefs: benthic biota associated with scuttled wrecks in Malta and implications for conservation” independently, all texts in this thesis are original, and all the sources have been quoted and acknowledged by means of complete references and according to Citation rules of the FTA.

In Prague, 12/04/2023

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Abstract

The purpose of this thesis was to investigate the conservation potential of shipwrecks scuttled as diving attractions to serve as artificial reefs in the coastal area of Malta. For this, the diversity of sessile biota and fish, as well as coverage of sessile biota was compared between artificial reefs (ARs) and natural reefs (NRs) in the Ċirkewwa Marine Park. The study found that NRs had higher diversity than ARs, likely due to the higher habitat heterogeneity in NRs. However, both NRs and ARs shared a high degree of similarity in terms of taxonomic composition, potentially due to similar environmental conditions. The overall cover of sessile biota in cm^2 per plot did not differ significantly between ARs and NRs, but the vertical plots on ARs showed a higher sum of cover in cm^2 than horizontal plots on ARs, and horizontal plots on NRs showed a higher sum of cover in cm^2 than vertical plots on NRs. However, the reasons for these differences in cover are not fully understood. Further research is needed to identify reasons for this pattern. The study of fish found the same richness on ARs and NRs, which indicates that ARs provide a suitable habitat for fish. Further, two fish species on the NRs are currently classified as vulnerable, whereas all recorded fish species on the ARs are classified as least concern, highlighting the importance of conservation of NRs. This study supports the idea, that wrecks as ARs have potential to provide additional habitat for sessile biota and fish.

Key words: Artificial reef, Biodiversity; Benthic Cover; Conservation; Fish: Malta; Natural Reef; Shipwreck; Sessile Biota; Wreck

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List of the abbreviations used in the thesis

Abbreviation	Definition
AR	Artificial Reef
NR	Natural Reef
SCUBA	Self-Contained Underwater Breathing Apparatus
WTO	World Tourism Organization
EU	European Union
Sp.	Species
n.d.	No date
Med	Mediterranean Sea
DIR-style	Doing it right – style
DD	Data Deficient
NE	Not Evaluated
LC	Least Concern

1. Introduction

The purpose of this thesis is to investigate the conservation potential for shipwrecks scuttled as diving attractions to serve as artificial reefs in the coastal area of Malta. It should serve as a guide for comprehending the potential for shipwrecks to resemble the natural reefs that are adjacent to them and, as a result, support the conservation of the marine habitat through habitat enhancement. Understanding the effect of these wrecks on the marine ecosystem is useful for marine conservation and the Maltese diving sector.

The world's seas and oceans play a major role in regulating the Earth's temperature, providing food, producing oxygen, providing clean water and protecting coasts (Sandifer & Sutton-Grier 2014).

The marine ecosystems are currently under threat from overfishing (Washington et al. 2016), climate change and pollution (Lu et al. 2018). Anthropogenic impacts and structural development have caused significant alterations to marine habitats. For instance, studies in the northern Red Sea found that the coastline has undergone significant environmental stress in recent decades due to recreational activities, especially tourism-related activities such as diving. These activities have caused substantial damage to inshore coral reefs, mangroves, and seagrasses, leading to heavy damage to marine ecosystems (Lakhouit 2020). Coastal development and population expansions have also contributed to this damage. Even deep oceans are not immune from the negative effects of human-produced waste and marine litter (Woodall et al. 2015).

For seas and oceans, the rate of increasing human impact is particularly alarming (Halpern et al. 2019). The degradation of coastal and ocean habitats, ecosystems and fisheries is a worldwide concern. Globally, one third of the world's coastlines are at high potential risk of degradation (Seaman 2007). Anthropogenic stressors have led to a habitat change, especially in coastal waters (Papadopoulou et al. 2017). Marine ecosystems are suffering through "increasing industrialisation, increasing use of resources, plus a build-up of populations on the coastline" leading to "habitat degradation" (Papadopoulou et al. 2017). Furthermore, invasive species are impacting the Mediterranean marine ecosystem, marine litter is increasingly detected and affecting the waters (Papadopoulou et al. 2017).

The island of Malta, located in the central Mediterranean Sea, is impacted by issues such as high population density, considerable tourism sector and exposure to the effects of climate change. Furthermore, as marine areas are a key component of the Maltese economy and culture, it is suspected that the marine coastal ecosystem around the island is suffering from degradation. The Maltese coast has transformed due to investment in hotels and housing (Xerri 2020). SCUBA diving represents one of the big tourism sectors on Malta: in 2018, almost 170.000 tourists visiting the island took part in SCUBA diving activities, which accounts to 6.5 percent of all tourists visiting Malta (Caruana & Sultana 2021).

The World Tourism Organization states coherently, in a worldwide perspective, that the SCUBA diving sector is growing as one of the mass tourism activities with significant economic importance. There are currently 5-7 million active certified divers worldwide, according to an estimate (Dowling 2008).

Scuttled wrecks create Artificial Reefs (hereafter ARs) in marine natural systems and serve as a SCUBA diving attraction. Dive tourism is today an important part of the global tourism industry and has become significant to local economies. Wreck diving has been rising in interest as well as demand, being a more varied and challenging kind of diving experience. Wreck divers are motivated by two main things: exploring the remains of sunken ships and artifacts from the past, and discovering marine life underwater (Edney et al. 2021).

In regard to the socio-economic and geographic context of the Maltese coastal area and the considered impacts on marine coastal ecosystems, as well as the popularity of Malta as destination for SCUBA diving, it is of interest to go beyond the use of wrecks simply as diving destinations by assessing the potential conservational value wreck-reefs create. The present thesis is meant to make a solid baseline assessment for deeper understanding of the role of artificial wrecks for biodiversity conservation.

1.1. The Mediterranean Sea

The Mediterranean Sea is an "intercontinental sea that stretches from the Atlantic Ocean on the west to Asia on the east and separates Europe from Africa" (Boxer et al. 2022).

It covers a total area of 2500000 kilometres (excluding the Black Sea) and a coastal extension of 46250 kilometres (excluding islands) (Trainito 2011). The maximum depth is

5267m, found at the Calypso Deep (36°34'00.0"N 21°08'00.0"E) (Fig. 1). The average depth of the Mediterranean is 1460 m (Kontoyiannis et al. 2016).

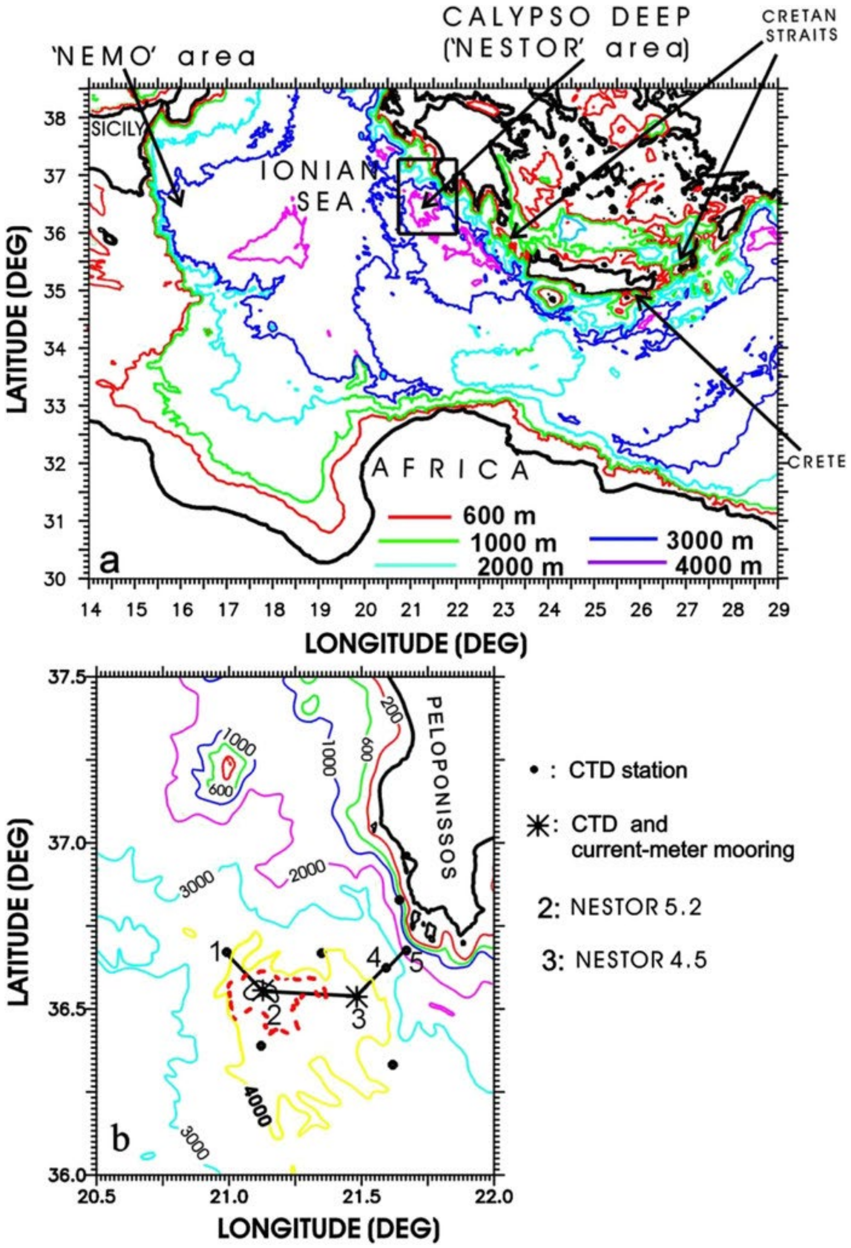


Figure 1: Map indicating the depth of the Mediterranean Sea (Kontoyiannis et al. 2016)

The Mediterranean Sea is unique, nearly landlocked, surrounded almost completely by land and bordered by three continents, Africa, Europe and Asia. As for the closeness of the Sea, particular conditions can be found in Mediterranean waters (Trainito 2011).

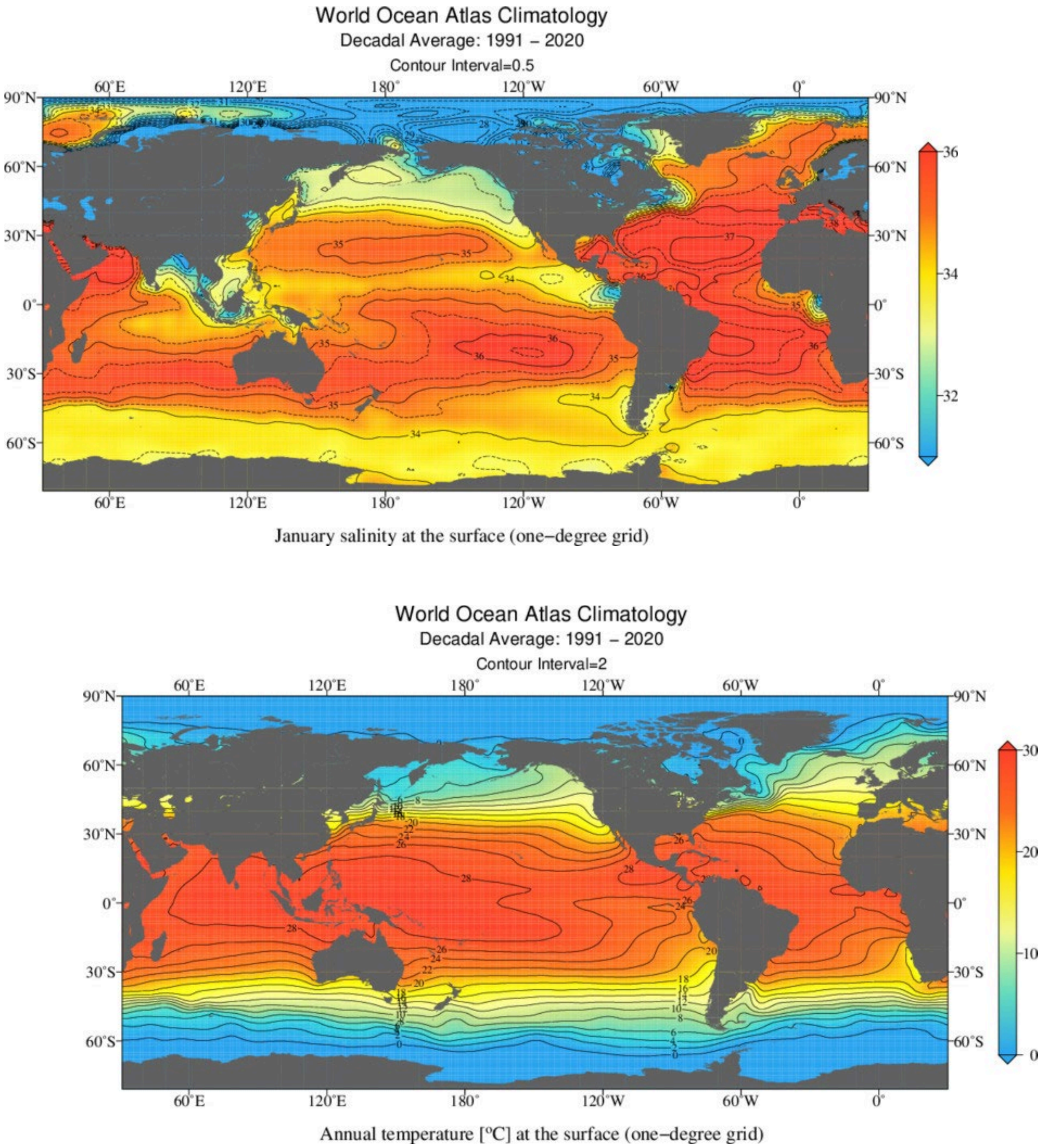


Figure 2: World annual salinity and temperature (Boyer et al. 2018)

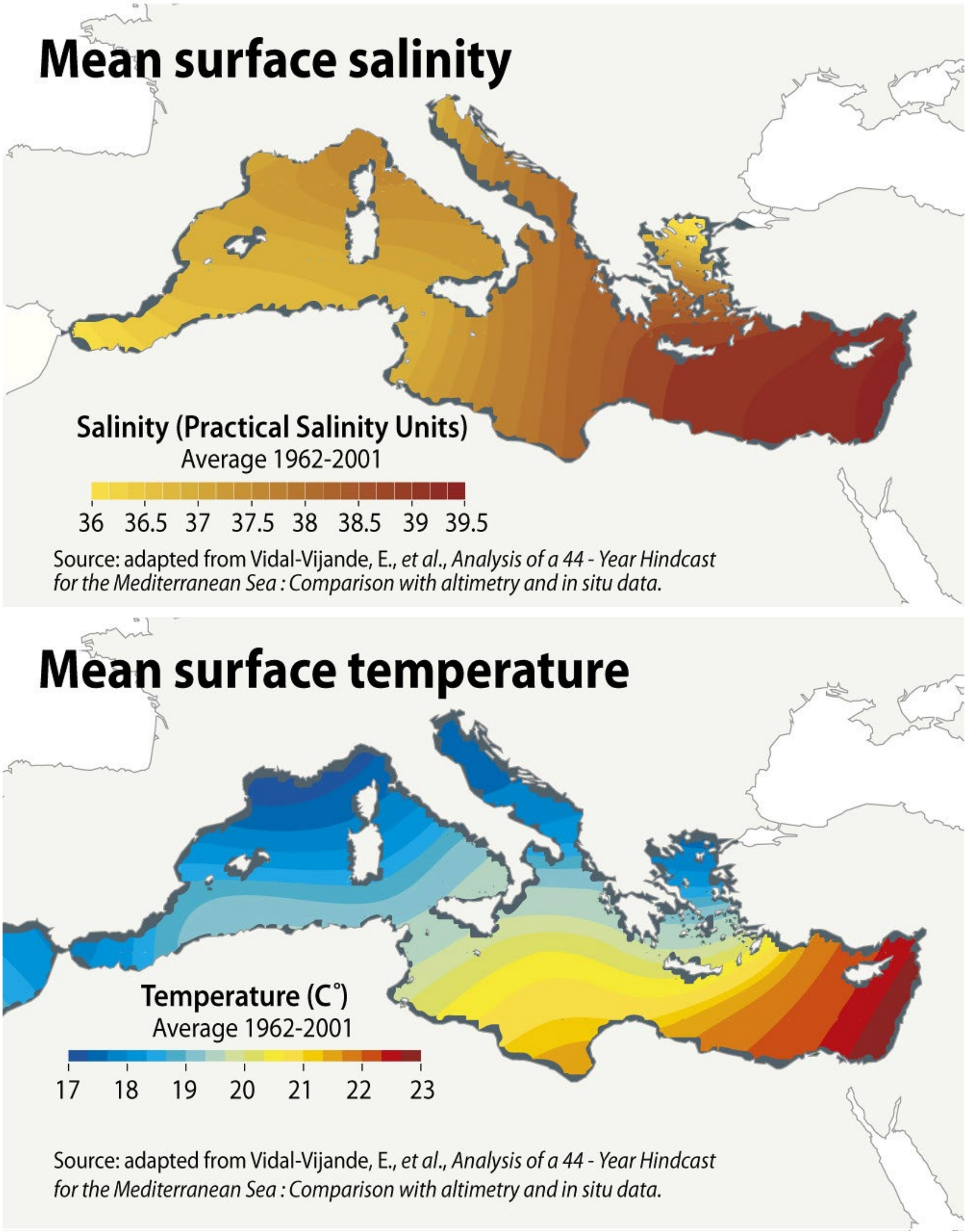


Figure 3: Mean surface salinity and temperature Mediterranean Sea (GRID-Arendal2013a, 2013b)

The average surface temperature of the Mediterranean exhibits a variation from 17°C in the western section to 23°C in the eastern region of the basin (GRID-Arendal 2013b).

The Mediterranean Sea comprises three main water masses with distinct salinity characteristics from 36.2 to 39.1 psu (GRID-Arendal, 2013).

As compared to other seas, the Mediterranean Sea has higher salinity levels and warmer temperatures, particularly in its eastern part (Boyer et al. 2018)(Fig. 2,3).

The Mediterranean connects to the Atlantic through the narrow Gibraltar Strait: the sea distance between the African coast and Europe is only 13 km (Fig. 4)(Boxer et al. 2022).

Figure 4: Strait of Gibraltar(Misachi John 2021)

The sea is connected with the Indian Ocean through the human-constructed Suez Canal, a 173 kilometre long communication channel (Fig. 5), and to the Black Sea via the Bosphorus Strait (Chaparro 2017; Boxer et al. 2022)

Figure 5: Suez Canal(Misachi John 2021)

The Mediterranean receives only one third of water by rivers. A continuous flow of water from the Atlantic Ocean feeds the Mediterranean Sea through the Strait of Gibraltar, which is the main source of surface water. After crossing the Gibraltar Strait, water flows east along the north coast of Africa. This current is the most stable part of the Mediterranean flow. It is strongest during summer, when evaporation in the Mediterranean is the highest. Small amounts of water also enter the Mediterranean from the Black Sea.

In summer, the Mediterranean surface water becomes more salty, due to high evaporation. This results in higher density, and the water sinks. The excess of groundwater then enters the Atlantic Ocean as subsurface flow, under the incoming current. The Mediterranean has been metaphorically described as breathing: inhaling surface waters from the Atlantic and exhaling deep waters from the bottom of the sea (Boxer et al. 2022). These are the factors, which lead to an overall trend of currents from west to east, along the coast of Africa and returning through the northern basins (Fig. 6)(Trainito 2011).

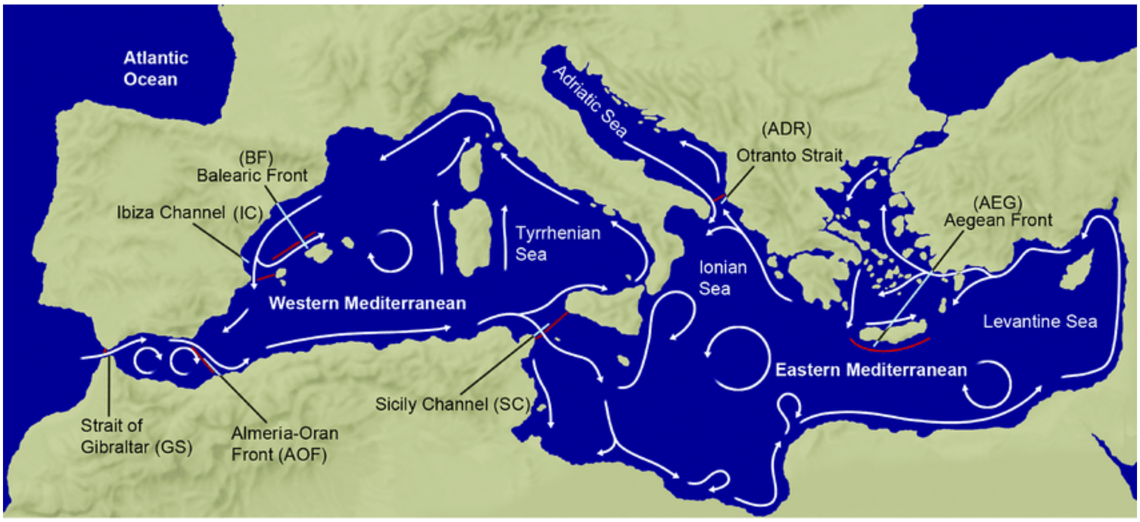


Figure 6: Currents of the Mediterranean (Pascual et al. 2017)

The Mediterranean is a sea characterized by insignificant tides, which do not have a major influence on the coast and on the organisms which inhabit the sea and its shore (Trainito 2011). The Mediterranean Basin is home to a diverse array of marine ecosystems including brackish water lagoons, estuaries, rocky shores, and pelagic systems, providing essential benefits to coastal inhabitants in the region (Gaetano Leone 2017).

The Mediterranean Sea can be structured following biogeographic variables into the Alboran Sea; Algerian and north Tunisian coasts; Tyrrhenian Sea; Balearic Sea to Sardinian Sea; Gulf of Lions and Ligurian Sea; northern Adriatic Sea; central Adriatic Sea; southern Adriatic Sea; Ionian Sea; Levant Sea; southern Aegean Sea; northern Aegean Sea (Bianchi et al. 2012).

Explicitly, the Mediterranean represents a biodiversity hotspot with a significant reserve of biological diversity, providing habitat to 4 % to 18 % of all identified marine species, while constituting only 0.82 % of the global ocean surface area (Coll et al. 2010; Monfort et al. 2021). The Mediterranean sea is an oligotrophic environment, low in nutrients, with limited primary productivity and high in oxygen levels(Jimenez et al. 2016). Nevertheless, the Mediterranean's coastal areas contain a high concentration of marine endemism (Pinnegar 2018).

In 2020, 8.33% of the Mediterranean Sea has been designated as a protected area by official statutes (Fig. 7). 97.33% of the total protected surface area in the Mediterranean is concentrated in the waters of EU member countries. 0.04% of the surface area in the

Mediterranean is a no-go, no-take, or no-fishing zone (MedPAN & UNEP/MAP-SPA/RAC 2021).

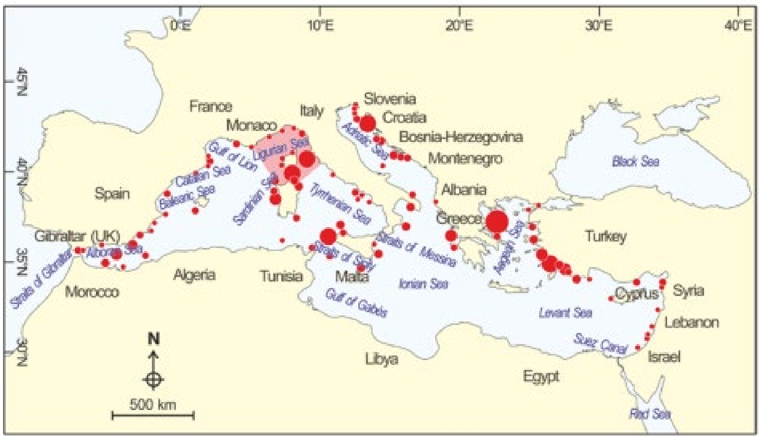


Figure 7: Geographical setting of the Mediterranean Sea. Red circles indicate coastal Marine Protected Areas, their diameter being roughly proportional to the surface protected (Bianchi et al. 2012)

As a reason of the geological evolution of the Mediterranean basin, and the drastic climatic fluctuations of the Quaternary, the Mediterranean evolved as a biodiversity hotspot, containing between 15000 and 20000 marine species (Bianchi et al. 2012).

The Annex II of the Barcelona Convention for the Protection of the Marine Environment and the Coastal Region of the Mediterranean lists the endangered and threatened species of the Mediterranean sea. Within the list are representatives of the Sponges (Porifera), which are filter feeders that help in maintaining water quality by filtering out organic matter and nutrients (Turon et al. 1998); representatives of the Cnidaria, which contribute to the construction of reefs and other structures in marine ecosystems (Goldstone 2008); Molluscs, which play a crucial ecological role by providing essential ecosystem services such as creating habitat structures for benthic organisms, acting as a filter to purify water, and serving as a significant food source for other organisms (Parker et al. 2013); as well es representatives of the Chordata (Fish, Sharks, Whales). Further, Green Algae (Chlorophyta), Red Algae (Rhodophyta) and Brown Algae (Phaeophyta) are endangered and threatened, and are primary producers in the marine ecosystem, producing oxygen and providing food for many organisms (“Ecological Importance of Algae - Algae Research Supply” n.d.).

One particular characteristic in the Mediterranean is the presence of *Posidonia* meadows, which are being altered by the alien species *Caulerpa taxifolia*, a seaweed which was introduced to Mediterranean waters through aquarium trade. Species of the genus *Caulerpa* are generally known as invaders of seagrass meadows, and *Caulerpa taxifolia* is also called killer alga showing a higher resistance to polluted waters then *Posidonia* sp. and is believed because of this, and amongst other reasons, to flourish (Meinesz et al. 2001; Glasby 2013).

1.2. Natural Reef Habitat Ecology

“A reef is a sedimentary rock aggregate, large or small, composed of the remains of colonial type organisms that lived near or below the surface of water bodies, mainly marine, and developed relatively large vertical dimensions as compared with the proportions of adjacent sedimentary rocks. The organisms, generally corals and algae and less commonly crinoids and bryozoans, creating the essential features of a reef, lived their mature lives on it and their hard parts remain in place there after death. Reefs tend to develop as mounds or ridges but also grow in irregular, asymmetrical forms. (…) Reefs are commonly characterized by lack of well-developed stratification” (Wilson 1950).

Natural reefs (referred as NRs here after) consist of rocks and / or skeletons of animals and the NRs around Malta, specifically, are dominated by rocky structures on which algae, such as *Cystoseira* sp. and *Dictyopteris* sp., are particularly popular (Stevens et al. 1995).

Rocky shore habitat is a biologically diverse environment that can include numerous habitat types, including platforms, rock pools, boulder fields, steep rocky cliffs, and more. Numerous small gastropods, polychaetes, and crustaceans inhabit temperate rocky reefs (Taylor 1998).

The Mediterranean rocky reefs express a variety of ecological functions due to their high habitat heterogeneity and diversity, which provide a variety of niches (Bevilacqua et al. 2021).

Different species have access to a variety of habitats on the rocky reef. The assemblages of temperate rocky reefs are highly dynamic and variable systems (Guidetti 2006). Temperate nearshore reefs are extremely diverse and productive habitats that provide fish and invertebrates with structure and shelter (Gunderson et al. 2008).

There are at least three ways that rock reef composition can affect reef ecology: the reef matrix's hardness, the relief pattern on the bottom, and the clarity of the water above the reefs. Shelter is one of the rocky reef habitats' most important functions. The shelter is crucial to juvenile and small species as well as to larger species that associate with the substrate. It is obvious that hard structures can physically protect animals from predators. Rock crevices, burrows, and caves may prevent a predator from getting to a potential prey in this area. Secondly, an immobile substrate can physically shield waves (Allen et al. 2006). Human activity has a significant impact on rocky reefs, as it does on other coastal marine habitats. For a number of marine species, rocky reef habitats are thought to be important areas for breeding, feeding, and nursery. Species related to rocky reefs have declined in abundance, and even without active and measurable degradation of reef structures, enhancement of the habitat is often aimed for by managers, as reefs provide valuable ecosystem services (Paxton et al. 2020).

Large quantities of organic matter, mostly in the form of *Posidonia oceanica* leaves but also in form of leaf epibiota (biota living on the leaf) and seaweeds that live on rhizomes, are exported by the *P. oceanica* ecosystem to adjacent ecosystems, particularly algae-dominated rocky reefs (Boudouresque et al. 2016).

Important subtidal canopy-forming seaweeds in shallow-water Mediterranean rocky habitats are members of the genus *Cystoseira*. These seaweed-dominated assemblies represent the highest level of Mediterranean seaweed complexity and share functional properties with kelp forests, despite their smaller size(Verdura et al. 2021). The reefs frequently show zonation, with light loving plants closer to the surface, and shade loving plants found deeper(Borg & Schembri 2002). Furthermore, Malta's coast is surrounded by limestone cliffs, which make up the Maltese coastline steep or vertical, punctuated by bays and inlets (Furlani et al. 2014).

One major contribution to the functioning of coastal ecosystems arises from vegetated rocky substrates. They participate in the cycle of nutrients, support marine coastal biota by providing food, nursery, and shelter, and encourage the diversity of important ecological compartments like microbiota and meiofauna. They are also primary sources and sinks of blue carbon (Bevilacqua et al. 2021).

Although extensive meadows of seagrass *Posidonia oceanica* can also be found on shallow horizontal hard substrates, macroalgae dominate the horizontal to almost vertical rocky bottoms. Belts of Fucales (*Cystoseira, Sargassum*) emerge in both sheltered and exposed locations. The interesting characteristics of Mediterranean macroalgal plants are the high biodiversity associated with the high miniaturization. A large number of species of algae can coexist on small surfaces (Bevilacqua et al. 2021).

The algae-coverage brings three-dimensional structure that makes it possible for very specific animal assemblages to develop as epiphytes, which are sessile organisms that grow on the surface of a plant. The diversity and distribution of these epiphytes is strongly influenced by the variations in macroalgal stands' spatial and temporal patterns (Fraschetti et al. 2006). Light intensity, nutrient availability, and biological interactions are some of the environment-dependent and depth-dependent factors that affect the distribution of aquatic plants (Sant & Ballesteros 2021). The various species that are associated with rocky reefs' processes of settlement and recruitment are influenced by the mineral composition of the rock-reef, which plays an important role in community assembly (Guidetti et al. 2004).

The spatial distribution of macroalgae and associated invertebrate assemblages reflects a combination of various bottom-up (e.g., energy inputs) and top-down (e.g., predation) processes. Besides and importantly, anthropogenic drives influence the distribution and growth of macroalgae (Power 1992).

Rocky reef fish connect various Mediterranean habitats via trophic transmission. This is the case with gregarious, small fish, e.g., the damselfish *Chromis chromis* and the cardinal fish *Apogon imberbis*. These species transport organic matter into depleted rocky reef habitats, mostly through the fecal pellets while migrating between the water column and the bottom as well as into and out of marine caves (Bevilacqua et al. 2021).

In terms of ecosystem services, which are at the foundation of the human well-being of most Mediterranean coastal nations, the extensive diversity of rocky reef species and the functional traits that are associated with them are responsible for a variety of human benefits (Bevilacqua et al. 2021).

For millennia, human activities have altered the Mediterranean Sea's coastal environments and NRs. Catastrophic events as large storms or thermal anomalies frequently shape shallow assemblages (Verdura et al. 2021).Reefs are deteriorating worldwide due to human disturbances such as climate change, overfishing, habitat destruction, nutrient enrichment and pollution (Sánchez-Caballero et al. 2021).

Several *Cystoseira* populations have experienced significant declines, primarily as a result of changes in water quality, just like other seaweeds that form canopies around the world (Verdura et al. 2021).

Overgrazing by sea urchins, which are the major benthic herbivore in the Mediterranean rocky bottom, and herbivorous fish significantly reduces algal biomass (Sala et al. 2012; Peleg et al. 2020). The decline of these algal communities may have a cascading effect on the community as a whole, as it could also have an impact on the recruitment rate of numerous rocky fishes that choose algae as their preferred habitat for settlement (Sala et al. 2012).

In most Mediterranean regions anthropogenic activities have resulted in significant changes in the composition of rock reefs (Coll et al. 2010). These impacts lead to changes in the habitat structure from a state in which species form canopy to alternative states, in the worst case to barren areas composed of filamentous and encrusting species(Thibaut et al. 2015). Furthermore, several benthic invertebrates and fish species that inhabit Mediterranean reefs have been threatened or are being threatened by overfishing, including commercial sponge and red coral collection, the consequence being a change of the structure of benthic assemblages on many reefs (Cattaneo-Vietti et al. 2016). However, the major drivers of shift cannot be clearly identified, and most studies speculate on multiple interacting anthropogenic stressors when determining ongoing reef change. There is an excessive degree of uncertainty concerning interactions between pressures and their cumulative impacts. As anthropogenic pressures have developed, it has slowly been acknowledged that the marine environment is a resource that may be harmed and degraded. The difficulty of working in the marine environment and the fact that underwater ecosystems are for many humans not directly visible and accessible by sight contributed to the reality that ecological restoration in marine ecosystems is far behind the restoration of terrestrial ecosystems (Papadopoulou et al. 2017).

1.3. Artificial Reef Ecology

“An artificial reef is one or more objects of natural or human origin deployed purposefully on the seafloor to influence physical, biological, or socioeconomic processes related to living marine resources”(Seaman 2000). Over the past 10-15 years, ARs have become a topic within recreational fishing, scuba diving, and scientific communities. Objects such as tires, old ships, and even oil platforms are being used as AR in the hope of improving productivity in the coastal area (Baynes & Szmant 1989). Sunken shipwrecks create AR structures and have become popular among divers for their ability to host large number of fish, which has led to increased numbers of ship-submersions around the world. Vessels are intentionally sunk to serve, among other purposes, as ARs and artificial habitats (Ilieva et al. 2019). Such artificial habitats have been installed and developed in many places to increase abundance and biodiversity as well to attract SCUBA diving tourism and restoration of weakened ecosystems (Seaman 2000). ARs are regarded as benefiting the marine hard bottom biota (Walker & Schlacher 2014).

ARs have become additions to reef ecosystems, sustaining established benthic communities (Perkol-Finkel & Benayahu 2004).

Based on the "global database of intentionally deployed wrecks to serve as artificial reefs" (Ilieva et al. 2019) there are 1907 purposefully scuttled shipwrecks around the world, not covering all existing data, but focusing on a "selection of available data either online or in publications in the English language" (Ilieva et al. 2019).

ARs were used in history predominantly for "enhancement of fishery harvests" by attraction and mirroring natural marine environments (Seaman 2000)and are nowadays part of restoration actions focused on recovering and re-building damaged habitat and ecosystem, following and addressing objectives: 1. Protection of habitats; 2. Enhancement of fisheries; 3. Economic viability. ARs may act as shelter from fishing and hence increase fish biomass; increase biological diversity of sessile and non-sessile organisms; restore and create habitat; enhance understanding of colonization processes on different substrate (Claudet & Pelletier 2004; Santos et al. 2005; Seaman 2007). ARs can be "any submerged structures places on substratum to mimic some characteristics of a NR, altering physical, biological and/or socio-economic variables related to marine resources" (Layman & Allgeier 2020).

Habitat restoration operations use ARs to enhance habitat (Paxton et al. 2020). AR can offer habitat for marine taxa, such as fishes, corals and other organisms (Anderson et al. 2019) and previous studies of ARs have suggested parameters such as currents and wave power influence the efficiency of ARs (Baynes & Szmant 1989). The additional substrate provided by ARs naturally gives more room for settlement of fouling, “resultant assemblages at any point in time of a colonisation process on a solid surface” assemblages (Svane & Petersen 2001; Layman & Allgeier 2020).

The research on ARs has shifted from main interest in fishery enhancement to use of AR for reconstruction of marine ecosystems (Lee et al. 2018). The importance of artificial habitats in aquatic systems remains a subject of debate in science (Seaman 2007).

1.4. Sunken Shipwrecks as Artificial Reefs

Sunken shipwrecks being a form of ARs may also impact mentioned objectives and affect advantageously the surrounding marine ecological system (Svane & Petersen 2001). Since 1942, more than 1907 ships have been scuttled worldwide to serve as ARs (Anderson et al. 2019).

Research has focused on studying different designs and materials to construct ARs for different purposes (Lima et al. 2019), while not much attention has been given to shipwrecks as ARs and the ecological connection and interaction between ARs and NRs.

As for the evaluation of the performance of ARs, Seaman provided a broad guide on the objectives an AR may have and strategies for measuring and evaluating the performance, concentrating on the documentation of benefits an artificial possesses towards human interest. While this guide serves as a good overview on the topic, it lacks the investigation of shipwrecks as ARs and the evaluation of their performance from the biodiversity conservation perspective. In fact, the keyword “wreck” is mentioned only once in the text (Seaman 2000).

Although successions of AR communities were thoroughly studied, little is understood about how ARs and NRs are linked with one another. One essential question that remains unanswered is whether AR communities can mimic NR groups which are adjacent. According to Baine (2001), the European Artificial Reef Research Network defines an AR as “a submerged structure placed on the substratum (seabed) deliberately, to mimic some characteristic of a natural reef”, defining the aim of an ARs as an imitation of NRs. The sessile fauna present on shipwrecks has been discussed by some studies since the 1960s (Jimenez et al. 2016).

Perkol-Finkel et al. (2006) propose three hypotheses concerning possibilities for ARs to mimic NR communities: Neighbouring ARs and NRs will (1) gain a comparable community structure given enough time; (2) be comparable at best in the event that they own comparable structural features; (3) usually differ, irrespective of age or structural features. To explore these hypotheses, the researchers investigated a 119-year old wreck and the adjacent NR. Their results supported the second hypothesis, meaningly that the communities differ if the AR and the NR possesses different features of orientation or complexity.

Walker and Schlacher (2014) suggest that the performance of ARs in mimicking and supporting devastated NRs is poorly understood and researched. They investigated the diversity of sessile invertebrate assemblages on a 3-year old wreck-reef in Eastern Australia and based the analysis on species abundance, richness and assemblages' composition. They found that the composition on the wreck differed fundamentally to assemblages in adjacent NRs and highlight the influence of time for colonisation of wrecks and underline the importance of taking the time shift into consideration when management considers scuttling a wreck for marine conservation (Walker & Schlacher 2014).

In an earlier published paper, Walker et al. (2007) studied the colonization of sessile organisms of a shipwreck in East Australia and found that barnacles, sponges, and bryozoans were predominant on the wreck and depth affected species abundance and diversity. They concluded that wreck reefs influence and increase the local diversity of the marine area. As support of previous research (Irving & Connell 2002) they found that vertical surfaces support a higher number of species and cover in comparison to horizontal surfaces, suggesting that this may be the reason of overlaying by sediment on horizontal surfaces, although their findings conclude that bivalves were more present on horizontal surfaces. The reason for this may be that this epifauna can grow above a sediment layer compared to less tough organisms and that bivalves have a high tolerance to predators and wave energy. Additionally, the authors found that fouling assemblages were less present inside of the wreck than outside of the wreck (Walker et al. 2007), which can be explained according to Baynes & Szmant (1989), as changed water current regimes inside the ship are likely to alter the supply of larvae and the availability of food. Jimenez et al. (2016) explain that the additional surface of a wreck placed underwater brings available areas for colonisation of organisms, which in case of lacking space for settlement would have been probably died off.

It is widely agreed that wrecks increase the available surface for settlement of fouling communities (Jimenez et al. 2016), and organism living on the surface of the ARs provide a food resource and secondary habitat for other marine organisms (Walker et al. 2007). Recruitment and colonization rates on ARs and shipwrecks are influenced among others by water depth and light intensity, waterflow, surface material and rates of sedimentation (Baynes & Szmant 1989; Svane & Petersen 2001; Boaventura et al. 2006).

Jimenez et al. (2016) describe how, by placing a shipwreck on a sandy bottom, the flow in water and current changes, enhancing the deposition of organic material next to the wreck. This in turn leads to an up-bringing of nutrient-rich water and hence to increased phytoplankton activity (Jimenez et al. 2016). Currents are crucial when it comes the transporting nutrients and organic matter. ARs may be more productive if their long axes are perpendicular to the prevailing current. The reason for this is that many resting invertebrates on these reefs consume organic particles and planktonic organisms present in the water column (Baynes & Szmant 1989). It is predicted that, under these circumstances, the AR will be colonised firstly by "algae and spores of a large number of epibenthic organisms" (Svane & Petersen 2001).

Baynes & Szmant (1989) found coherently during the study of a benthic community on a shipwreck off Florida that areas suspected with high water flow and low sedimentation show increased cover and higher species diversity then areas of the wreck which were confronted with more sedimentation and lower water flow.

Paxton et al. (2020) concluded after synthesising 39 studies in this field, that AR can mimic NRs in fish density, species richness and biomass. Nevertheless, they stress out that communities around and on ARs are highly dependent on the surrounding environment, namely the geographic setting (Paxton et al. 2020).

1.5. Role of Habitat Heterogeneity

There is a common understanding in wildlife research, that habitat heterogeneity and diversity is correlated with species diversity. Areas with a greater variety of different habitats tend to support a greater number and diversity of species. This is because different species have different habitat requirements, and areas with a diverse array of habitats can provide suitable living spaces for a broader range of species (Tews et al. 2004). In conservation biology, diversity of species has emerged as a significant issue (Chapin et al. 1998). It has been hypothesized that adding new habitats results not only in an increase in the heterogeneity of habitats, but also in the diversity of species (Rebele 1994). Conversely, the addition of habitats resulted in some cases in a decrease in the number of species and abundance of species (McGuinness & Underwood 1986).

It is believed that biological diversity and species cover density on a reef are impacted by the number of physical niches, which are the areas the marine organisms can settle or hide from possible predation. Productive marine environments are correlated with an existing high structural complexity of these environments (Todd & Turner 1986; Sebens 1991; Ecol et al. 1998) . Perkol-Finkel & Benayahu 2004 suggested that AR could increase the heterogeneity and surface availability and in turn may increase species diversity and production.

Stachowicz et al. (2007) discovered that taxon richness increases the resilience to pressures and rehabilitation from disturbances. Increased biological diversity will increase the value of the ecosystem, hence a shipwreck as AR may increase the resilience of the marine area it is placed.

Scuttled shipwrecks increase the habitat heterogeneity and thus the complexity of the marine area. The scientific landscape on the topic of ARs is increasing and research in this field is expanding. Nevertheless, broad knowledge about the usefulness of shipwrecks as ARs and the factors favouring or preventing the settlement of species is limited. The research lacks comparison of biotic assemblages on wrecks and the comparison to NRs because it concentrates mainly on fish assemblages on NRs and ARs (Perkol-Finkel & Benayahu 2005).

1.6. Fish Assemblages in Natural Reefs and Artificial Reefs

The literature landscape reveals greater investigation of fish on ARs and NRs than of sessile organisms, but there is nevertheless a shortage and mixed opinion in answering the question of similarity between ARs and NRs (Paxton et al. 2020).

ARs are recognized to aid nursery grounds and facilitate the abundance of predator fish densities. Nevertheless, studies have found that the communities on ARs are often less complex, compared to NRs, making them attractive for non-indigenous species and enhancing alien species over native (Paxton et al. 2020; Bevilacqua et al. 2021). Fish assemblages on artificial structures are generally less diverse and complex than those on natural hard substrates. This increases the risk of colonization by non-indigenous species (NIS) and to transform to a "stepping stone", disproportionately favouring the spread of alien species over native rocky reef species.

Carr and Hixon (1997) researched the effectiveness of small ARs in simulating fish assemblages associated with natural coral patch reefs in Bahamian waters and found that species richness and fish abundance were greater on NRs than on ARs.

Fowler and Booth compared wreck-reef fish assemblages with a coral patch in a tropical lagoon and found that fish abundance, species richness and diversity were comparable to those in NRs, but the composition of species differed, suggesting that increases in wreck-reef habitat may increase the abundance of particular species (Fowler & Booth 2012). Fish distribution around AR can be influenced by habitat variables, like reef age, structure, material, and substrate composition, as well as spatial orientation and heterogeneity (Sánchez-Caballero et al. 2021).

Folpp et al. (2013) evaluated fish assemblages on estuarine ARs and rocky-NRs and sand-flat in Australian waters using baited remote underwater video and concluded that ARs assemblages were significantly different from those at NRs.

Simon et al. (2013) compared fish assemblages at two metal wrecks and rocky-NRs and discovered that the assemblages at shipwrecks differ significantly in trophic structure from NRs. Furthermore, they suggested that shipwrecks should not be settled near NRs, as they could alter the community structure of natural habitats.

Sánchez-Caballero et al. (2021) compared ARs with NRs in the Gulf of California and found that the greatest richness was found on ARs, whereas species abundance was higher on NRs. In addition, the fish assemblages differed from those in the NRs.

One of the principles in the AR literature by which AR could lead to increase of fish-abundance are: "(a) providing additional food, (b) increasing feeding efficiency, (c) providing shelter from predation, (d) providing recruitment habitat for settling individuals that would otherwise have been lost to the population and (e) indirectly, because fish moving to artificial reefs create vacated space in the natural environment that allows replacement from outside the system“ (Layman & Allgeier 2020).

It is crucial to mention the „attraction hypothesis“, which stands opposite to the „production hypothesis “. The idea suggests that the increase in fish biomass around ARs may be caused by the depletion of fish populations in surrounding environments (Svane & Petersen 2001).

The biota of a NR may be impacted by various factors, including physical effects such as water currents and biological effects like the source of juveniles, larvae, and spores, as well as by the availability of food from surrounding waters and neighbouring NRs. The distance to the NRs may hence have a influence on the colonisation on the ARs (Schroeter et al. 2015).

The review of the literature reveals that the science to date concentrates on the comparison of NRs and ARs in terms of the group of fish but lacks the investigation of the abundance and richness of sessile organisms such as algae and sponges.

Higgins et al. (2022) found coherently during a systematic review of the literature on ARs, that benthic algae other than corals are the taxonomic groups, which are underrepresented in the studies on ARs.

1.7. Methods for the study of benthic organisms

Studying benthic organisms can be done by different methods. Overall, the data-collection should be done efficiently, coherent, and fit into the available budget. Scuba diving involves using scuba gear to observe and collect samples of organisms from the seafloor. Scuba diving is useful for studying organisms in their natural habitat, but it is limited to relative shallow waters. Remotely operated vehicles can be used to study organisms in deep water and in areas that are difficult or impossible to reach by other means. Video transects involves using cameras to survey the seafloor and record the organisms living there. ROV-methods typically find less small and cryptic species, compared to scientific scuba diving (Bull et al. 2023).

Optical sampling using photoquadrats has become widely used in marine-science, making it possible to extract quantitative data from the images using software like ImageJ, and recommended for marine assessment (Leujak & Ormond 2007).

Put simply, a photoquadrat is a quadratic area, which is photographed with a camera. Images taken by a digital underwater camera are the essential component providing the data. Surveys with photoquadrats have their advantages and limitations. Disadvantages are restrictions in the resolution which may complicate identification, even if the development in technology made improved image resolution possible. The advantages of photoquadrats are amongst others the non-destructive nature of this sampling method and decrease in bottom time and hence number of dives needed for a specific number of samples, decompression time as well as required gas, compared with measuring faunal and floral communities underwater and writing down in notes (Bravo et al. 2021). This method has become popular as it allows scientists to study the benthic communities in their natural habitat. "Photographic quadrat sampling is commonly used for the study of sessile benthic communities", and can be used to investigate a wide range of organisms, including sponges, algae, and other invertebrates (Trygonis & Sini 2012).

In comparison to other research methods photo-quadrat method is less time-consuming and hence makes it possible for scientists to obtain a higher amount of data (Foster et al. 1991).

2. Aims of Thesis

There is a lack of study on whether wreck-reefs can serve as a substitute for natural reefs (NRs) in terms of conservation and utilization. At my best knowledge, no study has compared the sessile organisms in the waters around Malta with those found on an NR in similar conditions. Given the degradation of marine ecosystems, it is important to compare biotic assemblages on wrecks and NRs to assess the suitability of wrecks for marine conservation. The thesis aim was to investigate whether scuttled wrecks can serve as artificial reefs (ARs) in Maltese coastal waters and contribute to filling the current gap in research on the topic.

2.1. Research Objectives

Objective #1 – To identify the species composition of biotic assemblages and to compare the biological diversity of sessile biota and fish between two wrecks and adjacent NR in similar environmental condition. Specifically using:

1. Alpha diversity
2. Tax. unit richness
3. Beta diversity
4. Shannon H Index

Objective #2 –To calculate the estimated cover of biotic assemblages on two wrecks and adjacent NR. Specifically, to compare:

1. Cover in cm² on ARs/NRs
2. Cover in cm² on vertical/horizontal direction
3. Mean cover of lowest taxonomic unit on ARs and NRs

Objective #3 – To review the conservation status of identified biota and fish on wrecks and NRs.

2.2. Research Questions

Perkol-Finkel et al. (2006) found in their comparison of benthic communities on a Wreck as AR and a nearby NR, that the AR does not mimic NR communities, unless it possesses similar structural characteristics. When AR and NR differ in structure, the communities vary. **Present work aimed to answer the question, if the assemblages on the AR in Ċirkewwa Marine Park differed with those on the nearby NR, when it comes to the presence of taxonomical units.**

Besides that, we aimed to clarify if the abundance **of taxonomical units,** in terms of the **cover, in the assemblages on the AR in Ċirkewwa Marine Park differed significantly when comparing to the abundance of taxonomic units in assemblages on the NR.**

As all NR, the NRs in the Ċirkewwa Marine Park allow for a longer time of colonization and establishment in the marine environment. The studied ARs were established in 1992 and 2007 and hence had a shorter colonization time. This lead to the assumption, that the biological diversity on an AR will differ and be lower than the biological diversity on a

NRs. Further, this assumption was strengthened, as the NRs were characterized by higher heterogeneity and structural complexity, than the wrecks. **We aimed to compare the biological diversity on ARs and NRs.**

After assessing these questions, we sought to review the conservation status of found **organisms and to conclude if scuttled wrecks in the Ċirkewwa Marine Park, as ARs, can support the local marine ecosystem and have importance in conservation of marine species and environment.**

3. Methodology

3.1. Study site

The study was conducted in the Ċirkewwa Marine Park (coordinates: 35°59.21544', 14°19.45812') in Malta which was established in 2021 (Fig. 8a,b,c). Due to the extensive reefs and habitats, the marine park contains high marine biodiversity and species of international conservation importance (Ċirkewwa Marine Park 2022).

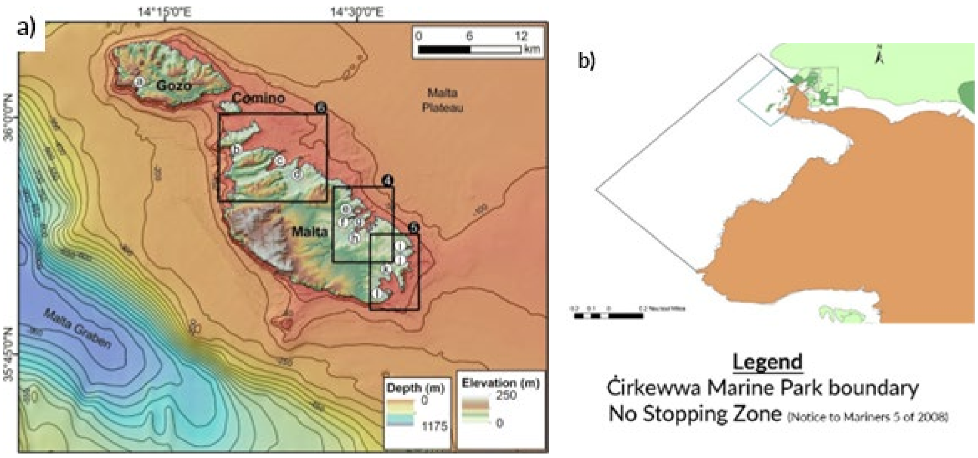
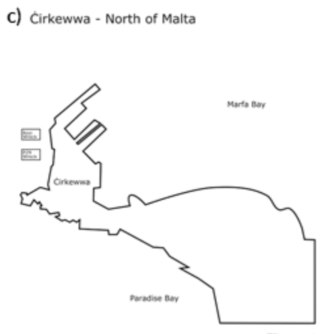


Figure 8: a) Map of Malta (Mueller et al. 2020) b) Ċirkewwa Marine Park boundaries (Ċirkewwa Marine Park 2022) c) Map of Ċirkewwa Marine Park



The tugboat MV Rozi, which was operating in Grand Harbour of Valletta, and the patrol boat P29 from the Maritime Squadron of the Armed Forces of Malta, were scuttled intentionally in 1992 and 2007, respectively, for diving purposes (Malta Independent 2013; Claudia Cuskelly 2016).

Both Wrecks lie at a depth of approximate 35 metres,relatively close to the shore, with a distance of c. 130 metres from the entry point to Rozi and c. 150 metres from the entry point to P29 (Fig.9, 10) . The two wrecks are situated at 100 metres from each other ("P29 wreck - Ċirkewwa - Malta Dive Sites" n.d.).

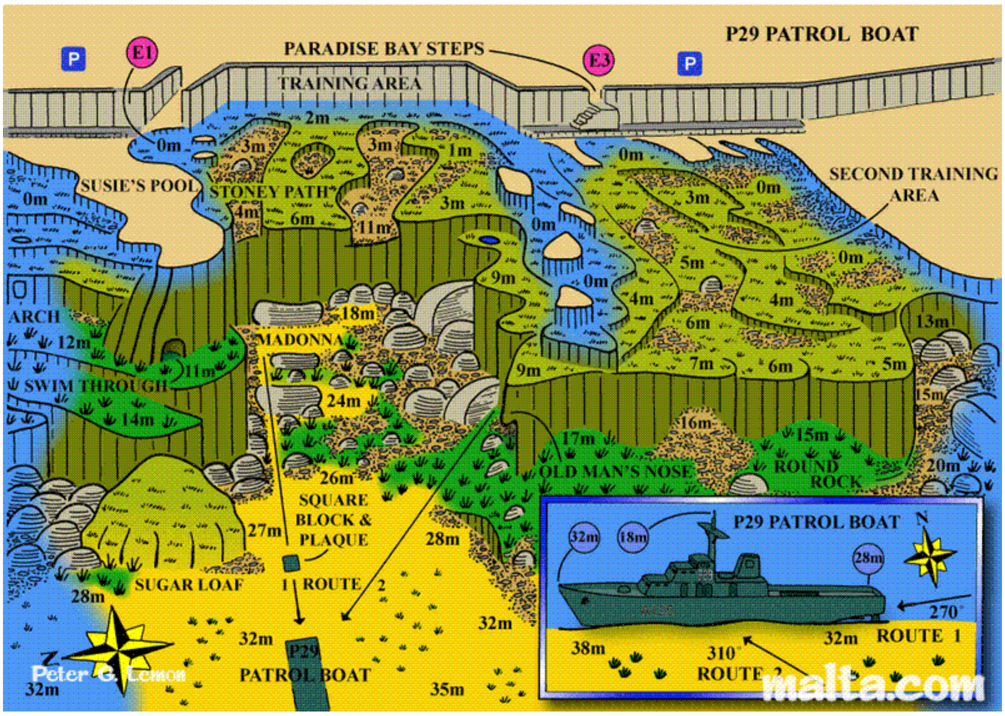


Figure 9: Dive-Site P29 (Lemon 2016)

The mean average sea surface temperature during the data collection was 21 °C and the mean average bottom temperature during the data collection was 18 °C.

Figure 10: Dive-Site Rozi (Lemon 2016)

3.2. Study design and data collection

The data was collected on the Shipwrecks P29 and Rozi and their adjacent reefs during the months of June, July, August and September 2022. For this, 28 dives of 60 min (average time) to maximum 36 meters depth were conducted.

Following tools were used by two divers during the data collection:

Complete equipment set-up in DIR-style: Dry-suit; two first stage and two second stage regulators with long hose and bungee for the alternative breathing source; diving computer and compass, 50 cm x 50 cm photoquadrat, GoPro7, GoPro8; Surface Marker Buoy with spool; Wetnotes; Main Mask; Back-up Mask; Fins; Data-loggers; diving lights with canister batteries (Fig. 11).

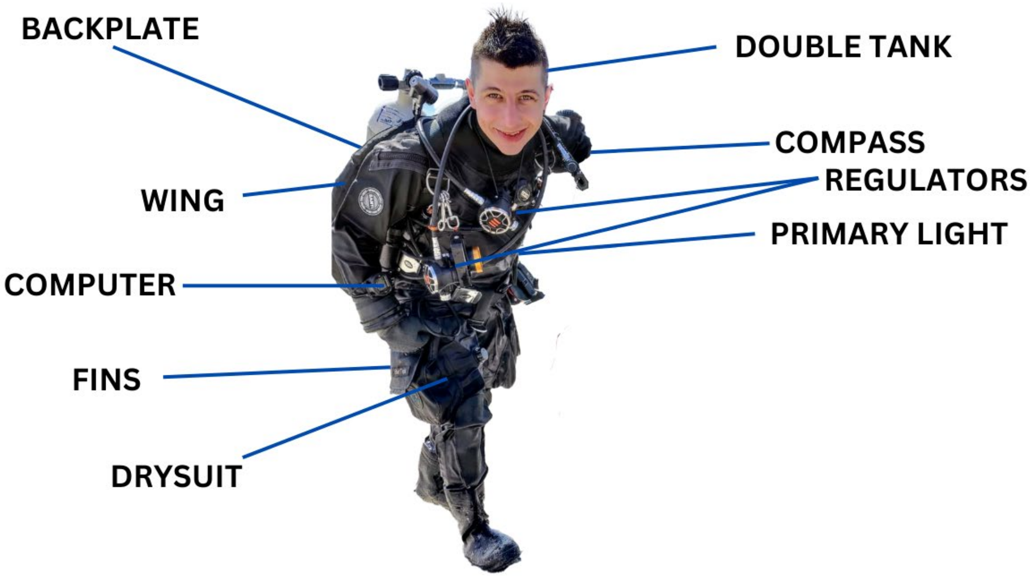


Figure 11: Survey Diver with equipment used during the data collection

3.2.1. Photoquadrat survey design

A total of 163 plots of photoquadrats with a size of 50 x 50 centimeters were collected. 110 data records were used after repeating a number of sample-takes because of blurred resolution, lack of light, and other technical reasons. This specific photoquadrat-size was chosen, as bigger quadrats would easily complicate the field work unnecessarily, and additional complications and risks of entanglement with diving gear was aimed to be minimized.

The final photoquadrat samples were taken with a GoPro8 and a GoPro7 camera. An additional canister-dive light was used to illuminate the organisms and aid following identification procedure. The photoquadrats were recorded once from a distance of approximately 40 centimetres and following with closeups of the specific organisms present.

To be consistent with the data collection the quadrats were taken in a similar scheme on both wrecks and NRs, with 27 quadrats on vertical areas and 6 quadrats on horizontal surfaces (Fig. 12, 13). Compared to ship-reefs, NRs have more diverse and varied structures, resulting in surfaces that are not always perfectly horizontal and vertical.

Figure 12: P29 Plot stations (Polanddivingphoto)

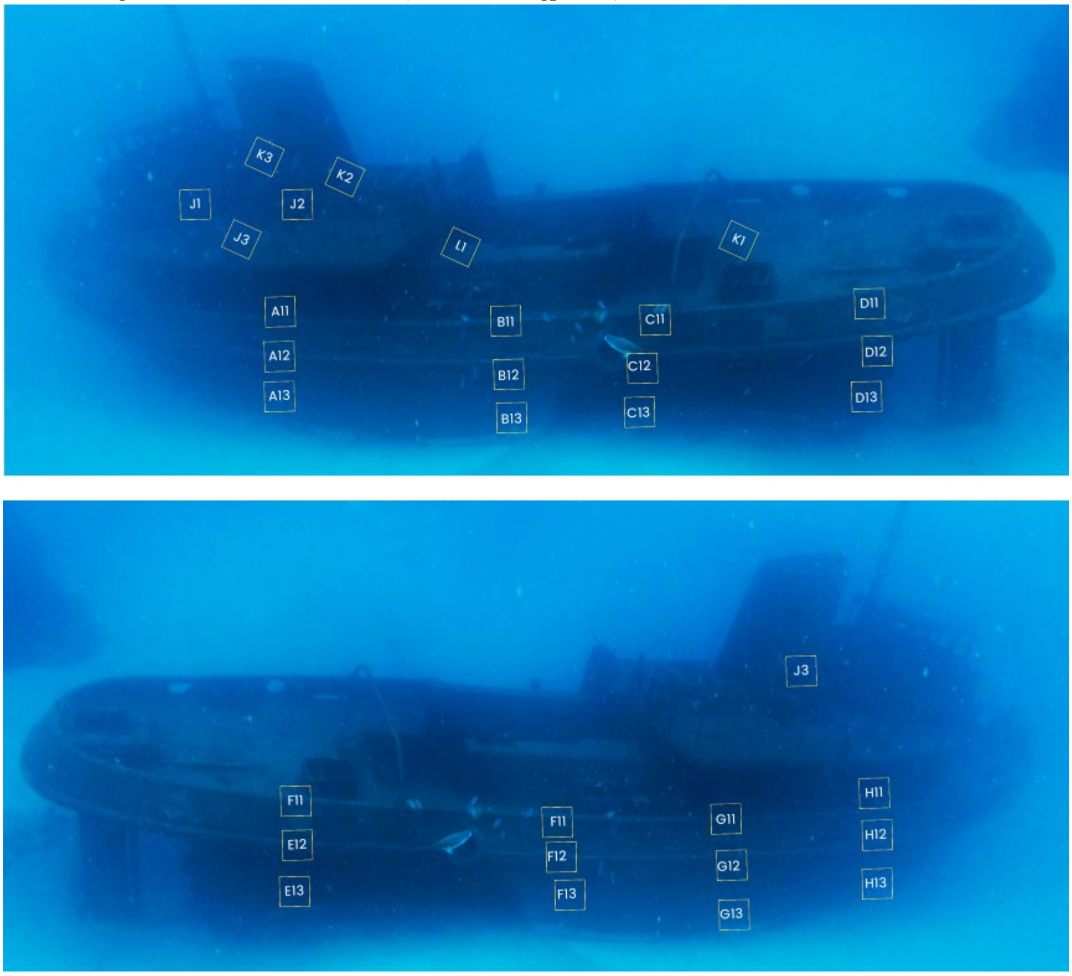


Figure 13: Rozi Plot Stations (Polanddivingphoto)

3.2.2. Fish survey

Additionally, video-transects were conducted, to assess the fish-species present on both ARs and NRs, using the same recording time and swimming pace and keeping a constant transect width of field of view of the camera. Two video-samples with a duration of 10 minutes were collected at each station with a time-distance of two weeks. This method was chosen over a stationary video-method as stationary recordings led to documentation of the same, or a small number of species, as different fish-species were accumulating and present in different areas of the shipwrecks and NRs. A total of six samples of 10 minutes were recorded, with two insufficient samples due to fogging camera lenses.

3.3. Data processing and statistical analyses

3.3.1. Species community composition and structure

The collected pictures of photoquadrats were sorted by site and station. First, the lowest taxonomic unit was investigated using mainly Atlante di Flora e Fauna del Mediterraneo: guida alla biodiversitá degli ambienti marini (Trainito 2011) to aid identification. Images of low quality, which were not applicable for data extraction were filtered out, whilst images suitable to identify biota were used for further analysis.

To compare the diversity of sessile biota between two wrecks and adjacent NRs in similar environmental conditions, alpha, beta, and Shannon diversity indices were used.

Alpha diversity, as an index of species richness, was calculated by counting the number of the lowest tax. unit per site. We conducted one-way ANOVA and a post-hoc HSD Tuckey test to test for differences in tax. unit Richness between NR and AR.

Beta-Diversity is an index of the variation in species composition or community structure between different ecosystems or habitats. Here we used the Sørensen Index, with the formula: $SI=(2*EC)/(E1+E2)$, whereby EC is the total number of species (tax. units) in common between the sets, E1 is the number of species (tax. units) in set 1 and E2 is the number of species (tax. units) in set 2 (Carass et al. 2020).

Then, the Shannon-H Index, which examines richness and evenness (relative abundance) was used (Fedor & Martina Zvarikova 2019; Konopiński 2020). Here, the Shannon-H

Index was calculated per plot using the lowest tax. unit found, instead of species, using following formula:

$$H = -\Sigma p_i * ln(p_i) \quad ,$$

where pi is the relative abundance of the ith species (tax. unit) in the sample containing S species (tax. units).

3.3.2. Cover of sessile biota

For each lowest taxonomic unit identified per photoquadrat, the coverage (in cm^2) was calculated from images of individual plots using ImageJ Software (Schneider et al. 2012). Coverage was extracted to Excel (Microsoft Corporation 2018) and sum of cover per plot (for all lowest tax. unit present per plot) was calculated. Additionally, mean cover of detected lowest taxonomic unit per plot was calculated.

To test for the differences between two wrecks and adjacent NR, in terms of the total cover of sessile biota, and cover of the lowest taxonomic unit, respectively, first, Kolmogorov-Smirnov tests of normality of data distribution were performed to test for meeting assumptions to use parametric tests. Then, factorial ANOVAs were applied, entering “Type” (AR/NR), “Direction” (V/H), and their interaction as independent categorical variables.
Statistical analyses were performed using TIBCO Statistica 14.0.1. (Data Science Workbench 2020).

3.3.3. Species richness and diversity of fish

To identify the species richness of fish, the lowest taxonomic units were identified using Atlante di Flora e Fauna del Mediterraneo: guida alla biodiversitá degli ambienti marini (Trainito 2011).

To compare the diversity of fish between two wrecks and adjacent NR in similar environmental conditions we used:

- Alpha-diversity (number of detected lowest taxonomic units) on ARs and NRs, as well as exclusively present units on ARs and NRs,
  and
- Beta-Diversity and the Sørensen Index, with the formula: *SI=(2*EC)/(E1+E2)*, whereby EC is the total number of species (taxon units) in common between the sets, E1 is the number of species (taxon units) in set 1 and E2 is the number of species (taxon units) in set 2 (Carass et al. 2020).

3.3.4. Conservation Status of Identified Sessile Biota and Fish on Artificial Reefs and Natural Reefs

For each taxonomical unit found, the conservation status was reviewed using the IUCN Red List and EUNIS species component (EUNIS -Species Database n.d.; IUCN 2022).

4. Results

4.1. Species community composition and structure

A total of 41 tax. units were identified at the study site, with 27 of them found on ARs and 34 on NRs. Among these units, 5 were exclusively present on ARs, and 12 were exclusively present on NRs.

Additionally, 1 group of unidentified sessile organism was found on ARs, and 2 groups of unidentified sessile organisms were found on NRs.

Furthermore, 15 tax. units were found on both ARs and NRs, with an additional discovery of 3 groups of unidentified sessile organisms present on both reef types (Table 1).

Table 1: List of sessile biota, their location and conservation status. X indicates the presence of the taxon, AR and NR indicate Artificial and Natural Reefs, respectively. Abbreviations for conservation status are: NE – Not evaluated, DD – data deficient.

Tax. unit	**AR**	**NR**	**Conservation status**
Acetabularia acetabulum	X	X	NE
Agelas oroides		X	NE
Amphiora sp.	X		DD
Bugula sp.		X	NE
Caulerpa cylindracea		X	NE
Caulerpa taxifolia		X	NE
Celleporina sp. / Schizomavella sp.	X	X	NE
Chrysymenia ventricose		X	NE
Clathrina sp.		X	NE
Crambe crambe	X	X	NE
Cystoseira sp.	X	X	NE
Dictyopteris sp.	X	X	NE
Dasycladus vermicularis	X		NE
Dictyota sp. 1/ Dictyota sp. 2	X	X	NE
Filograna sp.	X		NE
Flabellia petiolata	X	X	NE
Halocynthia papillosa		X	NE
Halopteris scoparia	X	X	NE
Hydrodrozoa sp.		X	NE
Liagora viscida	X	X	NE
Mesophyllum / Lithophyllum	X	X	NE
Myriapora truncata		X	NE
Padina pavonica	X	X	NE
Palmophyllum crassum	X	X	NE
Peyssonnelia sp.	X	X	NE

Phorbas tenacior	X		NE
Phoronis sp		X	NE
Protula sp.		X	NE
Reteporella sp.		X	NE
Sabella spallanzanii	X	X	NE
Sargassum sp. / Dictyota sp.	X	X	NE
Spirastrellidae sp.	X	X	NE
Tricleocarpa fragilis	X		NE
Unidentifed gelatenous alga		X	NE
Unidentified alga sp. 1		X	NE
Unidentified alga sp. 2	X	X	NE
Unidentified sponge	X	X	NE
Unidentified filamentous alga	X	X	NE
Unidentified red alga	X		NE
Zonaria tournefortii		X	NE

The tax. unit richness differed significantly between the ARs and the NRs, and was higher on NRs than on ARs ($F(1.\ 96)=30.90$, $p<0.001$). The tax. unit richness differed significantly between vertical and horizontally taken plots, whereby the richness on vertical directed photoquadrats was higher than the richness on horizontally directed plots ($F(1, 94)=5.61$, $p=0.029$). When taking the interaction of "direction" and "Type", there was no significant difference in the tax. unit richness ($F(1, 94)=0.165$, $p=0.69$) (Fig 14).

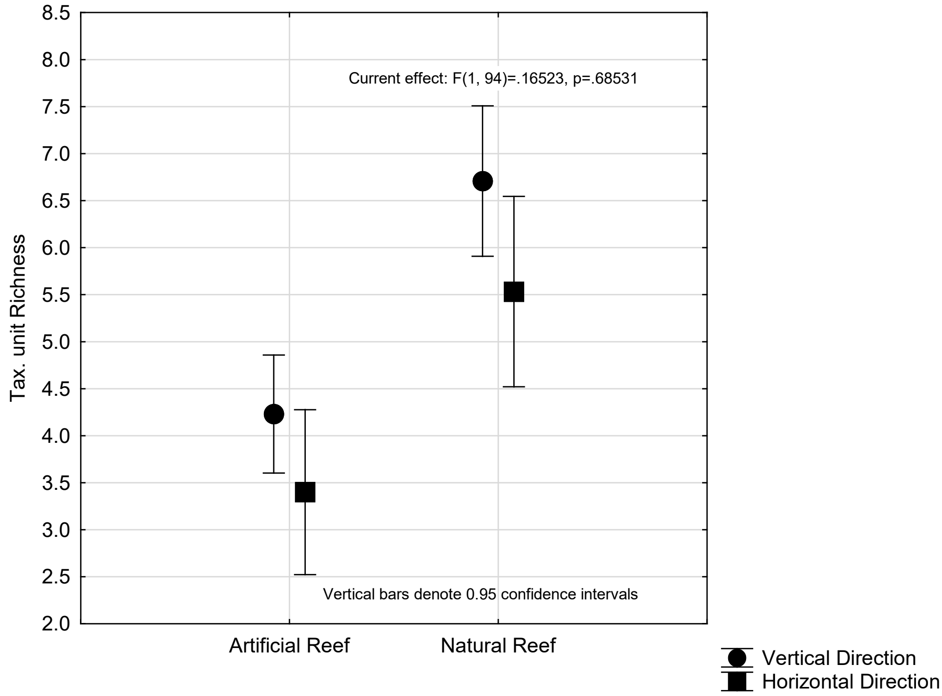


Figure 14: ANOVA (dependent variable: tax. unit richness; direction, independent variable: AR/NR)

The Beta Diversity, a proxy for the degree of community differentiation, using the Sørensen-index was an index of 0.72. The highest Shannon H Index was found on Rozi NR (1.65), followed by P29 NR (1.6) and P29 AR (1.33). The lowest Index was found on Rozi AR (0.52).

4.2. Cover of sessile biota

The tax. units in the communities covered 1499 cm^2 per plot on average (±92 cm2 SE). The mean cover of the tax. units was 1523 cm^2 per plot (±115 cm^2 SE) on ARs and 1463 cm^2 per plot (±153 cm^2 SE) on NR and was not different between sites ($F(1, 96)=0.098$, $p=0.754$). There was, however, a significant interaction between the "Direction" (vertical/horizontal) and the "Type" (AR/NR) ($F(1, 94)=15.461$, $p=.0002$). At ARs, vertical plots showed higher cover than on NRs, where horizontal plots had higher coverage (Fig. 15).

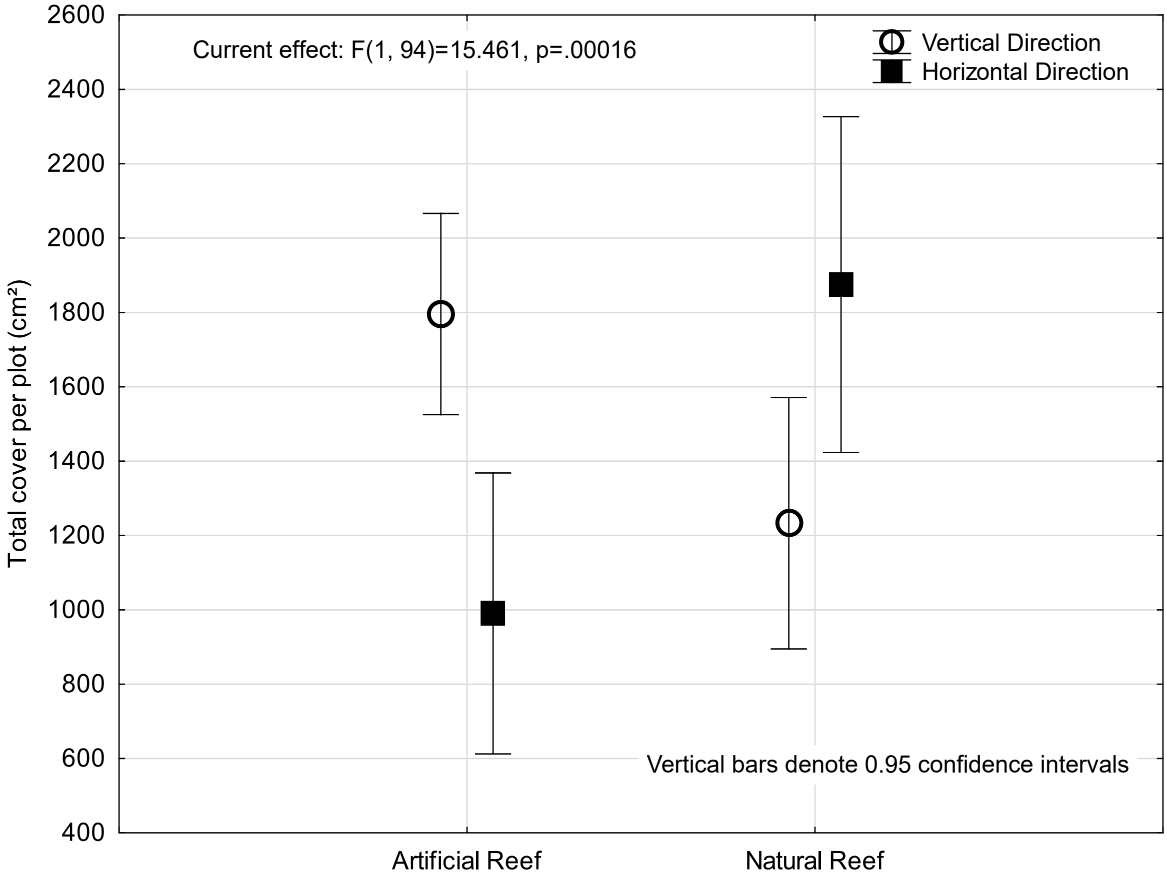


Figure 15: ANOVA (dependent variable: total cover per plot; direction, independent variable: AR/NR)

The unidentified filamentous alga had the highest total coverage per plot on both ARs and non-reserves NRs (Fig. 16).

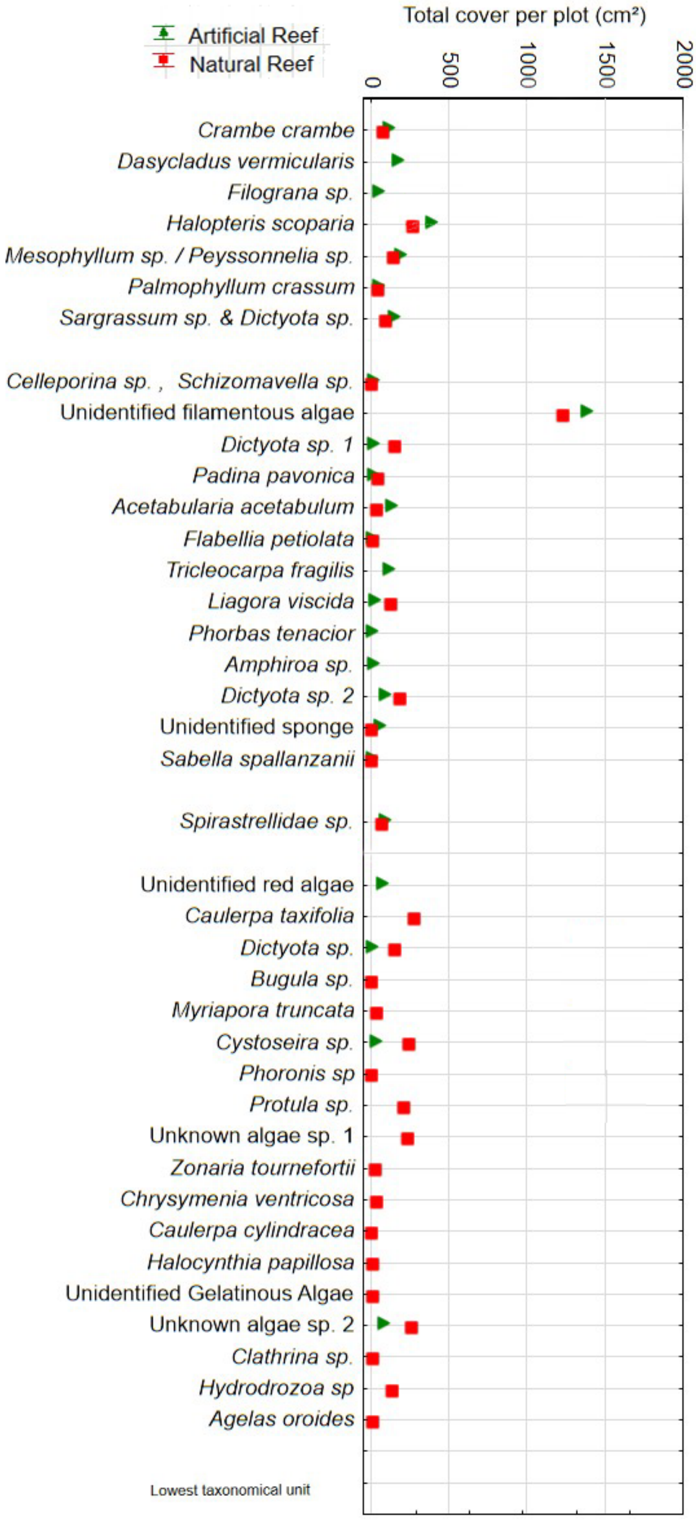


Figure 16: Graph for visualisation (lowest tax unit, AR/NR)

4.2.1. Description of Sessile Biota

4.2.1.1. Organisms found on both Artificial Reefs and Natural Reefs.

Follows a description of the organisms:

Acetabularia acetabulum was a species of green alga present on both AR and NR. This species is a single-celled organism, which belongs to the class Dasycladales (Fig. 17). *Acetabularia acetabulum* plays an important role in the Mediterranean ecosystem as a primary producer. It is a photosynthetic organism, and only visible in spring and summer, as the largest part of the green algae body grows in the summer-months. *Acetabularia acetabulum* is endemic to the Mediterranean and often associated with the sea slug *Elysia timida* ("Acetabularia acetabulum | DORIS" n.d.). We found a sum of 140.42 cm² cover of *Acetabularia acetabulum* on the NRs, and 1578.62 cm² on the ARs.

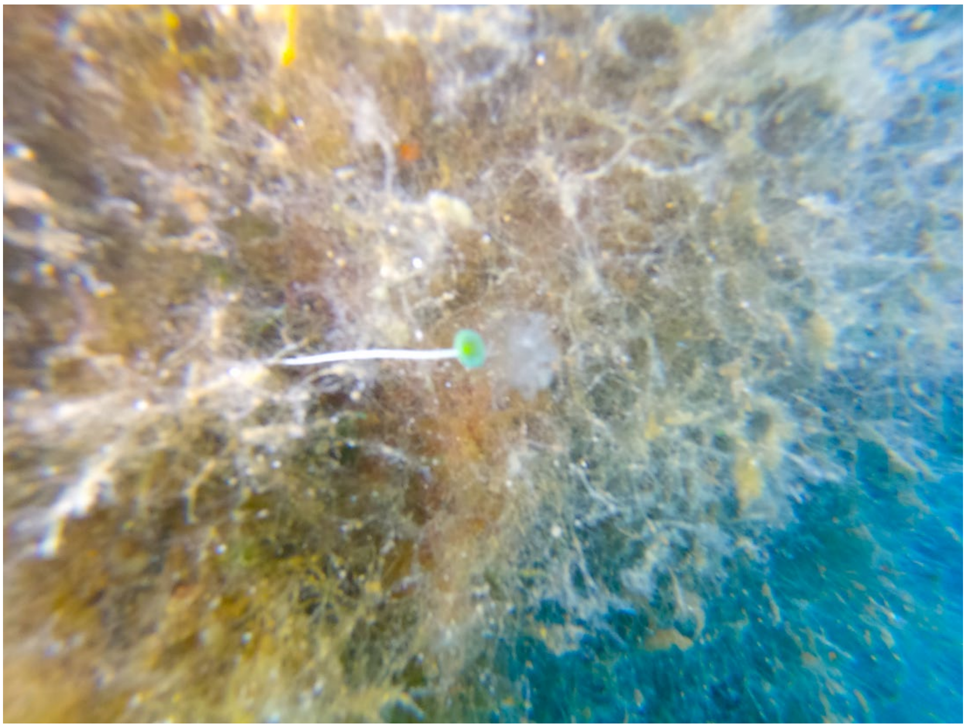


Figure 17: *Acetabularia acetabulum*

Crambe crambe is a widespread and common encrusting sponge in the Mediterranean Sea (Duran et al. 2004), found to a depth of 60 m. It grows on hard substrates and the colony of *Crambe crambe* can reach a up to 1m² (Fig. 18). The surface of the sponge is coarse and perforated by big oscules ("WoRMS - World Register of Marine Species - Crambe crambe (Schmidt 1862)" n.d.). *Crambe crambe* is predominantly present in shady areas, probably because the sponge faces less risk of mortality in shaded places. Marine sponges are important members of the benthic community in the Mediterranean sea, they play an important role in the ecosystem. They are filter feeders, providing habitat for other organisms, such as small fish and crustaceans (Turon et al. 1998). For this species, we found a higher coverage on the ARs than on the NRs, with 1536.33 cm² and 77.56 cm² respectively.

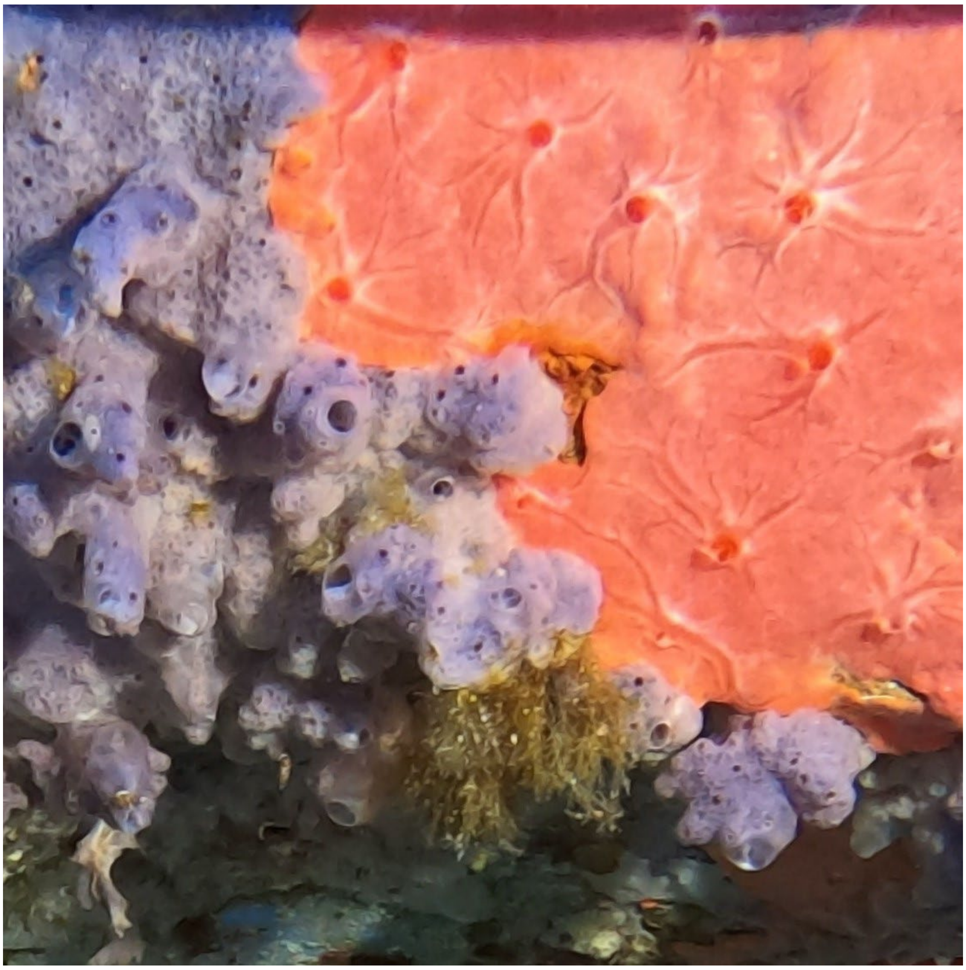


Figure 18: *Crambe crambe*

Species belonging to the genus *Cystoseira* play a major role in the Mediterranean sea, providing habitat, increasing habitat complexity and enhancing biodiversity. The species helps to stabilize the sediment and reduces the impact of waves on the coast and thus protecting coastal habitats. The *Cystoseira*-Ecosystem is threatened through overgrazing, deterioration of water quality, sedimentation and anthropogenic habitat change. The abundance of *Cystoseira* is declining in the Mediterranean, which leads scientists to encourage restoration of species belonging to the *Cystoseira* genus (de La Fuente et al. 2019). Present study found a cover of 2611.63 cm² on the NRs, and 33.4 cm² on the ARs.

Dictyopteris is brown alga, with a broad, worldwide distribution, excluding the polar regions. Nevertheless, most species of the genus inhabit warm-temperate waters, while the minority is found in cold waters (Bogaert et al. 2020). *Dictyopteris sp.* are known for their ability to resist their surface from biofouling, that is the attachment of other organisms (Othmani et al. 2014). *Dictyopteris* species are, like *Cystoseira sp.* known for their large size and complex branching structure, which provides habitat for a wide variety of other organisms. They also provide shelter, breeding and feeding grounds. Furthermore, *Dictyopteris sp.* act, as *Cystoseira sp.* as a physical barrier. During present study, we found that *Dictyopteris sp.* was present on both NRs and ARs, with 1629.02 cm² and 27.93 cm² respectively.

Dictyota sp. 1 was found on both NRs and ARs, with a cover of 905.55 cm² and 40.65 cm² respectively.

Dictyota sp. 2 was found on both NRs and ARs, with a cover of 365.9 cm² and 92.29 cm² respectively.

Celleporina sp. and *Schizomavella sp.,* are calcareous marine bryozoans belonging to the order *Cheilostomatida.* They are colonial animals forming encrusting colonies. These filter-feeders attach to rocks and algae, as well as to other structures. The bryozoans are important components of the benthic community, forming reef structures, cleaning the water and providing habitat for other organisms. We discovered the representatives of this order in both NRs and ARs, covering only 4.24 cm² on the NRs and a higher area of 45,58 cm² on the ARs.

Flabellia petiolata is the only species of the genus *Flabellia* (Fig. 19). The green algae inhabits rocky substrates of the Mediterranean Sea and the Atlantic Ocean and associates with other algae, such as *Dictyopteris spp*. and *Dictyota spp.* and is often found in areas, where *Posidonia sp.* flourishes (Gnavi et al. 2017). We found the green alga both on NRs and on ARs, with 68.5 cm² and 187.54 cm² coverage respectively.

Figure 19: *Flabellia petiolata*

Halopteris scoparia is dark brown algae, with cosmopolitan distribution in waters around Europe. It is a large, bushy brown seaweed that can be found in rocky shores and subtidal zones. The seaweed is a light-loving species, found in waters with high light intensity ("MACOI - Portuguese Seaweeds Website" n.d.). *Halopteris scoparia* was found on NRs and ARs, with 3486.61 cm² and 6953.629 cm² respectively.

Liagora viscida is a red algae, mainly inhabiting the Mediterranean, and was found both on NRs and ARs, with a cover of 126.21 cm² and 27.26 cm² respectively.

Mesophyllum, Lithophyllum are both genera of red algae that are commonly found in the Mediterranean Sea and the eastern Atlantic Ocean. They are member of the belong to the order Corallinales. The calcified algae embody calcium carbonate. They form a specific habitat by the accumulation of calcareous encrusting algae growing in dim light conditions" (Gibson et al. 2006). At the same time, they provide habitat, such as Rhodolith beds (complex communities of coralline algae with variable body size and shape) , which in turn act as nursery area for related organisms and are crucial for ecosystem function.

Coralline algae are furthermore significant in the carbon and carbonate cycles (Martin & Gattuso 2009; Kamenos et al. 2013). The habitat that coralline algae form is commonly considered to be of diverse and “the most important reef of the Mediterranean” (Foster et al. 2013; Basso et al. 2022). The coralline algae use calcium carbonate to build their cell walls and skeletons, which gives them a hard, encrusting or massive form. The calcifying properties makes them vulnerable to global change and the resulting ocean acidification (Martin & Gattuso 2009; Kamenos et al. 2013)

Peyssonnelia does not belong to the order Corallinales, but shares features, in that it is a calcifying algae and possesses similar appearance. Other studies have merged *Peyssonnelia* and Corallines in one group (Aponte & Ballantine 2001; González-Delgado & Hernández 2018).

Padina pavonica is distributed mainly in the Mediterranean Sea and the Atlantic Ocean, The thallus detaches in winter months and regrows in spring (Fig. 20). *Padina pavonica* is important and interesting, as it is next to *Newhousia imbricata* the only representative of brown algae, known the its calcification. The ocean acidification threatens *Padina*, as it also puts other calcifying organisms at risk to decalcify (Benita et al. 2018). The present study found *Padina pavonica* on both NRs and ARs, with a cover of 1087.31 cm² and 378,63 cm² respectively.

Figure 20: *Padina pavonica*

Palmophyllum crassum was found on both NRs and ARs, with a cover of 764.75 cm² and 1162.290 cm² respectively.

Sabella spallanzanii is a widely distributed and well-known member of the Mediterranean polychaetes (bristle worms), also known as Mediterranean fanworm (Giangrande et al. 2000). It is one of the largest members of *Sabellidae,* and can grow to 0.5 m (Read et al. 2011). The bristle worms were present on both NRs and ARs, covering 2.89 cm² and 4.82 cm² respectively.

Dictyota and *Sagrassum* species belong to the brown macroalgae (class *Phaeophyceae*). The genus *Sargassum* carries 335 taxa, from which only 9 species are known to be present in the Mediterranean. *Sargassum* carries some round, gas-filled structures, which enables it to float (Tara L. Casazza & Steve W. Ross 2022). *Sargassum sp.* are biomass producers, which are known for the broad beds they cover (Marquez et al. 2015). The seaweed forms three-dimensional habitats, supplying nursery, shelter and food for many species (Thibaut et al. 2014). *Dictyota sp*. belong to the family *Dictyotaceae* and are distributed in tropical and subtropical oceans. *Sagrassum* and *Dictyota* covered both NRs and ARs with 1142.588 cm² and 2220.56 cm² respectively.

The members of the Spirastrellidae marine sponges are generally colourful and grow in shady habitats such as caves, overhangs and the undersides of rocks. They are found in tropical and subtropical shallow water habitats (Rützler 2002). We found that *Spirastrellidae sp.* were both present on NRs and ARs, covering 1037.388 cm² and 1990.966 cm² respectively.

We classified a "unidentified filamentous alga" and found its high presence on both NRs and ARs, with 33102.46 cm² and 66361.36 cm² respectively. The alga was more abundant in ARs than in NRs. This biota had a soft, moss-like texture (Fig. 21).

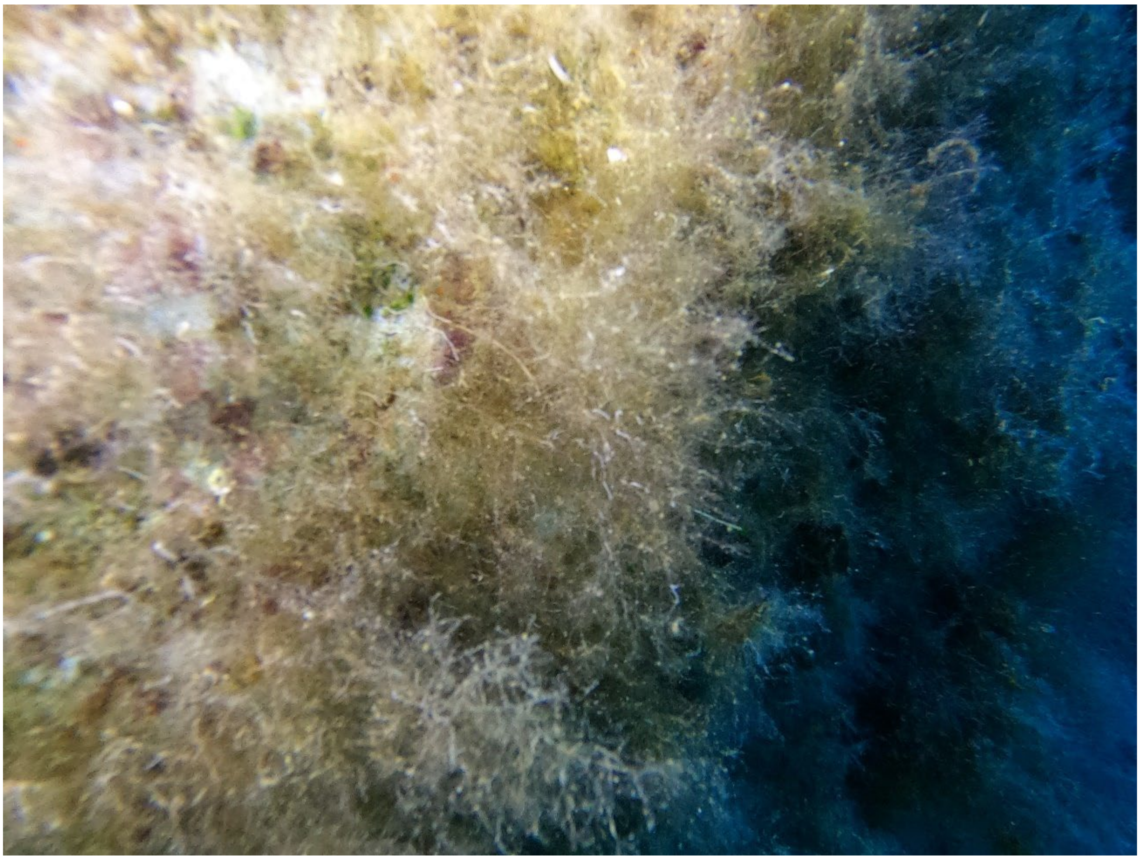


Figure 21: Unidentified filamentous alga

On both NRs and ARs we could find sponges but were unable to identify them. The coverage for them were 6.95 cm² and 187.85 cm² respectively.

On both NRs and ARs, we were unable to identify an alga, which we classified as "unidentified alga sp. 2", covering 256.86 cm² and 79.36 cm² respectively.

4.2.1.2. Organisms found exclusively on Artificial Reefs

Follows the descriptions of the organisms:

Amphiroa is a genus in the Corallinaceae order, occurring in tropical and temperate waters, but also inhabiting colder areas of the oceans. Currently, there a 55 species of red algae represented in the genus *Amphioroa* (Harvey et al. 2019) We detected this genus solely on the AR, with a cover of 19.97 cm².

Dasycladus vermicularis is a alga with cosmopolitan distribution ("Dasycladus vermicularis - Encyclopedia of Life" n.d.). The alga was only present on the photoquadrats taken on the ARS, with a total cover of 2494.77 cm².

The colonial tubeworm *Filograna sp.*was present on the AR, covering 48.3 cm².

Phorbas tenacior was found, covering 6.25 cm² of the photoquadrats taken on the ARs.

Tricleocarpa fragilis is a calcified red alga and was found to be present on the ARs, with a cover of 120.28 cm².

A red alga could not be identified on the AR, covering 145.39 cm².

4.2.1.3. Organisms found exclusively on Natural Reefs

Follow the description of the organisms:

Agelas oroides is a widely known sponge, distributed in the Mediterranean Sea and occurring up to a depth of 150 m. It is growing commonly in shady, sheltered habitats like crevices and caves. This sponge adds structural complexity to the ecosystem and provides habitat for other organisms (Idan et al. 2020). We found this species of sponge only on the NRs, covering 17.13 cm²of photoquadrats taken.

Representatives of the genus *Bugula* are Bryozoans, common in shallow tropical and temperate waters (Fehlauer-Ale et al. 2015). Bryozoans play a significant role in benthic habitats, as forming three-dimensional structures (Giampaoletti et al. 2020). We found this genus solely on the NR, covering 3.43 cm² of the taken photoquadrats.

The red algae *Chrysymenia ventricosa* was only found on the NR, covering 71.53 cm² of the taken photoquadrats.

Caulerpa cylindracea and *Caulerpa taxifolia var. distichophylla* are alien green algae, considered as invasive, fast spreading species in the Mediterranean (Montefalcone et al. 2015). The consequence of the invasion is the alteration of the structures in the Mediterranean Basin by reducing diversity (Piazzi et al. 2016). We found the algae only on NRs, covering 1659.31 cm², and not on the ARs.

Clathrina sp. has the "simplest organization among the sponges" (Ahyong et al. 2023). 68 species are represented in this genus (Klautau & Valentine 2003). The calcareous sponge was not represented on the ARs, but with a cover of 12.73 cm² on the NRs.

Halocynthia papillosa is a species of solitary tunicate, which occurs in depths from 2 to 100 m. The species is one of the most common Ascidians in the Mediterranean and grows mostly on coralline algae and shells (Klautau & Valentine 2003). We found the suspension feeder on the NR, covering 8.46 cm².

A cover of 132.11 cm² of other *Hydrodrozoa sp.,* as the ones already mentioned in the text, were found on the NRs.

Myriapora truncata is widely distributed in the Mediterranean Sea and is found on rocks and shells up to 130 m (Berning 2007). The "false coral" is a species from the phylum Bryozoa and covered 121.66 cm² of the photoquadrats taken at the NRs.

The tube worms *Phoronis sp.* are members of the Family Phoronidae. Five species are known in the Mediterranean. The suspension feeders live buried in soft sediments or settle on rocks and shells (Trainito 2011). We found *Phoronis sp.* on the NR, covering 0.25 cm².

Protula sp. are marine worms from the Family Serpulidae. The family name is derived from "serpula", latin for "small snake" and indicates the serpentine shape (Southward 1963). We found a cover of 1430.928 cm² on the NRs.

We found an interesting gelatinous alga on the NR, covering 14.57 cm² (Fig. 22).

Figure 22: Unidentified gelatinous alga

Reteporella is the largest genus in the family Phidoloporidae. *Reteporella sp*. are distributed from polar regions to the Tropics (Denisenko 2022). We found a cover of 2.35 on the NRs.

Zonaria tournefortii is a species of the genus Zonaria. The brown seaweed has been documented in the Mediterranean Sea and the South Africa's eastern coast (Montañés et al. 2006). It was present on the NRs, covering 90.57 cm².

Furthermore, there was an algal species with coverage of 2545.98 cm², which we were unable to identify and named "unidentified alga sp. 1" (Fig. 23).

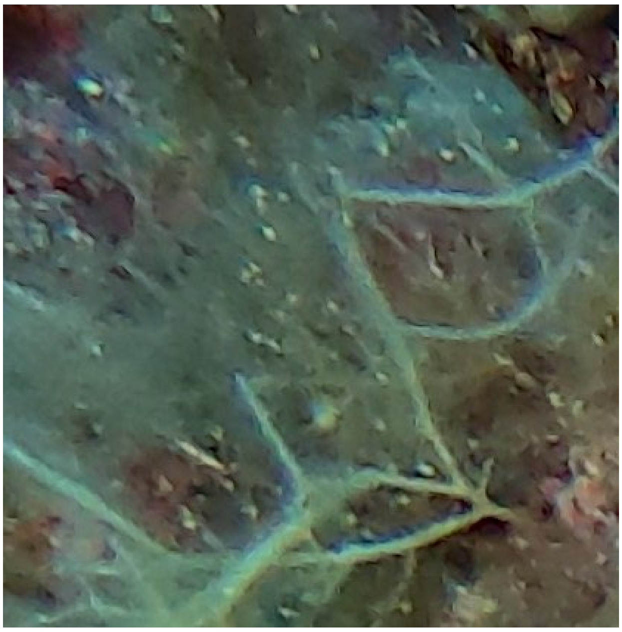


Figure 23: Unidentified alga sp. 1

4.3. Biological Diversity of Fish

The tax. units present overall was 29, while exclusively on ARs detected fish groups consisted of 7, and on NRs 10.

The Alpha Diversity, that is the diversity given in a particular area, on the ARs was 20, and on NRs 20 (Table 2).

Table 2: : List of fish, their location and conservation status. X indicates the presence of the taxon, AR and NR indicate Artificial and Natural Reefs, respectively. Abbreviations for conservation status are: LC-Least concearn, VU – vulnerable.

Tax. unit	AR	NR	Conservation status
Anthias anthias		X	LC
Apogon imberbis		X	LC
Boops boops	X	X	LC
Centrolabrus melanocercus		X	LC
Chromis chromis	X	X	LC
Coris julis	X	X	LC

Dentex dentex		X	VU
Diplodus puntazzo	X	X	LC
Diplodus sargus	X	X	LC
Diplodus vulgaris	X		LC
Epinephelus costae	X		LC
Epinephelus marginatus		X	VU
Hyporthodus haifensis	X		LC
Mullus surmuletus		X	LC
Muraena helena	X		LC
Mycteroperca rubra	X		LC
Oblada melanura	X	X	LC
Sarpa salpa		X	LC
Scorpaena porcus		X	LC
Seriola dumerili	X		LC
Seriola fasciata	X		LC
Serranus cabrilla	X	X	LC
Serranus scriba	X	X	LC
Siganus luridus		X	LC
Sparisoma cretense	X	X	LC
Spicara smaris	X		LC
Symphodus ocellatus		X	LC
Symphodus rostratus	X	X	LC
Thalassoma pavo	X	X	LC

As listed above, on the NR, we found *Dentex dentex* specimens which according to IUCN (2022) is classified as vulnerable (Fig. 24). The fish lives close to *Posidonia oceanica* beds, *Caulerpa* habitats and rocky bottoms. It appears until a depth to 200 m. *Dentex dentex* preys on cephalopods, molluscs and fishes (Morales-Nin & Moranta 1997; Stobart et al. 2012; Marengo et al. 2014). *Dentex dentex* is vulnerable to fishing and overexploitation, as for its large size, longevity and slow growth (García-Rubies et al. 2013).

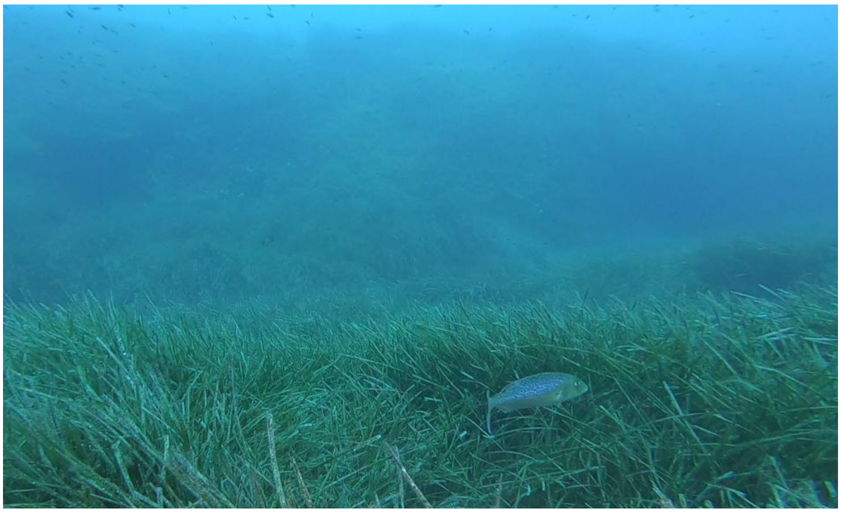


Figure 24: *Dentex dentex*

Another specimen found, the dusky grouper, *Epinephelus marginatus,* was lying on the rocky bottom of the NR during the survey (Fig. 25). This species is classified as vulnerable, accordingly to IUCN (2022). The dusky grouper is a protogynous hermaphroditic species, transitioning from female to male with growth. The grouper is associated with rocky habitats and may act as keystone species in the highest trophic level. It is a long lived, slow growing fish with complex life cycle (Condini et al. 2017). This species is listed as vulnerable and overexploitation is putting the dusky groupers at risk, accordingly to IUCN (2022).

Figure 25: *Epinephelus marginatus*

The Beta Diversity, using the Sørensen-index was an index of 0.55.

5. Discussion

5.1. Species community composition and structure

The Alpha diversity was higher on the NRs than on the ARs confirming the notion that rocky reefs naturally harbour high levels of biodiversity (Guidetti 2006; Walker & Schlacher 2014; Bulger et al. 2019; Bevilacqua et al. 2021).

It is important to note that alpha diversity is dependent on the characteristics of the ecosystem being studied. Therefore, it is not appropriate and of interest for this study to directly compare alpha diversity with reefs in different geographical regions or different ecosystems. Further, we did not find any study that used the same methods and a comparison of the alpha-index with studies using different methodology is incoherent. In our study, we measured the alpha diversity of the NRs and ARs to compare them with each other, which is a meaningful comparison because they are both located in the same general region and have similar environmental conditions.

Further, we found 5 tax. units present exclusively on ARs, and 13 tax. units present exclusively on NRs which suggests that the NRs provides higher amount of special niche characteristics, and hence host more organisms exclusively appearing on the NRs.

It is important to mention, that the longevity of NRs compared to shipwrecks has allowed them to undergo more successional stages and develop complex ecological niches, supporting a diverse and exclusive range of species. Additionally, the prolonged existence of rocky reefs has provided more opportunity for evolutionary changes, leading to the development of complex ecological niches. The significance of preserving the distinct niches found in NRs, as opposed to ARs, is emphasized by our findings. Additionally, it underscores the importance of replicating the structures of natural reefs in artificial reef constructions to facilitate the development of niche-specific life. Further, the NRs have a more complex architecture than the shipwrecks we investigated. The rocky reefs are characterised by diverse and complex structures such as holes and three-dimensional formations. The higher habitat heterogeneity on the studied NRs is likely a contributing factor to the higher tax. unit richness observed on NRs compared to ARs. The calculation of the Shannon H Index revealed accordingly the higher diversity on NRs than on ARs.

The Beta Diversity-Index shows that the taxonomic unit composition on NRs and ARs are not identical but share a high degree of similarity. This can be due to comparable environmental conditions, because the NRs and ARs are located close to each other, and this makes it possible that similar tax. units are able to survive and thrive. Additionally, there may be an exchange of organisms between the NRs and ARs through currents and historical events such as past oceanic condition, climatic changes could have shaped the taxonomic composition of NRs and ARs similarly.

The current study supports the hypothesis by Perkol-Finkel (2006), suggesting that ARs and NRs may host similar communities of organisms if they exhibit similar structural features. Of total 41 there were 15 common identifiable tax. units of sessile organisms on ARs and NRs.
The results of the review of the conservation status of identified biota on ARs and NRs indicates that most species are not evaluated for their conservation status. This highlights the need for further research on these species to determine their conservation status accurately.

5.2. Cover of Sessile Biota

The overall cover of sessile biota in cm² per plot in AR and NR did not differ significantly.

Interestingly, vertical plots on the ARs showed higher sum of cover in cm² than horizontal plots on ARs, while horizontal plots on the NRs showed a higher sum of cover in cm² than vertical plots on the NRs. This means, the orientation of the plots had a different effect on the coverage of organisms depending on whether they were on NRs or ARs.

The reasons for that are unknown, but one possibility could be that the vertical surfaces on ARs may experience water flow extremes, compared to the horizontal surfaces. The areas with high, perpendicular water flow could help to distribute nutrients more evenly and promote the growth of certain organisms. Baynes and Szmant (1989) also found higher cover of benthic organisms in areas of strong current. Vertical surfaces on ARs may be more conducive to the growth of certain encrusting organisms, such as sponges, who are better adapted to high water flow conditions. This is because the vertical orientation can provide better exposure to currents, allowing these organisms to capture food particles from the water column and to compete for space more effectively (Duckworth et al. 2004) Additionally, vertical surfaces that face directly the water flow, may receive more larvae and spores that can settle on the AR, while at the same time the other side of the AR would be totally sheltered from current, which would leave the sessile organisms on that side undisturbed and conserved, creating stable microhabitats. In contrast, vertical surfaces on NRs may not have the same level of water flow due to the complex and varied structure of the reef, resulting in lower coverage of sessile biota. The opposite is true for horizontal surfaces. In areas with high water flow, horizontal surfaces on natural reefs may be more favourable for the growth of certain filter-feeding organisms that can capture food particles from the water column. NRs offer a wide variety of orientations and surfaces that can be colonized by various organisms. However, on ARs such as shipwrecks, the horizontal surfaces may not be as exposed to water flow due to their location and structure, resulting in lower coverage compared to NRs.

The higher cover of horizontal plots on NRs than vertical plots could be explained by the amount of sunlight reaching the horizontal plots, and hence the growth of photosynthetic organisms, whereas, in comparison, vertical surfaces are penetrated less by sunlight. The NRs were facing to the west and horizontal plots received mostly sunlight. The ARs were scuttled in a more open space, where the sunlight reaches most of the day the wreck, but the vertical surfaces were, similarly to the NRs mostly shaded from the above-structure of the wreck. This could lead to the high cover of light-independent sessile organisms on the vertical plots of the wrecks. This work did not analyse statistically the community composition on vertical plots of the ARs and NRs, but it is likely that the vertical areas on the ARs were mostly covered by light-independent biota, and the horizontal surfaces on the NR by photosynthetic and light-dependent algae. The ARs feature simpler structural configurations and more uniform surfaces, which can create optimal conditions for the growth of these specific, light-independent organisms, such as sponges, whereas the horizontal surfaces of the NRs with their three dimensional structure may attract more algae. This hypothesis and the topic should be further investigated in following studies.

The observed patterns of cover may also be influenced by temperature, as the higher water current on ARs may result in different temperature conditions compared to the lower water current on natural reefs NRs. Water currents can significantly impact water temperature, particularly in areas with strong currents. Consequently, changes in water current can lead to changes in water temperature, which may have effects on the growth and distribution of organisms on both natural and artificial reefs. At the time of sampling there was no temperature difference between NRs and ARs (water temperature at time of sampling in sampling depth and area: 21 °C). A thorough investigation of the complex relationship between water current, temperature, and organism growth would necessitate long-term measurements of both variables over an extended period.

The lower cover of sessile biota on horizontal plots may be caused by sedimentation from water movement, as seen in Fig. 25. Our results align with the findings of Baynes & Szmant, which indicate that areas with higher levels of sedimentation (horizontal plots) exhibit lower coverage compared to areas with lower sedimentation (vertical plots), because sedimentation can negatively impact sessile benthic organisms by blocking pores, impeding feeding, limiting light to zooxanthellae, slowing nutrient exchange, increasing energy expenditure, and obstructing settlement (Baynes & Szmant 1989).

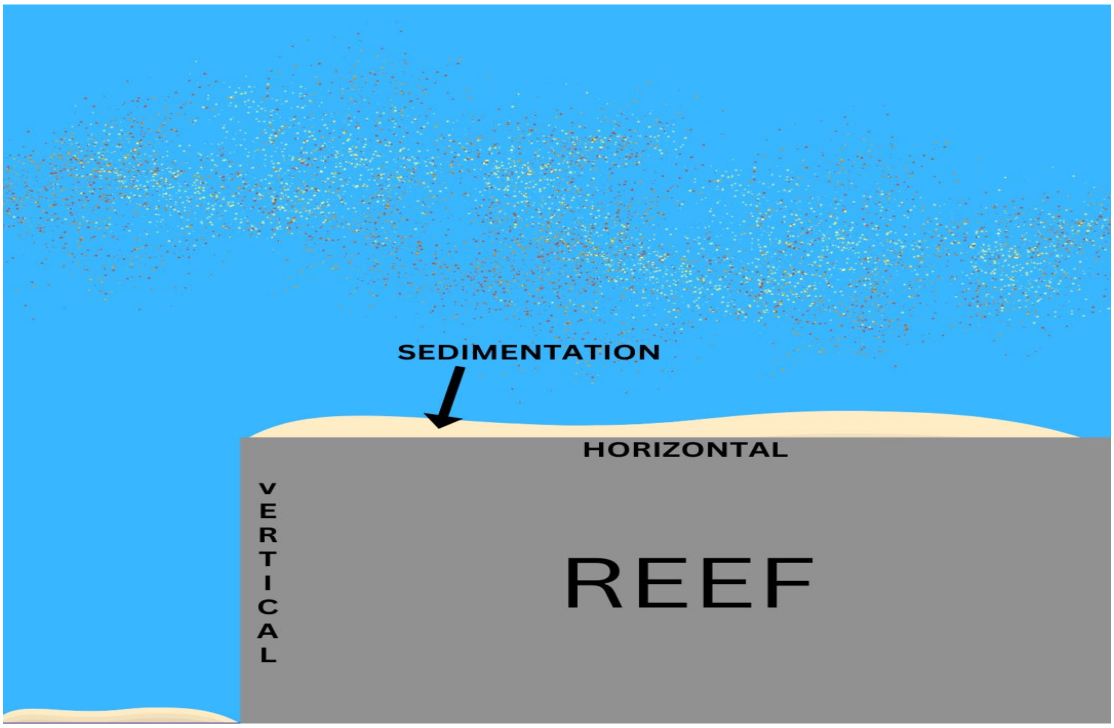


Figure 26: Sedimentation on horizontal surfaces

Besides the basic explanation given above, we do not fully understand how the cover of biota is affected by sedimentation on vertical and horizontal surfaces on any type of reef. On NRs, horizontal surfaces tend to have more complex structure which can trap sediment. We would have expected that this makes it harder for organisms to grow, compared to vertical oriented plots, but it may be also the case that the sediments make it easier for algae to attach. However, there are also factors that are not clear and may contribute to our findings. For example, it is unclear how the stronger currents on ARs affect the accumulation of sediment on horizontal surfaces, or if the structures on horizontal plots should lead to high sedimentation, as they act like holes and can be clogged easier. To understand this better, more research needs to be done.

Another factor that could contribute to our findings on the higher cover of vertical surfaces on ARs and horizontal surfaces on NRs is the disturbance by divers. Horizontal surfaces on the ARs may be imposed by a higher and more frequent amount of disturbances, as divers may tend to swim along and close to the superstructure of the wrecks (Fig. 27), also because divers are interested in visiting the gun on P29, which is laying on the deck, or in penetrating the wreck, whereas, as for the architecture of the NRs, divers may swim rather next to the reef, than over it.

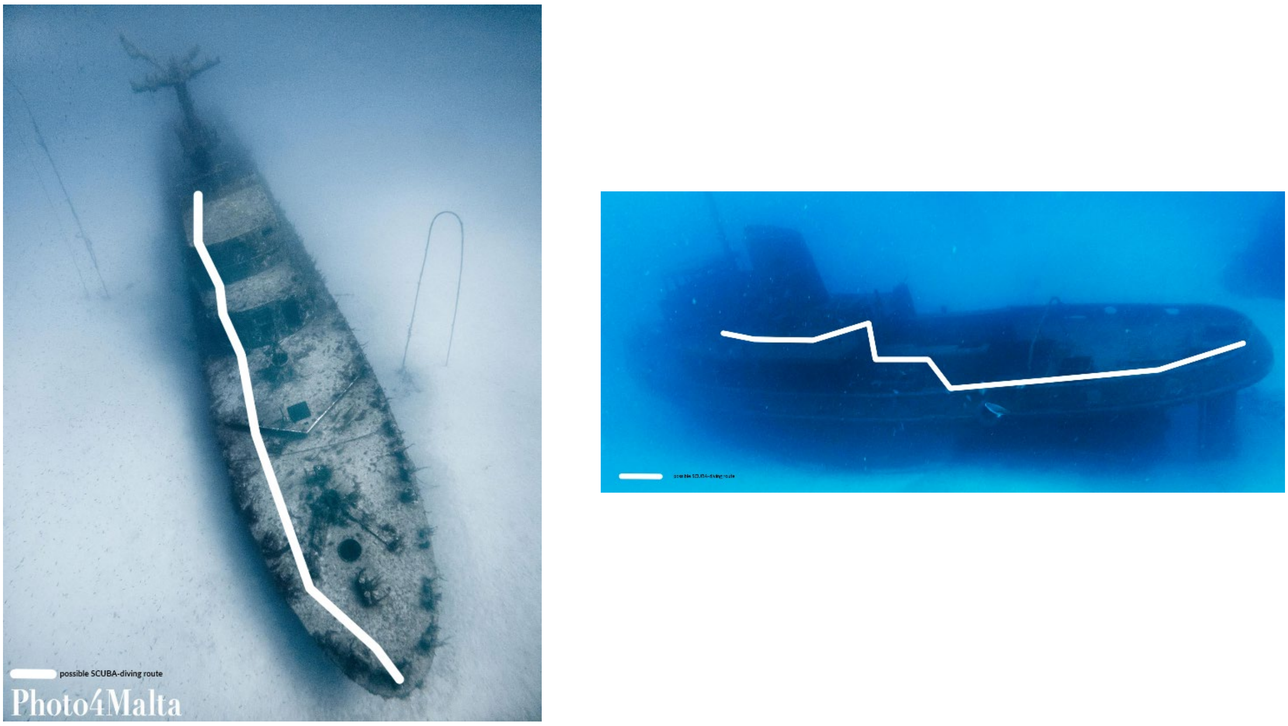


Figure 27: Possible SCUBA-diving route on P29 and Rozi (white path marks an example of frequent diving paths) (Polanddivingphoto)

In present study, we classified a unidentifiable algae as "unidentified filamentous alga". Despite the challenges we encountered in identifying this particular algae, it was evident that it played a crucial role in both natural and artificial reef environments, as it exhibited a high level of coverage. Such turf-forming algae are found to trap sediment and organic matter, thereby providing a favourable environment for detritivores (Bedini et al. 2015). Turf-forming algae may inhibit the recruitment of taller algae by preventing their spores from attaching to solid surfaces or, on the other hand, facilitate the recruitment of some kelps in the intertidal zone by creating suitable microenvironments and providing a refuge from grazing for microscopic stages (Connell et al. 2014). Turf-forming alga are known to compete with other alga such as *Cystoseira sp.* and *Sargassum sp.* and can prevent the recolonization of species that form canopies. Grazers were recognized as a significant facilitator of the substitution of canopy-forming algae with turf-forming algae (Connell et al. 2014). The identification of the role of this alga and the interactions with other sessile biota on the ARs and NRs is reserved for further study.

5.3. Species Richness and Diversity of Fish

When it comes to fish, our results concur with the conclusion of earlier mentioned authors, that ARs can effectively imitate NRs in terms of richness (Fowler & Booth 2012; Paxton et al. 2020). Our study found that there are no differences in alpha-diversity between ARs and NRs. This result is different to earlier mentioned results of Carr & Hixon (1997), who found a higher diversity of fish on NRs, than on ARs. Our study found, coherently to Carr & Hixon (1997), a moderate degree of similarity NRs and ARs. This suggests that ARs are effectively providing a suitable habitat for fish, like NRs. This could indicate that the ARs are functioning well as a substitute or additional habitat for fish.

Sessile organisms are part of the healthy reef-ecosystem and fish are dependent on their presence. These biota may also serve as a food source for fish, such as ascidians. Alternatively, epifauna living within the biota attached to structures, such as crustaceans, can also provide a food source for fish. The epifauna and flora may also provide shelter for small, cryptic species of fish, that live within them. Sessile organisms are part of the healthy reef-ecosystem and fish are dependent on their presence (Clynick et al. 2007).

Our study on the AR did not find any invasive or alien species. However, ARs could attract, because of the additional habitat availability, non-indigenous species and enhance the presence of alien species over native species (Paxton et al. 2020). The potential risks associated with introducing invasive or alien species to artificial reefs should not be disregarded, and ongoing monitoring and conservation efforts are necessary to ensure the protection of native species and biodiversity.

Regarding the identified fish on ARs and NRs, there are several species that are classified as of Least Concern (LC). On the other hand, two fish species found on NRs, Dentex

dentex and Epinephelus marginatus, are classified as vulnerable (VU) according to IUCN (2022). *Dentex dentex* is vulnerable to overexploitation due to its large size, longevity, and slow growth, while *Epinephelus marginatus* is a long-lived, slow-growing fish with a complex life cycle, making it vulnerable to overfishing. This underscores the need to protect NRs as essential habitats for the conservation of vulnerable species.

5.4. Limitations of this work

It is important to note that the findings of this study have limitations that should be considered. The study focused on two shipwrecks and two ARs, which may not be representative of all artificial and natural reef habitats. It should be noted that environmental conditions, such as water flow and light, were not controlled for in this study. This could potentially confound the results and limit the generalizability of the findings. Additionally, the potential influence of the divers on the environment and organisms of the artificial and natural reefs was not assessed in this study. The limited taxonomic resolution with several unidentified groups of the study may have also limited the ability to draw conclusions about specific species or groups of organisms and their response to different reefs and their orientations. Finally, the study focused on two-dimensional surfaces of the reefs and did not investigate the three-dimensional structure or the interactions between different parts of the reef, which could further limit the ability to fully understand the drivers of differences in cover and diversity between NRs and ARs. Limited resources in terms of funds and time prevent a deeper investigation of the topic at the moment of thesis production and are reserved for further research.

Another potential restriction of this study is the limited taxonomic resolution used in identifying the biota present on the reefs. The study may have overlooked species-level differences in the biota between ARs and NRs because of limited image resolution, limited sample-size and non-laboratory identification.

Marine conservation is a critical component of sustainable development. Despite this, debate about marine conservation is relatively limited, particularly compared to terrestrial conservation (Hillebrand et al. 2020). One reason for this is that the marine environment is difficult and expensive to study, due to factors such as depth and pressure. Additionally, many marine ecosystems are located in remote or hard-to-reach areas, which can make it challenging for researchers to access and study them. Another factor contributing to the limited research on marine conservation is a lack of funding and resources (Katsanevakis et al. 2015). Despite the protection of marine areas being included in the Sustainable Development Goals (SDG 14), research on whether wrecks can mimic natural reefs is also limited (THE 17 GOALS | Sustainable Development n.d.). One possible reason for this may be that marine conservation research may tend to prioritize the study of natural reefs. This could result in a lack of funding and resources being directed towards researching the potential of wrecks to support marine biodiversity. Additionally, wrecks may be more susceptible to damage from human activities, such as diving, than NRs, which can limit their potential as a conservation tool.

Furthermore, the present thesis did not compare wrecks as ARs with other forms of ARs.

5.5. Conclusion

We concluded that the studied ARs and NRs in Ċirkewwa Marine Park shared similar taxonomical compositions, possibly due to similar environmental conditions. The present study indicated that NRs hosted higher diversity than ARs, likely due to higher habitat heterogeneity. Although there was no significant difference in overall sessile biota cover between the two reef types, vertical plots on ARs showed higher cover than horizontal plots on ARs, while horizontal plots on NRs showed higher cover than vertical plots. The reasons for these differences were not fully understood and require further research. The study found that fish richness was similar on both NRs and ARs, suggesting that ARs could provide a suitable habitat for fish. However, the vulnerability status of two fish species on NRs highlighted the importance of conservation of NRs.

In conclusion, the importance of the present research resides in creating a baseline evaluation of potential of wrecks mimicking reefs for conservation of marine sessile biota. The findings of the study indicate that wrecks as ARs in the Ċirkewwa Marine Park provide additional habitat for sessile biota and fish and are therefore meaningful to take them in consideration in conservation planning and management. These findings need, however, to be supported by more research.

We conclude that the wrecks in the Ċirkewwa Marine Park can serve as artificial reefs, to support the marine environment. There are several factors that influence the effectiveness of a AR in mimicking NR, such as proximity to the NR and structures, but overall an additional area for organisms to colonize and seek shelter increases the available area for biota and hence reduces the pressure on an ecosystem by providing additional habitat. As we did not find significant differences in the cover of AR and NR, we can conclude that organisms will benefit through additional settlement possibilities. Even if the diversity was higher on NRs, the placement, use and conservation of wrecks as artificial reefs for marine restoration can contribute to the overall health of the ecosystem. Additionally, present study found that the communities on ARs and NRs share some degree of similarity, which can promote the existence of already present species.

The wrecks situated around Malta offer a significant contribution to the economy by attracting tourists and divers, while also providing non-material services of recreation and health to scuba divers through their surrounding marine environment. By emphasizing the beauty and significance of these ecosystems, diving can help boost awareness and encourage efforts towards their conservation. However, It is important to exercise caution with tourism and diving activities and take monitoring and reinforcement actions, such as limiting the amount of divers. A non-regulation could lead to significant pressure on the marine ecosystem. Additionally, scuttling new wrecks for conservation purposes must be done carefully and with awareness to prevent any damage to the environment. Sunken vessels may pose a risk to marine environment through pollution. The Mediterranean Sea, for instance, has 4% of the world's sunken wrecks (i.e. thousands of vessels, aircrafts and other commercial and military devices), which date back to Second World War and still promote uncontrolled leakage of toxic material and organic/inorganic pollutants (Medeiros et al. 2022). Nevertheless, the presence of shipwrecks needs to be evaluated in the immediate surrounding. Due to the presence, for instance trawling activities are naturally restricted. Further, intentionally scuttled shipwrecks, such as both studies wrecks in present study, were scuttled for diving purposes and hence should have been fully cleaned from oil before scuttling. To ensure proper disposal, the vessel that is to be sunk must first be emptied of any oil and debris (Devault et al. 2016).

In fact, the establishment of the Ċirkewwa Marine Park was a crucial step towards the protection of the marine environment in Ċirkewwa (Ċirkewwa Marine Park Management Plan 2022). Moreover, the wrecks can be utilized as educational resources to educate visitors on marine conservation, and promote sustainable tourism practices in the area. However, achieving the potential of the wrecks for conservation purposes would require a joint effort between various stakeholders, including local communities, government agencies, and conservation organizations. Bringing all actors on the same page could be challenging, given that divers may resist regulations that influence their activities, while fishermen may object to restrictions on their fishing actions. Nonetheless, finding common ground and developing effective conservation strategies that cater to the needs of all stakeholders is vital for ensuring the long-term sustainability of the wrecks and their associated ecosystems. By adopting a coordinated approach, the wrecks located around Malta can play an important role in promoting marine conservation and sustainable tourism in the region.

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