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ASSESSMENT OF THE ECOLOGICAL STATUS OF THE BENTHIC MACROFAUNA COMMUNITIES OF THE ALENTEJO COAST

Relatório de Dissertação de investigação do Mestrado em Engenharia Biológica e Química

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Abbreviations and symbols

AMBI	AZTI's Marine Biotic Index
BI	Biotic index
COI	Cytochrome c oxidase I
DNA	Deoxyribonucleic acid
EG	Ecological group
g	Gram
IPMA	Instituto Português do Mar e da Atmosfera
M-AMBI	Multivariate AZTI's Marine Biotic Index
ml	milliliter
PCR	Polymerase Chain Reaction
rDNA	Ribosomal deoxyribonucleic acid
µm	micrometer
12S	12S ribosomal Deoxyribonucleic Acid
16S	16S ribosomal Deoxyribonucleic Acid
ºC	Celsius degrees
TOM	Total Organic Mater

Resumo

Este trabalho incide sobre a avaliação do estado ecológico das comunidades de macrofauna bentónica devido ao seu papel fundamental nos sistemas aquáticos, equilíbrio ecológico, cadeias tróficas entre outros aspetos. Em 2019, zona costeira entre Sesimbra e Odeceixe, foram realizadas 156 colheitas de sedimento em 79 estações, entre 21 a 184 metros de profundidade. As amostras de sedimento foram peneiradas utilizando uma malha de 500 µm e os organismos macrobentócicos foram separados e identificados morfologicamente e, quando necessário, através da taxonomia integrativa. Foi também feita uma caracterização da granulométrica e do teor de matéria orgânica do sedimento de cada estação. Após a identificação dos espécimes, com base na granulometria, teor de matéria orgânica e entre parâmetros físicos, procedeu-se à categorização do estado ecológico de cada estação juntamente com a utilização de sistemas de informação geográfica. Para tal feito foi então executada análise com base em todos os resultados, uma avaliação do estado ecológico destas comunidades, usando índices bióticos e tendo em conta o impacto da pesca com ganchorra como outras atividades antropogénicas e também os locais geográficos das amostragens. Verificou-se que nos locais onde se encontra uma maior atividade humana, como locais de pesca e portos marinhos, acabam por possuir um pior estado ecológico devido ao stresse causado pelas atividades humanas, levando a um leve estado de perturbação. como foi o caso de uma parte das estações pertencentes aos setores MP2 e MP5. Apesar da diversidade de espécies encontradas entre as estações analisadas, algumas especies ocorreram com alguma frequência nomeadamente a *Aponuphis brementi* (Fauvel, 1916) e *Ampelisca brevicornis* (Costa, 1853) que ocorreram em mais de 30% das estações analisadas. No fim verificou-se que 1 dos 4 setores tem uma maior variação dos resultados que poderá se dever ao facto de ter sido o setor com menos amostras analisadas e ter grandes diferenças entre os diferentes pontos amostrados. Em geral a região analisada encontra-se com uma perturbação muito subtil e face aos estudos e as conclusões obtidas durante este trabalho é fundamental fazerem mais estudos e fazer uma monitorização da are de forma a verificar como evolui o estado ecológico ao longo dos anos.

Palavras-chave

Organismos Bentónicos, Diversidade, Costa Alentejana, Invertebrados marinhos, Índice biótico

Abstract

This work focuses on the assessment of the ecological status of benthic macrofauna communities due to their fundamental role in aquatic, ecological systems, trophic chains, among other aspects. In 2019, in the coastal zone between Sesimbra and Odeceixe, 156 sediment collections were carried out in 79 stations, between 21 and 184 meters deep. The sediment samples were sieved using a 500 µm mesh and the macrobenthic organisms were separated and identified morphologically and, when necessary, through integrative taxonomy. A characterization of the granulometric and organic matter content of the sediment at each station was also carried out. After identifying the specimens, based on granulometry, organic matter content and between physical parameters, proceed to the categorization of the ecological status of each station together with the use of geographic information systems. For this purpose, an analysis was then carried out based on all the results, an assessment of the ecological status of these communities, using biotic indices and taking into account the impact of hookfish fishing as well as other anthropogenic activities and also the geographic locations of the samplings. It was found that places where there is greater human activity, such as fishing grounds and marine ports, end up having a worse ecological status due to the stress caused by human activities, leading to a mild state of disturbance. as was the case with some of the stations belonging to the MP2 and MP5 sectors. Despite the diversity of species found between the permanent stations, some species occurred with some frequency, namely Aponuphis brementi (Fauvel, 1916) and Ampelisca brevicornis (Costa, 1853) which occurred in more than 30% of the transient stations. In the end, it was tolerated that 1 of the 4 sectors has a greater variation in the results, which could be due to the fact that it was the sector with the fewest presences present and that it had large differences between the different points sampled. In general, the follower region is experiencing a very subtle disturbance and, given the studies and how it has progressed during this work, it is essential to carry out more studies and monitor the area in order to verify how the ecological state has evolved over the years.

Keywords

Benthic organisms, Diversity, Alentejo oceanic coast, marine invertebrates, Biotic index.

1. Introduction

The present work was carried out in the Laboratory of Benthic Studies, belonging to the Division of Oceanography and Marine Environment (DivOA) of the Department of the Sea and Marine Resources (DMRM) and within the curricular unit of master's thesis of the master's degree in Biological and Chemical Engineering at the Barreiro School of Technology (EST-B) at the Polytechnic Institute of Setubal (IPS). The master's thesis was carried out in the scope of the progamme MarBIS – Marine Biodiversity Information System, funded by the Portuguese Government. The samples used in this work were collected during the oceanographic campaign of the project MINEPLAT - Determination of the potential in mineral resources on the Alentejo continental shelf and the natural constraints imposed by the uplift of the continental margin in the Plio-Quaternary, funded by European Union and national / regional public.

During the research time, four sectors, along the Portuguese coast from Sesimbra to Odeceixe, were analyzed with the main objective to characterize the benthic macroinvertebrate communities and the ecological status from the coast of Sesimbra to Odeceixe that belongs at the region to infralittoral and circalittoral zones. These areas have presented a high biodiversity of benthic macrofauna, fundamental for the trophic structure of marine ecosystems, ecological balance, and biological interactions (Henriques *et al.* 2015). All samples collected were from the infralittoral and circalittoral zone. These organisms are a key basis for all marine ecosystems (Diaz & Rosenberg 2008).

1.1. Characterization of the Host Institution

The Portuguese Institute for Sea and Atmosphere, I. P. (IPMA, IP) is a public institution, part of the indirect administration of State, endowed with administrative and financial autonomy and its own assets.

IPMA I. P. follows the responsibility of the Ministry of Economy and Sea and Ministry of Agriculture and Food, under supervision and guardianship of the respective ministers.

The strategic guidelines and goals for IPMA, I. P., as well as the monitoring of their implementation, are articulated between members of the Government responsible for the sea and science.

IPMA has responsibilities at national territory level in the areas of the sea and atmosphere. Focusing its research efforts on projects that accrue to direct applications for use in operating activities, pursuing continuous improvement in the information made available to its users whether for commercial use or public service and directed to the safeguard of people and property (ipma.pt/en/oipma/).IPMA I. P. is the state laboratory whose mission is to promote and coordinate scientific research, technological development, innovation and service on the seas and atmosphere, ensuring the implementation of national strategies and policies in their areas of expertise, thus contributing to the economic and social development. IPMA, IP, is the national authority in the fields of meteorology, aeronautical meteorology, climate, seismology, and geomagnetism.

IPMA, I.P. is an institution of reference at international level, for the accuracy and effectiveness with which it performs its technical and scientific duties of support for state action, as well as the relevance and impact of R & D that develops (www.ipma.pt).
IPMA has five departments with several divisions between them. The departments are as follows.

- Department of the Sea and Marine Resources (DMRM)
- Department of Weather Forecast and Geography (DMG)
- Department of Operations, Infrastructure and Technological Development (DOIDT)
- Regional delegation of Madeira (DRM)
- Regional Delegation of the Azores (DRA)

The organization chart is shown in figure 1.

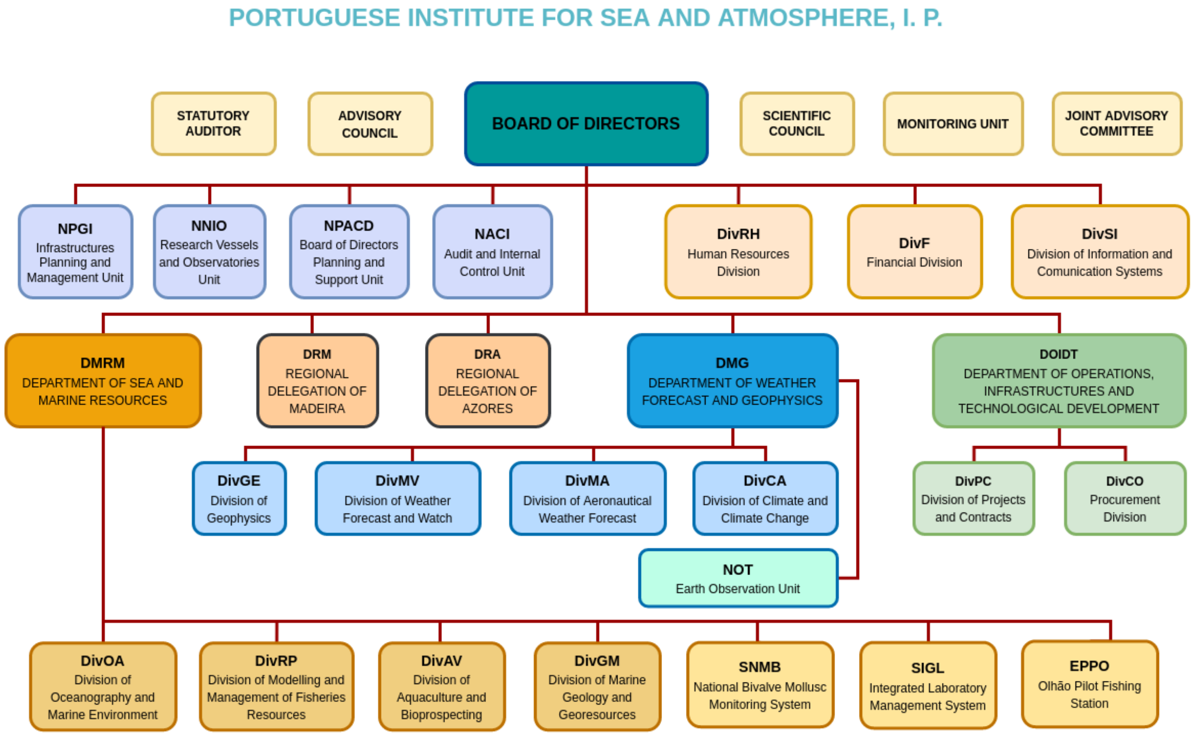

Figure 1 – IPMA organization chart.

The present work was carried out in the Laboratory of Benthic Studies, belonging to the Division of Oceanography and Marine Environment (DivOA) of the DMRM.

1.2. State of the art

Analyses related to macrofauna and benthic from the coastal ecosystems are important because of the vital ecosystem functions and services they provide (Gammal et al. 2017). Coastal environments are often heterogeneous, with a complex mosaic of different habitats and conditions that sustain high biodiversity. They are critical areas because of the vital ecosystem functions (primary production and nutrient cycling) and services (food provision and recreational opportunities) they provide (Levin et al. 2001; Barbier et al. 2011; Snelgrove et al. 2014). The integrity of coastal ecosystems is however under threat, from anthropogenic activities such as dredging, fishing and construction, eutrophication, and oxygen deficiency, among others that can impair biotic communities and alter ecosystem functions (Levin et al. 2001; Conley et al. 2011). Components such as abundance, biomass, and species richness of macrofauna communities can directly show us the ecological status of the habitat under study, together with the analysis of sediments we have great certainty of the status of the habitat (Gammal et al. 2017). The existence of sensitive or resistant organisms also shows us the disturbance status of the site, which is an index of the ecological status of the habitat (Masese et al. 2009).

The benthic organisms inhabit the bottom substrates of the waters (estuaries, rivers, seas, and oceans) and play a vital role in the maintenance of sediment and water quality (McLusky & Elliott 2004). Marine and estuarine benthic communities of macroinvertebrates are mostly composed of annelids, mollusks, and crustaceans, which live in the benthic zone/seafloor or that depend on it to live and are important constituents of ecosystems aquatic organisms, distributed mainly at the base of food webs, in close connection with pelagic food chains (Pearson & Rosenberg 1978; Dauer 1993).

The composition of these communities is an important indicator (bio-indicator) of environmental stress because they are particularly sensitive to pollutant exposure (Borg et al. 1998; Holland et al. 1987). This sensitiveness arises from the close relationship between benthic organisms and sediments which can accumulate environmental contaminants over time and the fact that these organisms are sessile or have reduced mobility, which means they receive prolonged exposure to contaminants in their immediate habitat (Sanders et al. 1980; Nixon et al. 1986).

The distribution of benthic macrofauna shows a clear relationship with the type of sediment, depth, amount of organic matter and distance from the coast and the temperate water habitats show a high diversity of organisms (Thorson 1966). The characterization of benthic communities and habitats, which includes the physic-chemical features of the substrate and the taxonomic identification of organisms, is important to understand marine ecosystems and their ecological status (Thorson 1966; Sanders et al. 1980; Nixon et al. 1986; Sánchez-Moyano et al. 2006). However, so far, routine assessments of macrobenthic invertebrates have been carried out using almost exclusively morphology-based approaches for species identifications. This is a time-consuming and skill-dependent approach, which has resulted in low-throughput in processing biomonitoring samples. Very often the specimens cannot be accurately assigned to species, either because morphology-based identifications are inherently difficult, or because of organisms' bodies that are damaged and missing diagnostic parts. In the case of immature stages, the species level identifications are extremely difficult or nearly impossible. Published data on macrobenthic surveys frequently report specimens assigned only to family or genus level (Lobo et al. 2017). Benthic communities are constantly changing due to changes in the ecosystem (type of sediment, organic matter, presence of contaminants) given the fact that they are very sensitive (Thorson 1966; Sanders et al. 1980; Nixon et al. 1986; Sánchez-Moyano et al. 2006). Based on the existing organisms in these communities it is possible to understand the ecological status of the habitats, using species as bioindicators, through their sensitiveness species or tolerance to changes and species that resist change. (Thorson 1966; Sanders et al. 1980; Nixon et al. 1986; Sánchez-Moyano et al. 2006; Leonardsson et al. 2009; Rosenberg et al. 2004; Dauvin et al. 2010).

The choice of benthic macroinvertebrates as study objects of environmental assessment is not only due to the significant role they play in marine systems, but also due to a variety of characteristics that make them susceptible to use in this type of studies (Pearson & Rosenberg 1978; Dauer 1993). The limited mobility of most of these organisms leads them to be more sensitive to local disturbances (Pearson & Rosenberg 1978; Dauer 1993). In addition, as they have life cycles relatively short and, are directly related to the sediment that can accumulate contaminants for long-term periods, they become an essential component in the study of changes in marine ecosystems (Pearson & Rosenberg 1978; Dauer 1993). The combination of these characteristics leds to the development of several indices that support the predictability of the organisms' responses to environmental alterations, and which are largely based on the paradigm of Pearson and Rosenberg (1978). This paradigm is based on the assumption that benthic communities respond to improvements in the quality of the environment by progressively following three steps: abundance increases, specific diversity increases, and dominant species change, from disturbance tolerant to disturbance sensitive. This pattern is very important when monitoring anthropogenic impacts because, as reported by Heip et al. (1992), human activity not only leads to changes in species associations, but as in other community attributes, such as density, biomass, and diversity, which can be used to assess their evolution.

Based on these general principles, several indices to study biological parameters were developed. For instance, AZTI's Marine Biotic Index (AMBI) is a biotic index capable of analyzing the ecological status of the site which ecological quality assessment allow classifying the sites based on the sensitivity/tolerance of the species to the disturbance (Borja et al. 2000). Other indices more complex, such as multivariate AMBI (M-AMBI) (Borja et al. 2004; Moxika et al. 2007) integrates functional and structural aspects of the benthos and is considered very promising as a measure of the quality of benthic habitats, due to the ability to integrate the physical structure of benthic community habitats (Borja et al. 2007). AMBI Calculates the Biotic Index by distributing the different taxa to different ecological groups. EGI is the most sensitive and EGV is the least sensitive. M-AMBI Use the AMBI result among other variables such as diversity and specific richness to give your final result. (Borja et al. 2007).

For a more accurate analysis other variables have to be taken into consideration as is the case of anthropogenic activities. Many types of fishing activities are extremely invasive impairing in the even the benthic organism’s incident or close to the fishing grounds. Anthropogenic activities such as intensive fishing non-selective end up negatively impacting ecosystems sea because they remove organisms from the sea that are not their target (Baptista 2012). An example is the use of traditional fishing bivalve dredge “ganchorra” to harvest bivalves with commercial interest that negatively influences benthic organisms given their high level of invasiveness (Duplisea et al. 2002). Knowledge of these impacts is mainly based on empirical data that describe biological differences between communities affected by distinct levels of drag disturbance (Thrush et al. 1998). Trawling and, in particular, traditional dredge fisheries lead to the death of benthic invertebrates because they are crushed directly by the net, because they expose the animals to predators or because they are caught in the nets (Duplisea et al. 2002; Thrush et al. 1998). The largest individuals suffer highest mortality when captured in the nets because they are not able to escape, whereas the smallest have a lower mortality rate. Although the size of the specimens has some bearing on the mortality rate, the main reason is their distribution in the sediment in depth. However, given that the most recurrent technique of bivalve fishery in the collection sites for this project is traditional fishing bivalve dredge “ganchorra” even animals hidden in the area are eventually caught as this technique is specific to the capture bivalves, which have negative effects on benthic communities (Gaspar et al. 2014). This is a type of fishing that captures the organisms that inhabit the most superficial areas of the sediment, namely bivalves. The minimum mesh is 30 mm, a maximum width of the "mouth" 1.5 m in the North West zone and 1 m in the other zones, the maximum length of the comb teeth is 55 mm for switchblade and razor, 15 cm. The minimum interval between teeth is 15 mm. During the use of this equipment, it is natural that there is a large amount of organisms of no commercial interest to be captured given the small mesh.. (dgrm.mm.gov.pt/ganchorra consulted a 21 of April 2022).

The identification of organisms can be done through morphological taxonomy, using diagnostic characters. However, many times errors in identification can be managed, or there cryptic species, or as the loss of diagnostic characters or the organisms are damaged, there are still situations of phenotypic variability and sexual dimorphism and in addition, some organisms have life cycles with distinct morphological phases, which makes it very difficult to identify. All these mishaps in this science led to the existence of many doubts in some species, family, genus and even in some phylum (Gewin 2002; Godfray et al. 2002; Mallett et al. 2003; Wheeler et al. 2004). Nowadays, morphological taxonomy is widely used to identify most organisms. However, whenever there are doubts or for some reason it was not possible to arrive at the species level, molecular biology should be used as an aid to the identification, combining genetic and morphological information (Dayrat 2005). The DNA Barcode is a unique nucleotide sequence for each individual with a universal reading code (Helbert et al., 2003). Each species has its DNA barcode that, although there are differences between organisms in the same species, there are places in the DNA that are highly conserved and that very rarely have significant alterations, as is the case of cytochrome c oxidase I (COI) widely used as a genetic marker for the identification of organisms. (Hebert et al., 2003; Dawnay et al., 2007). Animal species can be identified using a fragment of the mitochondrial cytochrome *c* oxidase subunit I (COI) gene, because this gene has a greater range of phylogenetic signal than any other mitochondrial gene and it is an extremely conserved gene with only a few alterations from organism to organism, making it more easier the identification (Hebert et al., 2003). In common with other protein coding genes, its third-position nucleotides show a high incidence of base substitutions, leading to a rate of molecular evolution that is about three times greater than that of 12S or 16S rDNA (Knowlton & Weigt 1998).

These two approaches of taxonomic identification should not be seen as exclusive, but rather as approaches to the same goal which simply differ in the characters, that are considered complementary. One system or the other may be used for particular cases, depending on the results it provides, but it can also be used simultaneously in a complementary way. Moreover, a DNA-based system of identification can only work if all the species have their ‘diagnostic’ sequences present in the database. An incomplete database will simply allow users to determine whether a given sequence is different from the others already stored. In other words, the species identification using DNA barcodes is dependent on morphological taxonomy. Also, in some cases, using both combined approaches (morphological and genetic) become the ideal recipe for species identification. The use of integrative taxonomy has also become an asset for the identification of new species, confirming the genetic and morphological differences (Lobo & Tuaty-Guerra 2017).

It's important that the collection of organisms is carried out with an appropriate methodology, in order to guarantee a good sampling for the type of analysis intended, promoting the best possible results. The study area of the present work is located from Sesimbra to Odeceixe, which includes a significant area where the traditional fishing bivalve dredge “ganchorra” is used to collect bivalves. It is extremely important to know the benthic communities and their status because the benthic communities are at the basis of ocean life, have a high biodiversity, being fundamental for the trophic structure of aquatic systems, ecological balance and biological interactions and changes in this community will potentiate changes in the entire ecosystem and many organisms have life cycles, identifying benthic organisms and communities also gives an indication of the status of plagic organisms and communities.. As mentioned before, benthic organisms are widely used for monitoring aquatic habitats (Pinto 2009; Reñones et al., 1997).

Coasts and estuaries are ecosystems of high environmental variability, biogeochemical transformations, and biological interactions, where dynamic exchanges of energy, mass and nutrients occur between benthic and pelagic habitats through different pathways, making them one of the richest ecosystems in the world (Pinto 2009). The implementation of effective management strategies that mitigate human-caused changes in these ecosystems requires a better understanding of how human pressures can cause changes in ecosystem structure and function (Pinto 2009). The traditional view of benthic-pelagic coupling has focused on the deposition of non-living organic material in benthic habitats and the release of inorganic nutrients from sediments. Anthropogenic pressures regulate benthic-pelagic coupling directly and indirectly through their effects on the physical (e.g., salinity, oxygen, temperature) and biological (e.g., species, communities, functional characteristics) components of ecosystems. In addition to anthropogenic activities with direct effects on communities, such as fishing, there are also other activities that end up indirectly affecting communities, such as the introduction of non-indigenous species that are often transported by ballast water (Pinto 2009).

1.3. Objectives

The main objective of this work was to assess the ecological status of the sea floor from the west coast of mainland Portugal, between the Professor Luiz Saldanha Marine Park to Odeceixe, through the analysis of the community's composition of the macrobenthic fauna and the relationship with environmental data and anthropogenic activities.

The specific objectives are as follows:

i. Identification of organisms collected in the scope of an oceanographic campaign, to the lowest possible taxonomic level, i.e., species level, using integrative taxonomy, when necessary.

ii. Analysis of the ecological status of benthic communities through ecological indices, granulometry and analysis of organic material

2. Material and methods

2.1. Sampling sites and sample collection

The biosedimentary material was collected from Sesimbra to Odeceixe, in Atlantic coast, during the oceanographic campaign MINEPLAT, on board the IPMA's research vessel. Noruega, for 15 days, between March and April of 2019. The samples were supposed to collect from a total of 79 sites distributed in four sectors: MP1, MP2, MP3 and MP5. The Sector MP1 was located near the traditional fishing bivalve dredge "ganchorra" area in between 38,07533 lat to 38,27123 lat; MP2 on or near to the area where traditional fishing bivalve dredge "ganchorra" is allowed area in between 38,38415 lat to 38,43219 lat; MP3 on or near to the southwest Alentejo and Costa Vicentina Natural Park between 37,47221 lat to 37,75616 lat; and MP5 around the harbor and city of Sines between 38,04011 lat to 38,78979 lat (Figure 2). Two replicates were taken in each location using a Smith Macintyre grab with a 0.05 m^2 sampling area. Sampling had the main objective of studying the ecological status of the stations and sectors sampled. Sampling stations are in the infralittoral and circalitoral area (from 21 to 184 meters).

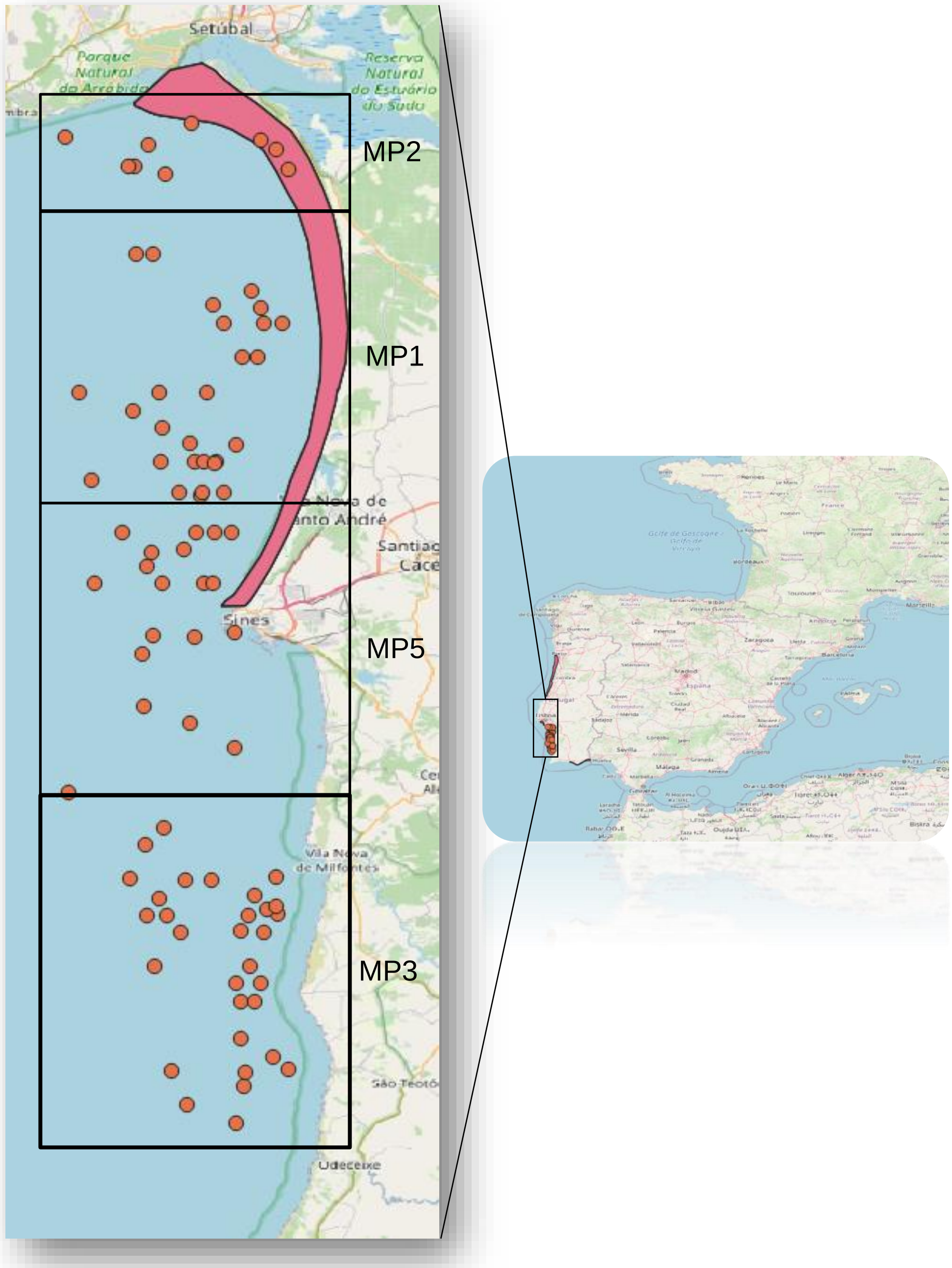

Figure 2 - Map with sectors and stations. The rectangular areas represent the sectors, the red bands represent the areas where traditional fishing bivalve dredge "ganchorra" takes place and the areas with a light green around them are protected areas.

In the first replicate, and after receiving the grab on the deck, a small sample of sediment (150 – 200 g.) was collected into a small round plastic box from the upper side before the jaws were opened, identified with the code of the sampling station and the date of collection. This sediment sample was used for determination of grain size and organic matter analysis. The remaining sediment of the grab was sieved through a 500 µm mesh to collect macrofauna. The soft-bodied organisms were fixed in 4% formaldehyde, and the remaining specimens in

70% ethanol. The second replicate was entirely sieved for macrofauna collection, and all organisms were fixed and conserved in 70% ethanol.

Due to lack of manpower on board, the sediment samples of sectors MP3 and MP5 could not be sieved on board, and they were stored in plastic bags for further analysis on land. The first replicate was stored with 4% formaldehyde, and the second frozen at -20ºC.

2.2. Sediment characterization

All sediment samples, with about 100 g each, were dried and then processed according to Gaudêncio et al. (1991): (i) sediment was washed through a 38 μm square mesh sieve to remove the fine fraction; (ii) the remaining sediment was dried in an oven at 100°C; (iii) using a RETSCH mechanical agitator, the sediment was sifted through a column of 17 square mesh sieves of the ASTM series (38 μm - 16000 μm) for sorting; and (iv) sedimentary fraction retained in each sieve was weighed. The grain size was classified according to the geometric scale of classification of Udden/Wentworth (Wentworth, 1922 in Kenny & Sotheran, 2013): gravels, composed of pebbles (4000 μm and 8000 μm) and fine gravel (2000 μm and 4000 μm), very coarse sand (1000 μm and 2000 μm), coarse sand (500 μm and 1000 μm), medium sand (250 μm and 500 μm), fine sand (125 μm and 250 μm), very fine sand (63 μm and 125 μm) and fine particles, composed of silt (38 μm and 63 μm) and clay (<38 μm).. The component fractions, medians, screening coefficients and classification of sedimentary types of surface sediment samples collected off the coast of Sesimbra to Odeceixe are presented in the annex 1.

For the calculation of the Total Organic Matter (TOM) of each site, approximately 100 g of sediment was placed in a crucible, dried in an oven at 100ºC, weighed and then grinded. Subsequently it was taken to muffle furnace (Nabertherm – 30 up to 3000ºC) at 450ºC during 2 hours for the incineration of organic matter. Then, each crucible was weighted, and the TOM was obtained calculating the difference between dry and incinerated sediment. This procedure was performed in duplicate for each sample and then the data obtained from the replicates were averaged in order to have a representative sample of the sites.

2.3. Collection of marine macrobenthic invertebrates

A total of 156 sediment samples, belonging to 79 stations, were sieved to separate the macrobenthic invertebrates (some sites do not have replicates). All samples were washed on a sieving table through a 0.5 mm screen in order to retain only the macrofaunal organisms. The organisms of each first replicate were fixed in 70% ethanol, with the exception of polychaetas and other soft-bodied organisms, that were fixed in 4% formaldehyde, to best preserve diagnosing characters) All specimens of each second replicate were fixed in 70% ethanol, The organisms were separated in large taxonomic groups, and placed in plastic recipients.

2.4. Taxonomic identification of macrobenthic communities

Macroinvertebrate specimens were identified to the lowest possible taxonomic level. For the identification of the organisms, the digital microscope Leica DMS 300, the stereo microscope Leica MZ12, and the light optical microscope Leica DM2000 LED were used to visualize morphological characters of the organisms. Tweezers, dissecting needles, and scalpels were used to manage the organisms and dissect when necessary. During the handling, organisms were always kept submerged in ethanol to avoid drying out, and after identification, they were properly placed in appropriate tubes or containers with ethanol 70% to preserve the samples. For morphological identification, taxonomic identification keys were used these being Identification key marine species, Hayward & Ryland (2017), Lincoln. (1979) and Gil. (2011) were used. The World Register of Marine Species (WoRMS) (https://www.marinespecies.org/) was used to verify the updated species names. Some problems occurred at this stage, many organisms were not identified until a lower taxonomic level because samples from the MP3 and MP5 sectors were not well conserved and some organisms became degraded.

After morphological identification, specimens with uncertain identification, damaged specimens with lack of diagnosis characters, non-indigenous species, and rare species were selected for genetic analysis. A portion of tissue of each specimen was taken and the total DNA was extracted using the E.Z.N.A.® Tissue DNA Kit, following the manufacturer's instructions. The equipment used were: finnpipette™ Thermo Scientific™ F2 micropipettes of 1000 µl, 100 µl, 10 µl and 5 µl and their respective tips, incubator with shaking ThermoMixer C (Eppendorf), Vortex Genie 2 (Scientific Industries SI), and a 5415 D centrifuge (Eppendorf) and microtubes. The genetic marker cytochrome *c* oxidase subunit I (COI-5P) was amplified, using the primer pair LoboF1 and LoboR1, following standard DNA barcoding protocols (Lobo et al. 2013). The equipment used for the amplification was the same as the previous ones, plus the thermocycler (Eppendorf). Positive and negative controls were also used to verify that the PCR was running well. Electrophoresis was performed with the positive and negative controls and with the marker (NZYTech), the Bio-Rad brand electrophoresis system, model PowerPac™ Basic. Finally, the gel was observed on the computer, using the BIO-RAD brand Doc™ XR+ Gel.

The amplified PCR products were bidirectionally sequenced (forward and reverse) by Eurofins Genomics (Ebersberg, Germany) using the same primers as for amplification, using the BigDye Terminator 3 kit on an ABI 3730XL DNA analyzer (Applied Biosystems™, Foster City, CA, USA). For each specimen, the chromatograms obtained were analyzed and both sequences aligned to create a consensus. Then the primers were removed from the sequence. All these bioinformatics steps are done on MEGA-X software (Kumar et al., 2018).

The obtained sequences were then placed in GenBank, and a blast was made to compare the DNA sequence with the public sequences, verifying the morphological identification.

2.5. Ecological status analysis – benthic indices

The taxonomic identifications obtained per each macrobenthic community were analyzed through the AMBI. AMBI is a software developed for assessing the quality of soft-bottom benthic macroinvertebrate communities. It includes a AMBI species list, with 10,638 taxa, covering the whole ocean. Using the same AMBI software, the two indices AMBI (in order to acquire the ecological level based on the sensitivity level of the species) and multivariate-AMBI (M-AMBI) (in order to verify the ecological status and sensitivity of the site based on species sensitivity, specific diversity, and specific richness) can be calculated. These indices explore the response of soft-bottom communities to artificially induced changes in water quality, integrating long-term environmental conditions. Currently, AMBI and M-AMBI are used worldwide in multiple applications and officially in many countries (Borja et al., 2020). Based on this software, it was possible to categorize species into 5 different ecological groups (EG), depending on their sensitivity to disturbances in the ecosystem. The ecological groups are:

EGI: Species belongs to this group are very sensitive to organic enrichment and are in abundance at unpolluted conditions.

EGII: These species are present in low densities and are relatively constant in abundance over time, unaffected by pollution.

EGIII: Species of this group are tolerant to high organic matter enrichment and are stimulated by an increase in organic pollution.

EGIV: Species belong to this group are second-order opportunistic species which are favored by unbalanced conditions.

EGV: This group species are first-order opportunistic species which are even more favored by unbalanced conditions.

The index value was calculated using the equation 1 which uses the percentage of organisms per group.

$$AMBI = \frac{[(0\times\%EGI)+(1{,}5\times\%EGII)+(\ 3\times\%EGIII)+(\ 4{,}5\times\%EGIV)+(6\times\%RGV)}{100} \qquad \text{Eq. 1}$$

The distributions of these ecological groups in AMBI were analyzed according to their sensitivity to pollution stress which provides a Biotic Index (BI) with eight levels 0 – 7 levels 0 (0,0 < mean AMBI < 0,2) and 1 (0,2 < mean AMBI < 1,2) represent an undisturbed BI, level 2 (1,2 < mean AMBI < 3,3) represents a slightly disturbed BI, level 3 (3,3 < mean AMBI < 4,3) and 4 (4,3 < mean AMBI < 5,0) a moderately disturbed BI, level 5 (5,0 < mean AMBI < 5,5) and 6 (5,5 < mean AMBI < 6,0) highly disturbed BI and finally level 7 (Azoic) extremely disturbed BI as is possible to see in figure 3 (Borja et al. 2020). The lower the value, the better the ecological status (Sivaraj et al., 2014; Borja et al., 2000). M-AMBI is a combination of Shannon's diversity, richness and AMBI, into a factor analysis multivariate approach which appears to be a suitable method to evaluate the ecological conditions using AMBI software. This method compares the results with reference conditions, to derive an M-AMBI value, which expresses the relationship between observed values and reference condition values. M-AMBI was qualified in five groups. The groups were: High quality, >0.77; Good, 0.53 – 0.77; Moderate, 0.39 – 0.53; Poor, 0.20 – 0.39; and Bad, <0.20 (Sivaraj et al. 2014). To use AMBI correctly it is important to consider the collection location boundaries. The AMBI 4.0 software uses as default the boundaries intercalibrated within the European benthic intercalibration exercise what its used for Northeast Atlantic (Borja et al. 2020). The figure 3 shows the different categories that it is possible to obtain with the AMBI and M-AMBI indices (Sivaraj et al. 2014).

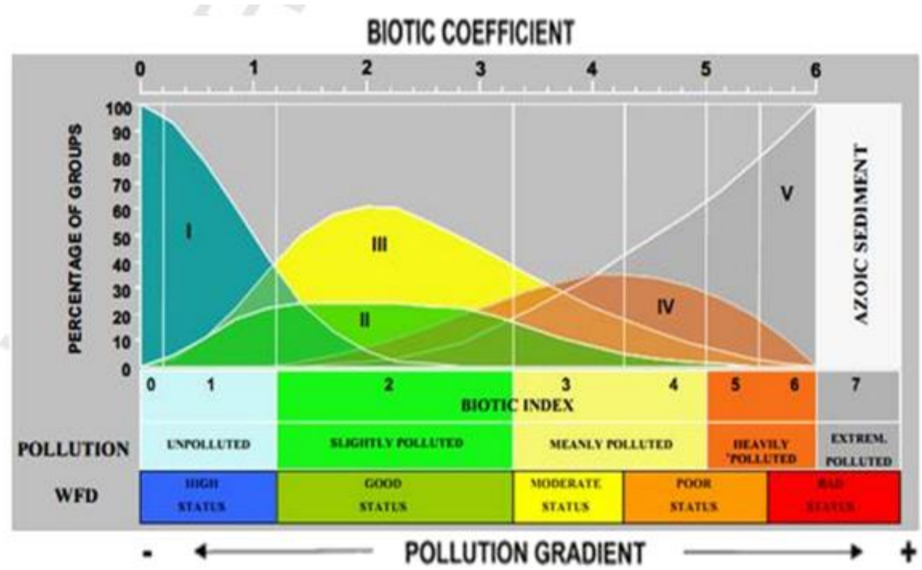

Figure 3 - Theoretical model, modified from Hily (1984), Hily et al. (1986) and Majeed (1987), which provides the ordination of soft-bottom macrofauna species into five ecological groups (EG IeV), according to their sensitivity to an increasing pollution gradient. The relative proportion of abundance of each group in a sample provides a discreet BI with eight levels (0 ± 7) and an equivalent continuous BC (values between 0 and 6).

3. Results

3.1. Sediment characterization

The sediment samples analyzed included coarse elements such as pebbles and biogenic gravel (mainly shell fragments of mollusks), sand and fine particles. The following sediment profile were observed:

1. **Very coarse gravelly sand** in the samples MP3-998 and MP3-999 well calibrated (1.3<So>1.4), with gravel content between 11% and 16 %, a fine particle content (<63 μm) up to 0.6% and with a median between 1087 μm and 1295 μm.
2. **Coarse gravelly sand** in samples MP1-984, MP1-987, MP2-976, MP2-DSS-03 and MP3-37, from well to medium calibrated (1.2<So>1.7), with gravel content of 8%, a fine content up to 0.4% and with a median ranging from 738 μm to 893 μm.
3. **Coarse, slightly gravelly sand** sand in samples MP1-981, MP1-982, MP1-993-3[a], MP3-997, MP5-02 and MP5-03, well calibrated (1.1<So>1.3), with gravel contents up

to 3% and fine elements up to 0.2%, respectively, and with a median ranging from 690 µm to 900 µm.

4. **Coarse muddy sand slightly pebbly** in sample MP1-022, medium calibrated (So=1.8), with gravel content of 1.4%, a fine content of 15% and with a median of 803 µm.
5. **Coarse sand** in samples MP1-992, MP5-06 and MP5-07, well calibrated (1.1<So>1.4), with a median ranging between 712 µm and 859 µm, without gravel content and a fine particle content up to 0.3%.
6. **Medium coarselly sand** in samples MP1-980 and MP3-40, medium calibrated (1.6<So>2.0), with a median ranging between 345 µm and 478 µm, and gravel contents between 7% and 11% and fine contents up to 1.8%, respectively.
7. **Medium sand slightly peeled** in MP1-036, MP1-037, MP1-993, MP3-996, MP5-12 and MP5-995 samples from well to poorly calibrated (1.4<So>2.7), with a median ranging between 285 µm and 374 µm and gravel contents between 0.1% and 1.6% and fine between 0.2% and 9%, respectively.
8. **Medium muddy sand slightly peeled** in samples MP1-026, MP1-991, MP3-23 and MP5-11, with a median oscillating between 258 µm and 363 µm, median to poorly<So>calibrated (2 3), with gravel content between 0.7% and 4.7% and a fine content between 10% and 25%.
9. **Mean sand** in sample MP1-028, without gravel content and a fine particle content of 2.5%, median calibrated (So=1.6) and with a median of 259 µm.
10. **Slightly peeled fine sand** in samples MP1-022A, MP1-978, MP1-979, MP2-975, MP2-977 and MP2-DSS-04, with gravel contents up to 2.3% and fine contents up to 7.3%, well to medium calibrated (1.2<So>1.8) and with a median ranging from 131 µm to 168 µm.
11. **Muddy sand slightly peeled** in samples MP1-006, MP1-021, MP1-034, MP1-983, MP1-989, MP3-18, MP5-13, MP5-20 and MP5-CSS-2, with gravel contents between 0.2% and 4.1%, fine between 11% and 21%, medium to poorly calibrated (1.6<So>2.7) and with a median ranging between 127 µm and 172µm.
12. **Fine sand** in samples MP1-985, MP1-990, MP2-DSS-02, MP3-37A, MP3-46, MP5-55, MP5-60, well to medium calibrated (1.2<So>1.7), with a median ranging from 125 µm to 197 µm, without gravel content and a fine particle content up to 4.3%.
13. **Fine muddy sand** in samples MP1-033, MP1-038 and MP5-14, without gravel content and a fine particle content between 10.6% and 19.7%, from well to medium calibrated (1.3<So>2.1), with a median ranging from 132 µm to 176 µm.

14. **Very thin sand slightly peeled** in samples MP3-39, MP3-49 and MP3-52, with gravel contents between 0.5% and 2.3% and fine between 4.5% and 7.8%, well to medium calibrated (1.2<So>1.5) and with a median of between 118 µm and 122 µm.
15. **Very fine muddy sand slightly peeled** on samples MP1-039, MP1-994, MP2-CSS-4, MP2-CSS-5, MP2-DSS-01, MP3-15, MP3-27, MP3-43, MP3-47, MP3-48, MP3-62, MP5-01, MP5-57 and MP5-CSS-1, with gravel contents up to 1.3%, fine between 10% and 47%, well to medium calibrated (1.4<So>2.0) and with a median ranging from 51 µm to 124 µm.
16. **Very fine muddy sand** in samples MP3-16, MP3-17, MP3-21, MP3-26, MP3-29, MP3-30, MP3-36, MP3-53, MP3-66, MP5 05 and MP5-09, without gravel content and a fine particle content between 10.6% and 30.5%, well the median calibrated (1.2<So>1.9), with a median ranging from 71 µm to 118 µm.
17. **Very fine sand** in sample MP3-28 and MP3-67, without gravel content and a fine content of 3%, well calibrated (So=1.3) and with a median between 115 µm and 116 µm.
18. **Muddy sand** in sample MP5-CSS-3, without gravel content and a fine particle content of 79.9%.

In the annex 2, the percentages of organic matter content (TOM) are presented. Stations showed TOM values between 0.3% and 6%. The sectors had an average of 2,7% of organic material. The MP1 sector had an average of 2,4% with a standard error of 0,17. The MP2 sector had an average of 2,4% with a standard error of 0,47, these values are due to the fact that in this sector there were significantly fewer stations and were significantly further apart, thus leading to an increase in the variation between the stations sampled The MP3 sector obtained a higher average of organic matter, obtaining 3,15% and with a standard error of 0,17. The low variation between the values of the MP3 sector is due to the fact that this sector was the most sampled. Finally, the MP5 sector has an average of 2,9% of organic matter composition present in the sediment, with a standard error of 0,31, which represents a high variation in the percentage of organic matter in the stations of this sector. Separating the stations by groups of 1% organic matter (Table 1)

Table 1 – Organic matter content range

TOM Range	Stations
0% to 1%	MP1-022, MP1-981, MP1-992, MP2-DSS-02, MP2-DSS-03, MP2-DSS-04, MP3-997, MP5-02, MP5-03 and MP5-06.
1,1% to 2%	MP1-38, MP1-982, MP1-984, MP1-987, MP1-989, MP1-990, MP1-993, MP2-976, MP3-37, MP3-40, MP3-46, MP5-07
2,1% to 3%	MP1-06, MP1-21, MP1-26, MP1- 28, MP1-33, MP1-34, MP1-36, MP1-37, MP1-38, MP1-39, MP1-978, MP1-979, MP1-980, MP1-992, MP1-994, MP2-975, MP2-977, MP2-DSS-01, MP3-36, MP3-28, MP3-48, MP3-52, MP3-53, MP3-66, MP3-67, MP3-996, MP5-05, MP5-09, MP5-12, MP5-14, MP5-55, MP5-57, MP5-60, MP5-995, MP5-CSS-01, and MP5-CSS-02.
3,1% to 4%	MP1-983, MP1-985, MP2-CSS-04, MP3-15, MP3-16, MP3-18, MP3-23, MP3-27, MP3-30, MP3-36, MP3-39, MP3-43, MP3-49, MP3-62, MP3-998, MP3-999, MP5-11, MP5-13, and MP5-20.
4,1% to 5%	MP2-CSS-05, MP3-21, and MP3-29
5,1% to 6%	MP5-CSS-03

3.2. Taxonomic identification of macrobenthic communities

A total of 3684 organisms were identified belonging to 324 different taxa, from those 1504 (41%) specimens were identified to the species or genus level. Several species of different phyla were found are very common in all sectors appearing at various sampling stations (table 2) and the main phyla relative to the number of specimens were Annelida, Arthropoda, Echinodermata, and Mollusca however, other phyla emerged in smaller amounts (Table 1).

Table 2 – Total number of organisms per phylum collected in the southwestern Atlantic coast.

Phylum	**nº of specimens**
Annelida	2455
Arthropoda	432
Bryozoa	2
Chordata	1
Cnidaria	64
Echinodermata	292
Mollusca	204
Nematoda	145
Nemertea	40
Platyhelminthes	32
Priapulida	1
Porifera	14

The complete list of bodies identified in the sectors can be found in annexes 3, 4, 5 and 6 respectively. The species with broader range of distribution, occurring from 13% to 53% of the sites, are shown in Table 3. the remaining species appeared with a lower occurrence (<=8%) between sampling stations

Table 3 – Percentage of occurrence, number of organisms and AMBI species group for species with broader distribution in samples collected in the southwestern Atlantic coast.

Phylum	Species	% of occurrence	nº of organisms	AMBI specie group
Annelida	*Aponuphis brementi* (Fauvel, 1916)	53	212	II
Arthropoda	*Ampelisca brevicornis* (Costa, 1853)	34	67	I
Echinodermata	*Ophiocten affinis* (Lütken, 1858)	33	114	II
Annelida	*Eclysippe vanelli* (Fauvel, 1936)	23	47	I
Annelida	*Nephtys hombergii* Savigny in Lamarck, 1818	20	22	II
Echinodermata	*Echinocardium cordatum* (Pennant, 1777)	19	41	I
Annelida	*Notomastus latericeus* Sars, 1851	18	22	III
Echinodermata	*Ophiura ophiura* (Linnaeus, 1758)	15	25	II
Annelida	*Glycera alba* (OF Müller, 1776)	13	17	IV

The most abundant species, and also present in 53% of the sites, was the polychaeta *Aponuphis brementi*. This species tolerates slightly disturbed habitats and is considered an organism of group II by AMBI. Within the nine most abundant species, four belonged to group II, and three to group I (species that inhabit undisturbed habitats). Moreover, *Notomastus latericeus*, with 22 specimens identified and *Glycera alba*, with 17 specimens identified, are organisms belonging to levels III and IV respectively, which means that they resist to disturbance in the ecosystem. Species found in EGIII are organisms that exist in ecosystems with mildly disturbed, moderate, and highly disturbed BI and the species present in EGIV are species present in places with moderate and mainly highly disturbed BI.

3.3. Ecological status analysis

AMBI and M-AMBI results are presented by sector respectively: MP1 in table 4 and 5, MP2 in table 6 and 7, MP3 in table 8 and 9 and MP5 in table 10 and 11.

Table 4 - AMBI results of the stations from the sector MP1.

Stations	I(%)	II(%)	III(%)	IV(%)	V(%)	AMBI	Not assigned (%)	BI	Disturbance Clasification
MP1-06	34,5	44,8	17,2	3,4	0	1,345	40,8	2	SD
MP1-21	26,8	46,3	12,2	4,9	9,8	1,866	30,5	2	SD
MP1-22	35,9	59,4	3,1	1,6	0	1,055	22,9	1	U
MP1-26	6,3	46,9	21,9	25	0	2,484	38,5	2	SD
MP1-28	37,8	43,2	10,8	8,1	0	1,338	26	2	SD
MP1-33	50	29,5	15,9	2,3	2,3	1,159	25,4	1	U
MP1-34	29,2	58,3	8,3	4,2	0	1,313	38,5	2	SD
MP1-36	27,3	52,3	20,5	0	0	1,398	35,8	2	SD
MP1-37	38,3	59,6	0	0	2,1	1,021	34,7	1	U
MP1-38	45,7	40	8,6	5,7	0	1,114	27,1	1	U
MP1-39	25,9	63	3,7	3,7	3,7	1,444	22,9	2	SD
MP1-978	19	59,5	9,5	2,4	9,5	1,857	36,4	2	SD
MP1-979	37,9	53,4	1,7	6,9	0	1,164	31	1	U
MP1-980	27,3	54,5	9,1	0	9,1	1,636	47,6	2	SD
MP1-981	12,1	57,6	18,2	12,1	0	1,955	61,2	2	SD
MP1-982	8,3	25	66,7	0	0	2,375	45,5	2	SD
MP1-983	41,1	51,8	7,1	0	0	0,991	45,6	1	U
MP1-984	38,5	28,2	2,6	0	30,8	2,346	39,1	2	SD
MP1-985	45	45	10	0	0	0,975	21,6	1	U
MP1-987	29,5	50,8	9,8	1,6	8,2	1,623	32,2	2	SD
MP1-989	26,8	64,3	7,1	1,8	0	1,259	16,4	2	SD
MP1-991	30,8	53,8	15,4	0	0	1,269	7,1	2	SD
MP1-992	0	70	20	10	0	2,1	60	2	SD
MP1-993	14,3	50	28,6	0	7,1	2,036	73,6	2	SD
MP1-994	10	45	15	0	30	2,925	16,7	2	SD

Order: U – Undisturbed, SD – Slightly Disturbed and BI – Biotic index

In the sector MP1 there are a total of 25 stations, 8 with BI level one (undisturbed) and 17 with BI level two (slightly disturbed) (Table 4). AMBI values are also in the middle of level's range, most stations being slightly disturbed, excepting for the station MP1-994, that is in the limit range (mean ambi of MP1-994 is 2,925 and the limit is 3,300) of the IB level two (slightly disturbed), the remaining sampling stations are within the parameters showing that the ecological status (considering only AMBI) is slightly disturbed, but still with 29,2% of level I species which is very sensitive to habitat changes and 51% of level II which is who are not so sensitive. It is important to mention that there are several taxa that were not identified to family level or below, which ends up interfering in the final results of the AMBI because when a taxon is at the family level or a less specific level, this taxon is often not assigned to an ecological group, which leads to greater imprecision of the final results. the more I specify the rate, the easier it is to assign an ecological group. In the Figure 4, the percentage of species per level of sensitivity together with the mean biotic indicator level (AMBI level) are showed for each station of the sector MP1.

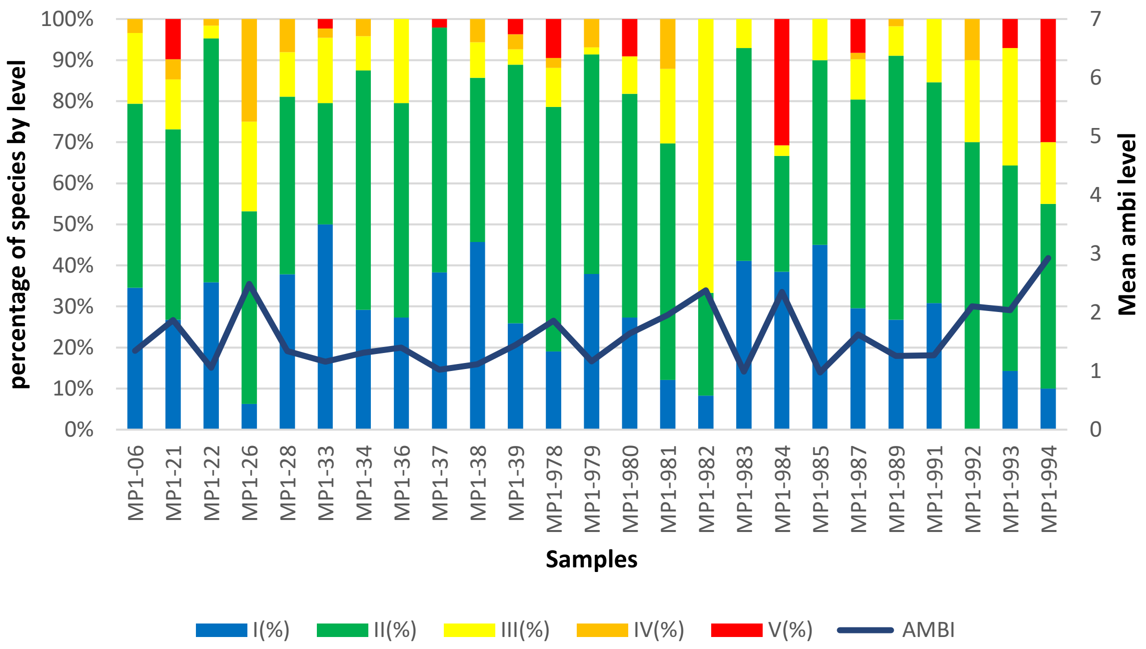

Figure 4 -Mean AMBI vs species percentage level per each stations from the sector MP1

The samples collected on the sector MP1, near a traditional fishing bivalve dredge “ganchorra”, showed a high quantity of organism of EGIV and EGV.

M-AMBI results, performed also by the AMBI software, match the values obtained from AMBI (Table 5).

Table 5 - M-AMBI results obtained by the AMBI software for stations from the sector MP1

Stations	AMBI	Diversity	Richness	M-AMBI	Status
Bad	6	0	0	0	Bad
High	0	4,51	42	1	High
MP1-06	1,3448	4,133	24	0,76677	Good
MP1-21	1,8659	4,3465	30	0,79411	High
MP1-22	1,0547	4,1004	29	0,81579	High
MP1-26	2,4844	3,6605	23	0,65519	Good
MP1-28	1,3378	3,9807	21	0,73451	Good
MP1-33	1,1591	4,5099	34	0,87635	High
MP1-34	1,3125	3,7388	21	0,71691	Good
MP1-36	1,3977	4,2275	36	0,85372	High
MP1-37	1,0213	4,0921	29	0,81711	High
MP1-38	1,1143	4,295	25	0,80008	High
MP1-39	1,4444	3,6639	26	0,73763	Good
MP1-978	1,8571	4,2474	29	0,77992	High
MP1-979	1,1638	4,3493	33	0,85652	High
MP1-980	1,6364	3,1057	11	0,57894	Good
MP1-981	1,9545	2,9964	18	0,59969	Good
MP1-982	2,375	1,7676	5	0,38834	Poor
MP1-983	0,99107	3,4735	23	0,72876	Good
MP1-984	2,3462	3,5778	19	0,6293	Good
MP1-985	0,975	4,3268	26	0,81771	High
MP1-987	1,623	3,8054	19	0,69004	Good
MP1-989	1,2589	4,3334	31	0,83586	High
MP1-991	1,2692	3,0931	10	0,59277	Good
MP1-992	2,1	2,6915	10	0,51195	Moderate
MP1-993	2,0357	2,4431	10	0,49615	Moderate
MP1-994	2,925	3,4073	12	0,53341	Good

The M-AMBI values compared to the AMBI values show that statistical results are coherent among sites (Table 5). Most of the sites are in a good status of quality, without disturbance or with a slightly disturbance. Most organisms found at the various stations belong to EGI and EGII. The sites MP1-982, MP1-992 and MP1-993 have a lower status (poor, moderate and moderate, respectively), even though they are considered by AMBI to be slightly disturbed. This is because M-AMBI uses several variables that contribute with a different weight. For instance, sites with similar AMBI index poses a quite different M-AMBI value, probably because of the specific richness and diversity.

The MP2 sector is the most northerly sector, it is the smallest sector containing only seven stations.

Table 6 - AMBI results of the stations from the sector MP2.

Stations	I(%)	II(%)	III(%)	IV(%)	V(%)	Mean AMBI	Not assigned (%)	BI	Disturbance Clasification
MP2-975	33,8	54,9	2,8	4,2	4,2	1,352	26	2	SD
MP2-976	46,7	40	13,3	0	0	1	65,9	1	U
MP2-977	26,7	66,7	4	2,7	0	1,24	26,5	2	SD
MP2-DSS-03	88,9	0	0	11,1	0	0,5	35,7	1	U
MP2-DSS-04	0	0	0	100	0	4,5	90,9	4	M
MP2-CSS-04	11,8	52,9	8,8	2,9	23,5	2,603	27,7	2	SD
MP2-CSS-05	19,2	78,8	1,9	0	0	1,24	7,1	2	SD

Order: U – Undisturbed, SD – Slightly Disturbed M – Moderate and BI – Biotic index

Four of the seven sites were slightly disturbed; however, they have a large diversity of organisms of distinct levels. The station MP2-DSS-04 was moderately disturbed (BI level four), however, 90% of the specimens were not assigned to an ecological group, probably because they were not identified to the species, genus, or family level (Figure 5).

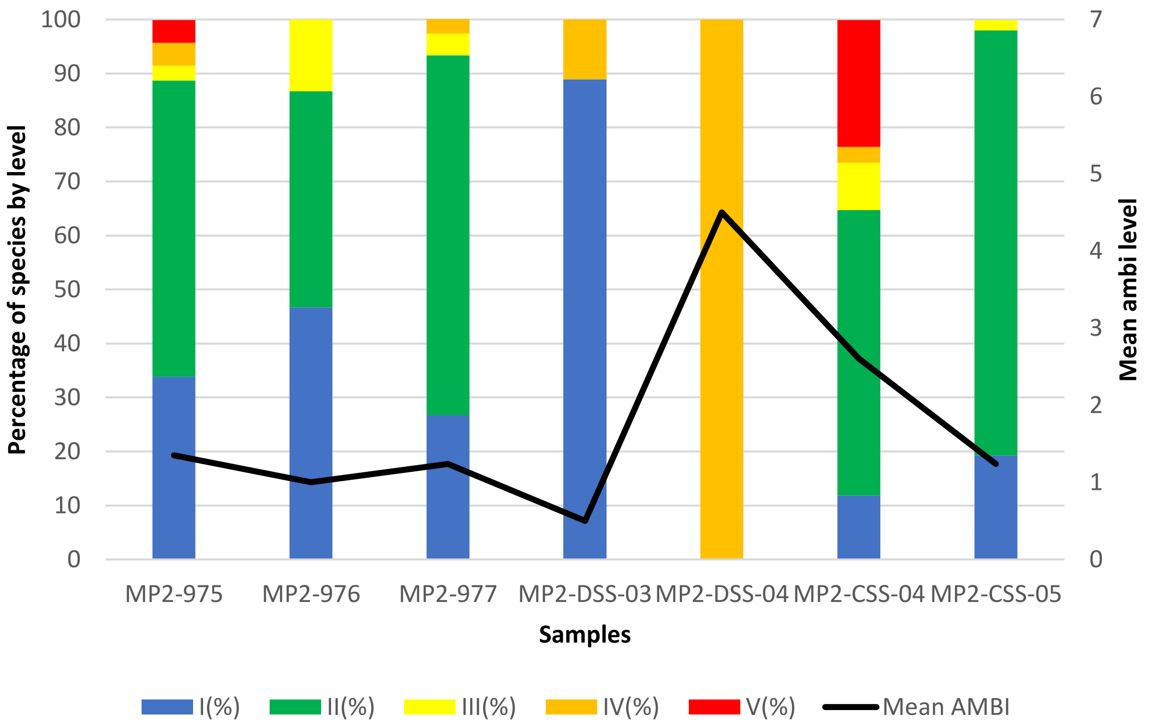

Figure 5 - Mean AMBI vs species percentage level per each station from the sector MP2.

All sites, excluding the stations MP2-DDS-04 and MP2-CSS-05, have at least good status (Table 7). The MP2 sector is close to the traditional fishing bivalve dredge “ganchorra” fishing area, with sampling stations that focus on the fishing area. The results show that the sampling stations are slightly disturbed (not including the two previous stations mentioned without a good status).

Table 7 - M-AMBI results obtained by the AMBI software for stations from the sector MP2

Stations	AMBI	Diversity	Richness	M-AMBI	Status
Bad	6	0	0	0	Bad
High	0	4,51	42	1	High
MP2-975	1,3913	4,2905	37	0,85938	High
MP2-976	1	2,4415	17	0,62125	Good
MP2-977	1,2568	4,3561	42	0,90507	High
MP2-DSS-03	0,47368	2,4452	8	0,59912	Good
MP2-DSS-04	4,5	1,2776	4	0,21963	Poor
MP2-CSS-04	2,4247	4,0573	30	0,7288	Good
MP2-CSS-05	1,2222	1,9094	12	0,53528	Good

M-AMBI results show that some sampling sites are in good environmental condition, in particular MP2-975 and MP2-977 with high status and MP2-976, MP2-DSS-03, MP2-CSS-04 and MP2-CSS-05 with good ecological status and at last the station MP2-DSS-04 as a poor ecological status. However, MP2-DSS-04 had a low rate of specimens identified at the species or genus level which led to a low specific richness and low diversity. This occurred because only 10% of the organisms were identified, making these results not reflect the season at the time of collection.

The results of the statistical analysis carried out with the AMBI software for the sector MP3 are shown in Table 8.

Table 8 - AMBI results of the stations from the sector MP3.

Stations	I(%)	II(%)	III(%)	IV(%)	V(%)	Mean AMBI	Not assigned (%)	BI	Disturbance Clasification
MP3-15	58,1	32,3	9,7	0	0	0,774	47,5	1	U
MP3-16	0	100	0	0	0	1,5	69,6	2	SD
MP3-17	13	73,9	4,3	8,7	0	1,63	53,1	2	SD
MP3-18	91,7	8,3	0	0	0	0,125	42,9	0	U
MP3-21	100	0	0	0	0	0	47,1	0	U
MP3-23	91,7	0	8,3	0	0	0,25	20	1	U
MP3-36	30,4	60,9	8,7	0	0	1,174	0	1	U
MP3-27	71,4	21,4	7,1	0	0	0,536	46,2	1	U
MP3-28	33,3	66,7	0	0	0	1	40	1	U
MP3-29	0	100	0	0	0	1,5	90,9	2	SD
MP3-30	21,2	78,8	0	0	0	1,182	31,3	1	U
MP3-36	44,4	55,6	0	0	0	0,833	40	1	U
MP3-37	26,7	73,3	0	0	0	1,1	28,6	1	U
MP3-39	100	0	0	0	0	0	85,7	0	U
MP3-40	83,3	16,7	0	0	0	0,25	62,5	1	U
MP3-43	33,3	66,7	0	0	0	1	76,9	1	U
MP3-46	61,1	38,9	0	0	0	0,583	52,6	1	U
MP3-47	25	0	75	0	0	2,25	33,3	2	SD
MP3-48	24,6	45,6	28,1	1,8	0	1,605	52,5	2	SD
MP3-49	54,2	29,2	0	8,3	8,3	1,313	60,7	2	SD
MP3-52	18,2	40,9	4,5	13,6	22,7	2,727	40,5	2	SD
MP3-53	54,5	36,4	0	0	9,1	1,091	51,1	1	U
MP3-62	0	75	12,5	12,5	0	2,063	57,9	2	SD
MP3-66	25	62,5	0	12,5	0	1,5	42,9	2	SD
MP3-67	50	50	0	0	0	0,75	66,7	1	U
MP3-996	58,8	23,5	17,6	0	0	0,882	45,2	1	U
MP3-997	0	50	50	0	0	2,25	66,7	2	SD
MP3-998	50	16,7	33,3	0	0	1,25	0	2	SD
MP3-999	71,4	14,3	14,3	0	0	0,643	36,4	1	U

Order: U – Undisturbed, SD – Slightly Disturbed, BI – Biotic index

The most frequent biotic index from the sites on the sector MP3 belong to BI level one and two (see Table 8 and Figure 6). The null numbers (0) of the biotic index (BI) from Mean AMBI means that there is biotic index level one with only EGI species. This is because in the equation 1, to calculate the value of the AMBI, the multiplication factor of the species is equal to zero, which causes the value at the end of the index to also be 0 given the absence of species of different EG. BI from Mean AMBI with value equal to 0 are means that the ecological status is undisturbed. Most stations have a percentage of not assigned organisms higher than 20% because many of them had not been identified to the species or genus level, and it was not possible to calculate its sensitivity level. It was not possible to identify these organisms down to the lowest taxonomic level "species or genus" because the samples were not in the best condition and some organisms end up being degraded and damaged.

Only five of the 29 MP3 stations (MP3-17, MP3-49, MP3-52, MP3-53, and MP3-63) had organisms of EGIV and EGV, which shows that the ecological status of the region is good. Despite many species of the station MP3-47 (60%) belonged to biotic level tree but the biotic index from mean AMBI remained on biotic level two. The Figure 6 shows the BI results with the line of mean AMBI.

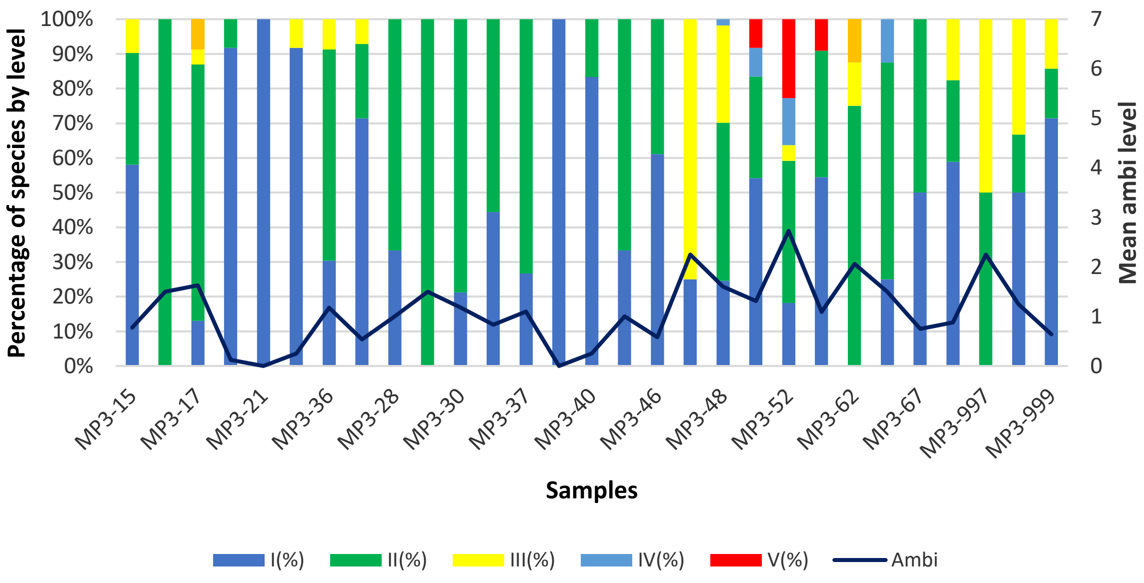

Figure 6 - Mean AMBI vs species percentage level per each station from the sector MP3.

Based on the values of the AMBI index, the ecological status of the seafloor in this sector is good and most stations are slightly disturbed or not disturbed. M-AMBI results, performed by the AMBI software are found in Table 8. The values obtained with the M-AMBI match the values obtained from AMBI (Table 9).

Table 9 - M-AMBI results obtained by the AMBI software for stations from the sector MP3

Stations	**AMBI**	**Diversity**	**Richness**	**M-AMBI**	**Status**
Bad	6	0	0	0	Bad
High	0	4,51	42	1	High
MP3-15	0,77419	3,4568	20	0,7304	Good
MP3-16	1,5	1,7713	6	0,46833	Moderate
MP3-17	1,6304	2,9942	17	0,62016	Good
MP3-18	0,125	2,9104	10	0,66943	Good
MP3-21	0	2,911	9	0,6714	Good
MP3-23	0,25	2,0402	6	0,57132	Good
MP3-36	1,1739	3,3773	12	0,64693	Good
MP3-27	0,53571	2,9322	11	0,65011	Good
MP3-28	1	3,1069	10	0,62576	Good
MP3-29	1,5	1,3486	3	0,41798	Moderate
MP3-30	1,1818	2,9148	10	0,59948	Good
MP3-36	0,83333	2,4226	7	0,56716	Good
MP3-37	1,1	3,2135	11	0,63337	Good
MP3-39	0	1,3788	3	0,51995	Moderate
MP3-40	0,25	2,4056	7	0,60469	Good
MP3-43	1	2,1416	6	0,52894	Moderate
MP3-46	0,58333	2,9413	15	0,67312	Good
MP3-47	2,25	2,2516	5	0,44758	Moderate
MP3-48	1,6053	3,2082	23	0,67592	Good
MP3-49	1,3125	3,4919	23	0,71634	Good
MP3-52	2,7273	3,8925	18	0,61997	Good
MP3-53	1,0909	3,8843	21	0,74731	Good
MP3-62	2,0625	2,4961	9	0,50362	Moderate
MP3-66	1,5	2,4067	7	0,52165	Moderate
MP3-67	0,75	2,2925	8	0,56947	Good
MP3-996	0,88235	2,9149	10	0,6194	Good
MP3-997	2,25	1,7925	4	0,40728	Moderate
MP3-998	1,25	1,9183	4	0,48306	Moderate
MP3-999	0,64286	2,914	8	0,6225	Good

The results of M-AMBI for most stations are congruent with the values obtained for the AMBI index. However, some divergences were found in few stations, MP3-16, MP3-29, MP3-39, MP3-43, MP3-47, MP3-62, MP3-66, MP3-997 and MP3-998 presented a moderate ecological status because although the results of the AMBI indicated slightly disturbance. However, M-AMBI as it has a very low specific richness and a very low divergence at the aforementioned stations, the station becomes a value much lower than the others. The stations MP3-17, MP3-49, MP3-52, MP3-53, and MP5-62 although they obtained organisms with EGIV and EGV, but even so the sampling stations ended up with good parameters because the organisms with a high and a higher EG had a low abundance. In general, the samples were in good ecological status.

Statistical analysis was then conducted using the AMBI software and the results for the MP5 sector are shown in table 10.

Table 10 - Mean AMBI vs species percentage level per each station from the sector MP5

Stations	I(%)	II(%)	III(%)	IV(%)	V(%)	Mean AMBI	Not assigned (%)	BI	Disturbance Clasification
MP5-CSS-01	33,3	44,4	11,1	11,1	0	1,5	62,5	2	SD
MP5-CSS-02	33,3	50	16,7	0	0	1,25	66,7	2	SD
MP5-CSS-03	23	41	19,7	11,5	4,9	2,016	30,7	2	SD
MP5-01	13	34,8	52,2	0	0	2,087	28,1	2	SD
MP5-02	0	0	100	0	0	3	66,7	2	SD
MP5-03	44,8	13,8	27,6	0	13,8	1,862	31	2	SD
MP5-05	30	30	10	30	0	2,1	63	2	SD
MP5-06	42,9	0	42,9	14,3	0	1,929	30	2	SD
MP5-07	25,6	65,1	0	0	9,3	1,535	41,1	2	SD
MP5-09	17,4	69,6	8,7	4,3	0	1,5	30,3	2	SD
MP5-11	0	77,8	11,1	11,1	0	2	40	2	SD
MP5-12	26,5	64,7	5,9	2,9	0	1,279	27,7	2	SD
MP5-13	0	100	0	0	0	1,5	50	2	SD
MP5-14	42,9	50	7,1	0	0	0,964	50	1	U
MP5-20	33,3	66,7	0	0	0	1	18,2	1	U
MP5-57	21,1	73,7	5,3	0	0	1,263	32,1	2	SD
MP5-60	20,8	70,8	8,3	0	0	1,313	27,3	2	SD
MP5-995	33,3	66,7	0	0	0	1	0	1	U

Order: U – Undisturbed, SD – Slightly Disturbed, BI – Biotic index

The MP5 stations, located near Sines, fit, in general, the biotic index level one and two. A total of four stations fitting the Biotic Index level one indicates an undisturbed state and fourteen stations fitting the Biotic Index level two what corresponds to a status slightly disturbed. Most stations have more than 20% of organisms not assigned to an ecological group, which can bias the results in order to increase the error in the attribution of the Biotic Index.

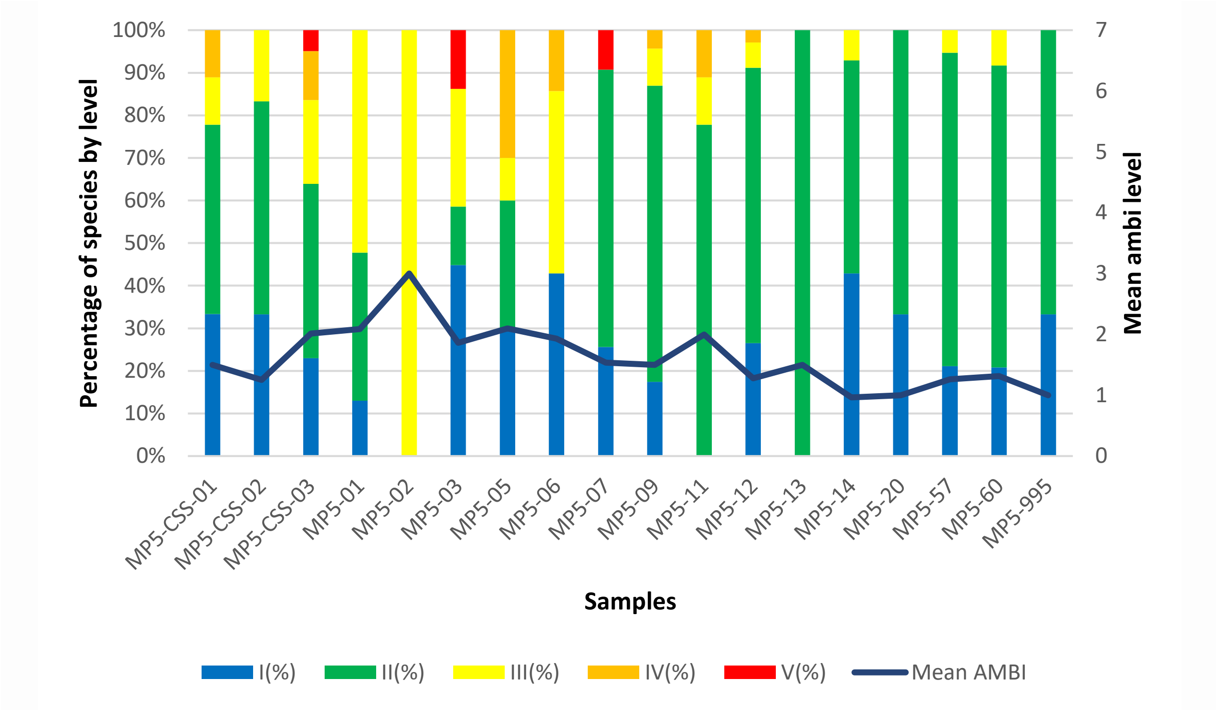

Figure 7 - Mean AMBI vs species percentage level per each station from the sector MP5

Table 10 and Figure 7 shows that most of the species collected in the sector MP5 belong to the EGI and EGII, only seven stations have specimens belonging to EGIV, and three stations to EGV. However, the results of the biotic index resulted in level one (MP5-05, MP5-14, MP5-20 and MP5-995) and two (the remaining stations), which tells us that most stations were only slightly disturbed. The station MP5-02 presented species belonging only to the EGIII, probably because 66.7% of the species were not assigned to an ecological group. This station had only three different taxa, making the result unreliable. M-AMBI results, performed by the AMBI software, are found in Table 11.

Table 11 - M-AMBI results obtained by the AMBI software at the MP5 stations.

Stations	AMBI	Diversity	Richness	M-AMBI	Status
Bad	6	0	0	0	Bad
High	0	4,51	42	1	High
MP5-CSS-01	1,5	2,4139	10	0,54132	Good
MP5-CSS-02	1,25	2,3627	8	0,54141	Good
MP5-CSS-03	2,0164	3,8892	26	0,71802	Good
MP5-01	2,087	2,4044	8	0,48884	Moderate
MP5-02	3	1,585	3	0,3357	Poor
MP5-03	1,8621	3,2971	11	0,58887	Good
MP5-05	2,1	2,5749	10	0,51332	Moderate
MP5-06	1,9286	2,171	5	0,46299	Moderate
MP5-07	1,5349	3,1661	12	0,60733	Good
MP5-09	1,5	3,2033	13	0,61877	Good
MP5-11	2	1,9656	5	0,44307	Moderate
MP5-12	1,2794	3,394	18	0,67941	Good
MP5-13	1,5	1,5	3	0,42916	Moderate
MP5-14	0,96429	2,8424	12	0,62135	Good
MP5-20	1	3,0958	9	0,61856	Good
MP5-53	1,2632	2,9488	12	0,60934	Good
MP5-60	1,3125	3,0592	13	0,6206	Good
MP5-995	1	0,9183	2	0,41306	Moderate

In general, more than half of the stations from sector MP5 presented a good ecological status, based on the taxonomic identifications. From eighteen stations, six (MP5-01, MP5-05, MP5-06, MP5-11, MP5-13, and MP5-995) because of the low species richness and the low mean AMBI result. Ten stations were in good ecological status which gives a total of 56%. In this way, a strong variation between the ecological status of the stations in this sector can be appreciated.

Standard deviation and standard error averages were calculated with the final M-AMBI values to understand the variation between the ecological status of each sector (Table 12). Stations in general have a low standard error, however the MP2 station has a higher error than the rest, this is due to the fact that this station only has 7 stations as compared to the rest having 18 or more.

Table 12 - Mean, standard deviation and standard error of the values obtained in the M-AMBI for each sector (MP1, MP2, MP3 and MP5) studied

M-AMBI	MP1	MP2	MP3	MP5
N	25	7	29	18
Average	0,705104	0,638361	0,59078	0,547635
Standard deviation	0,120724	0,212454	0,089681	0,098935
Standard error	0,023676	0,0803	0,016653	0,023319

According to Table 12, it is possible to see that, in general, the sectors are in a good ecological status (M-AMBI value > 0.53) and that the sector with the greatest variation (standard error) is MP2 with 8% error while the others does not exceed 2.4% standard error and the closer to the regions with higher anthropogenic activity, the greater the standard error, the sector with the smallest deviation (standard error) is the MP3.

4. Discussion

4.1. Sediment and organic matter

Generally, organic matter and sediment profile are closely related. Fine sediments, such as clay and silt, have a high tendency for the accumulation of organic matter (Hossain et al., 2014). Depth is another factor that influences the distribution of the sediment profile, because the further away from the coast and the deeper, the greater the tendency for the sediment to be thinner (mainly silt and clay) (Ewing et al., 1973). Marine ecosystems, where strong anthropogenic activity occurs, show high ecological disturbance, usually accumulating organic matter (Cotano & Villate 2006).

The MP1 sector has a very similar sediment profile between the stations sampled. Featuring an average of 60% medium, fine and very fine sand and a low content of clay and silt. None of the stations of the MP1 sector had more than 20% of clay or silt. The organic matter in MP1 sector also had an average of 2,4%, a very low value, which is considered normal given that the existence of clay and silt is very low (Hossain et al., 2014). Only station MP1-983 had a higher percentage of organic matter than normal for mostly sand sediment profile (Hossain et al., 2014). The station MP1-983 had 4% of organic material (the normal for sediment profiles, mostly sand, is <4%) (Hossain et al., 2014), leaving this station close to the limit. The MP1-983 station presents a sediment profile with finer sediment, with a percentage of 15,43% of clay and 23,1% of very fine sand and 16,43% of fine sand, which possibly led to the accumulation of a higher percentage of organic matter (Hossain et al., 2014).

The MP2 sector was very similar to MP1 sector, having a high content of very fine sand (36,6%) and fine sand (24,1%) with an average of 2,4% of organic matter. Two of the stations MP2-976 and MP2-DSS-03, present in MP2, had very coarse sediment profile that were located near and over the fishing region with traditional fishing bivalve dredge “ganchorra”, respectively. These stations also had low percentage of organic matter: 1,7% for MP2-976 and 0,9% for MP2-DSS-03. The MP2 sector showed the highest variance between stations. The MP2-CSS-05 station with 40.05% of clay, the MP2-DSS-03 station with coarse sediment (coarse sand and gravel that together exceeds 50% of the total analyzed sediment), the MP2-975 and MP2 stations -977 have a high amount of fine sand and medium sand, among other major differences in the stations of this sector that can be verified in Annex 1. (Hossain et al., 2014). The standard error of organic matter also reveals a large variation between seasons. Since this sector only has 7 stations, it may be influencing the large percentage of variation in results.

The MP3 sector was the most extensive with a total of 29 stations and presented some variance between them. The sediments in the sector were mainly composed by very fine sand and an average of 3,2% of organic matter, which justifies the existence of a greater amount of organic material present in this sector, as finer sediment leads to greater retention of organic material (Hossain et al., 2014). This sector has a lower anthropogenic activity as it is located on and next to a protected area (Figure 2). Some stations had levels above 4% of organic matter and the vast majority of these stations have a large amount of silt and clay (MP3-16, MP3-17, MP3-18, MP3-21, MP3-29, MP3-30, MP3-43, MP3-47 and MP3-62). Taking into account the type of sediment (15% and 30% in the sum between the percentages of silt and clay) it is natural that there is a greater amount of organic matter 4.1% according to the study by Hossain et al., (2014). The remaining stations end up having lower percentages of organic matter as they also have a lower amount of clay and silt. (Hossain et al., 2014).

The MP5 sector, located close to the port of Sines, had some variety of organic matter and sediment profile between stations. In general, the most abundant grain size fraction was composed by very fine sand. The station MP5-CSS-03 registered the highest amount of organic matter (6%) and the finest sediment, having 70,3% clay and 9,67% silt, probably due to its proximity to the port. The high intensity of anthropogenic activities occurring in this sector directly influenced the Sea floor, increasing the amount of organic matter (Cotano & Villate 2006). The vast majority of stations had a high value of organic matter (close to 4%) even with sediment profiles with larger grain size. The stations MP5-02, MP5-03, MP5-06, MP5-07 had a very low value of organic matter (less than 1,5%), probably due to they are located in the north of Sines far from the anthropogenic zone). In this way, we can understand that habitats with strong anthropogenic activity are more likely to have greater amounts of organic matter (Cotano & Villate 2006; Hossain et al., 2014).

4.2. Biotic index and geographic location

Firstly, it is essential to say that 41% of the organisms were identified up to the genus and 54% up to the family, which means that 46% of the organisms were not identified compared to the total number of organisms found in the samples. This situation is due to the fact that the taxonomic group with the highest incidence in the stations are the polychaetas with 62% of the total number of organisms found. More than half of the unidentified organisms up to the family were polychaetas representing 25% of the total and 54% of the unidentified organisms. The large percentage of organisms not identified until at least the family is due to the difficulty of identifying the taxonomic group (Alve et al., 2016). For instance, polychaetes and malacostraca, are organisms with various morphological characters that are easily damaged, turns the taxonomic identification to the species, genus, or family level, impossible to reach. Given this situation, a part of organisms was not assigned to an EG of AMBI, obtaining an average across all stations of 38,5%.

The more representative in terms of diversity and abundance taxonomic groups were annelids that have a wide spectrum regarding the sensitivity facilitating the perception of the ecological status (organisms with low tolerance to contaminants and organic matter, even organisms that have good resistance to them) to the presence of polychaetes which was the main class sampled (Dauvin & Ruellet, 2007). The second most abundant and whit most diversity phylum was Arthropoda, namely the order Amphipoda, which together with polychaetes are a good indicator of the ecological status of coastal and estuarine environments (Gomez Gesteira & Dauvin, 2000). When a local has sediments with high organic matter content, amphipods are one of the first groups to disappear from the ecosystem, and therefore, they are excellent indicators. Several families of amphipods, such as Ampeliscidae, Pontoporeidae, Melitidae and Gammaridae, are severely affected by hydrocarbons and can disappear completely given a large oil spill, for example (Dauvin, 1987, 1998 & 2000). A suitable number of stations had the presence of amphipods, namely from the Ampeliscidae family.

In the MP1 sector, although there was a notorious presence of organisms belonging to the EGIV and EGV (Figure 4), there was also a strong presence of amphipods in these regions. The stations MP1-26, MP1-978, MP1-984, MP1-987 and MP1-992 were the only ones with organisms of group IV or V with total absence of amphipods, however these stations were classified with a BI of level 2 and were only considered slightly disturbed. The total absence of Amphipods in these stations is most likely due to the type of sediment present in these places, since, based on the samples collected (annex 1), the previously mentioned stations have sediment with cursed sand and gravel, which is not the ideal sediment for the presence of amphipods (Biernbaum 1979). Another situation is that these stations are close to places with strong anthropogenic activity, was near traditional fishing bivalve dredge “ganchorra” area, which makes sensitive organisms disappear and more opportunistic organisms appear, as it was the case of polychaetas in other studies (Gomez Gesteira & Dauvin, 2000, Marques et al., 1997, 2003). Although the existence of organisms belonging to EGIV and EGV was perceived due to the proximity of an anthropogenic affected region, the stations previously mentioned were mostly undisturbed and slightly disturbed. According to the M-AMBI this stations are with high and good status Table 5, with the exception of stations MP1-992 and MP1-993, which are in the limit of the moderate to good status (transition value 0.53 between the range of good status to moderate status [Material and methods]) and station MP1-994 in between of good to moderate (Table 5). These values are so low probably because the stations previously mentioned are close to two places with strong anthropogenic activity, being located at the end of the MP1 sector near the fishing area with traditional fishing bivalve dredge “ganchorra” and close to the MP5 sector which is the sector around Sines what can influence the ecological state (place of high vessel traffic) (Thrush & Dayton, 2002; Thrush et al., 1998).

In the MP2 sector have a greatest variation of the ecological status between their stations, mainly due to the great distance between the MP2, MP2-DSS and MP2-CSS. Since MP2-DSS-03 AND MP2-DSS-04 are the stations with the greatest disturbance, that are located over an area with high anthropogenic activity, as is the case of traditional fishing bivalve dredge “ganchorra”, which end up having a greater disturbance in their ecosystem (Thrush & Dayton, 2002; Thrush et al., 1998). It is also important to mention that part of the MP2-DSS-04 station has a significant percentage of non-assigned organisms (90%) for calculating AMBI and M-AMBI. So to fully analyze this sector it is essential to complement the sediment and organic matter information along with the geographic location. The further away from places with anthropogenic activity, better will be the ecological status. (Thrush & Dayton, 2002; Thrush et al., 1998). Regarding the presence of amphipods like *Ampelisca brevicornis* (Costa, 1853) in relation to opportunist polychaetas like *Notomastus latericeus* Sars, 1851 and *Glycera* *alba* (O.F. Müller, 1776). in the stations MP2-DSS-04 and MP2-CSS-04, there is no presence of amphipods. However, in the station MP2-CSS-04 there is a large amount of polychaetas specimens from the family Capitallidae among other organisms of the EGV (Gomez Gesteira & Dauvin, 2000). Regarding the other stations (MP2-975, MP2-976 and MP2-977), given the existence of organisms belonging to the EGI (Melitidae Ampeliscidae, Amphithoidae) together with organisms from the EGIII and EGIV (opportunistic organisms such as *Glycera alba*) indicates that the ecosystem was slightly disturbed, (Borja et al., 2000) agreeing with the analysis of AMBI and M-AMBI.

In the MP3 sector there is a 63% of the stations have the presence of amphipods, namely belonging to the family Ampeliscidae, with a low amount of opportunistic (*Notomastus latericeus, Glycera alba*, *Polycirrus* sp. among others) organisms or with high tolerance to disturbances, appearing in 5 stations of the 29 stations present in this sector (MP3-17, MP3-48, MP3 -49, MP3-52, MP3-53, MP3-63 and MP3-62) whose percentage of species present combine to the EGIV and EGV does not exceed 15% except MP3-52 station with 22,7%. The sectors that present organisms that belong to EGIV and EGV also have amphipods among other organisms that are sensitive to disturbances in the ecosystems and the strong presence of organic and contaminated sediments (organisms that belong to ecological groups I and II) (Gomez Gesteira & Dauvin, 2000; Iliopoulou-Georgudaki et. al.,1997). Thus, we can say, according to AMBI, that the MP3 sector is generally slightly disturbed (34.5%) and undisturbed (65.5%) which makes it generally was undisturbed. The same agrees with the values obtained by the M-AMBI, giving in general a good ecological status (Figure 6). Another situation to take into consideration is that this sector is located next to a marine protected area, Professor Luiz Saldanha Marine Park (figure 2) which also reduces the stress on the communities due to low anthropogenic activities that occur in the area as demonstrated in other studies, such as Howell (2010).

Finally, the MP5 sector, which is located around the region of Sines, has a disturbance in some areas close to city of Sines and to the traditional fishing bivalve dredge “ganchorra” region. After the results obtained from AMBI, it was verified that 83,3% of the stations presente in this sector have a slightly disturbed level but the station MP5-02 was whit a high value which is almost transitioning to an IB level 3 (the value obtained in mean AMBI was 3 whose limit in IB level 2 is 3.3). Regarding the M-AMBI, 61,1% of the stations are in a good biological status, and 33,3% of the stations have a moderate status (MP5-CSS-02, MP5-01, MP5-06, MP5-11, MP5-13 and MP5-995) and poor status (MP5-02). The station MP5-02 is very close to the fishing area with traditional fishing bivalve dredge “ganchorra” and is also close to Sines, an area with a lot of anthropogenic activity which could influence the state of this station Thrush & Dayton, 2002; Thrush et al., 1998. Regarding the taxonomic groups present, the presence of amphipods is verified, but much less abundantly and with a lower diversity of species. Probably due to the fact that this sector is subject to a lot of anthropogenic activity that can generate some disturbance in ecosystems (Dauvin, 1987, 1998 & 2000).

In relation to the MP1 sector, the light content of average organic matter is 2.4%, which is a low value. Given the type of sediment in the sector which is poor in clay and silt percentages. (Hossain et al., 2014). The sediment is very thin despite the low presence of clay and silt, which makes this ecosystem more appropriate for many specimens (Hossain et al., 2014). Combining the results obtained using AMBI and M-AMBI together with the results of organic matter and granulometry the MP1 sector is slightly disturbed.

Comparing all sectors, MP2 showed the greatest variation in the results of the ecological status generated by M-AMBI, given the wide dispersion of stations in this sector compared to the remaining (Table 12) of its stations and the location of the same as previously mentioned. This sector in the stations MP2-DSS-03 MP2-DSS-04 taking into account the geographic location (place where there is strong anthropogenic activity such as traditional fishing bivalve dredge “ganchorra”)and the biological results, organic matter and granulometry, it is possible to say that they are in a moderately disturbed area, the rest can be considered slightly disturbed for the same reasons of the MP1 sector, are close to an area with high anthropogenic activity, presence of organisms of the ecological groups I and II together with organisms of groups IV and V.

The MP3 sector, located near a marine protected area (southwestern Alentejo and Costa Vicentina natural park). Regarding the organic matter and the type of sediment, is optimal given the parameters previously mentioned in the first chapter of the discussion. Regarding the AMBI and M-AMBI results, the vast majority of stations presented low disturbance, corresponding to what was expected (no disturbance) given that this sector is very close to a protected area, however, the ecological status of some areas (MP3-16, MP3-29, MP3-39, MP3-43, MP3-47, MP3-62, MP3-66, MP3-997 and MP3-998) was moderate (M-AMBI results) due to the specific richness be low. The final consideration in this sector is that it is undisturbed.

Comparing all sectors, MP5, located around the regions of Sines, had the worst ecological status (with the exception of the MP2-DSS-03 MP2-DSS-04 stations from MP2 sector) and the highest disturbance, possibly due to its proximity to anthropogenic activities, however this sector is still in a slightly disturbed state relative to the IB level. The results of the M-AMBI in turn reveal that 38.8% of the stations are in moderate (33.3%) and poor (4.5%) status, given the low richness, low diversity and the values previously obtained by the AMBI. Given all the variables, it can be said that this sector is on the edge of a slightly disturbed sector and can easily migrate to a moderately disturbed sector, finding itself in the opposite situation of the MP3 sector.

5. Conclusion

The MP1, MP2 and MP5 sectors are closer to areas with anthropogenic activity, which leads to a tendency to accumulate organic matter in the sediments and a higher disturbance, making the ecological status worse. The macrobenthic communities of many stations in the aforementioned sectors were composed of organisms belonging to ecological groups IV and V, which are organisms more resistant to disturbances in ecosystems. The MP3 sector, which is located in a region with less anthropogenic activity and close to a protected area, presented an ecological status without disturbance. However, although the MP3 sector was generally in good ecological condition, the macrobenthic communities showed low specific richness. It is concluded that places with greater proximity to anthropogenic activities have a greater presence of organic matter in the sediment and greater disturbance of ecosystems. It is also verified that the greater the number of samples, the more representative the sampling becomes. Finally, it is concluded that in this way, it is necessary to carry out monitoring over the years in order to verify the evolution of the ecological status of these communities, namely the MP5 sector, located in an area of greater human activity.

6. Conclusão

Os setores MP1, MP2 e MP5 possuem uma maior proximidade a áreas com atividade antropogénica, o que leva a uma tendência a acumular matéria orgânica nos sedimentos e uma perturbação mais elevada, fazendo com que o estado ecológico piore. As comunidades macrobentónicas de muitas estações dos setores anteriormente referidos foram compostas por organismos pertencentes aos grupos ecológicos IV e V, que são organismos mais resistentes a perturbações nos ecossistemas. O sector MP3, que se encontra numa região com uma menor atividade antropogénica e próximo a uma área protegida, apresentou um estado ecológico sem perturbação. No entanto, embora o setor MP3 se encontrasse com um bom estado ecológico em geral, as comunidades macrobentóncias apresentaram uma baixa riqueza especifica. Conclui-se que os locais com maior proximidade a atividades antropogénicas, apresentam maior a presença de matéria orgânica no sedimento e maior perturbação dos ecossistemas. Verifica-se também que quanto maior o número de amostras, mais representativa se torna a amostragem. Por fim conclui-se que desta forma, é necessário efetuar uma monitorização ao longo dos anos de forma a verificar a evolução do estado ecológico destas comunidades, nomeadamente do setor MP5, localizado numa área de maior atividade antrópica.

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Annexes

Annex 1 Granulometry results

stations	Sediment profile											
	Pebble	**Gravel**	**Very coarse sand**	**Coarse sand**	**Medi-um sand**	**Fine sand**	**Very fine sand**	**Silt**	**clay**	**M**	**So**	**Type of sediment**
MP1_006	1,14	1,39	1,26	3,37	23,21	34,45	18,15	6,02	11,01	160	1,9	Fine muddy-sand slightly with gravel
MP1_021	0,96	0,31	0,46	0,94	19,34	40,88	19,65	5,04	12,42	152	1,7	Fine muddy-sand slightly with gravel
MP1_022	0	1,38	5,63	53,34	15,89	6,68	1,96	0,61	14,52	803	1,8	Fine muddy-sand slightly with gravel
MP1_022A	0	0,33	1,98	4,85	8,64	44,02	35,62	4,00	0,56	138	1,4	Coarse muddy-sand slightly with gravel
MP1_026	0	1,09	0,77	9,91	39,67	8,72	15,03	6,38	18,44	258	3,0	Medium muddy-sand
MP1_028	0	0,00	0,22	6,66	46,97	23,40	20,26	2,21	0,27	259	1,6	Medium sand
MP1_033	0	0	0,20	1,72	34,37	22,57	21,50	5,65	14,00	169	2,1	Fine muddy-sand
MP1_034	0	0,21	0,62	3,48	29,80	25,59	20,48	4,48	15,34	160	2,0	Fine muddy-sand slightly with gravel
MP1_036	0	0,75	3,04	30,04	31,26	13,48	19,33	1,95	0,16	374	2,1	Medium sand with a little of gravel
MP1_037	0	1,61	5,82	22,85	35,82	15,01	15,93	2,45	0,50	335	2,0	Medium sand with a little of gravel
MP1_038	0	0	0,33	0,86	6,17	49,15	32,92	3,06	7,51	132	1,3	Fine muddy-sand
MP1_039	0	0,77	0,27	1,38	10,00	36,59	38,94	2,65	9,41	124	1,4	Very fine muddy-sand with a little of gravel
MP1-978	0	1,49	2,34	3,53	9,07	41,57	34,69	1,25	6,07	134	1,3	Fine mud sand slightly with gravel
MP1-979	0	0,63	3,76	5,07	5,47	40,62	42,33	1,68	0,45	131	1,2	Fine mud sand slightly with gravel
MP1-980	4,56	6,49	9,97	14,89	34,52	18,52	9,28	1,48	0,29	345	2,0	Medium sand with gravel
MP1-981	0,65	1,16	5,57	76,08	15,41	0,79	0,26	0,07	0,01	832	1,3	Coarse sand with a little of gravel
MP1-982	0,77	2,33	23,81	56,77	15,67	0,36	0,25	0,05	0,00	882	1,3	Coarse sand with a little of gravel
MP1-983	0	0,38	3,55	17,13	18,87	16,43	23,10	5,12	15,43	161	2,6	Fine mud sand slightly with gravel
MP1-984	2,82	3,80	11,34	60,20	18,07	2,42	0,95	0,26	0,14	849	1,3	Coarse sand with gravel
MP1-985	0	0	0,68	4,42	28,03	41,61	21,01	3,51	0,74	194	1,5	Fine sand
MP1_987	1,62	4,98	17,26	62,88	11,19	1,50	0,43	0,12	0,02	868	1,2	Coarse sand with gravel
MP1-989	0	0,55	0,65	2,05	9,55	38,77	29,91	5,31	13,23	127	1,6	Fine muddy-sand with a little of gravel
MP1-990	0	0	2,98	16,84	15,86	43,08	17,31	3,10	0,84	197	1,7	Fine sand
MP1-991	0	0,71	8,78	19,25	28,32	16,12	12,84	3,85	10,12	288	2,3	Medium muddy-sand with a little of gravel
MP1-992	0	0	2,33	75,85	19,25	1,51	0,79	0,24	0,04	767	1,3	Coarse sand
MP1-993	0	0,14	2,88	33,73	19,43	11,82	28,39	3,44	0,17	309	2,7	Medium sand with a little of gravel
MP1-993-3ª dredge	0,87	1,47	9,83	73,79	11,68	1,35	0,84	0,18	0,00	842	1,2	Coarse sand with a little of gravel
MP1-994	0	0,62	2,42	11,69	10,32	18,16	31,55	8,35	16,89	100	2,4	Very fine muddy-sand with a little of gravel
MP2-975	0	0,623	0,577	7,043	19,323	33,047	36,622	2,323	0,442	152	1,7	Fine sand sland slightly with gravel
MP2-976	1,47	5,55	21,37	43,32	24,04	3,51	0,45	0,18	0,12	847	1,6	Coarse sand with gravel
MP2-977	0	2,25	6,48	8,02	20,35	24,14	36,02	2,43	0,31	168	1,8	Fine mud sand slightly with gravel

MP2-CSS-4	0	0,42	0,88	4,35	14,44	15,11	41,73	4,23	18,83	90	2,0	Very fine muddy-sand with a little of gravel
MP2-CSS-5	0,93	0,34	0,15	0,49	1,92	6,48	42,53	7,11	40,05	51	-	Very fine muddy-sand with a little of gravel
MP2-DSS-01	0	0,59	1,07	2,26	6,06	30,49	49,54	2,18	7,82	113	1,4	Very fine muddy-sand with a little of gravel
MP2-DSS-02	0	0	0,68	3,46	13,38	57,27	25,18	0,04	0	161	1,3	Fine sand
MP2-DSS-03	3,52	4,47	14,81	39,64	34,68	2,27	0,61	0,00	0,00	738	1,6	Coarse sand gravel
MP2-DSS-04	0,25	0,24	2,36	3,54	10,49	45,00	38,01	0,10	0,01	144	1,4	Fine mud sand slightly with gravel
MP3_15	0	0,21	0,45	0,81	1,02	4,90	65,25	12,58	14,80	69	1,5	Very fine muddy-sand with a little of gravel
MP3_16	0	0	0,44	0,60	3,16	13,20	52,13	11,47	18,99	71	1,6	Very fine muddy-sand
MP3_17	0	0	2,08	4,18	14,00	20,24	40,30	7,65	11,55	100	1,9	Very fine muddy-sand
MP3_18	0	0,39	0,75	2,09	23,27	33,37	29,12	3,28	7,74	158	1,7	Fine muddy-sand slightly with gravel
MP3_21	0	0	0,84	4,07	11,84	20,06	40,45	8,48	13,89	95	1,9	Very fine muddy-sand
MP3_23	0	2,73	6,56	13,15	33,59	17,75	16,17	2,52	7,54	267	2,0	Medium muddy-sand with a little of gravel
MP3_26	0	0	0,19	0,54	1,55	17,40	68,22	3,94	7,97	100	1,3	Very fine muddy-sand
MP3_27	0,14	0,75	1,60	2,22	6,76	34,28	44,19	3,29	6,78	119	1,5	Very fine muddy-sand with a little of gravel
MP3_28	0	0	2,73	1,99	3,96	31,05	57,29	2,32	0,67	116	1,3	Very fine sand
MP3_29	0	0	0,44	0,95	2,74	11,14	58,03	9,55	17,16	71	1,5	Very fine muddy-sand
MP3_30	0	0	0,36	0,55	4,76	20,63	54,01	7,24	12,46	89	1,5	Very fine muddy-sand
MP3_36	0	0	0,54	0,92	2,27	34,18	47,19	5,95	8,97	108	1,5	Very fine muddy-sand
MP3_37	1,36	6,20	28,71	35,49	23,30	4,02	0,84	0,09	0,00	893	1,7	Coarse sand slightly gravel
MP3_37A	0	0	0,45	2,23	3,78	52,56	39,67	1,12	0,19	134	1,2	Fine sand
MP3_39	0,45	1,85	4,37	5,14	5,33	31,12	47,04	4,24	0,45	122	1,5	Very fine muddy-sand with a little of gravel
MP3_40	1,07	6,11	12,18	26,83	51,49	1,85	0,44	0,03	0	478	1,6	Medium sand with gravel
MP3_43	0	0,49	1,56	1,49	1,79	8,92	62,63	7,80	15,32	84	1,5	Very fine muddy-sand with a little of gravel
MP3_46	0	0	0	1,51	2,97	45,91	48,67	0,79	0,15	125	1,2	Fine sand
MP3_47	0	0,96	0,89	0,97	2,16	21,22	55,16	5,36	13,29	102	1,4	Very fine muddy-sand with a little of gravel
MP3_48	0	0,15	0,34	0,53	1,49	24,58	61,39	3,40	8,12	109	1,2	Very fine muddy-sand with a little of gravel
MP3_49	0,58	1,16	4,77	3,54	3,94	29,90	51,61	4,06	0,44	118	1,3	Very fine sand with a little of gravel
MP3_52	0,19	0,26	0,42	0,75	1,67	43,54	45,36	2,65	5,16	122	1,2	Very fine sand with a little of gravel
MP3_53	0	0	0,34	1,27	4,16	36,39	47,25	2,51	8,08	118	1,3	Very fine muddy-sand
MP3_62	0,10	0,03	0,17	1,00	12,77	27,19	44,38	3,92	10,45	106	1,7	Very fine muddy-sand with a little of gravel
MP3_66	0	0	0,04	0,25	0,94	11,19	76,38	4,59	6,61	93	1,2	Very fine muddy-sand
MP3_67	0	0	0,25	1,33	3,13	35,32	56,61	2,95	0,41	115	1,3	Very fine sand
MP3-996	0	0,78	5,24	19,99	68,03	5,35	0,44	0,13	0,04	340	1,4	Medium sand with a little of gravel
MP3-997	0	1,91	4,21	73,94	19,32	0,42	0,17	0,03	0	690	1,3	Coarse sand slightly gravel
MP3-998	1,74	9,52	44,02	41,50	1,85	0,66	0,51	0,17	0,03	1087	1,3	Very coarse sand with gravel
MP3-999	0,65	15,54	53,96	26,26	1,78	0,49	0,87	0,32	0,12	1295	1,4	Very coarse sand with gravel
MP5_01	0,71	0,57	0,40	0,98	1,89	9,89	65,82	6,94	12,80	83	1,4	Very fine muddy-sand with a little of gravel
MP5_02	0	1,97	12,25	71,02	14,06	0,63	0,06	0,00	0,00	864	1,2	Coarse sand slightly gravel

MP5_03	0	2,25	24,14	62,91	10,45	0,16	0,10	0,00	0,00	900	1,1	Coarse sand slightly gravel
MP5_05	0	0	0,33	1,18	3,42	31,44	48,43	4,78	10,42	108	1,4	Very fine muddy-sand
MP5_06	0	0	3,43	69,81	26,33	0,19	0,23	0,01	0,00	712	1,4	Coarse sand
MP5_07	0	0	2,61	89,47	6,42	0,82	0,53	0,13	0,01	859	1,1	Coarse sand
MP5_09	0	0	0,54	1,57	3,43	18,37	57,08	6,45	12,57	94	1,4	Very fine muddy-sand
MP5_11	0	4,74	4,39	20,04	36,71	4,89	11,35	3,50	14,40	363	2,7	Medium muddy-sand with a little of gravel
MP5_12	0	0,68	4,89	20,63	32,87	13,21	18,70	2,97	6,05	314	2,2	Medium sand with a little of graveç
MP5_13	0,28	0,04	0,42	1,87	18,53	31,69	33,21	3,83	10,14	131	1,7	Fine mud sand slightly with gravel
MP5_14	0	0	0,23	4,12	35,19	26,45	22,37	4,03	7,60	176	1,7	Fine muddy-sand
MP5_20	0	0,27	0,31	2,19	31,33	24,78	27,17	4,37	9,58	172	1,9	Fine muddy-sand with a little of gravel
MP5_55	0	0	0	0,613	14,942	55,378	26,378	2,689	0	159	1,4	Fine sand
MP5_57	0	0,16	0,30	1,26	6,23	35,09	45,68	5,06	6,23	115	1,4	Very fine muddy-sand with a lithe of gravel
MP5_60	0	0	1,02	3,88	21,76	29,11	42,66	1,35	0,22	140	1,6	Fine sand
MP5-995	0	0,47	1,31	16,50	45,97	13,66	14,65	2,21	5,23	285	1,7	Medium sand whit some gravel
MP5-CSS-1	0	0,25	0,88	2,03	12,74	33,64	32,42	5,25	12,80	124	1,7	Very fine muddy-sand with gravel
MP5-CSS-2	0	4,08	3,72	20,26	13,75	14,21	30,84	3,70	9,45	160	2,7	Fine mud sand slightly with gravel
MP5-CSS-3	0	0	0,40	1,43	5,00	3,24	10,00	9,67	70,27	-	-	Mud sand

Annex 2 Organic matter results

Stations	Date	MOT Replicate 1 (%)	MOT Replicate 2 (%)	MOT (%)
MP1_006	31/03/2019	2,6	2,5	2,5
MP1_021	31/03/2019	2,0	2,1	2,1
MP1_022	31/03/2019	1,0	1,0	1,0
MP1_022A	31/03/2019	2,4	2,6	2,5
MP1_026	01/04/2019	2,9	2,9	2,9
MP1_028	01/04/2019	2,0	2,1	2,1
MP1_033	01/04/2019	3,2	2,7	3,0
MP1_034	01/04/2019	2,8	2,7	2,8
MP1_036	02/04/2019	2,5	2,8	2,7
MP1_037	02/04/2019	2,7	2,6	2,7
MP1_038	01/04/2019	1,9	1,8	1,9
MP1_039	02/04/2019	2,5	2,9	2,7
MP1-978	02/04/2019	2,4	2,2	2,3
MP1-979	02/04/2019	2,7	2,6	2,6
MP1-980	02/04/2019	3,0	2,4	2,7
MP1-981	02/04/2019	0,8	0,7	0,8
MP1-982	02/04/2019	1,1	1,0	1,1
MP1-983	01/04/2019	4,0	4,0	4,0
MP1-984	01/04/2019	0,9	1,2	1,1
MP1_985	01/04/2019	3,0	3,7	3,3
MP1_987	01/04/2019	1,1	1,8	1,5
MP1-989	31/03/2019	1,9	1,9	1,9
MP1-990	31/03/2019	0,9	1,6	1,2
MP1-991	31/03/2019	3,0	2,5	2,7
MP1-992	31/03/2019	0,4	0,5	0,5
MP1-993	31/03/2019	1,3	0,9	1,1
MP1-993_3ª dredge	31/03/2019	1,3	1,6	1,4
MP1-994	31/03/2019	3,0	2,9	2,9
MP2-975	03/04/2019	2,9	3,1	3,0
MP2-976	03/04/2019	2,1	1,2	1,7
MP2-977	03/04/2019	2,4	2,4	2,4
MP2-CSS-04	04/04/2019	3,5	3,0	3,2
MP2-CSS-05	04/04/2019	4,4	5,0	4,7
MP2-DSS-02	03/04/2019	0,9	0,8	0,8
MP2-DSS-03	03/04/2019	0,8	0,8	0,8
MP2-DSS-04	03/04/2019	0,8	1,0	0,9
MP2-4DSS-01	03/04/2019	2,8	2,4	2,6
MP3_15	27/03/2019	3,1	3,0	3,1
MP3_16	27/03/2019	3,9	3,9	3,9
MP3_17	27/03/2019	4,0	4,1	4,0

MP3_18	**27/03/2019**	3,6	3,9	**3,8**
MP3_21	**26/03/2019**	4,7	4,7	**4,7**
MP3_23	**26/03/2019**	3,6	3,6	**3,6**
MP3_26	**29/03/2019**	2,2	2,3	**2,2**
MP3_27	**27/03/2019**	3,5	3,5	**3,5**
MP3_28	**29/03/2019**	2,5	2,5	**2,5**
MP3_29	**27/03/2019**	4,2	4,0	**4,1**
MP3_30	**27/03/2019**	3,8	3,7	**3,8**
MP3_36	**29/03/2019**	3,4	3,5	**3,5**
MP3_37	**29/03/2019**	1,4	1,4	**1,4**
MP3_37A	**29/03/2019**	1,2	1,2	**1,2**
MP3_39	**29/03/2019**	3,3	3,2	**3,2**
MP3_40	**29/03/2019**	1,5	1,7	**1,6**
MP3_43	**28/03/2019**	3,5	3,9	**3,7**
MP3_46	**28/03/2019**	1,6	1,8	**1,7**
MP3_47	**28/03/2019**	3,1	3,1	**3,1**
MP3_48	**28/03/2019**	3,1	2,8	**3,0**
MP3_49	**28/03/2019**	3,8	3,4	**3,6**
MP3_52	**28/03/2019**	2,7	2,9	**2,8**
MP3_53	**28/03/2019**	2,4	2,8	**2,6**
MP3_62	**26/03/2019**	3,4	3,6	**3,5**
MP3_66	**26/03/2019**	3,1	3,0	**3,0**
MP3_67	**28/03/2019**	2,8	2,6	**2,7**
MP3-996	**29/03/2019**	2,2	2,4	**2,3**
MP3-997	**29/03/2019**	0,9	0,8	**0,8**
MP3-998	**29/03/2019**	3,9	3,9	**3,9**
MP3-999	**29/03/2019**	3,3	3,1	**3,2**
MP5_01	**25/03/2019**	3,3	3,3	**3,3**
MP5_02	**25/03/2019**	0,6	0,5	**0,6**
MP5_03	**25/03/2019**	0,4	0,4	**0,4**
MP5_05	**24/03/2019**	2,9	3,0	**2,9**
MP5_06	**24/03/2019**	0,2	0,3	**0,3**
MP5_07	**24/03/2019**	1,2	1,1	**1,1**
MP5_09	**24/03/2019**	2,9	3,0	**2,9**
MP5_11	**25/03/2019**	3,7	3,2	**3,5**
MP5_12	**24/03/2019**	2,1	2,0	**2,1**
MP5_13	**25/03/2019**	3,7	3,6	**3,7**
MP5_14	**23/03/2019**	2,5	2,6	**2,6**
MP5_20	**25/03/2019**	3,5	3,5	**3,5**
MP5_55	**24/03/2019**	2,6	2,7	**2,6**
MP5_57	**30/03/2019**	2,6	2,6	**2,6**
MP5_60	**26/03/2019**	3,0	3,0	**3,0**
MP5-995	**30/03/2019**	2,9	2,9	**2,9**
MP5-CSS-01	**24/03/2019**	2,9	2,9	**2,9**
MP5-CSS-02	**25/03/2019**	3,1	2,5	**2,8**

MP5-CSS-03	**25/03/2019**	6,0	6,0	**6,0**

Annex 3 - identification table of organisms present in sector MP1

				Sector	MP1																								
Phylum	**Class**	**Order**	**Family**	**Species/Stations**	**06**	**21**	**22**	**26**	**28**	**33**	**34**	**36**	**37**	**38**	**39**	**978**	**979**	**980**	**981**	**982**	**983**	**984**	**985**	**987**	**989**	**991**	**992**	**993**	**994**
Annelida	Clitellata				1								1	1															
	Polychaeta				10	13	16	18	9	12	11	29	19	7	13	12	15	6	39	40	37	16	5	10	8	1	12	11	3
		Amphinomida	Amphinomidae	*Chloeia venusta* Quatrefages, 1866											1														
		Eunicida																	5					8	1			4	2
			Eunicidae						4			20					1	1						2	4				
				Eunice dubitata Fauchald, 1974					2																				
				Lysidice unicornis (Grube, 1840)																							8		
				Palola siciliensis (Grube, 1840)								5												7					
				Paucibranchia sp.																						1			
			Lumbrineridae			1				1		4						2					1		1	1			
				Lumbrinerides amoureuxi Miura, 1981		2											1												
				Lumbrineris latreilli Audouin & Milne Edwards, 1833					1													4							
				Scoletoma laurentiana (Grube, 1863)									6																
			Oenonidae	*Arabella iricolor* (Montagu, 1804)						1		1	2	3			1												
				Arabella sp.												1					1								
			Onuphidae				1											1											
				Aponuphis brementi (Fauvel, 1916)	3	3	6	3	6	3	1		3	2	22	8	2		4	4			2		3		8	2	
				Aponuphis grubii (Marenzeller, 1886)																		2							
				Aponuphis sp.												1					1								
				Hyalinoecia tubicola (O.F. Müller, 1776)		1					1	1										1							
				Nothria conchylega (Sars, 1835)			12	2																					
				Nothria sp.						1															11				
				Onuphis eremita Audouin & Milne Edwards, 1833													1												
				Onuphis sp.	1					1						5	3												
				Paradiopatra calliopae Arvantidis & Koukouras, 1997													6												
				Paradiopatra quadricuspis (M. Sars in G. O. Sars, 1872)					1														6		2				
				Paradiopatra sp.		3				1																			
		n/a	Arenicolidae				1																						
				Arenicola marina (Linnaeus, 1758)							1																		
			Capitellidae			3				1			1			1		1				12		5					3
				Capitella sp.		1										2												1	3
				Heteromastus filiformis (Claparède, 1864)			1	3																					
				Notomastus latericeus Sars, 1851	2	1		1	2		1	1		2	1	1							2			2			
				Notomastus sp.					2	1												1			1				1
			Chaetopteridae			1																							
			Magelonidae																				1						
			Magelonidae	*Magelona mirabilis* (Johnston, 1865)						1																			
			Maldanidae			1	2				1				2						5	1	2		1			1	
				Heteroclymene robusta Arwidsson, 1906											2														
				Macroclymene santanderensis (Rioja, 1917)					1																				
				Maldane glebifex Grube, 1860																					1				
				Maldane sarsi Malmgren, 1865																									
				Maldane sp.															1							1			
				Maldanella sp.												1													

				Sector	MP1																								
Phylum	**Class**	**Order**	**Family**	**Species/Stations**	**06**	**21**	**22**	**26**	**28**	**33**	**34**	**36**	**37**	**38**	**39**	**978**	**979**	**980**	**981**	**982**	**983**	**984**	**985**	**987**	**989**	**991**	**992**	**993**	**994**
				Notoproctus oculatus Arwidsson, 1906																					1				
				Praxillella affinis (M. Sars in G. O. Sars, 1872)																									1
			Orbiniidae		3			1			1	1	3			1	1		1				1						
				Orbinia latreillii (Audouin & H. Milne Edwards, 1833)																					1				
				Naineris sp.		2																							
				Orbinia sp.			4				1				1										1				2
			Oweniidae								1												1						
				Owenia fusiformis Delle Chiaje, 1844																	1								
				Owenia sp.													2						4						
			Sabellariidae																		2								
				Sabellaria alveolata (Linnaeus, 1767)	1											1													
				Sabellaria sp.			1														3								
				Sabellaria spinulosa (Leuckart, 1849)								2	1									1							
			Scalibregmatidae	*Scalibregma inflatum* Rathke, 1843																		1							
		Phyllodocida				2						4									4		1		1				
			Aphroditidae													1	1												
				Laetmonice hystrix (Savigny in Lamarck, 1818)			1				1					1													
				Laetmonice sp.												1													
				Lepidasthenia sp.																					1				
				Pontogenia chrysocoma (Baird, 1865)								4				1									1			1	
			Glyceridae										1								5								
				Glycera alba (O. F. Müller, 1776)	1			1			1			1	1										1				
				Glycera lapidum Quatrefages, 1866							1		1																
				Glycera sp.			1																						
				Glycera tesselata Grube, 1863						1		1																	
				Glycera unicornis Lamarck, 1818			1						1																
			Goniadidae	*Goniada emerita* Audouin & H. Milne Edwards, 1833						1																			
				Goniada maculata Örsted, 1843			1								1														
				Goniada sp.																			1		1				
			Nephtyidae	*Nephtys cirrosa* Ehlers, 1868	1		1				1												1						
				Nephtys hombergii Savigny in Lamarck, 1818	5	1	1	1	1	1				1					1						1				
			Nereididae									1																	
			Phyllodocidae			1											1				5		1						
				Mysta barbata Malmgren, 1865								1																	
			Sigalionidae	*Pholoe inornata Johnston, 1839*																			1						
				Pholoe sp.																									1
				Sigalion sp.																							4		1
				Sthenelais sp.													2												
			Syllidae														1												
			Syllidae	*Syllis gracilis* Grube, 1840				1																					
		Sabellida	Sabellidae	*Branchiomma bombyx* (Dalyell, 1853)									1																
			Serpulidae	*Protula tubularia* (Montagu, 1803)																					1				
			Spionidae			1																							
				Dipolydora flava (Claparède, 1870)				1																					
				Malacoceros sp.		1																							

				Sector	**MP1**																								
Phylum	**Class**	**Order**	**Family**	**Species/Stations**	**06**	**21**	**22**	**26**	**28**	**33**	**34**	**36**	**37**	**38**	**39**	**978**	**979**	**980**	**981**	**982**	**983**	**984**	**985**	**987**	**989**	**991**	**992**	**993**	**994**
				Prionospio sp.										1															
				Scolelepis (Scolelepis) foliosa (Audouin & Milne Edwards, 1833)				1																					
			Magelonidae	*Magelona* sp.						1																			
		Terebellida										4	1				5							4					
			Ampharetidae		2	1		1	2	1	1			2		5	1		4				2		2				
				Sosane sulcata Malmgren, 1866	1																								
				Amage sp.								1																	
				Ampharete sp.			1																						
				Auchenoplax crinita Ehlers, 1887												1													
				Eclysippe vanelli (Fauvel, 1936)	3		2		6	5			5	2	2	1		3	1		4		1		1				
				Ampharete octocirrata (M. Sars, 1835)					1																				
				Zatsepinia rittichae Jirkov , 1986	1																								
			Cirratulidae			1		1	3					1			3		3								4		
				Monticellina		1											1												
			Flabelligeridae					1																					
			Melinnidae	*Melinna cristata* (M. Sars, 1851)				1								1													
				Melinna palmata Grube, 1870				2																					
				Melinna sp.															1										
			Pectinariidae							1									4										
				Amphictene auricoma (O. F. Müller, 1776)								1	1								1								
				Auricoma anficteno (O. F. Müller, 1776)												1													
				Lagis koreni Malmgren, 1866				1		1						1													1
			Terebellidae			1	4				3	1	1	2	3	4	1				1		1		3				
				Nicolea sp.									1	2															
				Pista sp.						1					1									1					
				Proclea graffii (Langerhans, 1884)					1																				
				Streblosoma sp.		1																							
				Thelepus cincinnatus (Fabricius, 1780)			2					1																	
				Thelepus setosus (Quatrefages, 1866)											1														
			Trichobranchidae	*Trichobranchus sp.*										2	1														
				Terebellides stroemii Sars, 1835		1								1	1						1				2				
	Sipuncula	n/a	Aspidosiphonidae	*Aspidosiphon (Aspidosiphon) muelleri* Diesing, 1851																				1					
			Sipunculidae	*Sipunculus (Sipunculus) nudus* Linnaeus, 1766				1				1			1						1			1				1	
Arthropoda																						4							
	Malacostraca																					4							
		Amphipoda			3							5		1	2	3		3				4					44	24	
			Iphimediidae																									4	
			Ampeliscidae										1																
				Ampelisca brevicornis (Costa, 1853)	3	1	6		3	5	1	1	1		1		7		1				4		1				
				Ampelisca gibba G.O. Sars, 1883													1												
				Ampelisca typica (Spence Bate, 1856)													2												
			Aoridae										1																
				Lembos websteri Spence Bate, 1857			3																						
			Aristiidae	*Perrierella audouiniana* (Spence Bate, 1857)								1																	
			Atylidae	*Nototropis falcatus (Metzger, 1871)*						1																			

				Sector	MP1																								
Phylum	**Class**	**Order**	**Family**	**Species/Stations**	**06**	**21**	**22**	**26**	**28**	**33**	**34**	**36**	**37**	**38**	**39**	**978**	**979**	**980**	**981**	**982**	**983**	**984**	**985**	**987**	**989**	**991**	**992**	**993**	**994**
			Dexaminidae															1											
			Iphimediidae	*Iphimedia minuta* G.O. Sars, 1883										1															
			Leucothoidae	*Leucothoe procera* Spence Bate, 1857	1																								
			Phoxocephalidae	*Harpinia antennaria* Meinert, 1890											1														
		Cumacea			1					1												4							
			Bodotriidae							1								1											
				Bodotria scorpioides (Montagu, 1804)					2																				
				Iphinoe trispinosa (Goodsir, 1843)					1						1														
		Decapoda										1					1							8					
			Atelecyclidae	*Atelecyclus rotundatus* (Olivi, 1792)									1																
			Axiidae	*Calocaris macandreae* Bell, 1846	1																								
			Callianassidae						1																				
				Callianassa sp.																					1				
				Callianassa subterranea (Montagu, 1808)	1					1																			
				Gilvossius tyrrhenus (Petagna, 1792)				2																					2
			Hippolytidae														1												
			Leucosiidae	*Ebalia cranchii* Leach, 1817				1							1														
			Paguridae																						1				
			Paguridae	*Pagurus prideaux* Leach, 1815						1																			
			Pasiphaeidae	*Pasiphaea sivado* (Risso, 1816)									1																
			Polybiidae	*Liocarcinus pusillus* (Leach, 1816)				1																					
			Porcellanidae	*Pisidia longicornis* (Linnaeus, 1767)							1																		
		Isopoda																									4		
			Arcturidae																4										
				Cyathura carinata (Krøyer, 1847)								1																	
			Cirolanidae	*Cirolana cranchii* Leach, 1818					1									1		8									
				Eurydice pulchra Leach, 1815													1					2							
				Natatolana borealis (Lilljeborg, 1851)																						1	4		
		Tanaidacea	Tanaididae																								4		
			Tanaissuidae	*Tanaissus bamberi* Esquete, 2015			1																						
Briozoa																1													
Cnidaria	Anthozoa												10																
		Alcyonacea	Primnoidae	*Callogorgia verticillata* (Pallas, 1766)										1															
		Penicillaria	Arachnactidae	*Isarachnanthus* sp.				1																					
Echinodermata	Asteroidea	Paxillosida	Astropectinidae	*Astropecten irregularis* (Pennant, 1777)																		1							
	Echinoidea	Echinolampadacea	Fibulariidae	*Echinocyamus pusillus* (O.F. Müller, 1776)																		1							
		Spatangoida	Brissidae	*Brissopsis lyrifera* (Forbes, 1841)						1					2						2		1						
			Spatangidae	*Spatangus purpureus* O.F. Müller, 1776								1																	
			Loveniidae	*Echinocardium cordatum* (Pennant, 1777)	1					1			1	1										14		1			
	Holothuroidea	Apodida	Synaptidae	*Oestergrenia digitata* (Montagu, 1815)		1																							
	Ophiuroidea	Amphilepidida	Amphiuridae	*Amphiura chiajei* Forbes, 1843	1							2					7					2							
				Acrocnida brachiata (Montagu, 1804)								1													2				
				Amphipholis squamata (Delle Chiaje, 1828)										4			1												
			Ophiactidae	*Ophiactis balli* (W. Thompson, 1840)									4																
		Ophiurida														1	1		8										

				Sector																MP1									
Phylum	**Class**	**Order**	**Family**	**Species/Stations**	**06**	**21**	**22**	**26**	**28**	**33**	**34**	**36**	**37**	**38**	**39**	**978**	**979**	**980**	**981**	**982**	**983**	**984**	**985**	**987**	**989**	**991**	**992**	**993**	**994**
			Ophiuridae	*Ophiocten affinis* (Lütken, 1858)		6	8	6		1	6	8	3		5	5	10				15	4	2	1	9	4			4
				Ophiura ophiura (Linnaeus, 1758)		2				1		1	6		1				1				6						
				Ophiura robusta (Ayres, 1852)							2	1		6							2								
				Ophiura sp.																				4					
Molusca	Bivalvia										1		1			1					1								
		Carditida	Astartidae	*Astarte sulcata* (da Costa, 1778)									2																
		Cardiida	Cardiidae	*Cerastoderma edule* (Linnaeus, 1758)																			1						
				Papillicardium minimum (Philippi, 1836)		1						6																	
				Parvicardium sp.													1												
			Semelidae	*Scrobicularia plana* (da Costa, 1778)											1														
			Tellinidae	*Asbjornsenia pygmaea* (Lovén, 1846)															1				1		1				
				Fabulina fabula (Gmelin, 1791)						1																			
				Macomangulus tenuis (da Costa, 1778)										2															
				Moerella donacina (Linnaeus, 1758)																		2							
		Lucinida	Lucinidae	*Lucinoma borealis* (Linnaeus, 1767)								8																	
		Myida	Corbulidae	*Varicorbula gibba* (Olivi, 1792)															1					1					
			Pholadidae																					8					
		n/a	Cuspidariidae	*Cardiomya costellata* (Deshayes, 1835)																						1			
				Cuspidaria obesa (Lovén, 1846)						2																			
		Nuculida	Nuculidae	*Nucula nucleus* (Linnaeus, 1758)																				1					
				Nucula sulcata Bronn, 1831						1											4	1							
		Pectinida	Pectinidae	*Mimachlamys varia* (Linnaeus, 1758)																	1								
		Venerida	Arcticidae	*Arctica* sp.																				2					
				Diplodonta rotundata (Montagu, 1803)													1				2								
	Cephalopoda	Sepiida	Sepiolidae	*Sepietta neglecta* Naef, 1916			1																						
	Gastropoda	Caenogastropoda	Turritellidae	*Turritellinella tricarinata* (Brocchi, 1814)			1																						
		Cephalaspidea	Cylichnidae	*Cylichna cylindracea* (Pennant, 1777)		1																	1						
		Littorinimorpha	Eulimidae	*Melanella polita* (Linnaeus, 1758)										1															
		Neogastropoda	Buccinidae	*Buccinum humphreysianum* Bennett, 1824			1				1																		
	Scaphopoda	Dentaliida	Dentaliidae	*Antalis entalis* (Linnaeus, 1758)																						1			
				Antalis vulgaris (da Costa, 1778)																4									
Nematoda					1	3	2			4		4		1		2		1		32				4	1			4	
Nemertea	Anopla				1							10							5		4		1						
Nemertea										1																	8		
Platyhelminthes							1								1	1								8					
Porifera												2																	

Annex 4 - identification table of organisms present in sector MP2

Sector				MP2						
Class	**Order**	**Family**	**Species/Stations**	**975**	**976**	**977**	**DSS-03**	**DSS-04**	**CSS-4**	**CSS-5**
				1						
Clitellata					1					
Polychaeta				20	53	19	8	32	19	3
	Phyllodocida	Polynoidae	*Lepidonotus squamatus* (Linnaeus, 1758)	1						
			Malmgrenia castanea McIntosh, 1876			1				
	Eunicida					1				
		Lumbrineridae							2	1
			Lumbrinerides sp.						1	
			Scoletoma laurentiana (Grube, 1863)	1						
		Oenonidae	*Arabella iricolor* (Montagu, 1804)	1		1				
		Onuphidae	*Aponuphis brementi* (Fauvel, 1916)	14		23			16	
		Onuphidae	*Aponuphis* sp.						4	
		Onuphidae	*Hyalinoecia tubicola* (O.F. Müller, 1776)			2			2	
		Onuphidae	*Onuphis* sp.	12					1	
		Onuphidae	*Paradiopatra* sp.	1						1
		Onuphidae							1	
	n/a	Capitellidae							12	
			Capitella sp.	1					3	
			Heteromastus filiformis (Claparède, 1864)			1				
			Leiocapitella sp.						1	
			Notomastus latericeus Sars, 1851	1					2	
		Chaetopteridae			4					
		Magelonidae	*Octomagelona bizkaiensis* (Ceberio & Fiege, 2001)						2	
		Maldanidae		4						1
			Chirimia bíceps (M. Sars, 1861)	1						
			Euclymene sp.			1				
			Maldane sarsi Malmgren, 1865	1						
			Maldanella sp.	2						
		Orbiniidae							3	1
		Orbiniidae	*Orbinia* sp.	1						
		Sabellariidae	*Sabellaria spinulosa* (Leuckart, 1849)			2				
	Phyllodocida								1	
		Aphroditidae	*Aphrodita aculeata* Linnaeus, 1758		1					
			Laetmonice filicornis Kinberg, 1856		4					
			Lepidonotus squamatus (Linnaeus, 1758)	1						
		Glyceridae	*Glycera alba* (O. F. Müller, 1776)	1						
			Glycera tridactyla Schmarda, 1861			2				
		Goniadidae	*Goniada* sp.		4					
		Nephtyidae	*Nephtys cirrosa* Ehlers, 1868	1						
			Nephtys hombergii Savigny in Lamarck, 1818			1			1	
		Nereididae				1				
			Hediste diversicolor (O.F. Müller, 1776)			1				
		Phyllodocidae		1						
		Sigalionidae	*Sigalion* sp.						1	

			Sector					MP2		
Class	**Order**	**Family**	**Species/Stations**	**975**	**976**	**977**	**DSS-03**	**DSS-04**	**CSS-4**	**CSS-5**
	Sabellida	Sabellidae	*Chone sp.*			1				
	Terebellida			1		1				
		Ampharetidae		2		1				
			Sosane sulcata Malmgren, 1866						3	
			Amage sp.							1
			Ampharete acutifrons (Grube, 1860)			1				
			Ampharete finmarchica (M. Sars, 1865)			2				
			Ampharete sp.	1		1				
			Eclysippe vanelli (Fauvel, 1936)	3					4	
		Cirratulidae		2						
		Pectinariidae	*Amphictene auricoma* (O. F. Müller, 1776)	1		1				
		Terebellidae				1				
		Terebellidae	*Polycirrus caliendrum* Claparède, 1869			1				
		Trichobranchidae	*Trichobranchus sp.*			1				
			Terebellides sp.	1						
			Terebellides stroemii Sars, 1835			2				
Sipuncula	n/a	Sipunculidae	*Sipunculus (Sipunculus) nudus* Linnaeus, 1766			2				
Malacostraca	Amphipoda					1	1			
		Ampeliscidae		2						
			Ampelisca brevicornis (Costa, 1853)		2	3				4
			Ampelisca sp.	1						
		Ampithoidae				1				
		Ampithoidae	*Pleonexes gammaroides* Spence Bate, 1857			1				
		Melitidae	*Melita palmata* (Montagu, 1804)			1				
		Oedicerotidae					1			
	Cumacea	Bodotriidae	*Bodotria scorpioides* (Montagu, 1804)			1				
			Iphinoe sp.	1						
			Iphinoe trispinosa (Goodsir, 1843)			1				
	Decapoda			1						
		Callianassidae	*Gilvossius tyrrhenus* (Petagna, 1792)	1						
		Inachidae	*Inachus dorsettensis* (Pennant, 1777)		1					
		Pasiphaeidae	*Pasiphaea sivado* (Risso, 1816)		1					
		Porcellanidae	*Pisidia longicornis* (Linnaeus, 1767)		1					
	Euphausiacea	Euphausiidae	*Nyctiphanes couchii* (Bell, 1853)			2				
	Isopoda	Cirolanidae	*Cirolana cranchii* Leach, 1818		1	1				
Anthozoa										39
	Actiniaria			1						
	Pennatulacea	Virgulariidae	*Virgularia mirabilis (*Müller, 1776)						1	1
Echinoidea	Spatangoida	Brissidae	*Brissopsis lyrifera* (Forbes, 1841)	2		1			2	
		Loveniidae	*Echinocardium cordatum* (Pennant, 1777)	1	4				1	
Holothuroidea									1	
Ophiuroidea	Ophiurida	Ophiuridae	*Ophiocten affinis* (Lütken, 1858)	4	1				3	
			Ophiura ophiura (Linnaeus, 1758)			2				1
			Ophiura sp.			8				
	Amphilepidida	Ophiactidae	*Ophiactis balli* (W. Thompson, 1840)			1				

			Sector					**MP2**		
Class	**Order**	**Family**	**Species/Stations**	**975**	**976**	**977**	**DSS-03**	**DSS-04**	**CSS-4**	**CSS-5**
Bivalvia								4	1	
	Cardiida	Tellinidae							1	
			Asbjornsenia pygmaea (Lovén, 1846)				1			
			Macomangulus tenuis (da Costa, 1778)			1				3
			Moerella donacina (Linnaeus, 1758)		1					
	Myida	Corbulidae	*Varicorbula gibba* (Olivi, 1792)				2	4		
	Mytilida	Mytilidae						4		
	n/a	Cuspidariidae	*Cuspidaria rostrata* (Spengler, 1793)			2				
	Venerida	Arcticidae	*Arctica islandica* (Linnaeus, 1767)						1	
		Veneridae	*Dosinia exoleta* (Linnaeus, 1758)				1			
Gastropoda	Caenogastropoda	Cerithiidae	*Bittium reticulatum* (da Costa, 1778)	2						
	Littorinimorpha	Caecidae	*Caecum trachea* (Montagu, 1803)				5			
Scaphopoda	Dentaliida	Dentaliidae	*Antalis vulgaris* (da Costa, 1778)				9			
					4	1			1	
									2	
					1					
				3	4					
n/a	Priapulomorpha	Priapulidae	*Priapulus caudatus* Lamarck, 1816							1

Annex 5 - identification table of organisms present in sector MP3

				Sector	MP3																												
Phylum	**Class**	**Order**	**Family**	**Species/Station**	**15**	**16**	**17**	**18**	**21**	**23**	**26**	**27**	**28**	**29**	**30**	**36**	**37**	**39**	**40**	**43**	**46**	**47**	**48**	**49**	**52**	**53**	**62**	**66**	**67**	**996**	**997**	**998**	**999**
Annelida	Clitellata																		1														
	Polychaeta				22	14	23	6	4	1		10	2	5	10	6	4	4	6	6	16	1	49	24	5	10	9	5	12	4			
		Eunicida				1	5				5	2			3	1					1		15		1		3	4		1			8
			Eunicidae	*Marphysa* sp.									1																				
				Palola siciliensis (Grube, 1840)																		1			1								
				Paucibranchia fallax (Marion & Bobretzky, 1875)	2																												
			Lumbrineridae		1		2						4								1			1									
			Onuphidae				2								9		4								1		1						
				Arabella iricolor (Montagu, 1804)	2							3																					
				Aponuphis bilineata (Baird, 1870)	6		1								2	2													2				
				Aponuphis brementi (Fauvel, 1916)		4	1				3		1		2	3	2			1	2		3	2	2	2	1						
				Aponuphis sp.	1		2	1																1	2	2			1				
				Hyalinoecia tubicola (O.F. Müller, 1776)	3																												
				Leptoecia apalpata (Hartmann-Schröder, 1975)			1														1												
				Nothria maremontana André & Pleijel, 1989											11													1					
				Nothria sp.			3														1												
				Onuphis sp.																			1		1								
				Paradiopatra quadricuspis (M. Sars in G. O. Sars, 1872)																	1												
				Paradiopatra sp.					2																								
		n/a	Capitellidae																					2	5	2							
				Notomastus latericeus Sars, 1851																					1								
				Notomastus sp.																				1	2								
			Magelonidae																					5									
				Magelona filiformis Wilson, 1959																	7												
				Magelona johnstoni Fiege, Licher & Mackie, 2000				1	4																								
			Maldanidae																		2		1			1							
				Maldane sarsi Malmgren, 1865																						1							
			Orbiniidae		3			2									1		1				3		1								
				Naineris laevigata (Grube, 1855)																			2										
			Oweniidae		2																												
				Galathowenia oculata (Zachs, 1923)																		1											
		Phyllodocida			4				1					5						3	1		2	1		6			4	10	8		8
			Aphroditidae		1			1			1													1		1							
			Glyceridae	*Glycera alba* (O. F. Müller, 1776)			1																										
			Goniadidae	*Goniada maculata* Örsted, 1843																	1												
				Goniada sp.																													
			Hesionidae																2														
			Microphthalmidae	*Hesionides* sp.																													
			Nephtyidae	*Nephtys cirrosa* Ehlers, 1868																			4		1	2			1				
				Nephtys hombergii Savigny in Lamarck, 1818													1						1	1									
			Sigalionidae	*Sigalion* sp.																	1												
		Terebellida					1												1				2		3								16
			Ampharetidae				1	1															4				1	1					
				Auchenoplax crinita Ehlers, 1887																				1									

			Sector		MP3																												
Phylum	**Class**	**Order**	**Family**	**Species/Station**	**15**	**16**	**17**	**18**	**21**	**23**	**26**	**27**	**28**	**29**	**30**	**36**	**37**	**39**	**40**	**43**	**46**	**47**	**48**	**49**	**52**	**53**	**62**	**66**	**67**	**996**	**997**	**998**	**999**
				Eclysippe vanelli (Fauvel, 1936)			1																2			1							
			Cirratulidae				1																1	2	2								
			Pectinariidae								2																						
				Amphictene auricoma (O. F. Müller, 1776)																									1				
				Petta pusilla Malmgren, 1866	1																												
			Terebellidae						1												1		4	3	4	1	1						
				Pista sp.																			2										
				Polycirrus sp.																					1								
	Sipuncula	n/a	Aspidosiphonidae	*Aspidosiphon (Aspidosiphon) muelleri* Diesing, 1851															4									1		2		4	
			Golfingiidae									2																					
				Golfingia (Golfingia) elongata (Keferstein, 1862)							2	1																					
				Nephasoma (Nephasoma) minutum (Keferstein, 1862)									1																				
				Onchnesoma steenstrupii Koren & Danielssen, 1876				5				1			4																		
				Thysanocardia procera (Möbius, 1875)																						2							
			Sipunculidae	*Sipunculus (Sipunculus) nudus* Linnaeus, 1766																1													
Arthropoda	Malacostraca								2	2			1											1									
		Amphipoda				2		2				1	2		5		2	2						3		1							
			Ampeliscidae										1				1									1							
				Ampelisca brevicornis (Costa, 1853)	2			1			2												4	5	3	1							
				Ampelisca sp.																			1										
			Dexaminidae	*Dexamine* sp.																										1			
			Ischyroceridae	*Ericthonius* sp.													1																
			Leucothoidae																					1									
			Phoxocephalidae	*Phoxocephalus holbolli* (Krøyer, 1842)																								1					
				Phoxocephalus sp.																						1							
		Cumacea														1																	
			Bodotriidae	*Bodotria* sp.		1																											
			Diastylidae	*Diastylis lucifera* (Krøyer, 1837)							2																						
		Decapoda				1	2									1																	
			Callianassidae	*Callianassa subterranea* (Montagu, 1808)																		2											
				Gilvossius tyrrhenus (Petagna, 1792)			1																										
			Goneplacidae						1																								
			Leucosiidae	*Ebalia* sp.							1																						
			Paguridae								1																						
			Paguridae	*Pagurus pubescens* Krøyer, 1838										1																			
		Isopoda										1												1									
			Cirolanidae	*Eurydice pulchra* Leach, 1815					1			2																		4			16
		Tanaidacea	Apseudidae	*Apseudes spinosus* (M. Sars, 1858)								2																					
			Sphyrapodidae																					1									
			Tanaididae																											2			
	Ostracoda	Podocopida	Pontocyprididae										1																				
	Thecostraca	Scalpellomorpha	Scalpellidae	*Scalpellum scalpellum* (Linnaeus, 1767)																			15										
Briozoa															1																		
Chordata	Ascidiacea									1																							
Cnidaria	Anthozoa																													1			

			Sector		MP3																												
Phylum	**Class**	**Order**	**Family**	**Species/Station**	**15**	**16**	**17**	**18**	**21**	**23**	**26**	**27**	**28**	**29**	**30**	**36**	**37**	**39**	**40**	**43**	**46**	**47**	**48**	**49**	**52**	**53**	**62**	**66**	**67**	**996**	**997**	**998**	**999**
Cnidaria										8																2							
Echinodermata	Echinoidea	Spatangoida																						1									
			Loveniidae	*Echinocardium cordatum* (Pennant, 1777)																	1					1				4			8
	Holothuroidea	Apodida	Synaptidae	*Leptosynapta* sp.																						1							
		Holothuriida	Holothuriidae																				1										
	Ophiuroidea	Amphilepidida	Amphiuridae	*Amphiura (Ophiopeltis) securigera* (Düben & Koren, 1846)																			1	1									
				Amphiura chiajei Forbes, 1843													1																
				Amphipholis squamata (Delle Chiaje, 1828)																													16
		Ophiurida									2												1										
			Ophiuridae																							1						4	
				Ophiocten affinis (Lütken, 1858)	1												3							1									
				Ophiura ophiura (Linnaeus, 1758)																					1								
Molusca	Bivalvia																				1												
		Galeommatida	Lasaeidae	*Kellia suborbicularis* (Montagu, 1803)																									1				
		Carditida	Astartidae	*Astarte sulcata* (da Costa, 1778)	1					2			1																				
		Cardiida	Tellinidae	*Asbjornsenia pygmaea* (Lovén, 1846)																				1									
				Macomangulus tenuis (da Costa, 1778)	2										1																		
				Moerella donacina (Linnaeus, 1758)																												8	
								1			1																						
		Venerida	Ungulinidae	*Diplodonta rotundata* (Montagu, 1803)	1																												
		Lucinida	Lucinidae	*Loripes orbiculatus* Poli, 1795													1																
		Myida	Corbulidae	*Varicorbula gibba* (Olivi, 1792)																							1						
		Nuculida	Nuculidae	*Nucula sulcata* Bronn, 1831												1																	
		Nuculanida	Nuculanidae	*Lembulus pella (Linnaeus, 1758)*																								1					
		Pectinida	Pectinidae	*Palliolum tigerinum* (O. F. Müller, 1776)					1																								
		Venerida	Veneridae																			1											
	Gastropoda	Caenogastropoda	Turritellidae																	1													
		Littorinimorpha	Naticidae	*Euspira nitida* (Donovan, 1803)	1																												
		Neogastropoda	Muricidae	*Trophonopsis muricata* (Montagu, 1803)						1																							
	Scaphopoda	Dentaliida	Dentaliidae	*Antalis entalis* (Linnaeus, 1758)							1																						
				Antalis vulgaris (da Costa, 1778)														1															
Nematoda					2																		1				1			2	8	8	8
Nemertea					1							1																					
Platyhelminthes																			1	1							1		2		8		
Porifera																										5							

Annex 6 - identification table of organisms present in sector MP5

				Sector	MP5																	
Phylum	**Class**	**Order**	**Family**	**Species/Station**	**CSS-01**	**CSS-02**	**CSS-03**	**01**	**02**	**03**	**05**	**06**	**07**	**09**	**11**	**12**	**13**	**14**	**20**	**57**	**60**	**995**
Annelida	Clitellata									4												
	Polychaeta				13	9	23	9		1	13	24	16	8	6	12	2	12	2	6	8	
		Eunicida			1		8	3		4	2		4			8	1				6	4
			Eunicidae													1						
			Lumbrineridae				4						6									
			Lumbrineridae	*Lumbricalus* sp.			4												1		1	
			Onuphidae											1				2				
				Aponuphis bilineata (Baird, 1870)				1								1						
				Aponuphis brementi (Fauvel, 1916)									2	8	5	9		4		9	8	
				Aponuphis sp.	1		1												1	4	2	
				Nothria conchylega (Sars, 1835)			4															
				Nothria sp.										3								
		n/a	Capitellidae				2						4									
				Capitella sp.			1															
			Magelonidae	*Magelona mirabilis* (Johnston, 1865)												1						
			Maldanidae					1														
			Orbiniidae													1				1		
				Naineris sp.						4												
		Phyllodocida					1		4	4			10							1	1	
			Acoetidae			1																
				Panthalis oerstedi Kinberg, 1856																1		
			Aphroditidae				1							2								
			Glyceridae				1									2						
				Glycera alba (O. F. Müller, 1776)			1					8										
			Goniadidae	*Goniada* sp.													1			1	1	
			Microphthalmidae	*Hesionides* sp.	1																	
			Nephtyidae				2															
				Nephtys hombergii Savigny in Lamarck, 1818			3							1								
			Syllidae							4												
		Sabellida	Sabellidae													1					1	
		Spionida																			1	
			Spionidae	*Malacoceros fuliginosus* (Claparède, 1868)						4												
			Spionidae		1															1		
		Terebellida																1				
			Ampharetidae								1											
			Cirratulidae		1		1									1						
			Pectinariidae				2															
			Terebellidae													1		1				
	Sipuncula	n/a	Aspidosiphonidae	*Aspidosiphon (Aspidosiphon) muelleri* Diesing, 1851						4			2									
			Golfingiidae	*Onchnesoma steenstrupii* Koren & Danielssen, 1876	2	1										1		1	1			2
Arthropoda	Malacostraca	Amphipoda			2		1		4		2			2								
			Ampeliscidae	*Ampelisca brevicornis* (Costa, 1853)				1								1		2	1			
				Ampelisca sp.	1						2											
			Dexaminidae	*Dexamine spinosa (Montagu, 1813)*							1											

				Sector							MP5											
Phylum	**Class**	**Order**	**Family**	**Species/Station**	**CSS-01**	**CSS-02**	**CSS-03**	**01**	**02**	**03**	**05**	**06**	**07**	**09**	**11**	**12**	**13**	**14**	**20**	**57**	**60**	**995**
			Leucothoidae																	1		
		Cumacea					1													1		
			Bodotriidae	*Bodotria arenosa* Goodsir, 1843			2				1											
				Bodotria scorpioides (Montagu, 1804)										1								
				Iphinoe trispinosa (Goodsir, 1843)										1								
		Decapoda											16									
			Processidae	*Processa* sp.						4												
		Isopoda				2																
			Anthuridae				1															
			Cirolanidae	*Eurydice pulchra* Leach, 1815																	1	
				Eurydice spinigera Hansen, 1890								16										
			Gnathiidae	*Gnathia maxillaris* (Montagu, 1804)							1											
	Ostracoda	Podocopida	Loxoconchidae	*Loxoconcha rhomboidea* (Fischer, 1855)							1											
Echinodermata	Echinoidea	Spatangoida	Loveniidae	*Echinocardium cordatum* (Pennant, 1777)						1								1				
	Ophiuroidea	Amphilepidida	Amphiuridae	*Amphipholis squamata* (Delle Chiaje, 1828)														1			1	
		Ophiurida												1				1				
			Ophiuridae	*Ophiocten affinis* (Lütken, 1858)		1								2		1						
				Ophiura albida Forbes, 1839	1											1						
				Ophiura ophiura (Linnaeus, 1758)		2													1			
				Ophiura robusta (Ayres, 1852)															1			
				Ophiura sp.															1			
Molusca	Bivalvia						2						4							1		
		Cardiida	Solecurtidae										1									
		Limida	Limidae	*Limaria* sp.									4									
		Carditida	Astartidae	*Astarte sulcata* (da Costa, 1778)		1	3											1				
		Cardiida	Semelidae	*Abra alba* (W. Wood, 1802)			12															
			Tellinidae	*Macomangulus tenuis* (da Costa, 1778)			1						4									
		Myida	Corbulidae	*Varicorbula gibba* (Olivi, 1792)			5															
		Nuculida	Nuculidae	*Nucula nucleus* (Linnaeus, 1758)			1															
		Nuculanida	Nuculanidae	*Lembulus pella (Linnaeus, 1758)*							3			1	1							
		Venerida	Ungulinidae	*Diplodonta rotundata* (Montagu, 1803)				2														
	Gastropoda	Cephalaspidea	Scaphandridae	*Scaphander lignarius* (Linnaeus, 1758)												1						
	Scaphopoda	Dentaliida	Dentaliidae	*Antalis entalis* (Linnaeus, 1758)																1		
				Antalis vulgaris (da Costa, 1778)								8				2					1	
Nematoda						1		12		8		24		2		2		1				
Nemertea	Anopla																					
Nemertea									4						1						1	
Platyhelminthes								3							2				2			
Porifera																						
Priapulida	n/a	Priapulomorpha	Priapulidae	*Priapulus caudatus* Lamarck, 1816																		