Biofouling on plastic waste as indicator for source origin
Macroplastics in the Delta project

Final report

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Thesaurus

OSPAR = International cooperation for the protection of the marine environment in the North-East Atlantic.
Macroplastics = Large plastic particles with a size of over 5 mm.
Biofouling = the attachment of organisms to natural (rocks, shells) or unnatural (ship hulls, wood, plastic) surfaces.
State of degradation = The state and quality of a certain item.
Non-indigenous species = Species introduced outside their natural range.
Bioaccumulation = The accumulation of a substance or chemical in an organism.
Biodegradability = The capacity for biological degradation of organic materials.
Metamorphosis = The biological process of the physical development of an organisms after hatching/birth.

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Abstract

Information about the sources of plastic litter found in the Zeeuwse Delta is still scarce. This problem is addressed by this study with the aim to identify the possible sources of plastics that washed ashore on a selection of beaches in the Zeeuwse Delta. For this study the source and origin of the plastics is tried to be identified with the use of biofouling organisms that are attached to the beached plastic litter. In order to identify the sources based on biofouling, plastic containing biofouling was collected on the selected beaches of Vlissingen-Dishoek, Oostkapelle, Neeltje Jans and Kwade Hoek. Important parameters studied during this project were: type of species that is found as biofouling and where they originate from, type of plastic the biofouling is found on and age of the plastic based on state of degradation and percentage of coverage of the fouling organism. The result were presented in graphs and discussed based on the stated parameters and compared to the results of earlier done project by Wageningen University & Research. The main results of this study were that all identified fouling organisms were native to the North Sea, with only the *A. modestus* found as invasive species, which has been found in the North Sea since World War II. The same amount of hard and soft plastics were found with higher numbers of bryozoans on soft plastics, while barnacles were more abundant on hard plastics. Most of the plastics had a high degradation state indicating that the plastics were in the marine environment for a longer period. However most of the plastic were covered for 0-20% by fouling organisms. Based on the larval and metamorphosis stage it was found that the plastic litter had to be in the marine environment for at least a couple of months before biofouling could be observed. It was not possible to indicate the exact time since different conditions like temperature and wave/current action influence the rate of degradation of the plastic and the growth probabilities of the fouling organisms. The study concluded that since the identified fouling organisms were native to the North Sea and most plastic were presumably only in the water for a couple of months, the plastic originate from sources around the North Sea and close to the Zeeuwse Delta. Further studies to the metamorphosis and growth rates of the fouling organisms can be useful to get an exact indication for how long the fouling organisms has been on the plastic litter.

Table of contents

1. Introduction

1.1 Background

Plastic marine pollution is a worldwide problem that has been growing since the 1950's when plastic became a widely used product. The plastic waste has caused challenges and severe impacts on marine species, ecosystems and even human lives. It has become more important to identify and understand the problem behind plastic pollution, with interest on the sources and pathways of the plastic pollution with hope to tackle this devastating problem. In the last decade, several studies have been done to identify the sources of the plastic pollution. The Macroplastics in the Delta Project is new projects with the goal to find the sources of the plastic pollution in the Zeeuwse Delta, The Netherlands.

The Macroplastics in the Delta project is a project based on the Greenland Marine Litter Project and the Griend Marine Litter Project done under Wageningen University & Research. Both projects focussed on washed up marine litter on the beaches in West Greenland for the Greenland Marine Litter Project and on the beach of the Dutch Wadden island of Griend for the Griend Marine Litter Project. Both project were done to identify the sources and potential pathways of the marine pollution and how the litter eventually ends up on the shoreline.

The plastic items that were collected during both projects were used for a Litter-ID session to analyse the litter. Litter-ID is a method that is used to analyse the marine litter. A Litter-ID analysis can be used to determine the sources, origin and underlying reasons of how the litter ended up in the area. During Litter-ID sessions the litter is categorized within the 120 categories of the OSPAR Beach Litter Monitoring Guideline. After the litter is categorized in the OSPAR categories, the items are then categorized with the use of a special protocol to get a better insight in the type of plastics and the possible origins. The items are also counted and weighted to get a good insight to the ratios between the different categories (Strietman et al., 2021).

The Macroplastics in the Delta project is a research project done by Wageningen Marine Research located in Yerseke. Wageningen Marine Research is an independent research institute that does research on marine, coastal and freshwater areas, not only in the Netherlands but also in other parts of the world. Wageningen Marine Research delivers advice on different marine related issues supported by scientific research to help and contribute of sustainable management, usage and protection of the project areas. Wageningen Marine Research is part of Wageningen University & Research.

1.2 Aim of this study

The aim of the Macroplastics in the Delta project is to identify the possible sources of plastic pollution that are washed ashore on the beaches in Zeeland, The Netherlands. This is necessary to minimize the amount of marine litter found in the Zeeuwse Delta. The Zeeuwse Delta is exposed to the open North Sea and to some of the major rivers like the Haringvliet and the Schelde. Due to the open and flat space of the beaches, there is a lot of influence from the wind and waves onto the beaches. The overall project will focus on what types of plastics are found and in which amounts, where the plastics originate from, what type of plastic/packaging is collected here and how old the plastic is.

Figure 1.1 *Marine litter that has washed ashore on Kwade Hoek, The Netherlands. Photos: G. Faasse.*

The aim for this study is to identify the possible sources of marine litter based on biofouling of marine organisms such as bryozoans, barnacles and calcifying tube worms. It is currently unknown to what extent plastic litter contains biofouling and what it can tell about the origin of the plastic waste. The aim of this study is therefore to get a better insight into the biofouling of flora and fauna on plastics that are washed ashore on the Zeeuwse coast. Several important parameters in this study are studied. One of the parameters is the type of species that is found as a biofouling organism. This is important to try and identify from what area the species originate from and if this can help with the identification of the source origin of the plastic. Another parameter is the age of the plastic in relation to the state of degradation and percentage of coverage of the species on the examined plastic items. This can help to predict a global time of how long the examined plastic items were in the marine environment and to relate this back with where it came from. A last parameter that is looked into is the type of plastic biofouling is more common on. Here a difference between hard and soft plastic may indicate a different preference for the different type of fouling organism and if there is a difference in deposition of the different type of plastics.

These parameters around the biofouling can help with the identification of the potential source origin of the plastics that were collected. If the sources can be identified, measures can be done to minimize the plastic pollution and help with decreasing with the plastic problem in the Zeeuwse Delta. A side parameter is the fact that within on the observed biofouling organism, non-indigenous species can be observed. This can indicate the source origin of the plastic as well as the possible invasiveness and impacts of the observed species.

Figure 1.2 *Fouling organisms on beached marine litter (Barnacles left, bryozoans right). Photos: C. Kleijn*

1.3 Research questions

The following research question were studied in this final thesis:

1. Can biofouling (flora and fauna) on plastic items collected on a selection of beaches in the Zeeuwse delta be an indicator of source origin of the plastic?

The following sub questions were drafted to help with answering the main research question.

a. What is the total coverage percentage of fouling organisms on the plastic items?
b. How old is the collected plastic with fouling organisms?
c. Where does the plastic items originate from based on the fouling organisms?
d. On what type of plastic is biofouling more common?

1.4 Hypothesis

A hypothesis for the main research question can be based on earlier studies on biofouling of organisms on plastic in the aquatic environment in general as well as on the earlier studies on the plastic on beaches of Greenland and Griend, where the presence of fouling organisms was also assessed. During these studies multiple items with biofouling were found. DNA identification found that not all the observed fouling organisms were native to the areas (Strietman et al., 2020). However the observed non-native species have been found in these area for a much longer period of time. It is therefore expected that the identified fouling organisms found during this study are native to the North Sea and the plastic source origin is from the North Sea area.

It is expected that fouling organisms will be observed on plastic at all four project locations of Vlissingen-Dishoek, Oostkapelle, Neeltje Jans and Kwade Hoek. This is based on the fact that all project locations are exposed to the North Sea and to the dominant wind directions from the south, west and north here in the Netherlands. It is expected that more plastic will be found during or after a storm, when there is more movement towards the beaches with wind and waves.

Barnacles are expected to be the most abundant species to be found as biofouling, based on the preference of these species for hard substrates. Harder and bigger plastic items are more likely to be deposited on the beaches whereas smaller plastic will be easier carried back to the sea or are blown away (Weslawski, 2018). Bryozoa are also expected to be found as biofouling on hard and soft plastics since they grow on hard and softer substrates in the natural environment (Ernst et al, 2011).

New non-native biofouling organisms are expected to be found in smaller numbers. This is because most of the fouling organisms are expected to be either native to the North Sea or have already settled here for a much longer period of time. The identification on non-native species may be hard to pinpoint based on visual inspections alone. All identification within this project will be done using own knowledge or expert opinions. Additional DNA analysis will be conducted after the graduation period.

1.5 Overview of the paper

Chapter two will give information on the marine plastic pollution and the fouling of aquatic organisms on plastic. In Chapter three the methods used during this study are described for the collection of the plastic waste, the analysis and the sampling of the plastic litter containing fouling organisms. In Chapter 4 the results of the study and the conducted Litter-ID session are presented. Chapter 5 reflects upon the results of the study and the Litter-ID session with comparison to the Greenland and Griend Marine Litter Projects. In Chapter six and seven the main conclusions are summarized and recommendations are given for future research.

2. Theoretical framework

2.1 Marine plastic pollution

Plastic is a widely used material in the modern world and industry. Over 380 million metric tons of plastics is produced every year, of which over 50 percent is single-use products like plastic bags and packaging. Only 9 percent of the produced plastics is recycled after use, while the other part is either incinerated, stored or ends up in the environment (POI, 2021). It is estimated that up to 12 million metric tons of plastic end up in our oceans and seas every year. Over 80 percent of the annual plastics input comes from the movements of plastic pollution from land into the marine environment caused by wind, rain or via the rivers, while the remaining part comes from plastics released at sea due to the fishing activities (Jambeck et al., 2015; Sherrington, 2016).

Of the plastic that enters the ocean, 94 percent eventually ends up on the seabed, while 5 percent of the plastic washes ashore on beaches all over the world. The remaining 1 percent is the plastic that actually floats in the ocean surface. In comparison to the huge amount of plastics on the seafloor, the amount of plastic that ends up on the beaches seems minor but looking at the total amount of plastic that ends up in the oceans every year, it still accounts for 0.6 million metric tons of plastic that eventually washes ashore (Sherrington, 2016). The worldwide plastic pollution in our oceans is now a recognized problem due to the detrimental impacts on the marine biodiversity and ecosystems. Some of the problems that the plastic pollution gives to organisms is the ingestion and entanglement, as the plastic is mistaken for food. The ingestion and entanglement often leads to marine animals dying from the injuries, bioaccumulation and stress (Gregory, 2009).

Figure 2.1 *Entangled sea turtle in a fishnet; Dead albatross following the ingestion of plastics. Photos: 1. J. Chais; 2. C. Jordan*

Over 100 million marine animals die each year from the complication of plastic waste. It is predicted that by the year 2050 there will be more plastic than fish in our oceans and that the numbers of dead marine animals will grow significantly with an increased chance of extinct animals as result (Rhodes, 2018).

Most of the plastic are persistent and buoyant. It is easily transported by the wind and ocean currents and can end up in even the remotest regions of the planet (Barnes et al, 2010). The concentration of plastic debris in the ocean's surface ranges from 0 to 150 items per hectare as where it is unknown where they eventually end up (Katsanevakis & Katsarou, 2004; Barnes & Milner, 2005). Therefore, there is still a gap in knowledge for the actual movements and the buoyant factor of the marine litter and where it eventually ends up. With the fast-growing development of the technology sector, more options become available for the modelling and predictions of the transport of marine litter in the oceans. This is especially of importance for the lack of knowledge about the transport and behaviour of plastics when they end up in the marine environment. With the help of models, the sources of the plastic pollution and movements of the plastic waste can be identified, giving the option to more and better solution to the problem (Kaandorp et al., 2020).

2.2 Biofouling on plastic

Another problem that comes with the plastic pollution of the oceans is the fact that plastic is a new transport vector for fouling organisms. For millions of years natural debris has floated and dispersed across the open oceans. The floating natural debris have been known to transport flora and fauna all over the oceans with records dating back to the medieval times (Van Duzer, 2004; Gregory, 2009). The attachment of organisms onto natural or artificial substrates is called biofouling.

Plastic is highly persistent due to the low biodegradability in the natural environment. Several studies have shown that marine litter doubles the opportunity for flora and fauna to travel along. This is caused by the hydrophobic layer on the plastics that differs much from natural substrates (Barnes, 2002; Garcia-Vazquez et al., 2018). The floating plastic make an excellent raft for attaching and colonization of aquatic organisms like barnacles, mollusc or algae (Engler 2012; Kiessling et al., 2015; Weslawski et al., 2018). The increases in plastic concentrations in the oceans over the last decades have augmented the available surface and habitat for flora and fauna to attach to. Some of the found fouling organisms can be identified as non-indigenous species. Non-indigenous species are species that are introduced outside their natural habitat. With the use of the plastic non-indigenous species can travel to all parts of the world. This gives the non-indigenous species the chance to establish in habitats unlike their own. If the non-indigenous species can establish in the new habitat successful, they can impact and damage the current ecosystems and can become an invasive species (Hellman et al., 2008; Garcia-Vazquez et al., 2018).

The first report of invasive biofouling was on the invasion of the bryozoan, *Membranipora tuberculate*, in the waters of New Zealand (Gregory, 1978). During the study it was found that the *M. tuberculate* travelled along on plastic substrates and successfully invaded the environment (Gregory, 2004). Since this report the subject of plastic and fouling organisms became more interesting and more studies followed on the biofouling on plastic and the distribution of invasive flora and fauna in combination with marine litter. The importance of plastic as transportation vector of flora and fauna was reported with research done by Barnes (2002) and Barnes & Milner (2005). Observations were done over a latitude from the North Atlantic to the South Atlantic while exposed island along the line were examined for beached marine litter. The colonization on the marine litter was different depending on the water temperature and conditions of the period (Barnes & Milner, 2005). Both Weslawski & Kotwicki (2018) and Garcia-Vazquez (2018) found that especially hard plastics were inhabited by flora and fauna, with a higher abundance of barnacles, molluscs and algae. Until this day there is still a lot unknown about biofouling on plastic.

3. Methodology

3.1 Locations

For the collection of plastic with fouling a total of four beaches across the province of Zeeland, The Netherlands have been selected. The beaches are selected on different parameters like position, dominant wind direction, presence of tourism and influences from the outflowing rivers. The different influences on the four beaches can give a better insight in the effects on the amount of plastics that are found on the beaches and may also highlight a difference in the amount and found biofouling. A map with all four project locations have been highlighted in figure 3.1 below.

Figure 3.1 *Locations of the beaches where plastic was collected*
Illustration: Wageningen University & Research; map: Google Maps.

The different influences on the four beaches can give a better insight in the effects on the amount of plastics that are found, the different influences are visible in the table below.

Beach	Important wind direction	Presence of river	Presence of tourists	Other	Number of visits
Vlissingen-Dishoek	South/west	X (Westerschelde)	X		6
Oostkapelle	South-west-north				5
Neeltje Jans	South-west-north			Storm surge barrier	6
Kwade Hoek	West-north	X (Haringvliet)			4

Table 1. Overview of project location with different influencing parameters.

3.1.1 Kwade Hoek

Kwade Hoek is a beach located in the north-western part of the delta on the island of Goeree-Overflakkee. The dominant wind directions that influence Kwade Hoek are winds from the west and north. The beach has presence from the Haringvliet which flows out in the North Sea and a lot of ship trafficking in the area due to the harbour of the Maasvlakte. There is also a lot of dredging activity in the area. The beach is mostly protected for the breeding birds that use the nature area and is not popular among tourists. The main and only activity on the beach is from hiking. There is a lot of activity of the volunteers of beach clean-up group NLGO, since it is one of the most polluted beaches in the delta. They have been very helpful in the gathering of plastic with biofouling for this study.

There is a lot of deposited seaweeds and seashells on the tide line, where a lot of smaller parts of plastic like wires are entangled with. The beach is wide with the dunes and hinterland starting about 100 meters back. With spring tide the water reaches all the way to the back of the hinterland.

figure 3.2 *Beach Kwade Hoek*
Photos: C. Kleijn

3.1.2 Neeltje Jans

Neeltje Jans is a beach that was raised when the Oosterscheldekering, a storm surge barrier, was built as part of the Delta works. The beach is under influence from winds that come from the north, south and west. With the storm surge barriers located on each side of the beach, there is a special current going past the beach with in- and outgoing tide. The beach is a popular destination for tourism in the summer months due to the great open space for wind surfing and swimming. There is less activity for beach clean-ups but there are plans to make it a green beach.

The beach has a rather flat profile and is close to the dunes. There are less deposited sea weeds but more seashells which may indicate that lighter items are taken away more easily. The beach lays between two groynes, creating deposit hotspots in the corners of the beach as can be seen in figure 3.3.

Figure 3.3 *Beach Neeltje Jans*
Photos: C. Kleijn

3.1.3 Oostkapelle

Oostkapelle is a beach located at the west side of the island Walcheren. It is the only beach among the four project beaches that is only under the influence of the North Sea. This means the only flow of plastic towards the beach is from waves, currents and wind. The dominant wind directions are north, south and west. Oostkapelle is a popular beach for tourism in the summer months just like Neeltje Jans, with only people going for a walk along the beach during the monitoring period. Towards the summer months beach houses are placed along the dune line of the beach, removing a big part of the area that is searched for plastic waste that ends up along the dunes caused by the wind. The area borders to an existing OSPAR beach monitored by Stichting de Noordzee. The beach has rows of pole heads to break the waves. The beach is known for the strong currents with outgoing tide. This may lead to lower amounts of plastic between the tide line and the water line.

The beach has small sand banks between the rows of pole heads. This creates a small dip where after the beach slowly hells before the dune line begins. There is a lot of deposition of seashells with some smaller chunks of sea weeds.

Figure 3.4 *Beach Oostkapelle*
Photos: 1. C. Kleijn; 2, B. Oerlemans

3.1.4 Vlissingen-Dishoek

Vlissingen-Dishoek is the beach next to the Boulevard beach of Vlissingen. The beach is directly influenced by winds from the south and west. Especially with strong storm winds from the south a lot of wave action happens towards the beach. This creates a thick line of foam from the movements of the water and waves. The stronger winds also leads to the increased deposition of natural debris like reeds and sea weeds. The natural debris is collected once a year and gets deposited along the dune line so that the natural debris can decompose. This leaves behind a lot of smaller plastics. The beach is divided in sections with the use of pole heads to decrease the wave and currents towards the beach. The beach quickly transitions into the dunes. There is a lot of tourism throughout the year, with an increase in activity in the summer months. The beach has influence of the outflowing Western Scheldt, which is the main shipping routes to the harbour of Terneuzen, Antwerp and Ghent.

The beach is flat and narrow. There is a lot of deposition of natural debris especially in storm periods (figure 3.5). The pole heads lead to less wave and current action. The high amount of tourism activity on the beach can lead to a higher amount waste that has not arrived from the sea but left behind by people.

Figure 3.5 *Beach Vlissingen-Dishoek*
Photos: C. Kleijn

3.2 Beach visits

For the collection of data, the beaches of Vlissingen-Dishoek and Neeltje Jans were visited for a total of six times while Oostkapelle was visited five times and Kwade Hoek four times. The data that was collected led to an overview on the amount of plastic items with fouling found during the project period. This can be related back to the certain condition like a storm or spring tide that can occur during the period. Potential differences between beach locations can give a better understanding on the influence of different conditions on the amount plastic waste in general.

The beaches were monitored and plastic was collected by walking along the tide line and the dune line. The tide line was expected to be the best spot to find plastic with fouling, since this spot has the newest plastics being washed ashore with the incoming tides. All plastics were collected and plastics with fouling organisms were kept separate in zip lock bags. All other plastics were collected and used in another part of the Macroplastics in the Delta project, where the focus was on finding the origin of plastics based on the use of labels, text and language (Oerlemans, 2021).

3.3 Collection and identification of fouling

Plastic with biofouling was collected separately from the other plastics. Back home the fouling organisms were photographed for identification purposes. A photograph was taken of both the entire plastic item as well as a close-up photo of the all the fouling organisms. This was done so that experts could examine the photos and help with the identification of the fouling. To prevent contamination between the different items with biofouling, each item was placed in separate sealable bags during collection. Some of the items with biofouling from Kwade Hoek were additionally collected by volunteers of beach clean-up group NLGO. These items were stored in a freezer to preserve the fouling organisms for possible DNA identification before they were picked up and analysed as described above.

After the beach monitoring, the items were examined more closely to identify what kind of species was found as biofouling, whether the biofouling was native or invasive, on what type of plastic the biofouling was found, what the total coverage percentage of biofouling was and how old the plastic was approximately. After this examination, the biofouling was sampled, preserved in >97% Ethanol and stored for further DNA analysis and identification in the laboratory. The results of this additional DNA analysis is beyond the scope of this thesis. The samples were marked with information such as what beach it originated from, on what date the sample was found and what presumed species were sampled. The marking was done with a pencil to prevent the text from fading out by possible leakage of the 97% Ethanol.

The sampled biofouling was photographed next to the plastic items and were documented in a file to have a clear overview of the samples and the respective item it was found on (see Appendix X?). All samples were placed straight in a cool box to preserve the samples and prevent the Ethanol to evaporate.

Figure 3.6 *Fouling sample together with the sampled plastic item*
Photos: C. Kleijn.

The origin of the fouling organisms was tried to be identified based on the photos that were taken of all species. The photos are shown in the accompanying file (filename Photos_Biofouling_Corne) The photos were examined by taxonomy experts Emiel Brummelhuis (Wageningen Marine Research), Douwe van de Ende (Wageningen Marine Research), Reindert Nijland (Wageningen University) and Marco Faasse (Acteon Marine Biology Consultancy). This further identification of species was needed to assess where the organisms originated from and if this information could be related to the origin of the plastic. The identification of possible non-indigenous species could additionally be applied to assess possible invasiveness of the species at the locations.

After identification of species, additional information of the identified species was collected from the literature to obtain information on settling speed and growth of these fouling organisms. This was needed to get some insight into roughly the time needed for the organism to become visible as fouling on the marine litter. This can be used as additional parameter as minimum time that the item was present in the water.

3.4 Data analysis

For the collected plastic items with fouling organisms, the quantity, type of material, type of fouling, state of degradation (age), total coverage, species origin and possible invasiveness was determined. This was done by the examination of the plastic items, fouling organisms and expert opinions.

3.4.1 Quantity

The quantity or amount of plastic items with biofouling were analysed per beach. The amount of items was compared to the other beaches in a graph. This was done to get an insight on the difference in amount of plastic with fouling on all four monitored beaches. The differences were related back to the location of the beach, the influence of outflowing rivers, impact of tourism and the dominant wind directions as well as storms or spring tide that happened during the monitoring period.

3.4.2 Type of material

For the type of materials, a difference was made between hard and soft plastics. Also, other materials were taken into account to show potential differences or comparisons of substrate on which fouling was found. The type of materials was analysed with the fouling organisms that were found during the monitoring period. This provided information on what type of material was more common to have biofouling and if organisms prefer certain type(s) of material. This was compared with other studies that were done to fouling on plastic.

3.4.3 Type of fouling

Also, the type of biofouling was further analysed. The species were determined and as mentioned in the section above, it was put against the type of plastic. The amount of times the fouling organism is found has also been highlighted to see what type of organism is more common on what type of plastic.

3.4.4 State of degradation

The plastic items were also categorized in states of degradation. This was done to possibly determine the age of the plastic. The exact degradation times cannot be assessed, but the applied method can give some information on how long the plastic items were in the marine environment before washing ashore on the beaches. The state of degradation was determined to decolouration, cracks and marks. The following categories were used to determine the state of degradation; Heavily damaged, damaged, slightly damaged, almost no damage. This method is based on the method used during a cap analysis study of Jan Mayen and Svalbard (Schravesande, 2019).

3.4.5 Total coverage

In relation to the determination of the age of the plastic items, the total coverage of the fouling organism was determined. A larger extent of coverage pointing at a longer time being present in the marine environment. The total coverage was determined by taking the whole item as 100% and making a rough estimate of the total coverage. The following categories were used for the coverage; 0-20%, 20-40%, 40-60%, 60-80%, 80-100%.

3.5 Additional activities

During the graduation period multiple additional activities have been done to get a better understanding in the world of plastic marine litter.

3.5.1 Litter ID Kwade Hoek

One of the additional activities was the Litter ID session on the collected marine litter in Kwade Hoek. A litter ID session is an analysis of beach litter in an interactive way developed by Wageningen University & Research. The goal of the litter ID session in Kwade Hoek was to try and point out the sources, origin and if possible the solutions. A litter ID session was also done for the Griend and Greenland Marine Litter project.

Since June 2020 beach litter was collected with the help of beach clean-up group NLGO. For the litter ID session 30 kilograms was selected for every month from June 2020 until March 2021 from the total of 3500 kilograms of beach litter that has been collected over this period. For the litter ID session, the months were put together into quarters to get a better view on the influence of the seasons. Due to the Covid-19 restrictions there were no interactions with stakeholders and only a small group worked on the session. This led to the fact that only two quarters were completed in the three day period. The two quarters were Q1 – January, February, March and Q2 – July, August, September.

The items of these quarters were sorted, counted, weighted and put under the categories of OSPAR. This is an international method for the categorisation of marine plastic waste. During the litter ID session plastic with biofouling organisms were photographed and documented. No intensive work was done for finding items with biofouling. The results on biofouling will be worked out in the results, the other results of the litter ID session are not allowed to be shared and will therefore not be included.

Figure 3.7 *Litter ID session Kwade Hoek*
Photos: M. Boonstra

3.5.2 OSPAR screening Veere

Stichting de Noordzee runs an initiative throughout the Netherlands to monitor the changes on the amounts of marine litter that end up on the beaches. This is done with the use of the OSPAR Beach Litter Monitoring Guidelines. For the screening of Veere a 100 meters wide perimeter is set. The beach of Veere borders to the beach of Oostkapelle, which is one of the project beaches. The perimeter is walked back and forward beginning on the dune line and ending near the water line. All marine litter that is found in the perimeter is collected and categorized in one of the 120 OSPAR categories. All the items with biofouling were photographed and documented. The results are worked out and compared to the data from Oostkapelle.

4. Results

4.1 Results

First, the results of the fouling analysis of this study will be discussed in first subchapter. The following subchapter will focus on the age of the plastic with the results of the state of degradation and coverage of the fouling organism. All raw data is presented in appendix A.

4.1.1 Fouling analysis

The total amount, including plastics without fouling organisms, is visualized in figure 4.1. On the beach of Vlissingen-Dishoek the highest number of plastic items was found with a total of 1004 while only 244 items were found on Kwade Hoek. It has to be noted that Kwade Hoek was visited four times in comparison to the six times for Vlissingen.

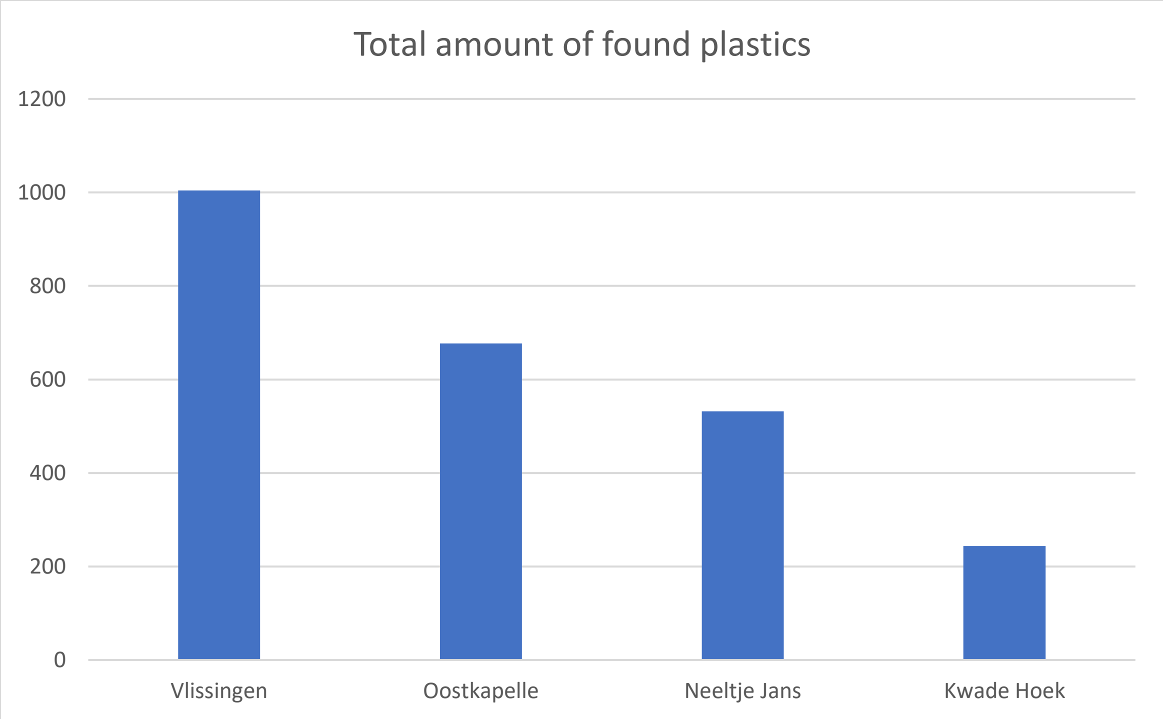


Figure 4.1 Total number of plastics found per beach during the project, including items without fouling organisms. Number of visits per beach were: Vlissingen 6 visits, Oostkapelle 5 visits, Neeltje Jans 6 visits, Kwade Hoek 4 visits.

In total 73 plastic items with biofouling were collected during the monitoring process. Of these, 53 were collected on the beach of Kwade Hoek (73%), 13 were collected on Oostkapelle, 6 on Neeltje Jans and 2 on Vlissingen (figure 4.2). A total of seven items were collected on the beach of Kwade Hoek by volunteers of beach clean-up group NLGO.

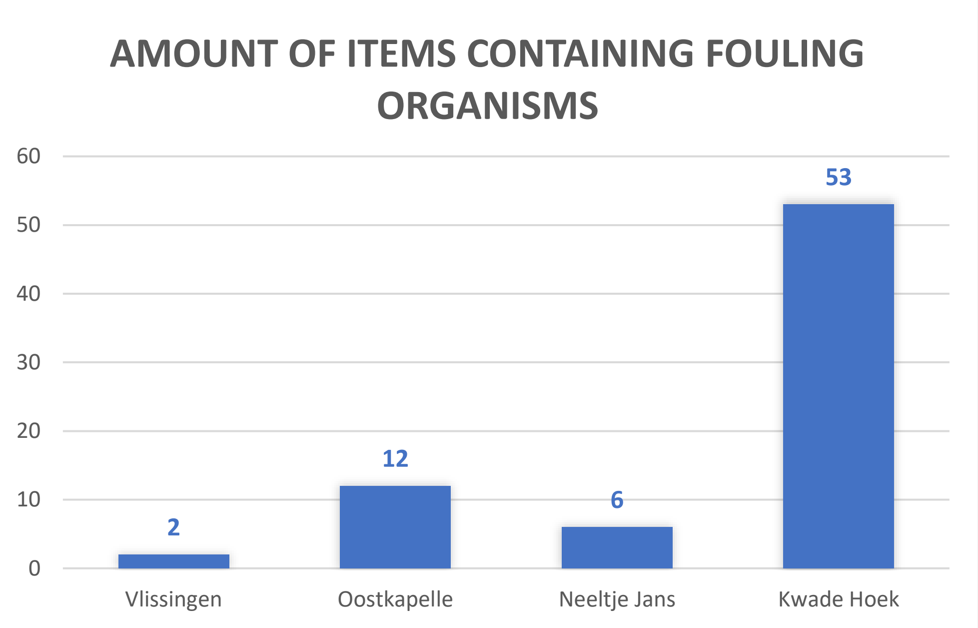


Figure 4.2 Number of plastic items containing biofouling that were found on the studied beaches in Zeeland (February-May 2021). Number of visits per beach were: Vlissingen 6 visits, Oostkapelle 5 visits, Neeltje Jans 6 visits, Kwade Hoek 4 visits.

In total 110 fouling organisms were found on the 73 plastic items (figure 4.3). This is because some of the items contained multiple types of fouling organisms. The observed fouling species consisted of bryozoans (Bryozoa), barnacles (Cirripedia), pipe hydroids (Cnidaria), calcifying worms (Annelida), and anemones (Cnidaria). Bryozoans were the most common fouling species found with 62 observations. As for the barnacles 34 observations were noted in total on the items. The other fouling organisms were found in smaller numbers.

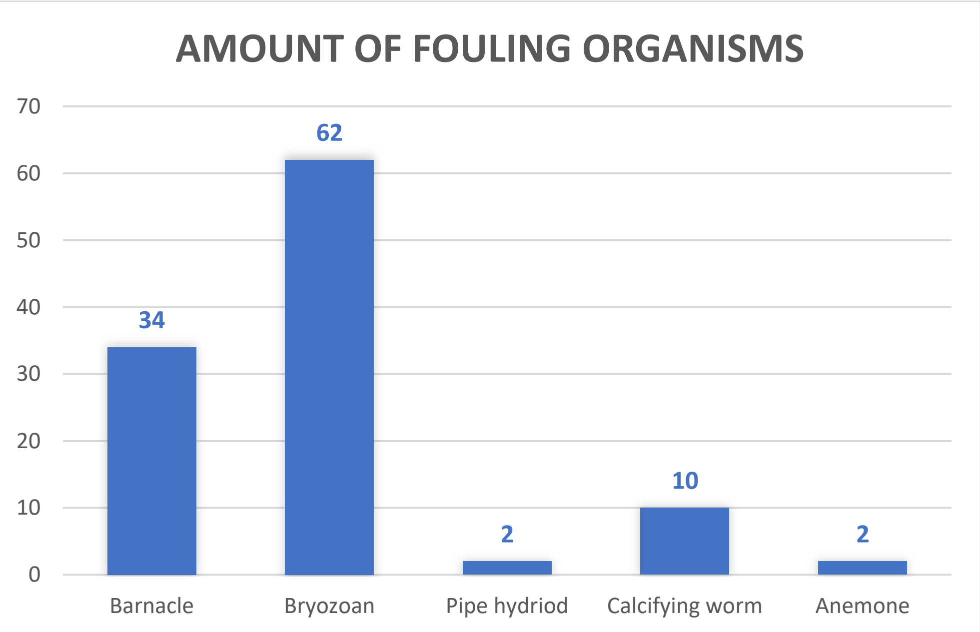


Figure 4.3 Number of fouling organisms observed on plastic items that were found on the studied beaches in Zeeland (February-May 2021).

The following biofouling organisms were identified by experts with the use of photo documentation.

Figure 4.4 Photos of the observed and identified biofouling organisms: upper left – calcifying worm (Spirobranchus triqueter), upper right - bryozoans (Conopeum reticulum), middle left - barnacles (Austrominius modestus), middle right – barnacles (Semibalanus balanoides) lower left - anemone (Metridium dianthus), lower right - pipe hydroids (Ectopleura larynx).

The biofouling distribution between the four locations was very different (figure 4.5). Most of the fouling organisms were registered at Kwade Hoek, with the highest amount of observed number of bryozoans and the only place where pipe hydroids and anemones were found. At the other three locations bryozoans, barnacles and calcifying worms were observed.

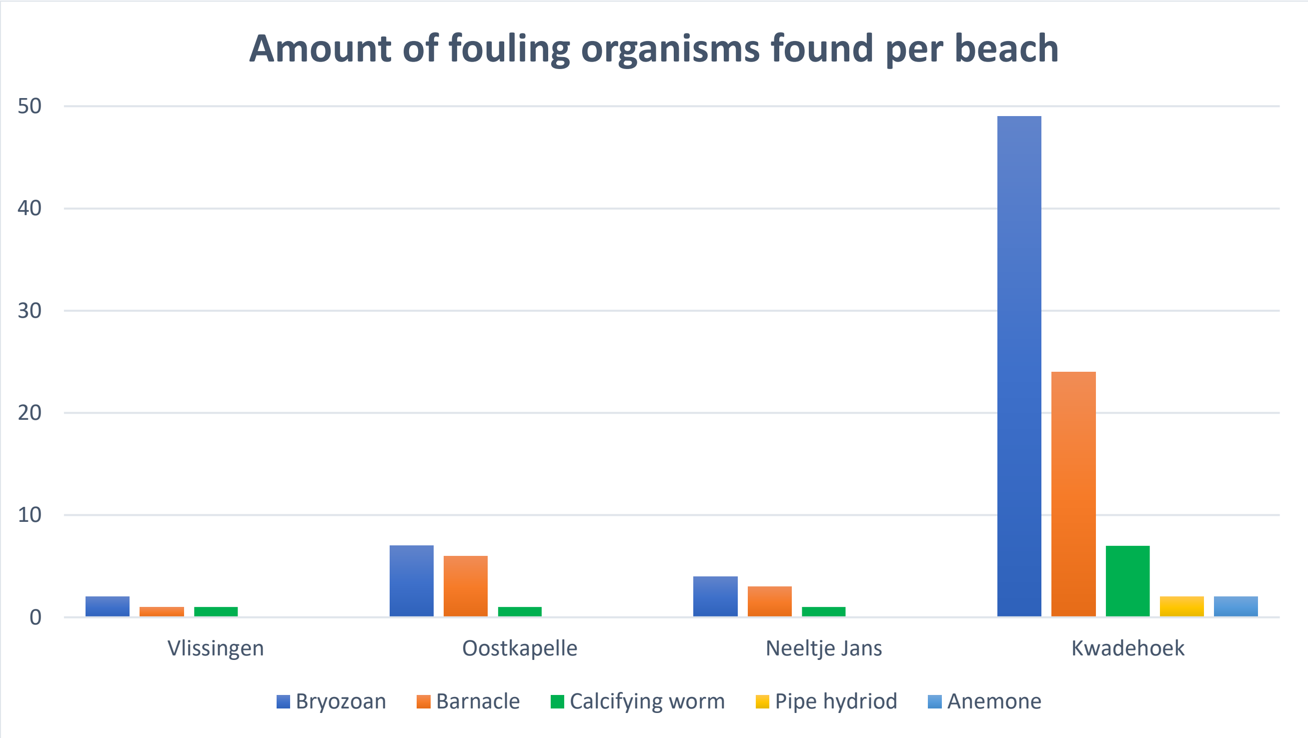


Figure 4.5 Number of types of fouling organisms observed on plastic items that were found on the four studied beaches in Zeeland.

As for the type of plastic containing biofouling that was found on the four beaches, an equal amount of hard and soft plastics was observed (figure 4.6). Other materials, textiles and metal cans, with fouling organisms have also been included since they also form part of the OSPAR Beach Litter Monitoring Guidelines.

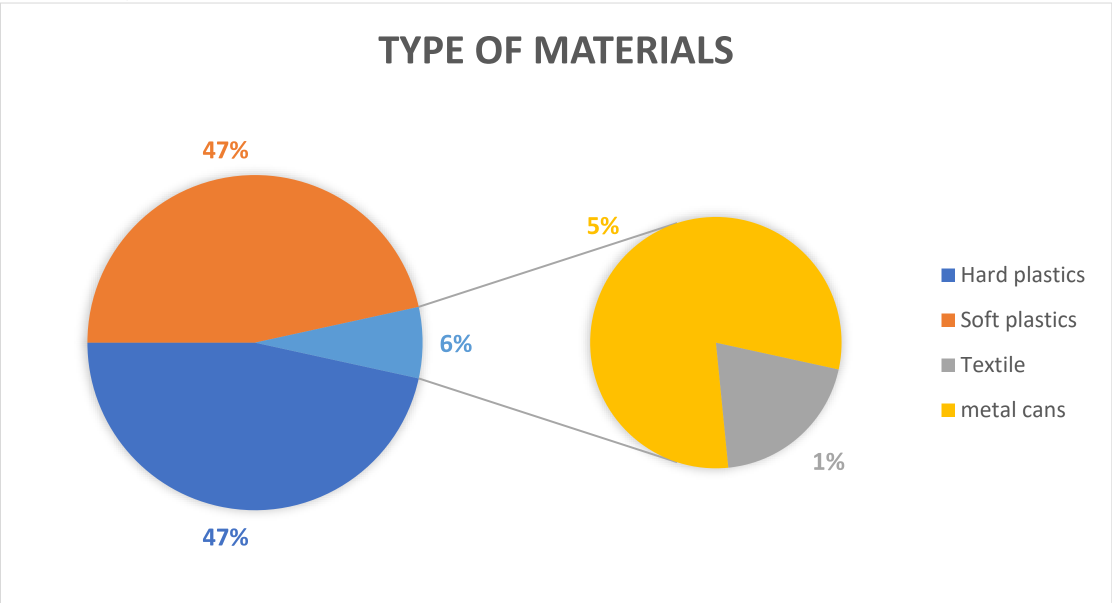


Figure 4.6 Type of materials (with 6% non-plastic) of the collected items containing biofouling during the monitoring of the project locations. Number of visits per beach were: Vlissingen 6 visits, Oostkapelle 5 visits, Neeltje Jans 6 visits, Kwade Hoek 4 visits.

A distinct difference in type of materials containing fouling was observed on each of the four beaches (figure 9.9 appendix B). On Kwade Hoek most of the plastic items that contained biofouling consisted of soft plastics. Also fouling on other materials, such as textile and metal cans, was observed. The plastic items found on Vlissingen-Dishoek and Oostkapelle were all hard plastics.

The type of fouling was different on each of the materials (figure 4.7). Bryozoans were most common on soft plastic, while barnacles were most often found on hard plastics (25) and less on soft plastic (20). With regards to the soft plastic, the barnacles were more abundant on industrial sheeting and not on the fragile softer plastics. Hard plastics contained a similar amount of bryozoans and barnacles, and had the largest share of calcifying worms. The hard plastics was also the only material where anemones were found on as biofouling. On the textile only barnacles were found, while all four metal cans contained both bryozoans and barnacles.

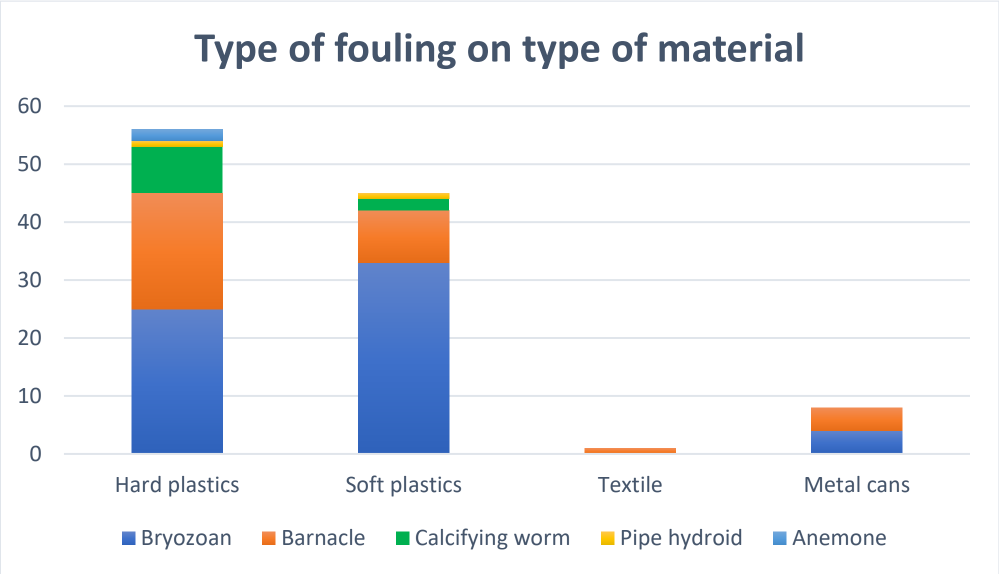


Figure 4.7 The type of fouling that was found on each of the found materials.

4.1.2 Age of the plastics

The age of plastic was assessed using of the state of degradation of the plastics and the total coverage of the fouling organisms on the plastic items. Almost 50% of the plastic items with fouling was heavily damaged (figure 4.8). Plastic items with fouling that had almost none to no damage was found the least. Since most of the plastic, 53 of the 73 items, were found on Kwade Hoek, a separate graph was also made for this particular location on the state of degradation of these plastic items (see figure 9.10 appendix B). 62% of the plastic items found on Kwade Hoek was classified as heavily damaged, while only 2% had almost none to no damage. The slightly damaged and damaged were close to the same in both graphs.

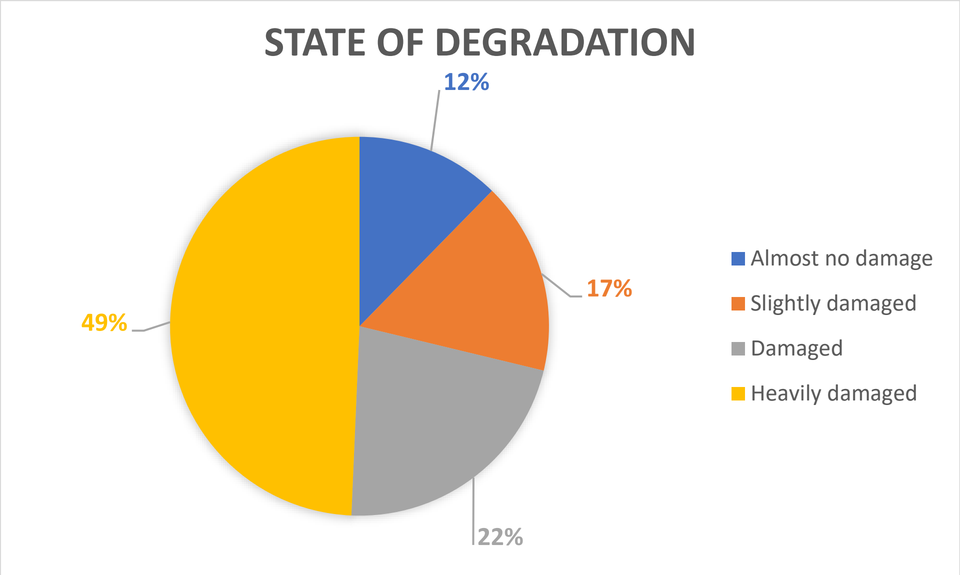


Figure 4.8 State of degradation of the plastic items.

The coverage of the fouling organisms is important to get an indication on how long the plastic items were in the marine environment. If an item contained multiple fouling organisms the coverage was summed into one category. Over half of the items contained only 0-20% biofouling coverage. With the increasing percentage of coverage, the amount of plastics gets less with only 1% of the items being covered for 80-100%.

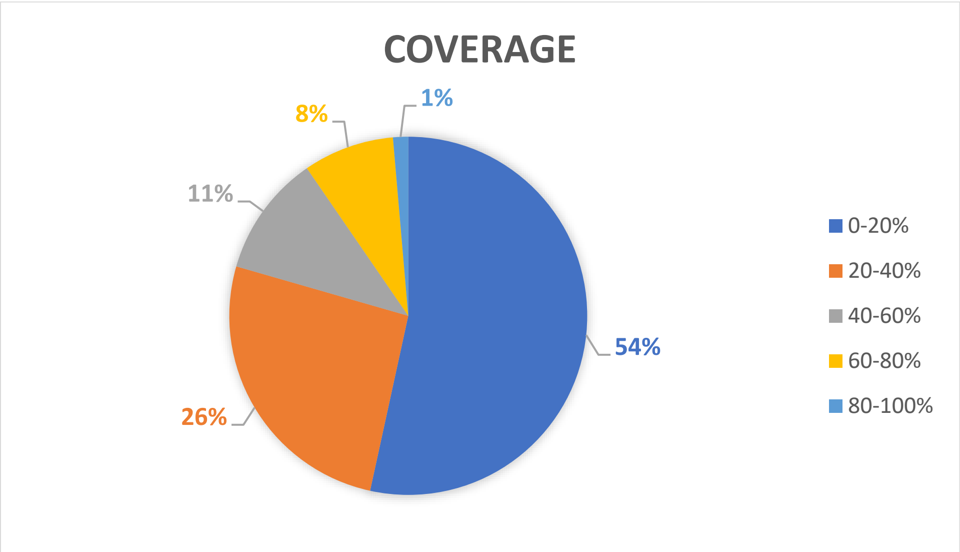


Figure 4.9 Coverage in % of the fouling organisms.

For the coverage, a separate graph has been made for the plastic item that were found on Kwade Hoek, just like done with the state of degradation (see figure 9.11 appendix B). On this beach, 55% of the plastic were covered for 0-20%. Also, all the items categorized in the 60-80% and 80-100% categories were found on Kwade Hoek.

4.2 Results litter ID session Kwade Hoek

During the litter ID session on Kwade Hoek more than 3100 plastic items were sorted and categorized. In total 29 litter items with fouling organisms were found during the litter ID session on Kwade Hoek (figure 4.10). 62% of the litter items with fouling organisms consisted of soft plastic and 28% of hard plastics. The remaining materials were 3% textile and 7% rubber.

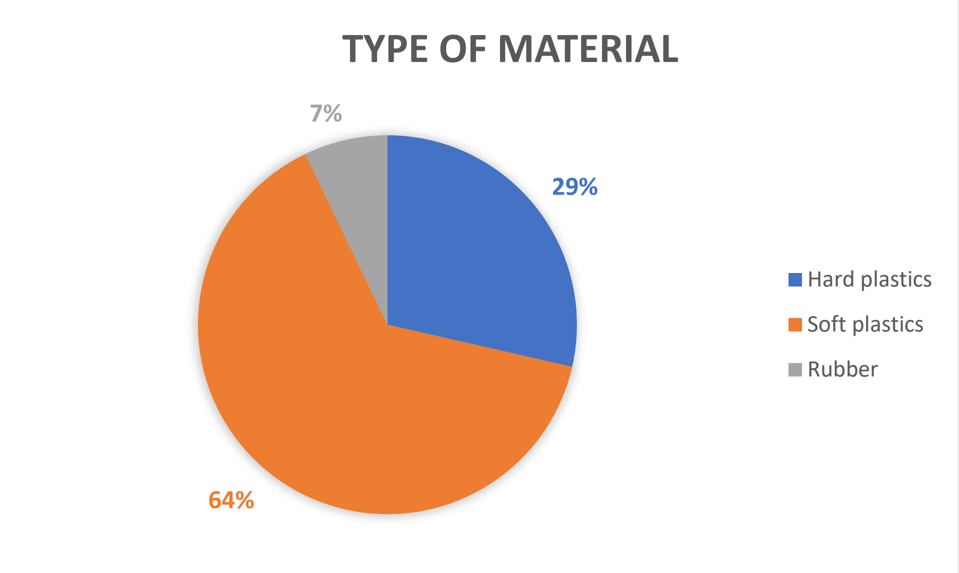


Figure 4.10 Type of materials of the beach marine items from the litter ID session of Kwade Hoek, April 2021.

Of the total 38 fouling organisms that were found on the litter items, 22 were bryozoans and 8 items with barnacles were found (figure 4.11). There was also a low amount of calcifying worms, pipe hydroids and netted dog whelk eggs found.

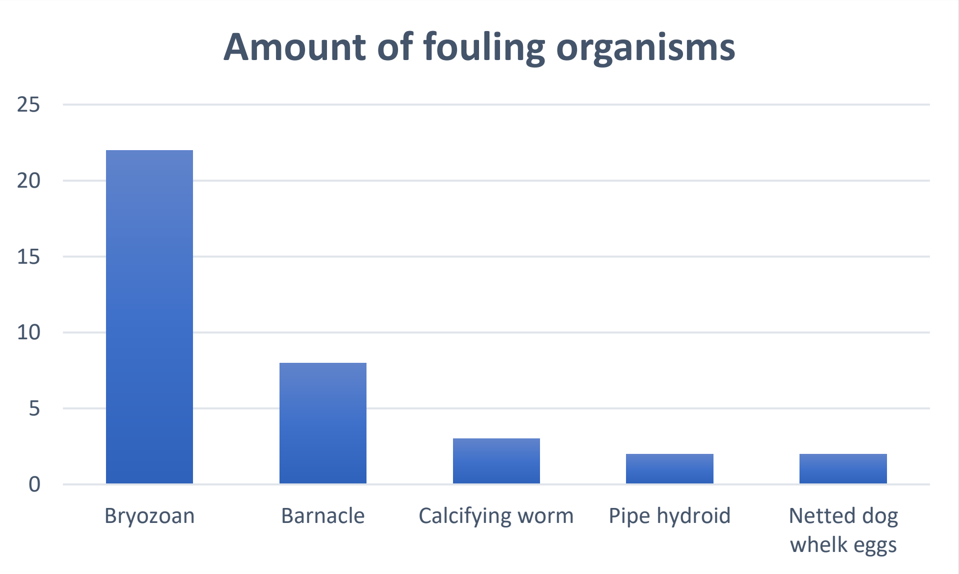
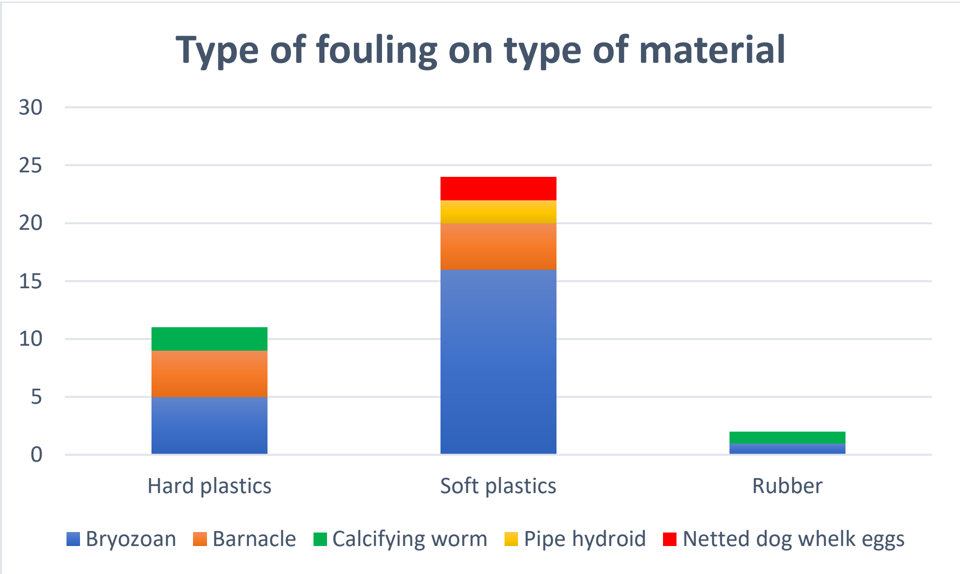


Figure 4.11 Amount of the fouling organisms found on beach litter items during the litter ID session of Kwade Hoek, April 2021.

With the use of photographs the species were tried to be identified. For the Litter ID the bryozoan Conopeum reticulum was identified. For the barnacles both the *Austrominius modestus* and the *Semibalanus balanoides* were identified. The calcifying worms were of the *Spirobranchus triqueter* and the hydroid was the *Ectopleura larynx.* These species were the same as the found organisms during the monitoring (figure 4.4.). However, during the Litter ID session netted dog whelk eggs (*Tritia sp.)* were found (figure 4.12).

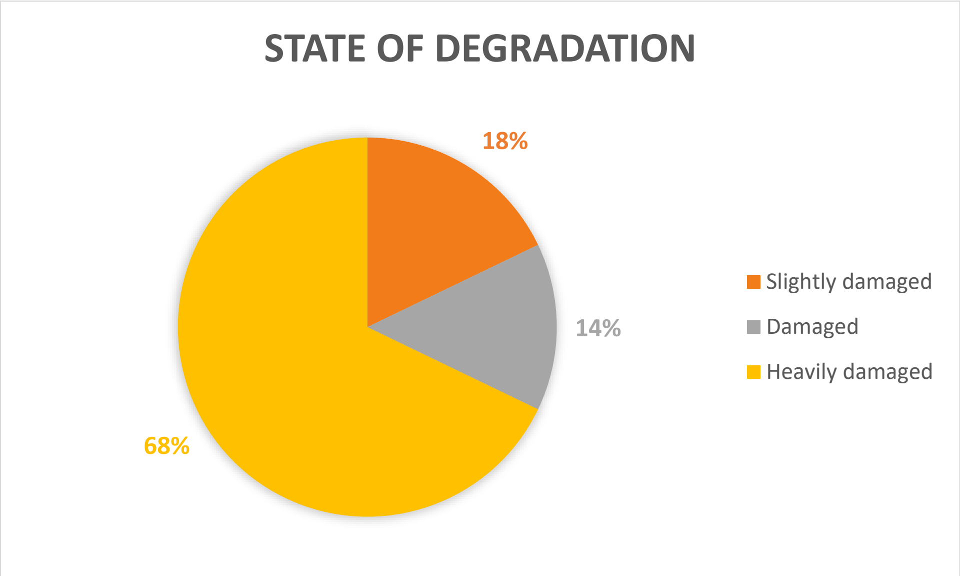
Figure 4.12 photo of the netted dog whelk eggs (Tritia sp.) during the Litter ID session of Kwade Hoek, April 2021.

The type of fouling was different on each of the materials (figure 4.13). Bryozoans were more common on soft plastic while barnacles were found equal on soft and hard plastics. The soft plastic also contained two colonies of hydroids. A rare sighting were the eggs of the Netted dog whelk that were present on two pieces of soft plastic items. The hard plastics contained more bryozoans than barnacles. The hard plastics and rubber contained the calcifying worms.



4.13 The type of fouling that was found on each of the found materials during the litter ID session.

68% of the litter items were heavily damaged with only 14% damaged and 18% slightly damaged. In comparison to the state of degradation on Kwade Hoek during the monitoring (figure 4.8), most items were heavily damaged with around the same amount of damaged and slightly damaged items as observed before.



4.14 State of degradation of the plastic items found during the litter ID session.

Half of the items were covered for 0-20% by fouling organisms while 25% was covered for 20-40% (figure 4.15). Only one item was covered for 80-100%. Figure 4.15 is comparable to figure 4.9 which showed that most items were covered for 0-40% and only a few items were covered for 40-100%.

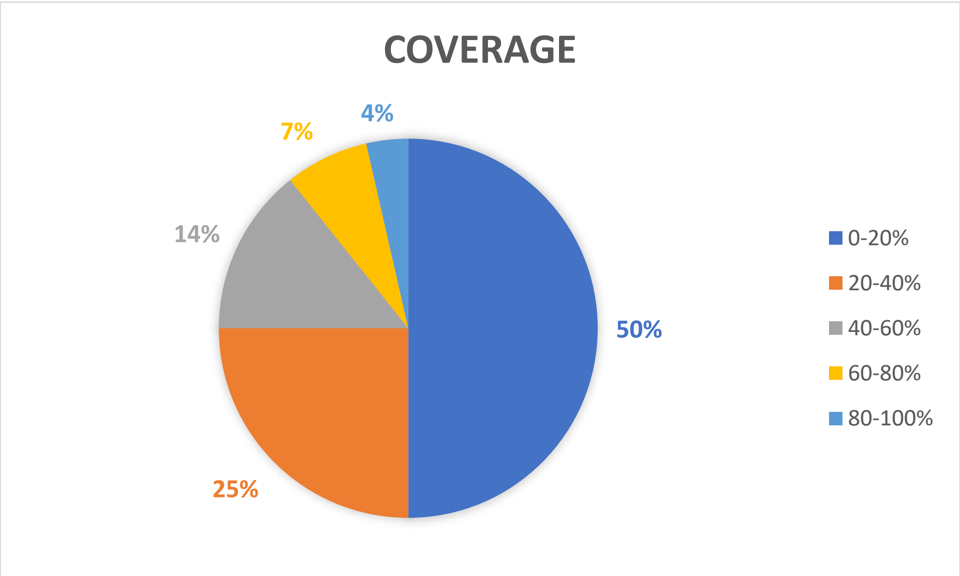


Figure 4.15 Coverage in % of the fouling organisms on the items found during the litter ID session.

4.3 Results OSPAR screening

During the OSPAR screening on beach Veere only three items were found that contained biofouling. The items were found close to the dune side and not on the water or tide line. All three items were different materials with one soft and one hard plastic item and a metal can (table 4.16). The soft plastic contained bryozoans while the hard plastic contained barnacles. The metal can contained both bryozoans and barnacles. All three items were heavily damaged and were only covered for 0-40%.

Material	Fouling organism	State of degradation	Coverage
Soft plastic	Bryozoa	Heavily damaged	20-40%
Hard plastic	Barnacle	Heavily damaged	0-20%
metal can	Bryozoa,Barnacle	Heavily damaged	20-40%

Table 4.16 Table with the items and information found during the OSPAR screening of beach Veere.

5. Discussion

5.1 Observed number of plastics with biofouling

It was expected that there would be plastic containing biofouling found on all of the four project locations. Most of the items that contained fouling organisms were found on Kwade Hoek while the least amount of items was found on Vlissingen with only 2 items. The high amount of plastic items on Kwade Hoek can be related to multiple causes. Kwade Hoek is located near the Maasvlakte 2 so there is a lot of ship traffic in the area. There is also a ship anchor point of the coast. For the shipping routes there is a lot of dredging activities in the area to keep the shipping channel deep enough for the ships to pass through. Most of the items that were found in Kwade Hoek were heavily damaged and older. This may indicate that there is a lot of plastic in the sediments around the beach of Kwade Hoek and are released due to the high dredging activity in the area or that the plastics are longer in the marine environment. Resulting in a longer period for fouling organisms to attach and grow on the plastics.

The beach of Kwade Hoek is also more open to the open North Sea where a lot of the currents and wind influence the area. The beach is also located closer to England than the other locations which may indicate that there is more plastic coming from England and end up on the beach. The influence of the river Haringvliet can also be a possibility for the higher amount of plastic items found, since a big part of the plastics in the marine environment can come from the rivers (Lebreton et al., 2017).

The fact that less plastic with biofouling was found on Vlissingen-Dishoek, Oostkapelle and Neeltje Jans cannot be related back to the total amount of plastic items that was found on the beach, since most plastics were found on Vlissingen and the least on Kwade Hoek (see figure 4.1). Only 0,2% of the plastic item found on the beach of Vlissingen-Dishoek contained fouling organism while this was 22% on Kwade Hoek. A possibility can be that the plastic items that wash ashore on these beaches were in the marine environment for a shorter period of time. This can also be confirmed by figure 4.8 and figure 9.10 in appendix B, that shows that the plastic found on these three beaches were less damaged than the plastics found on Kwade Hoek. The plastic items also had a lower percentage of fouling coverage than the plastic items on Kwade Hoek.

On Vlissingen and Oostkapelle and mainly also on Neeltje Jans, only hard plastics were found while on Kwade Hoek more soft plastics were found. This difference can be explained by the fact that more of the soft plastics are carried back into the sea with the outgoing tide and the wind that blows away from the beaches. The higher number of plastics found on Vlissingen-Dishoek beach may be related to a higher amount of plastic that comes from the Western Scheldt, the river Schelde or the high shipping traffic in the area.

5.2 Biofouling organisms

As far as the fouling organisms that were found, the bryozoans were the most found organism. Bryozoans are invertebrates that commonly grow on rocks, shells, seaweeds and poles. The bryozoans that were found were most common on soft plastics followed by the hard plastics. Soft plastics are more comparable to the seaweeds, while the hard plastics relate to the rocks, shells and poles. With the barnacles it is the other way around. Barnacles were more abundant on the hard plastics with some found on softer plastics. In this case the soft plastics consisted mainly of industrial sheeting and not the fragile plastics as was seen with the bryozoans. Barnacles are commonly found on rocks and poles but also on ship hulls.

The calcifying worm is a worm that creates a calcareous tube around him and are mainly found on hard substrates like rocks. The calcifying worms were found on hard plastics and in one case on soft plastics. This was however industrial sheeting which is a harder type of soft plastic. Anemones are also common on rocks and poles which related to the fact that both anemones found were on hard plastics. The hydroids can be found on hard substrates in general but was in this case found on both hard and soft plastics.

5.3 Identified species and invasiveness

A side focus of this study was to indicate the presence of the non-indigenous species that can be found as a fouling organism. This can help with unravelling the origin of the plastic as well as the impact of the plastic on the environment, for instance the invasiveness of the observed species. These findings could also help with future research into invasive species in the Zeeuwse delta, to find the origin of the organism and a possible solution to the invasion of the found fouling organism.

Based on the photographs of the fouling organisms species were identified. Most of the identified species turned out to be either native to the North Sea or a non-native species that was observed in the area for a much longer period of time.

The bryozoan that could be identified to species level was *Conopeum reticulum*, the seamat. This species of bryozoan is common in the North Sea along the Dutch and Belgium coast (Tyler-Walters et al., 2005).

For the barnacles the following species could be further identified: the New Zealand barnacle, *Austrominius modestus*, and the smooth acorn-shell, *Semibalanus balanoides. A. modestus* originates from New Zealand but has been commonly found in the North Sea since the second World War. It is assumed that *A. modestus* was introduced via fouling on the hull of ships and as larvae in bilge water (Avant, 2007). It is a successful invasive species in the North Sea since it adapted to the different conditions. *S. balanoides* is a common barnacle in the North Sea and can be found on hard substrates similar to *A. modestus* (White, 2008).

The calcifying worms that were found as fouling were all identified as *Spirobranchus Triqueter. S. triqueter* is a commonly found calcifying worm in the North Sea (Gosselin & Sewell, 2013).
The hydroid that was found was identified as *Ectopleura larynx. E. larynx* is commonly found around the British Isles and the North Sea (Baxter et al., 2012).

Of the identified species none were new to the Zeeland delta region. Of the fouling organisms that were not identified, it may be possible that there are new non-indigenous species among them, although this is not expected based on the visual inspections. This will become more clear when the DNA identification is completed in the future.

Based on the identification of the species further information could be collected on settling time and metamorphosis of these species (table 5.1). This indicates the time needed between larval stage and the time for the settlement to a substrate like plastic. The metamorphosis is also given where the time is shown for the organism to end the larval stage. This gives an indication on at least how long the plastic had to be in the aquatic environment to obtain the biofouling. Based on these species, plastic had to be in the water for at least a couple of months regarding the larval stage and metamorphosis time to settle and attach to plastic.

Assessing the age of the plastics with the use of the total coverage of the plastic items was also hard to conduct. If plastic occurs in the marine environment for a short period of time, there is not a lot of time for fouling organisms to attach to the item and expand. This may explain why the largest part of the plastic items were only covered for 0-20%. Looking at figure 5.1 the bryozoans (*Conopeum Reticulum)* and barnacles (*Austrominius Modestus & Semibalanus balanoides)* it takes multiple months for these organisms to go from the larval stage to fouling on a substrate. This means that the plastic had to be in the marine environment for at least that period of time. With the tubeworms (*Spirobranchus Triqueter)* and the pipe hydroid (*Ectopleura larynx)* it is harder to predict since the settling time and metamorphosis period is relatively short.

Species	Settling time & metamorphosis	Reference
Conopeum reticulum	Larval stage: 1-6 months Metamorphosis: 30 days Growth rate: 1000 zoids/30 days*	Tyler-Walters et al., 2005
Austrominius modestus	Larval stage: 2-3 month Metamorphosis: 1-3 day Growth rate: 0,5 - 1 mm/month*	CABI, 2020 Avant, 2007
Semibalanus balanoides	Larval stage: 1-2 moths Metamorphosis: 1-3 day Growth rate: 0,5 - 1 mm/month*	White, 2007
Spirobranchus triqueter	Larval stage: 11-30 days Metamorphosis: 1-2 weeks Growth rate: 1.5mm/month*	Riley, 2005 Gosselin & Sewell, 2013
Ectopleura larynx	Larval stage: 1 week Metamorphosis: 2-3 weeks Growth rate: -	Somodevilla, 2020 Baxter et al., 2012

Table 5.1 Table with the identified fouling organisms and information about the settling time from larval stage and the metamorphosis time.
**= All growth rates were found during studies but differ under different conditions like temperature and wave/current action.*

5.4 Age of the plastics

The age of the plastic turned out to be hard to determine based on de state of degradation. Most of the items were heavily damaged (figure 4.8). This high amount of heavily damaged plastics may indicate that the plastics were in the marine environment for a longer period of time. This is an assumption made based on this methodology, although the information on how fast plastic wears out is currently lacking. Plastics that are in the sediments are preserved for longer. The plastic may degrade faster under increased UV light intensity or higher wave action but there is no sufficient data to support these hypothesis. The age of the plastic can be determined based on the identified fouling organisms with the settling time and metamorphosis time (table 5.1)

Only three items that contained both fouling and text were predominantly coming from The Netherlands, with only one metal can containing the Greek language (see figure 5.2). This metal can was not damaged very much so it is possible that it came from a fishing boat in the North Sea region

Figure 5.2 Metal Coca Cola can with Greece text containing bryozoans, found on the beach of Kwade Hoek, 12th of May 2021.

As part of the macroplastics in the delta project besides the identification of the source origin of the plastic based on biofouling, the origin of plastic was also identified with the use of text, labels and language done by Brendan Oerlemans from WMR. This together helped with modelling exercises that were done by Bram van Duinen from Utrecht University on the travel path of plastic when in the marine environment.

5.5 Comparison to the Griend and Greenland Marine Litter Project

During the litter ID session of the Griend Marine Litter Project six items with fouling organisms were registered. These fouling organisms consisted of barnacles and calcifying worms. All six items with fouling were hard plastics. The results of the Macroplastics in the Delta project also showed that the collected plastic items contained barnacles and calcifying worms. These organisms were more abundant on hard plastics but were also found on soft plastics. Since on Griend all items containing fouling organisms were hard plastics the results are comparable to the Delta results. In the Zeeland Delta additional species groups were found, such as the bryozoans (most dominant group found), hydroids and anemones.

During the litter ID session for the Greenland project nine items were found with fouling organisms. The fouling organisms that were registered were calcifying worms, barnacles and bryozoans. All the items were hard plastics, just like in Griend. Some of the items on Greenland contained multiple fouling organisms. Similar species groups were found in Greenland as in the Delta project.
This means that the type of fouling species from all three locations were rather similar, consisting of mainly bryozoans, barnacles and calcifying worms, and on top of that in Zeeland also hydroids and anemones. The number of plastics with fouling found in the Delta was higher than that on Griend and Greenland, but the effort of finding plastics with fouling was also much higher.

5.6 Litter ID session Kwade Hoek

During the session items containing fouling animals could have been lost in the process as some of the categories were collected in one pile. The items contained bryozoans, barnacles, calcifying worms and pipe hydroids, the same as were observed on the items collected during the monitoring period in Kwade Hoek. Two plastic items contained dog whelk eggs (*Tritia sp.*). Trails of this species were not found in the earlier monitoring sessions and was therefore a new find. The *Tritia sp*. Is a specie that is native to the North Sea (Tyler-Walters, 2007). Further DNA identification could not be done, since the items were collected at an earlier stage and were withered.

5.7 OSPAR screening Veere

During the regular OSPAR screening on beach Veere (near the location of the Oostkapelle monitoring) a total of three plastic items were found that contained fouling organisms. The fouling consisted of barnacles and bryozoans. These items with biofouling were found higher up the beach compared to the monitoring on Oostkapelle when items with fouling were found closer to the sea.

6. Conclusion

The goal of this research was to find the possible sources and origin of plastic that wash ashore in the Zeeuwse Delta using information on biofouling, with the following research question: Can biofouling (flora and fauna) on plastic items collected on a selection of beaches in the Zeeuwse delta be an indicator of the source origin of the plastic? For answering of this research question, the type of fouling organisms, total coverage of the fouling organisms and the state of degradation were studied.

In the hypothesis it was stated that barnacles and bryozoans were expected to be found most often as fouling organism, with especially more barnacles since it was expected that harder items would be deposited more easily. However, the same amount of hard and soft plastics containing biofouling were found during the monitoring of the beaches. Bryozoans were far more abundant on soft plastics but were also found in an equal amount to barnacles on hard plastics. Other organisms that were observed as fouling organisms were calcifying worms, hydroids and anemones. Barnacles, bryozoans and calcifying worms were also found during earlier Marine Litter Projects on Griend and in Greenland.

With the state of degradation most of the plastic items with biofouling were heavily damaged indicating that the plastics have been in the water for a longer period of time. This is however harder to indicate with no sufficient information on degradation rates of plastic in relation to UV light or wave/current action. The plastics also could have been in the water for a long period of time before any biofouling attached to it. Most of the items were covered by fouling organisms for 0-40%. The metamorphosis rate from larval state to actual fouling organism indicated that in most situations the plastic had to be in the environment for at least a month for the bryozoans, a couple of weeks for the tubeworms and hydroids and only a day for the barnacles. Important with this is the fact that growth rate and settlement highly depend on conditions like temperature and wave/current action.

In conclusion, biofouling of flora and fauna can be an additional indicator for the origin of the plastics. The life cycle of the fouling organisms and the age of the plastic regarding the state of degradation and coverage could be used to predict how long the plastic has been in the environment. They show that plastic has been in the water for at least a couple of months based on the metamorphosis and growth rate of the identified species. Together with modelling exercises done by Bram van Duinen and the identification of language on packages done by Brendan Oerlemans, fouling provides information on the duration and potential origin of plastic items found on the marine shores. As based on biofouling the source origin of the plastics are most likely to be areas around the North Sea and close to the Zeeuwse Delta.

DNA identification is further needed to obtain the exact species information on the natural origin of the fouling organisms. The use of total coverage and state of degradation to predict the plastic origin was found more difficult to apply since it is not possible to indicate how fast certain species grow and how fast plastic degrades over time, especially if plastics end up in the sediments.

7. Recommendations and improvement for further research

Since this research was meant to develop a method for future studies on biofouling on plastic, multiple recommendations and improvements can be done.

- DNA identification of the sampled fouling organisms is recommended. This will not be done until after this graduation internship due to the covid-19 pandemic. An additional recommendation is to do the DNA identification already during the study. This will give more clarity on what species are found and can give a much better answer to the research question where the plastic originates from.
- A substantial difference was observed between the amount of plastics with fouling organisms found on Kwade Hoek and the other three beaches. This was even the case when the 6 items collected by volunteers of NLGO were excluded. This made a comparison between the four beaches a bit harder. For next time it can maybe help to set a fixed number of plastics containing biofouling per beach. This way you get an equal number for all beaches. This makes it easier to compare and work out within the results.
- A lot of the fouling organisms were withered because the items were on the beach for a long period and were therefore not suitable for DNA identification. More information about the species is therefore needed besides making pictures of the fouling organisms. A possibility for next time is to keep all the plastic with biofouling and try to identify the organisms together with experts already at location. This way all the experts can have a closer look to the fouling organisms and the experts can discuss their indication. This can lead to better identification of the organisms. This is of course only possible when the covid-19 pandemic is over.
- A lot of information can be obtained from the growth rate of the fouling organisms to get an indication on how long it had to take for the species to grow. Therefore more research can and need to be done on the growth rate of the organisms to get an exact time. It is important that all items with fouling organisms are kept, as the growth rate can be used to calculate the exact time for how long the plastic had to be in the water. This was not possible with the limited space and no access to a lab or bigger workspace.

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Appendix

Appendix A Raw data results

Beach	Amount
Vlissingen	2
Oostkapelle	12
Neeltje Jans	6
Kwade Hoek	53
	73
Type of fouling	**Amount**
Barnacle	34
Bryozoan	62
Pipe hydriod	2
Calcifying worm	10
Anemone	2
	110

Figure 9.1 Amount of plastic with biofouling found per beach and the amount and type of fouling.

Beach	Bryozoan	Barnacle	Anemone	Pipe hydriod	Calcifying worm
Vlissingen	2	1	0	0	1
Oostkapelle	7	6	0	0	1
Neeltje Jans	4	3	0	0	1
Kwadehoek	49	24	2	2	7

Figure 9.2 Amount and type of fouling found per beach.

Date	Beach	Amount of items	Type of material	Bryozoan	Barnacle	Calcifying worm	Pipe hydriod	Anemone
24-mrt	Vlissingen	1	Hard plastics	1	1	1	0	0
5-mei	Vlissingen	1	Hard plastics	1	0	0	0	0
4-mrt	Oostkapelle	1	Hard plastics	0	1	0	0	0
19-mrt	Oostkapelle	3	Hard plastics	2	2	0	0	0
24-mrt	Oostkapelle	3	Hard plastics	3	0	0	0	0
9-apr	Oostkapelle	4	Hard plastics	2	2	0	0	0
5-mei	Oostkapelle	1	Hard plastics	0	1	1	0	0
x	Oostkapelle							
4-mrt	Neeltje Jans	1	Soft plastics	1	0	0	0	0
4-mrt	Neeltje Jans	1	Hard plastics	1	0	0	0	0
16-apr	Neeltje Jans	1	Hard plastics	0	1	0	0	0
30-apr	Neeltje Jans	2	Hard plastics	1	1	0	0	0
12-mei	Neeltje Jans	1	Soft plastics	1	1	1	0	0
18-mrt	Kwade Hoek	10	Soft plastics	10	4	0	1	0
18-mrt	Kwade Hoek	4	Hard plastics	4	2	0	1	0
31-mrt	Kwade Hoek	7	Soft plastics	6	1	1	0	0
31-mrt	Kwade Hoek	5	Hard plastics	5	4	4	0	2
30-apr	Kwade Hoek	1	Soft plastics	1	0	0	0	0
30-apr	Kwade Hoek	1	Hard plastics	1	0	0	0	0
12-mei	Kwade Hoek	14	Soft plastics	14	3	0	0	0
12-mei	Kwade Hoek	6	Hard plastics	4	5	2	0	0
12-mei	Kwade Hoek	1	Textile	0	1	0	0	0
12-mei	Kwade Hoek	4	Metal cans	4	4	0	0	0

Figure 9.3 Overview of the dates and visited beaches with the type and amount of plastics and found fouling organisms.

Beach	Hard plastics	Soft plastics	Textile	metal cans
Vlissingen	2	0	0	0
Oostkapelle	12	0	0	0
Neeltje Jans	4	2	0	0
Kwade Hoek	16	32	1	4

Figure 9.4 Type of materials found per beach.

Type of material	Bryozoan	Barnacle	Calcifying worm	Pipe hydroid	Anemone
Hard plastics	25	20	8	1	2
Soft plastics	33	9	2	1	0
Textile	0	1	0	0	0
Metal cans	4	4	0	0	0

Figure 9.5 Type of biofouling on the type of material.

Beach	Total Amount
Vlissingen	1004
Oostkapelle	677
Neeltje Jans	532
Kwade Hoek	244

Figure 9.6 Total amount of found plastics including plastics without biofouling.

State	Amount
Almost no damage	9
Slightly damaged	12
Damaged	16
Heavily damaged	36
	73

Coverage	Amount
0-20%	39
20-40%	19
40-60%	8
60-80%	6
80-100%	1
	73

Figure 9.7 The amount of all plastics categorized for the state of degradation and percentage of coverage.

State	Amount
Almost no damage	1
Slightly damaged	9
Damaged	10
Heavily damaged	33
	53

Coverage	Amount
0-20%	29
20-40%	11
40-60%	6
60-80%	6
80-100%	1
	53

Figure 9.8 The amount of plastics found on Kwade Hoek categorized for the state of degradation and percentage of coverage.

Appendix B additional figures results

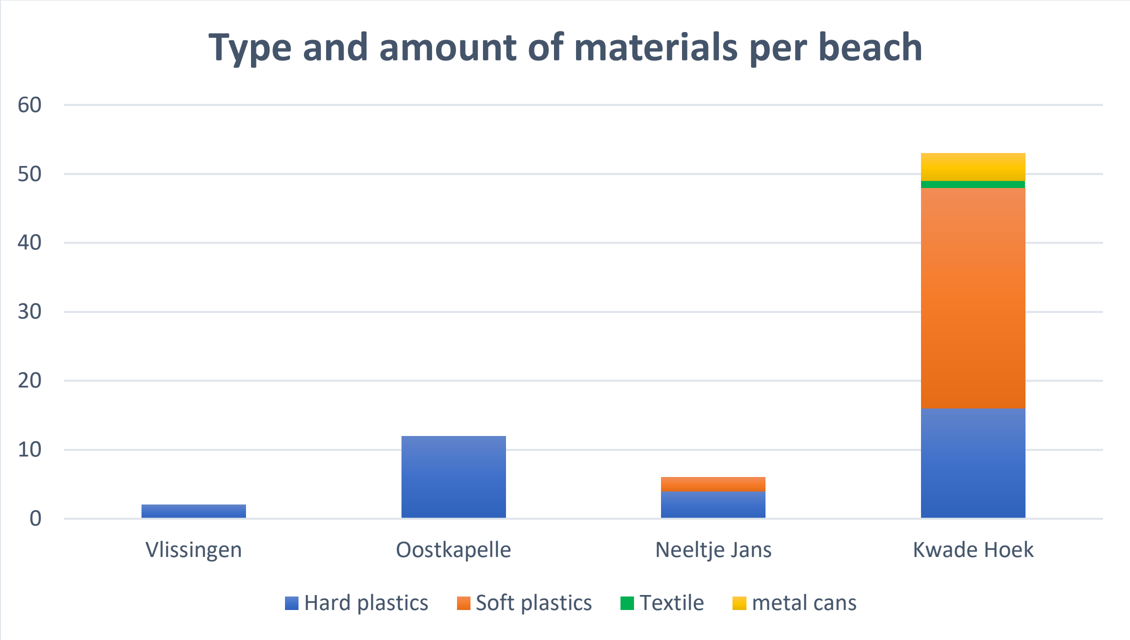

Figure 9.9 Type and amount of the plastic items with fouling found per beach.

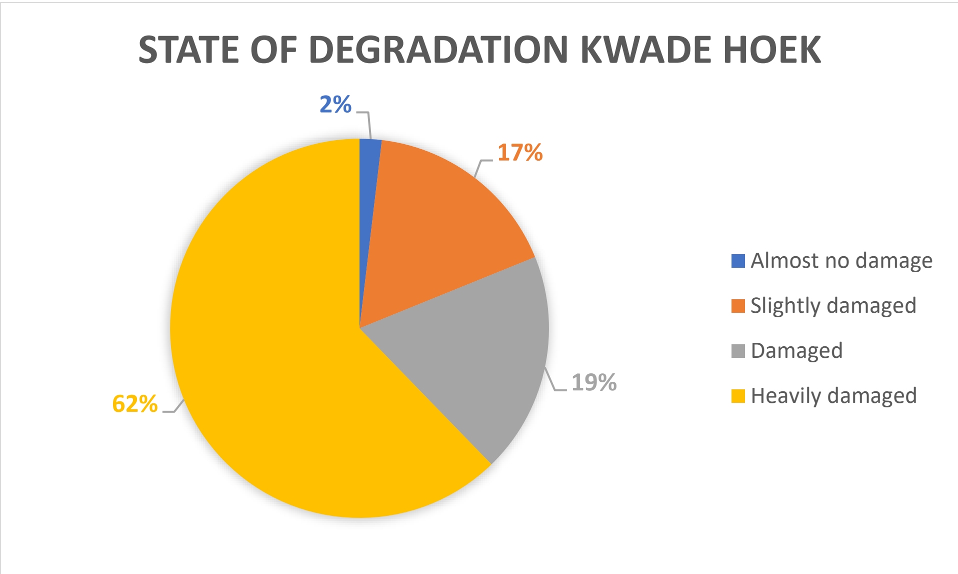

Figure 9.10 State of degradation of the plastic items found on Kwade Hoek.

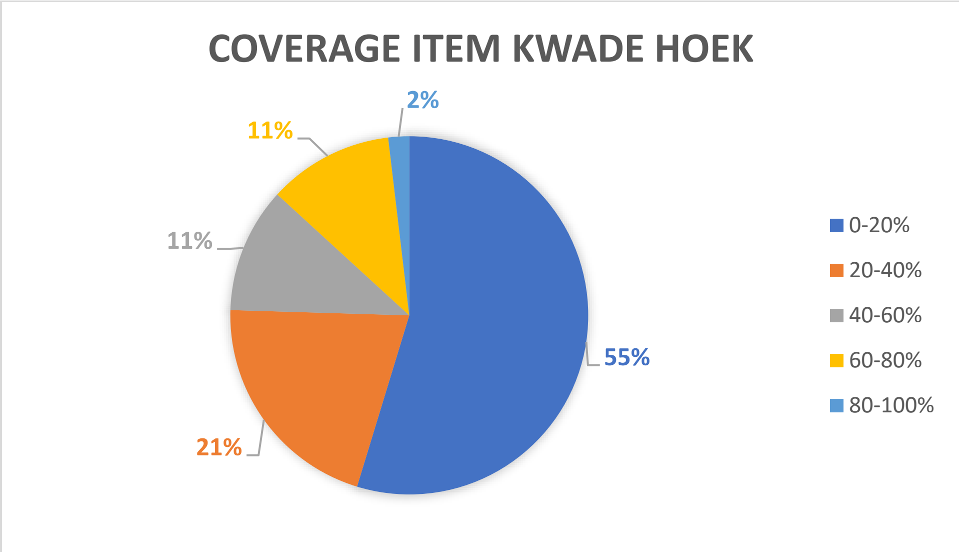

Figure 9.11 Coverage in % of the fouling organisms on the items found in Kwade Hoek.