Dover to Folkestone MCZ 2016 Monitoring Report

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Dover to Folkestone MCZ 2016 Monitoring Report

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Foreword

Natural England commission a range of reports from external contractors to provide evidence and advice to assist us in delivering our duties. The views in this report are those of the authors and do not necessarily represent those of Natural England.

Background

Following designation, Natural England started a baseline monitoring programme across all marine protected areas.

This report was commissioned as part of an inshore benthic marine survey of the Dover to Folkstone MCZ.

Acknowledgements

We thank the Marine Protected Areas Group (MPAG) representatives for reviewing earlier drafts of this report.

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Executive Summary

This report is one of a series of Marine Protected Area (MPA) characterisation and monitoring reports delivered to Defra by the Marine Protected Areas Group (MPAG). The purpose of the report series is to provide the necessary information to allow Defra to fulfil its obligations in relation to MPA assessment and reporting, in relation to current policy instruments, including the Oslo-Paris (OSPAR) convention, the UK Marine and Coastal Access Act (2009) and other relevant Directives (e.g. the Marine Strategy Framework Directive). This characterisation report is informed by data acquired during a dedicated Day Grab survey carried out at the Dover to Folkestone Marine Conservation Zone (MCZ) (during 2016) and will form part of the ongoing time series data and evidence for this MPA.

Dover to Folkestone MCZ is an inshore site located on the south eastern coast of England within the 'Eastern Channel' Charting Progress 2 (CP2) sea area. A number of features of conservation importance (FOCI), including both habitats and species, are designated for protection within the Dover to Folkestone MCZ. This report provides a characterisation of a number of Broadscale Habitats (BSHs) ('A5.1 Subtidal coarse sediment', 'A5.2 Subtidal sand', 'A5.3 Subtidal mud' and 'A5.4 Subtidal mixed sediments'), and species FOCI (Native Oyster *Ostrea edulis*) designated within the MCZ.

The extent and distribution of designated BSHs was in line with expectations based on previous information. 'A5.4 Subtidal mixed sediments' was the most frequently occurring BSH across the site. The infaunal assemblage was typical of this soft sedimentary habitat and considered healthy. *Sabellaria spinulosa* communities were also present throughout the site, although upright 'reefy' tube structures were not observed. The infauna of the other BSHs were also typical of those habitats and were therefore considered healthy. The Infaunal Quality Index confirmed that the status of all these habitats was either 'Good' or 'High'.

Although the MCZ is designated for Native Oyster (*Ostrea edulis*), no specimens were recovered during the survey, however this should not be interpreted as the species is absent from the site.

Contaminant levels were generally below OSPAR Background Affect Concentrations, but a station in close proximity to the port of Dover did have elevated levels of heavy metals, polycyclic aromatic hydrocarbons and polychlorinated biphenyls. This could indicate localised contamination that may be exerting biological impacts on the site.

There are a number of monitoring recommendations made within the report for future surveys. The similarity of 'A5.4 Subtidal mixed sediments' outside the MCZ compared to those inside will allow the use of a BACI style approach for assessing the efficacy of management measures in this BSH.

1 Introduction

Dover to Folkestone Marine Conservation Zone (MCZ) is part of a network of sites designed to meet conservation objectives under the UK Marine and Coastal Access Act (2009). These sites will also contribute to an ecologically coherent network of Marine Protected Areas (MPAs) across the North-east Atlantic, as agreed under the Oslo-Paris (OSPAR) Convention and other international commitments to which the UK is a signatory.

Under the Marine and Coastal Access Act (2009), Defra is required to provide a report to Parliament every six years that includes an assessment of the degree to which the conservation objectives set for MCZs are being achieved. In order to fulfil its obligations, Defra has directed the Statutory Nature Conservation Bodies (SNCBs) to carry out a programme of MPA monitoring. The SNCB responsible for nature conservation inshore (between 0 nm and 12 nm from the coast) is Natural England (NE) and the SNCB responsible for nature conservation offshore (between 12 nm and 200 nm from the coast) is the Joint Nature Conservation Committee (JNCC). Where possible, this monitoring will also inform assessment of the status of the wider UK marine environment; for example, assessment of whether Good Environmental Status (GES) has been achieved, as required under Article 11 of the Marine Strategy Framework Directive (MSFD).

This characterisation report primarily explores data acquired from the first dedicated monitoring survey of Dover to Folkestone MCZ, which will form the initial point in a monitoring time series against which feature (and site) condition can be assessed in the future. The specific aims of the report are discussed in more detail in Section 1.2.

1.1 Site overview

Dover to Folkestone MCZ is an inshore site on the south eastern coast of England (Figure 1). Dover to Folkestone MCZ was recommended as a MCZ by the 'Balanced Seas' regional stakeholder group project. It is located in the jurisdictional area of the Kent and Essex Inshore Fisheries and Conservation Authority (IFCA) and falls within the wider 'Charting Progress 2' (CP2) area 'Eastern Channel'. The site is neighboured by the Dover to Deal MCZ and Folkestone Pomerania MCZ (Figure 1). The site overlaps with the 'Kent South' Water Framework Directive (WFD) Waterbody. There is a WFD water quality sampling point at the Dover long sea outfall inside the site, and a Bathing Waters Directive monitoring site at Folkestone Beach just outside the western boundary.

The MCZ extends approximately 3 km from the shoreline, ranging from the intertidal to a water depth of approximately 30 metres below sea level (chart datum), and covers approximately 20 km^2. At the time of writing, there were no byelaws in place restricting fishing activity within the site, although inshore sightings maps show there is some low intensity mobile and static gear fishing taking place over and close to the site (Vanstaen and Breen, 2014).

The west of the site sits within the Dover Harbour Authority area. The Dover dredge disposal site is to the southwest of the site, just south of the Dover harbour entrance. Due to the proximity of the site to the port, the western end of the site is subject to high levels of shipping passaging over or close to the site, with an estimated annual average of >10,000 vessels passaging over the western site boundary. This decreases to an annual average of up to 200 vessels in the north east of the site at Deal (MMO, 2014). A historic subsea telegraph cable passes through the site and makes landfall at St Margaret's Bay (Figure 1).

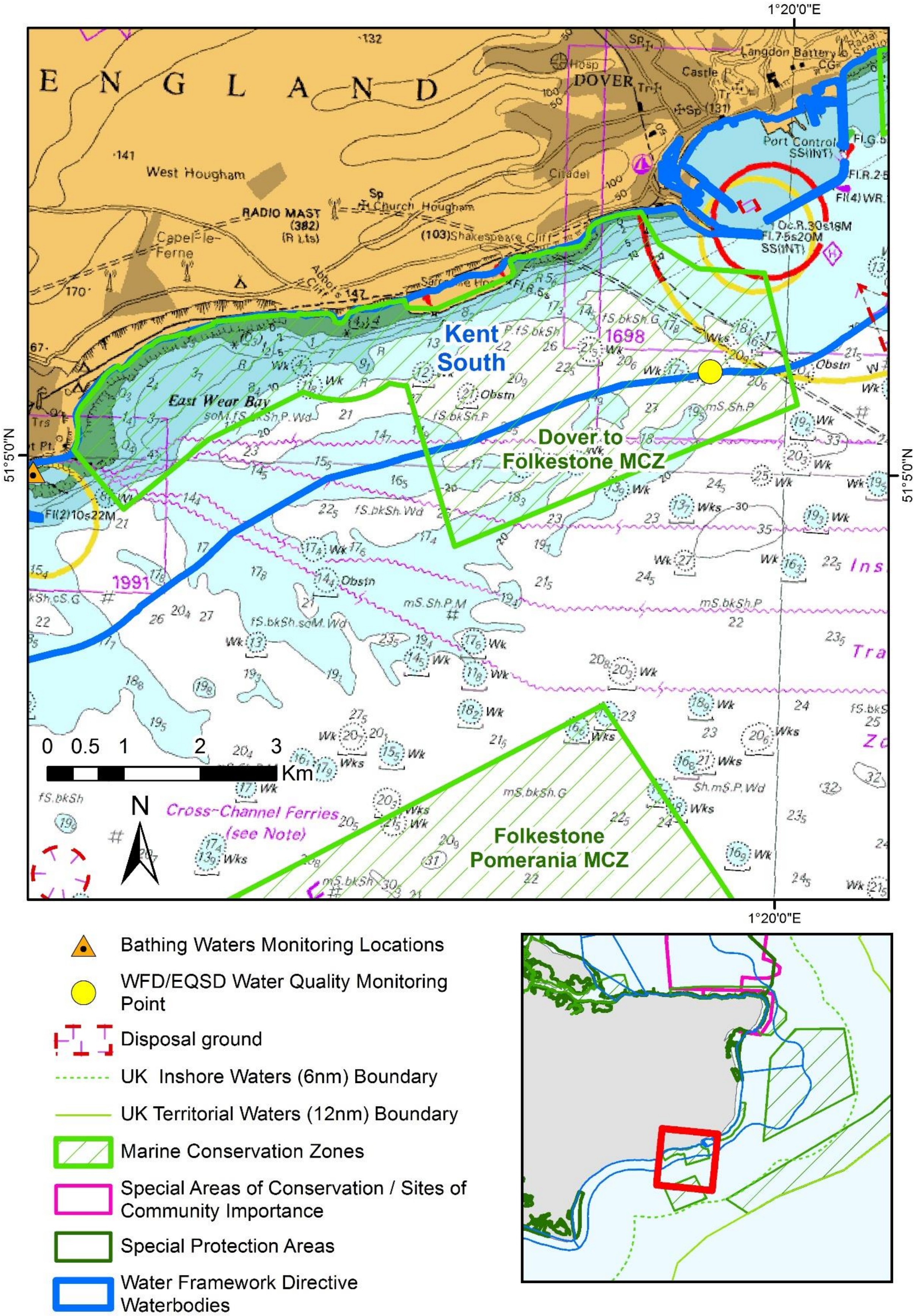

Figure 1. Location of the Dover to Folkestone MCZ in the context of Marine Protected Areas and management jurisdictions proximal to the site.

The site was designated[1] due to the presence of a wide variety of benthic habitats, including mixed subtidal sediments that support populations of brittlestars, squat lobsters, crabs, fish and molluscs. Native oysters are also found in the area. In addition to the soft sediment habitats, the rocky and chalk seashores are notable for their diverse intertidal underboulder communities.

1.2 Aims and objectives

1.2.1 High-level conservation objectives

High-level site-specific conservation objectives serve as benchmarks against which the efficacy of the General Management Approach (GMA) in achieving the conservation objectives (i.e. maintaining designated features at, or recovering them to, 'favourable condition') can be assessed and monitored.

As detailed in Dover to Folkestone MCZ designation order[1], the conservation objectives for the site are that the designated features (Table 1):

a) So far as already in favourable condition, remain in such condition; and

b) So far as not already in favourable condition, be brought into such condition, and remain in such condition.

It should be noted that 'maintain' GMAs may have been applied based on an indirect or proxy assessment, as opposed to being based on empirical monitoring evidence (i.e. direct observations).

1.2.2 Definition of favourable condition

Favourable condition, with respect to a habitat feature, means that:

a) Its **extent and distribution** is stable or increasing;

b) Its **structures and functions**, including its quality, and the composition of its characteristic biological communities, are such as to ensure that it remains in a condition which is healthy and not deteriorating; and

c) Its natural **supporting processes** are unimpeded.

The extent of a habitat feature refers to the total area in the site occupied by the qualifying feature and must also include consideration of its distribution. A reduction in feature extent has the potential to alter the physical and biological functioning of sedimentary habitat types (Elliott *et al.*, 1998). The distribution of a habitat feature influences the component communities present and can contribute to the condition and resilience of the feature (JNCC, 2004).

[1] http://www.legislation.gov.uk/ukmo/2016/6/contents/created

Structure encompasses the physical components of a habitat type and the key and influential species present. Physical structure refers to topography, sediment composition and distribution. Physical structure can have a significant influence on the hydrodynamic regime operating at varying spatial scales in the marine environment, as well as influencing the presence and distribution of associated biological communities (Elliott *et al.,* 1998). The function of habitat features includes processes such as: sediment reworking (e.g. through bioturbation) and habitat modification, primary and secondary production and recruitment dynamics. Habitat features rely on a range of supporting processes (e.g. hydrodynamic regime, water quality and sediment quality) which act to support their functioning as well as their resilience (e.g. the ability to recover following impact).

For species features, favourable condition means that:

a) The quality and quantity of its habitat are such as to ensure that the population is maintained in numbers which enable it to thrive;

b) The composition of its population in terms of number, age and sex ratio are such as to ensure that the population is maintained in numbers which enable it to thrive; and

c) Its natural supporting processes are unimpeded.

1.2.3 Report aims and objectives

The primary aim of this characterisation report is to explore and describe the attributes of the designated features within the Dover to Folkestone MCZ (Table 1), to enable future assessment and monitoring of feature condition. The results presented will be used to develop recommendations for future monitoring, including the operational testing of specific metrics which may indicate whether the condition of the feature has been maintained, is improving or is in decline.

The broad objectives of this monitoring report are provided below:

1) Provide a description of the **extent[2], distribution, structural** and (where possible) **functional** attributes, and the **supporting processes**, of the designated features within the site (see Table 2 for more detail), to enable subsequent condition monitoring and assessment;

2) Note observations of any Habitat or Species FOCI not covered by Designation Order as features of the site;

[2] Note that where current habitat maps are not available, extent will be described within the limits of available data.

3) Present evidence relating to marine litter (Descriptor 10), to satisfy requirements of the Marine Strategy Framework Directive;

4) Provide practical recommendations for appropriate future monitoring approaches (e.g. metric selection, survey design, data collection approaches) for both the designated features and their natural supporting processes with a discussion of their requirements.

1.2.4 Feature attributes and supporting processes

To achieve report Objective 1, the report will present evidence on a number of feature attributes and supporting processes, as defined in supplementary advice on conservation objectives currently being developed by Natural England for the designated features (Table 1) within the Dover to Folkestone MCZ [3]. It should be noted that it was not possible to address all feature attributes in the monitoring characterisation survey, given the comprehensive nature of the attribute lists for each feature. The feature attributes were therefore rationalised and prioritised, resulting in a smaller sub-set.

The list of selected feature attributes and supporting processes considered in this report is presented in Table 2, alongside the generated outputs for each.

[3] when published, the Conservation Advice package will be available from: https://designatedsites.naturalengland.org.uk/Marine/MarineSiteDetail.aspx?SiteCode=UKMCZ0033andSiteName=DoverandcountyCode=andresponsiblePerson=andSeaArea=andIFCAArea=

Table 1. Dover to Folkestone MCZ site overview, including General Management Approach (GMA) for designated features (© Natural England and Environment Agency 2022).

Charting Progress 2 Region[4]	Eastern Channel	
Spatial Area (km²)	20	
Water Depth Range (m)	0-30	
Existing Data and Information	**Fraser, M. and Easter, J. (2017).** Dover to Folkestone MCZ 2016 Baseline Survey Report. 86 pp. **Colenutt, A., Grewcock, G. and Evans, J. (2015).** Dover to Folkestone rMCZ Post-survey Site Report. 62 pp. **Godsell, N., Meakins, B. and Jones, N. (2013).** Dover to Folkestone rMCZ. 53 pp.	
Current and Proposed Management Measures	None	
Features Present (BSH)	**Designated**	**GMA**
A1.1 High energy intertidal rock*	✓	Maintain
A1.2 Moderate energy intertidal rock*	✓	Maintain
A1.3 Low energy intertidal rock*	✓	Maintain
A2.1 Intertidal coarse sediment*	✓	Maintain
A2.2 Intertidal sand and muddy sand*	✓	Maintain
A3.1 High energy infralittoral rock	✗	N/A
A3.2 Moderate energy infralittoral rock	✓	Maintain
A4.2 Moderate energy circalittoral rock	✗	N/A
A5.1 Subtidal coarse sediment	✓	Maintain
A5.2 Subtidal sand	✓	Maintain
A5.3 Subtidal mud	✓	Maintain
A5.4 Subtidal mixed sediments	✓	Maintain
A5.5 Subtidal macrophyte dominated sediment	✗	N/A
Features Present (Habitat FOCI)		
Intertidal Underboulder Communities*	✓	Maintain
Littoral Chalk Communities*	✓	Maintain
Peat and Clay Exposures	✗	N/A
Ross Worm (*Sabellaria spinulosa*) Reef	✗	N/A
Blue Mussel Beds	✗	N/A
Subtidal Chalk	✗	N/A
Subtidal Sands and Gravels	✗	N/A
Features Present (Species FOCI)		
Native Oyster (*Ostrea edulis*)**	✓	Maintain
Short Snouted Seahorse (*Hippocampus hippocampus*)**	✗	N/A
Geology Present		
Folkestone Warren	✓	Maintain

** The characterisation survey reported here did not extend into the intertidal.*

***The characterisation survey was not specifically designed to target species FOCI.*

[4] http://webarchive.nationalarchives.gov.uk/20141203170558tf_/http://chartingprogress.defra.gov.uk/

Table 2. Feature attributes and supporting processes addressed to achieve report Objective 1, for the Dover to Folkestone MCZ (© Natural England and Environment Agency 2022).

Feature attributes	Features	Outputs
Extent and distribution	A5.1 Subtidal coarse sediment A5.2 Subtidal sand A5.3 Subtidal mud A5.4 Subtidal mixed sediments	Particle Size Distribution (PSD) derived from seabed sediment samples.
Sediment composition and distribution	A5.1 Subtidal coarse sediment A5.2 Subtidal sand A5.3 Subtidal mud A5.4 Subtidal mixed sediments	PSD derived from seabed sediment samples.
Presence and spatial distribution of biological communities **Presence and abundance of key structural and influential species** **Species composition of component communities**	A5.1 Subtidal coarse sediment A5.2 Subtidal sand A5.3 Subtidal mud A5.4 Subtidal mixed sediments	Biological communities (and derived biotopes) derived from each sedimentary BSH.
Non-indigenous species (NIS)	Dover to Folkestone MCZ	Location of samples where NIS were recorded.
Presence and distribution of the species	Native Oyster (*Ostrea edulis*)	Location and density of individuals recorded in Day Grab samples
Supporting processes:		
Sediment contaminants	A5.2 Subtidal sand A5.4 Subtidal mixed sediments	Assessment of sediment contaminant concentrations against OSPAR thresholds.

2 Methods

2.1 Data sources

Data used to inform this characterisation report have been compiled from a survey carried out at Dover to Folkestone MCZ in August 2016 by the Environment Agency (Fraser and Easter, 2017). Locations of grab samples collected during the 2016 survey are shown in Figure 2.

The 2016 survey also collected still images to assess the rock features of the MCZ using a SeaSpyder drop camera system but the images were not interpretable due to poor visibility due to high turbidity, and are not reported or discussed here. Further information is available in the Fraser and Easter (2017). In addition, the Hamon Grab sampling in 2012 (Godsell *et al.,* 2013) collected four infaunal samples used to inform the acoustically-derived habitat map (Colenutt *et al.,* 2015).

2.2 Survey Design

To achieve report Objective 1, the BSH map produced by the 2012 verification survey (and additional data from the Natural England evidence database) were used to inform the selection of 35 Day Grab sampling stations within the MCZ (Fraser and Easter, 2017). The 2016 sampling stations included the four stations sampled in 2012 and another 31 stations nested within the mapped habitats to ensure spatially sufficient sampling of the BSHs (Fraser and Easter, 2017). Due to the low number of grab samples collected in the 2012 verification survey, it was not possible to use power analysis to plan the sampling strategy for this survey.

Bathymetry and existing sediment data were used to identify another 19 stations outside and to the west of the MCZ. The stations were placed above the 20 m depth contour in order to sample within the same depth range as the stations located inside the MCZ (Fraser and Easter, 2017).

In combination, these stations conformed to a Before-After-Control-Impact (BACI) design and the 2016 data provides the initial point of reference for detecting impacts to the features within the MCZ (Fraser and Easter, 2017).

Additionally, four stations within the MCZ were also chosen for sediment contaminant analysis (Fraser and Easter, 2017).

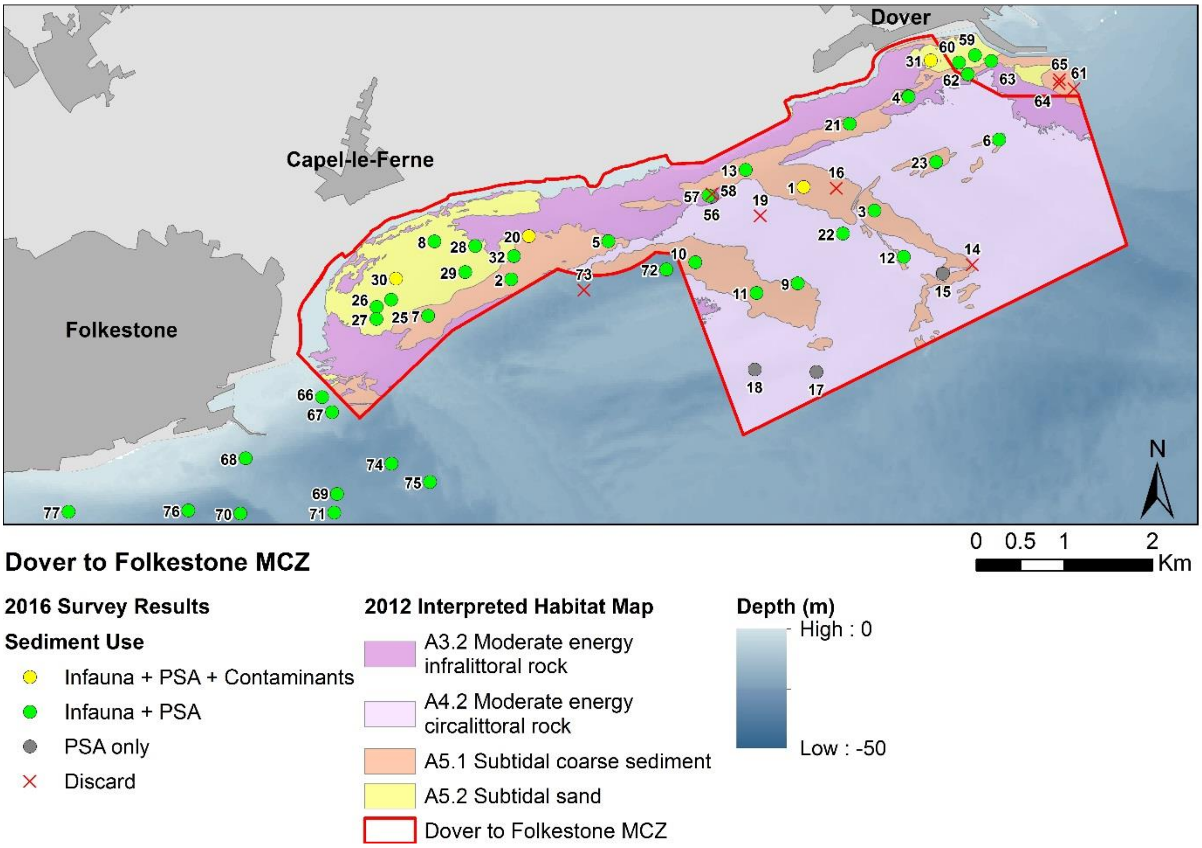

Figure 2. Location of ground truth samples collected from the Dover to Folkestone MCZ in 2016. (© Natural England and Environment Agency 2022).

2.3 Data acquisition and processing

2.3.1 Seabed sediments

Sediment samples for PSA and benthic infauna analyses were collected using a 0.1 m^2 Day Grab as described in the Environment Agency Water Framework Directive (WFD) operational instructions 104_10 (2012) and 009_07 (2014).

A 500 ml sub-sample was taken from each grab sample and stored at -20°C prior to determining the Particle Size Distribution (PSD). Sediment samples were processed by the Environment Agency's National Laboratory Service following the recommended methodology of the North East Atlantic Marine Biological Analytical Quality Control (NMBAQC) scheme (Mason, 2011). The less than 1 mm sediment fraction was analysed using laser diffraction and the greater than 1 mm fraction was dried, sieved and weighed at 0.5 phi (ϕ) intervals. Sediment distribution data were merged and used to classify samples into sedimentary broad scale habitats.

The faunal fraction was sieved over a 1 mm mesh, photographed, and then fixed in buffered 4% formaldehyde. Faunal samples were processed by the Institute of Estuarine and Coastal Studies to extract all fauna present in each sample. Fauna were identified to species level where possible, enumerated and weighed (blotted wet weight) to the nearest 0.0001 g following the recommendations of the NMBAQC scheme (Worsfold *et al.,* 2010). To achieve report Objective 3, any marine litter fragments >1 mm present in the residues were extracted and counted for each sample.

At four stations, additional grabs were collected to retrieve material for contaminant analysis, providing a record of the most recent contaminant levels deposited in the sediment. Surface sediment scrapes were sampled to a maximum depth of 1 cm (avoiding the anoxic layer), following the methodology detailed in the Environment Agency Operational Instruction 10_01 (2016).

2.4 Data preparation and analysis

2.4.1 Sediment particle size distribution

Sediment particle size distribution data (half phi classes) were grouped into the percentage contribution of gravel, sand and mud derived from the classification proposed by Folk (1954). In addition, each sample was assigned to one of four sedimentary BSHs using a modified version of the classification model produced during the Mapping European Seabed Habitats (MESH) project (Long, 2006).

2.4.2 Physico-chemical properties

Sediment dry weight contaminant concentrations were normalised to 5% aluminium (for heavy metals) and 2.5% total organic carbon content (for organics) to take account of the variation between sediment types (OSPAR Commission, 2015) for comparison.

Results were compared against Oslo-Paris (OSPAR) Convention Background Assessment Criteria (BAC) and Environmental Assessment Criteria (EAC) / Effects Range Low (ERL) thresholds for heavy metals, polycyclic-aromatic hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs), above which concentrations may chronically impact marine fauna.

2.4.3 Biological community data preparation

Benthic macrofauna data sets were checked to ensure consistent nomenclature and identification policies. Any discrepancies identified were resolved using expert judgement following the truncation steps presented in Annex 3. Invalid taxa and fragments of countable taxa were removed from the data set while the presence of colonial taxa was changed to a numerical value of one. Records were combined where a species was identified correctly both by using its binomial name and by using its binomial name with a qualifier e.g. *Lumbrinereis cingulata* 'aggregate'. Records labelled as 'juvenile' were combined with adults of the same genus/species/family.

The infaunal species abundance data were cross-referenced against a list of 49 non-indigenous target species which have been selected for assessment of Good Environmental Status (GES) in GB waters under MSFD Descriptor 2 (Stebbing *et al.,* 2014; Annex 6). The list includes two categories; species which are already known to be present within the assessment area (present) and species which are not yet thought to be present but have a perceived risk of introduction and impact (horizon). An additional list of taxa, which were identified as invasive in the 'Non-native marine species in British waters: a review and directory' (Eno *et al.,* in 1997) was also used to cross reference against all taxa observed (Annex 6).

2.4.4 Statistical analyses

The truncated macrofauna abundance data were imported into PRIMER v6 (Clarke and Gorley, 2006) to enable multivariate analysis and the derivation of various metrics for univariate analysis. Species classification information and a number of relevant factors/indicators were also assigned to the data at this stage. The number of taxa (S), total abundance of enumerable individuals (N), Shannon (H'(Log^e)), species evenness (1-λ') and Hills (N1) diversity metrics were derived for each sample using the DIVERSE function within PRIMER v6. The Infaunal Quality Index (IQI), an assessment of benthic faunal condition, was calculated using the 11/03/2014 update of the Environment Agency's IQI Excel workbook (Phillips *et al.,* 2014).

Nonmetric multidimensional scaling (nMDS) ordination, analysis of similarity (abundance square-root transformed species data and Bray-Curtis similarity) between (ANOSIM) and dissimilarity within (SIMPROF with associated SIMPER) groups were conducted in PRIMER v6 to explore differences in biological community composition for (a) between the habitat features and (b) between examples of comparable features located within and outside of the MCZ boundary.

Summary statistics, data interpretation/manipulation and appropriate statistical tests were performed on the univariate sample level metrics to test for and explain any significant differences between habitat and spatial groups (Minitab 18.1, 2017). Because the univariate sample statistics are not independent of one another, and assumptions of multivariate ANOVA are unlikely to be met, a global test of statistical significance was first performed using ANOSIM on normalised univariate indices and Euclidean distance.

Comparisons were then undertaken, separately for each univariate statistic, using ANOVA and post-hoc Tukey tests. Data transformation was applied to overcome non-normality. Biomass was square root transformed. The assumption of equality of variance was often not met but the implications in using ANOVA were thought not to be too severe. Conclusions were sense-checked against box-plots and existing knowledge regarding the influence of sedimentary structure on benthic communities.

3 Results and Interpretation

3.1 Site overview

The Dover to Folkestone MCZ 2016 subtidal baseline characterisation survey was completed in August 2016, and identified all four designated sediment BSHs, 'A5.1 Subtidal coarse sediment', 'A5.2 Subtidal sand', 'A5.3 Subtidal mud', and 'A5.4 Subtidal mixed sediments' (Table 3). The distribution of BSHs was in good agreement with the 2012 interpreted habitat map on which the survey was designed with the exception of areas predicted to be coarse sediment from the habitat map (Figures 3 and 4). The 2016 particle size analysis results showed that areas predicted to be 'A5.1 Subtidal coarse sediment' to the north and west of the site were 'A5.4 Subtidal mixed sediments'. 'A5.1 Subtidal coarse sediment' was recorded to the south east of the site in agreement with the predicted habitat map, and also outside the MCZ to the west of the Dover Harbour wall, in areas predicted to be 'A5.2 Subtidal sand'.

The percentage contribution of gravel, sand and mud of the 2016 samples is shown in Figures 4 and 5. 'A5.4 Subtidal mixed sediments' were spatially distributed both inside and outside the MCZ, including in areas predicted to be sublittoral rock (Figure 3). The infauna in this BSH was the most diverse and numerous (Table 4, Figures 6 and 7), and the communities inside and outside the MCZ were similar. The Ross Worm (*Sabellaria spinulosa*) was often observed in the mixed sediment grab samples, present in an encrusting form attached to pebbles/cobbles as opposed to tube clusters forming erect 'reefy' structures.

Table 3. Number of samples collected in each BSH inside and outside of the MCZ boundary (© Natural England and Environment Agency 2022). Does not include station 47 (only infauna collected).

Broadscale Habitat (BSH)	Grab – PSA and Infauna		Grab – PSA only	
	In	Out	In	Out
'A5.1 Subtidal coarse sediment'	4	2	0	1
'A5.2 Subtidal sand'	7	0	0	0
'A5.3 Subtidal mud'	1	1	0	0
'A5.4 Subtidal mixed sediments'	14	12	3	0

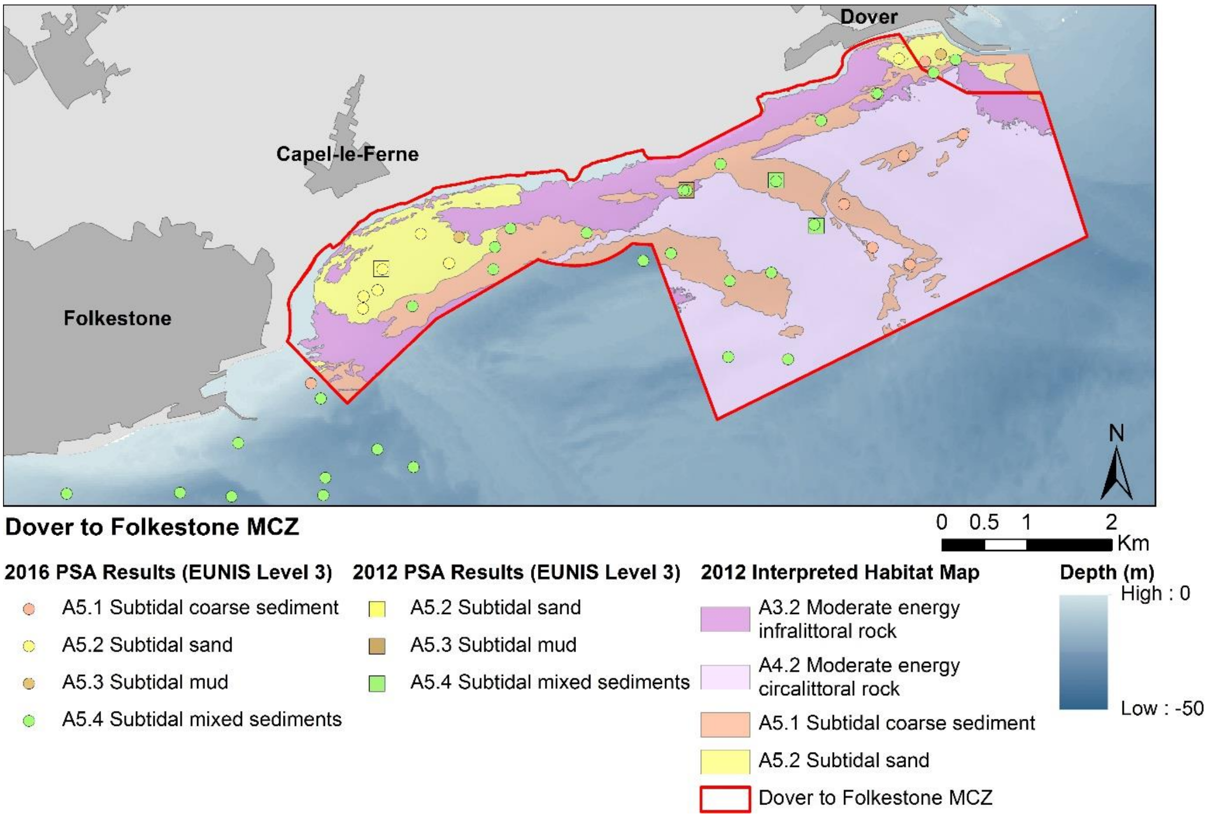

Figure 3. Distribution of Broadscale Habitats (BSHs) derived from 2016 Particle Size Analysis (PSA) data compared to the 2012 interpreted habitat map (© Natural England and Environment Agency 2022).

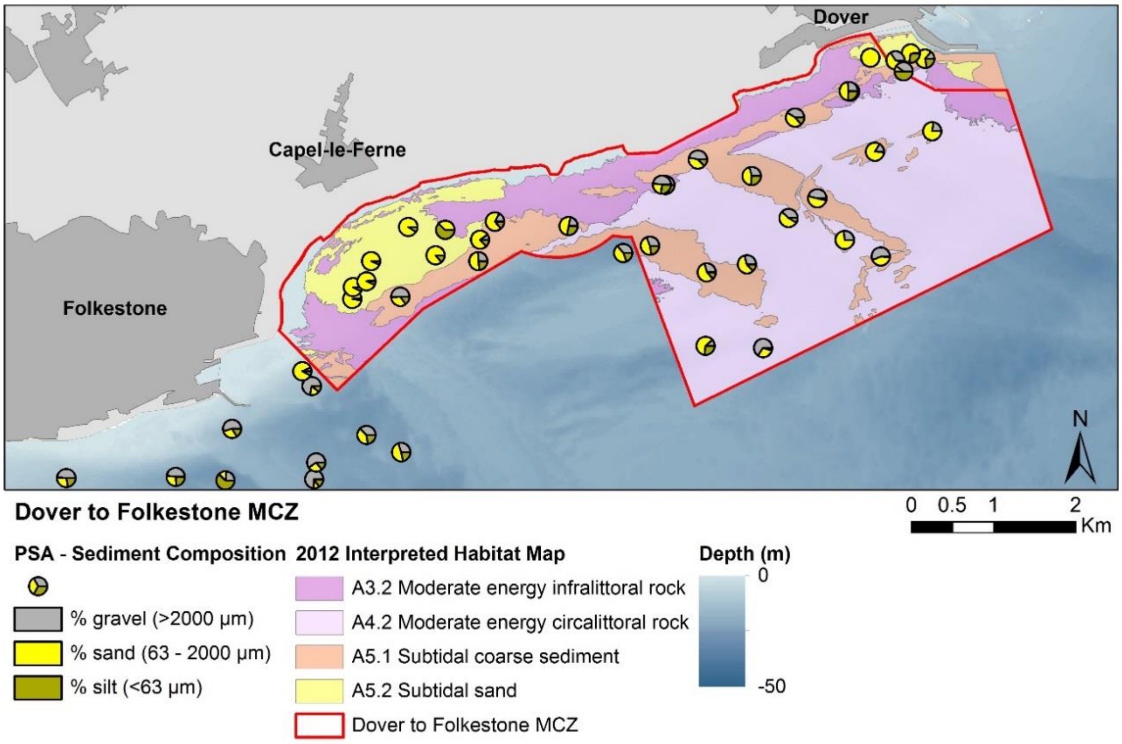

Figure 4. The proportion of mud, sand and gravel in Particle Size Analysis (PSA) samples collected in 2016 (© Natural England and Environment Agency 2022).

Table 4. Mean (± standard error) macrobenthic species abundance, richness, total biomass, infaunal quality index (IQI) and other univariate indices of the 0.1 m^2 Day Grab samples for the four different Broadscale Habitats (BSHs) collected outside and within the Dover to Folkestone MCZ in 2016 (sieved to 1 mm) (© Natural England and Environment Agency 2022). For metrics of stations sampled inside the MCZ that had data suitable for ANOVA, BSH that do not share a superscript letter are significantly different for that metric. Note that Biomass data had a square root transformation applied before ANOVA.

		Sample number	Total taxa	Abundance (n sample^{-1})		Taxa Richness (S sample^{-1})		Biomass (g)		Shannon Index H'(log^e)		Simpson's Evenness (1-λ')		Hill's N1		Infaunal Quality Index (IQI)	
				Mean	±S.E.	Mean	±S.E.	Mean	±S.E.	Mean	±S.E.	Mean	±S.E.	Mean	±S.E.	Mean	±S.E.
'A5.1 Subtidal coarse sediment'	Inside	4	195	372	159	74.5[a]	19.8	1.03[b]	0.58	3.19[a]	0.38	0.89[a]	0.05	29.1[a]	8.5	0.75[a,b]	0.02
	Outside	2	69	288	266	35.5	28.5	3.51	3.38	2.14	0.51	0.82[a]	0.02	9.6	4.5	0.62	0.06
'A5.2 Subtidal sand'	Inside	7	61	162	50	18.7[b]	2.0	5.07[a,b]	2.12	1.72[a]	0.12	0.70	0.05	5.8[a]	0.5	0.67[b]	0.01
	Outside	0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
'A5.3 Subtidal mud'	Inside	1	22	287	-	22.0[a,b]	-	12.81[a,b]	-	1.11[a]	-	0.47[a]	-	3.0[a]	-	0.78[a,b]	-
	Outside	1	14	50	-	14.0	-	0.51	-	2.12	-	0.82	-	8.4	-	0.70	-
'A5.4 Subtidal mixed sediments'	Inside	14	287	1208	309	90.0[a]	8.1	7.08[a]	1.19	2.67[a]	0.30	0.74[a]	0.07	22.9[a]	5.5	0.77[a]	0.02
	Outside	12	283	1332	294	96.5	9.8	29.3	20.7	2.51	0.26	0.74	0.05	17.8	4.6	0.79	0.03

3.2 Subtidal sediment BSH

Sediment composition and biological communities

Sixty-seven percent of the sediment samples were classified as 'A5.4 Subtidal mixed sediments' (Figures 4 and 5). Overall, there were significant differences between infaunal community species composition and sediment type (ANOSIM, global R = 0.808, p < 0.01). This is further illustrated by the the nMDS plot (Figure 8) where infaunal communites are generally grouped in ordination space in relation to the BSH from which they were sampled. Finer, less heterogenous sediments being relatively species poor and low in abundance. In addition, aggregations of the Ross Worm (*Sabellaria spinulosa*) tubes were often found within the mixed sediments, a key species which exerts a modifying structural and biological effect on the rest of the community. Across all BSH, there was no significant difference in the Infaunal Quality Index (IQI) inside and outside the MCZ boundary (t = -0.68, p = 0.507).

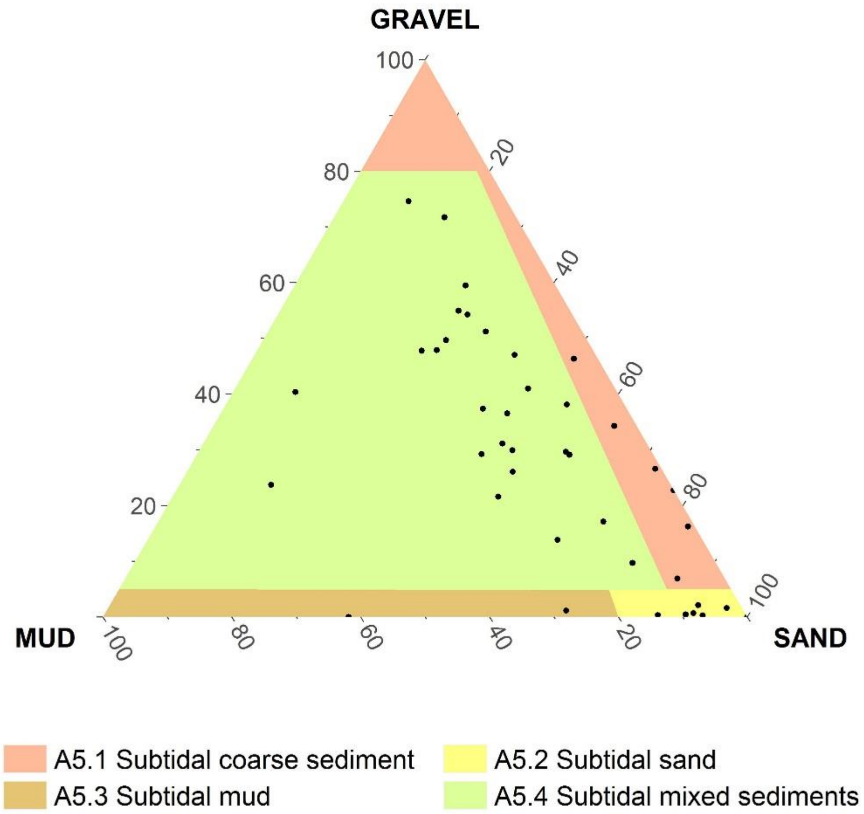

Figure 5. Classification of particle size distribution (half phi) information for each of the 2016 Dover to Folkestone MCZ 0.1 m^2 Day Grab samples (closed black circles) into one of the sedimentary Broadscale Habitats (coloured areas) The points are plotted on a true scale subdivision of the Folk triangle into the simplified classification for UKSeaMap (Long, 2006; Folk, 1954) (© Natural England and Environment Agency 2022).

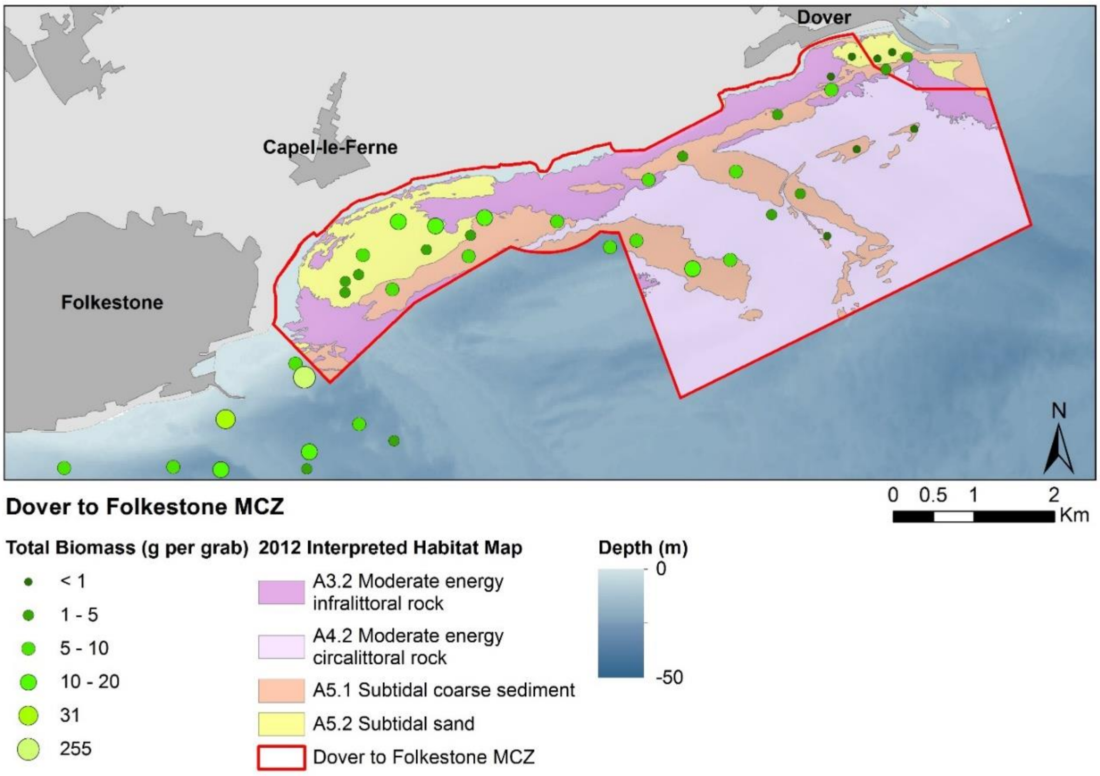

Figure 6. Spatial pattern of biomass (g wet weight) by Broadscale Habitat for Day Grabs sampled in the 2016 Dover to Folkestone MCZ survey (© Natural England and Environment Agency 2022).

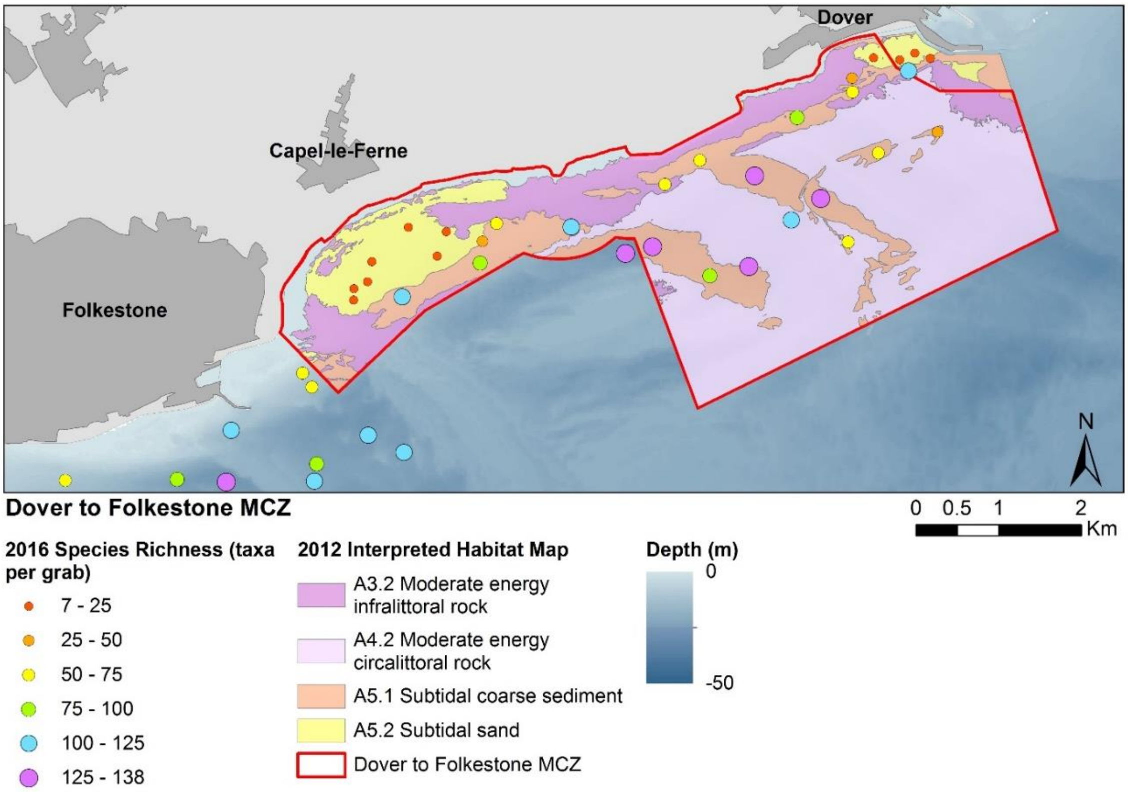

Figure 7. Spatial pattern of species richness (number of taxa per grab) by Broadscale Habitat for Day Grabs sampled in the 2016 Dover to Folkestone MCZ survey (© Natural England and Environment Agency 2022).

3.2.1 Subtidal coarse sediment

The mean percentage mud, sand and gravel content of samples in this BSH were 3%, 72% and 26% respectively. The samples from this BSH recorded 47% (n = 195) of all taxa (n = 419) identified within the MCZ.

There was a low mean similarity (16%) among benthic communities assigned to 'A5.1 Subtidal coarse sediment' within the MCZ. The small brittlestar *Amphipholis squamata*, the amphipod *Socarnes erythrophthalmus* and the polychaete *Glycera lapidum* were the three greatest contributors to similarity (Table 5). They were consistently present across the four stations but the infaunal assemblages did vary considerably, as evidenced by the spread of points in the MDS (Figure 8).

Clustering with associated SIMPROF analysis showed that the stations assigned to 'A5.1 Subtidal coarse sediment' inside the MCZ grouped into three separate communities. Adjacent stations DVFK06 and 23 were characterised by *A. squamata* and *S. erythrophthalmus* (cluster *e*, Figure 8, Table 6), whilst DVFK12 was a distinct community with high abundances of the as the Ross Worm *Sabellaria spinulosa* and the encrusting barnacles *Verruca stroemi* and *Balanus crenatus*. DVFK03 was part of a separate cluster, associated with adjacent 'A5.4 Subtidal mixed sediments' stations characterised by the ascidian *Dendrodoa grossularia* and polychaete *Praxillella affinis* (cluster *i*) (Figure 9)

Although the number of samples was too low for a robust comparison of this BSH inside and outside the MCZ, the two stations outside the MCZ were >1 km from the coarse sediment samples inside the MCZ (Figure 3). Clustering with associated SIMPROF analysis showed that the samples collected outside the MCZ were not associated with any 'A5.1 Subtidal coarse sediment' communities inside the MCZ, and instead had communities that were similar to adjacent samples of a different BSH (Figure 8, Table 6). These samples were characterised by species associated with sandy communities, such as the polychaetes *Spiophanes bombyx* and *Lanice conchilega*. Of the six samples assigned as 'A5.1 Subtidal coarse sediment', only three of the associated infaunal samples were characterised as coarse sediment biotopes, indicating that the biotope characterisations stretch across the PSA-defined BSH boundaries (Figure 9).

The infauna of stations from this BSH were had a significantly higher taxa richness than samples from the 'A5.2 Subtidal sand' BSH (one-way ANOVA, $F_{[3,\ 22,]} = 11.35$, $p<0.001$), although there was a large variation in the number of samples collected in each BSH (Table 4).

Three out of the four stations within the MCZ were classified as "High" using the IQI index, with one being at "Good" status, suggesting low levels of chemical or organic enrichment pressures in these areas. Notably, site DVFK60 outside the MCZ was one of only two individual stations with an IQI of 'Moderate' status (IQI = 0.56, 'Moderate' status is between 0.54 – 0.64). The other station was the adjacent DVFK63 (IQI = 0.57). Both stations were close to Dover harbour wall (Figure 2).

Table 5. The top five species that characterise each BSH (sampled inside and outside the Dover to Folkestone MCZ site boundary), assessed using SIMPER analysis on untransformed abundance data (© Natural England and Environment Agency 2022). Groups with two or less samples were not included.

A5.1 Subtidal coarse sediment – inside MCZ		**A5.2 Subtidal sand**	
Species	**% contribution to characterisation**	**Species**	**% contribution to characterisation**
Amphipholis squamata	15.57	*Nucula nitidosa*	49.89
Socarnes erythrophthalmus	6.59	*Magelona johnstoni*	19.16
Glycera lapidum	6.38	*Spiophanes bombyx*	14.83
Sabellaria spinulosa	5.03	*Fabulina fabula*	5.42
Nemertea	4.57	*Nephtys hombergii*	2.41
A5.4 Subtidal mixed sediments – inside MCZ		**A5.4 Subtidal mixed sediments – outside MCZ**	
Sabellaria spinulosa	26.16	*Sabellaria spinulosa*	42.91
Lumbrineris aniara/cingulata	6.62	*Lumbrineris aniara/cingulata*	5.28
Mediomastus fragilis	6.18	Actiniaria	4.37
Ampelisca diadema	6.00	*Molgula manhattensis*	4.31
Abra alba	4.61	*Ampelisca diadema*	3.06

a)

b)

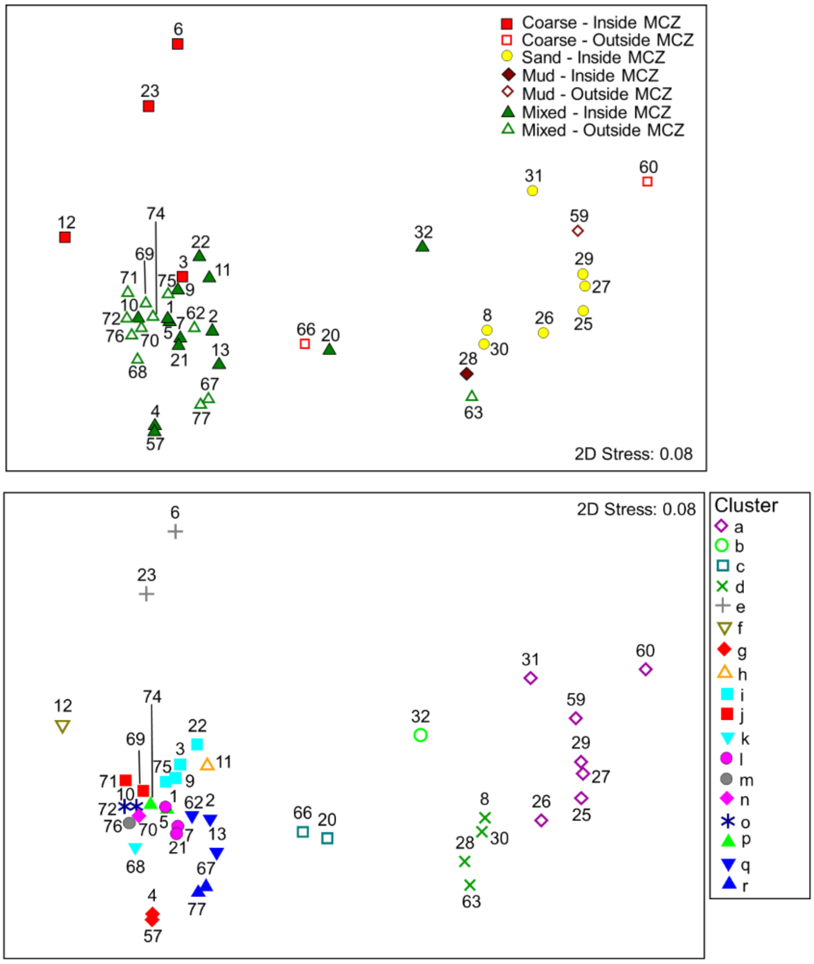

Figure 8. Nonmetric multidimensional scaling (nMDS) plot of infaunal communities (sieved to 1.0 mm) sampled in the 2016 Dover to Folkestone MCZ survey, grouped by (a) assigned sediment Broadscale Habitats, and (b) groupings of stations with significantly different community structure, derived from SIMPROF analysis (© Natural England and Environment Agency 2022). The point labels indicates the station number (minus the DVFK prefix).

Table 6. The top five species that characterise each community defined by SIMPROF analysis, assessed using SIMPER analysis on untransformed abundance data from the 2016 Dover to Folkestone MCZ survey (© Natural England and Environment Agency 2022). SIMPROF-defined communities composed of one sample are not listed.

Group 'a' (Sand, inside MCZ; Mud and Coarse sediment, outside MCZ) (n = 7)		**Group 'c' (Mixed sediments, inside MCZ; Coarse sediment, outside MCZ) (n = 2)**	
Species	**% contribution to characterisation**	**Species**	**% contribution to characterisation**
Magelona johnstoni	34.38	*Mytilus edulis*	27.17
Nucula nitidosa	28.83	*Spiophanes bombyx*	26.45
Spiophanes bombyx	9.18	*Lanice conchilega*	17.03
Nephtys cirrosa	6.73	*Nucula nitidosa*	4.35
Chaetozone christiei	5.71	*Abra alba*	4.35
Group 'd' (Sand and Mud, inside MCZ; Mixed sediments, outside MCZ) (n = 4)		**Group 'e' (Coarse sediment, inside MCZ) (n = 2)**	
Nucula nitidosa	48.14	*Amphipholis squamata*	15.56
Spiophanes bombyx	37.20	*Socarnes erythrophthalmus*	11.11
Nephtys hombergii	2.67	*Glycera lapidum*	8.89
Abra alba	2.52	*Goodallia triangularis*	4.44
Magelona johnstoni	2.10	*Lagotia viridis*	2.22
Group 'g' (Mixed sediments, inside MCZ) (n = 2)		**Group 'i' (Mixed sediments, inside and outside MCZ' Coarse sediment, inside MCZ) (n = 4)**	
Ampelisca diadema	94.02	*Dendrodoa grossularia*	9.46
Abra alba	1.40	*Praxillella affinis*	8.55
Jasmineira elegans	0.79	*Lumbrineris aniara/cingulata*	8.29
Lumbrineris aniara/cingulata	0.55	*Sabellaria spinulosa*	6.52
Sabellaria spinulosa	0.55	*Ampelisca spinipes*	4.89
Group 'j' (Mixed sediments, outside MCZ) (n = 2)		**Group 'l' (Mixed sediments, outside MCZ) (n = 3)**	
Sabellaria spinulosa	33.82	*Sabellaria spinulosa*	65.53
Lumbrineris aniara/cingulata	8.67	*Molgula manhattensis*	3.03
Amphipholis squamata	6.94	*Unciola crenatipalma*	2.50
Molgula manhattensis	6.94	*Dendrodoa grossularia*	2.32
Actiniaria	5.49	*Lumbrineris aniara/cingulata*	2.27
Group 'o' (Mixed sediments, inside and outside MCZ) (n = 2)		**Group 'p' (Mixed sediments, inside and outside MCZ) (n = 2)**	
Sabellaria spinulosa	62.46	*Sabellaria spinulosa*	22.04
Molgula manhattensis	8.48	*Molgula manhattensis*	9.72
Lumbrineris aniara/cingulata	2.60	*Lumbrineris aniara/cingulata*	7.82
Actiniaria	2.31	*Mediomastus fragilis*	4.74
Amphipholis squamata	1.73	*Ampharete lindstroemi*	3.79
Group 'q' (Mixed sediments, inside and outside MCZ) (n = 3)		**Group 'r' (Mixed sediments, outside MCZ) (n = 2)**	
Sabellaria spinulosa	62.46	*Sabellaria spinulosa*	32.00
Mediomastus fragilis	8.48	*Mytilus edulis*	26.77
Ampharete lindstroemi	2.60	*Balanus crenatus*	18.46
Abra alba	2.31	*Mediomastus fragilis*	6.77
Molgula manhattensis	1.73	*Phyllodoce mucosa*	2.15

‘A5.1 Subtidal coarse sediment’ (SS.SCS.ICS.MoeVen)

***Moerella spp.* with venerid bivalves in infralittoral gravelly sand © Environment Agency and Natural England 2016.**

‘A5.1 Subtidal coarse sediment’ (SS.SCS.CCS.MedLumVen)

***Mediomastus fragilis, Lumbrineris spp.* and venerid bivalves in circalittoral coarse sand or gravel.**

Figure 9. Example images of benthic samples in the 0.1 m^2 Day Grab and on the sieve assigned to the ‘A5.1 Subtidal coarse sediment’ feature within the Dover to Folkestone MCZ (© Environment Agency and Natural England 2016).

3.2.2 Subtidal sand

The mean percentage mud, sand, and gravel content of samples in this BSH were 7%, 93% and 1% respectively. The samples from this BSH accounted for 15% of all taxa (n = 61) recorded within the MCZ and this BSH was not recorded outside the MCZ. Representative images of samples from this BSH are shown in Figure 10.

The taxa richness of samples from this BSH were significantly lower than samples from ‘A5.1 Subtidal coarse sediment’ and ‘A5.4 Subtidal mixed sediments’ habitats (one-way ANOVA, $F_{[3, 22,]} = 11.35$, $p < 0.001$), although there was a large variation in the number of samples collected in each BSH (Table 4).

‘A5.2 Subtidal sand’ (SS.SSA.CMuSa.AalbNuc)

***Abra alba* and *Nucula nitidosa* in circalittoral muddy sand or slightly mixed sediment. ©**

‘A5.2 Subtidal sand’ (SS.SSA.IFiSa.NcirBat)
***Nephtys cirrosa* and *Bathyporeia* spp. in infralittoral sand.**

Figure 10. Example images of benthic samples in the 0.1 m2 Day Grab and on the sieve assigned to the ‘A5.2 Subtidal sand’ feature within the Dover to Folkestone MCZ (© Environment Agency and Natural England 2016).

There was a moderate mean similarity (38%) among benthic communities assigned to ‘A5.2 Subtidal sand’ within the MCZ. The Nut Shell *Nucula nitidosa*, and the polychaetes *Magelona johnstoni* and *Spiophanes bombyx* were the species that best characterised the BSH and were the three greatest contributors to similarity (Table 5). Clustering with associated SIMPROF analysis showed that there were two distinct communities within the BSH, defined by differing abundances of *N. nitidosa* and *M. johnstoni* (clusters *a* and *d*, Figure 8, Table 5). The MNCR biotopes assigned to the samples followed a similar distinction in community structure (Figure 13).

All seven samples were classified as “Good” using the WFD IQI index. This suggests limited levels of chemical or organic enrichment pressures in these areas. The IQI scores of this BSH were the lowest of the BSHs within the MCZ and were significantly lower than those from ‘A5.4 Subtidal mixed sediments’ (Table 4; one-way ANOVA, $F_{[3, 22,]} = 4.23$, $p < 0.05$).

‘A5.3 Subtidal mud’ (SS.SSA.CMuSa.AalbNuc)

***Abra alba* and *Nucula nitidosa* in circalittoral muddy sand or slightly mixed sediment.**

‘A5.3 Subtidal mud’ (SS.SSA.IMuSa.FfabMag)

***Fabulina fabula* and *Magelona mirabilis* with venerid bivalves and amphipods in infralittoral compacted fine muddy sand.**

Figure 11. Example images of benthic samples in the 0.1 m^2 Day Grab and on the sieve assigned to the ‘A5.3 Subtidal mud’ feature within the Dover to Folkestone MCZ (© Environment Agency and Natural England 2016).

3.2.3 Subtidal mud

Two samples were assigned to the ‘A5.3 Subtidal mud’ BSH, one outside and one inside the MCZ boundary. The mean percentage content of mud, sand, and gravel in this BSH was 45%, 55% and 1% respectively.

The sample from this BSH accounted for 6% (n = 22) of all taxa recorded within the MCZ. Clustering with associated SIMPROF analysis showed that the station inside the MCZ, DVFK28, was associated with adjacent stations in the northwest of the MCZ assigned as ‘A5.2 Subtidal sand’ communities (cluster *d*) and characterised by *N. nitidosa* and the polychaete *S. bombyx.* The ‘A5.3 Subtidal mud’ station sampled outside of the MCZ was also associated with a cluster of stations mostly composed of ‘A5.2 Subtidal sand’ samples (cluster *a*) (Figure 8, Table 6). This association with sand samples is also reflected in both samples being assigned muddy sand MNCR biotopes (Figure 13). Some representative images of samples from this BSH are shown in Figure 11.

The infauna from this BSH had a lower taxa richness than samples from the ‘A5.1 Subtidal coarse sediment’ and ‘A5.4 Subtidal mixed sediments’ habitats (Table 4; one-way ANOVA, $F_{[3, 22,]} = 11.35$, $p < 0.001$). The IQI scored station DVFK28 inside the MCZ boundary as “High” status using WFD threshold values, and the sample outside (DVFK59) at “Good” status.

In the 2012 verification survey, one station was recorded as ‘A5.3 Subtidal Mud’. This station (DVFK56) was resampled in 2016 and was recorded as ‘A5.4 Subtidal Mixed Sediments’.

3.2.4 Subtidal mixed sediments

The mean percentage content of mud, sand, and gravel in this BSH was 18%, 49% and 34% respectively. The samples from this BSH accounted for 69% of all taxa (n = 287) recorded within the MCZ

There was greater heterogeneity in the sediment composition of this BSH outside the MCZ compared to within which was attributable to two stations (DVFK62 and DVFK70) with relatively high proportions of mud (51% to 62%), and another station (DVFK67) with a low proportion of mud (11%). Overall, there was a statistical difference (ANOSIM, global $R = 0.099$, $p = 0.03$) but given the low R value the sediments are considered comparable, on average.

The infauna in stations assigned to ‘A5.4 Subtidal mixed sediments’ had a significantly higher taxa richness than stations from ‘A5.2 Subtidal sand’ and ‘A5.3 Subtidal mud’ habitats (Table 4; one-way ANOVA, $F_{[3, 22,]} = 11.35$, $p < 0.001$).

Eleven out of the fourteen samples within the MCZ were classified as “High” using the WFD IQI, with the other three being at “Good” status. This suggests low levels of chemical or organic enrichment pressures in these areas. The IQI scores of this BSH were statistically higher than the sand habitat (Table 4; one-way ANOVA, $F_{[3, 22,]} = 4.23$, $p < 0.05$).

There was a low mean similarity (22%) among benthic communities assigned to ‘A5.4 Subtidal mixed sediments’ within the MCZ (Table 5). The Ross Worm *Sabellaria spinulosa*, and the polychaetes *Lumbrineris* sp. and *Mediomastus fragilis* were the three greatest contributors to similarity. Many samples within this BSH were assigned to a *S. spinulosa* reef biotope (SS.SBR.PoR.SspiMx, A5.611, Figures 12 and 13). The mean dissimilarity between ‘A5.4 Subtidal mixed sediments’ and ‘A5.1 Subtidal coarse sediment’ was 87%, predominately due to the higher abundances of *S. spinulosa* and the amphipod *Ampelisca diadema* at Mixed stations, and higher abundances of the Baked Bean Ascidian *Dendrodoa grossularia* and the barnacle *Balanus crenatus* at Coarse stations.

‘A5.4 Subtidal mixed sediments’ (SS.SBR.PoR.SspiMx)

Sabellaria spinulosa **on stable circalittoral mixed sediment.**

‘A5.4 Subtidal mixed sediments’ (SS.SMX.CMx.OphMx)

Ophiothrix fragilis **and/or** ***Ophiocomina nigra*** **brittlestar beds on sublittoral mixed sediment.**

‘A5.4 Subtidal mixed sediments’ (SS.SMU.ISaMu.AmpPlon)

Ampelisca spp., Photis longicaudata **and other tube building amphipods and polychaetes in infralittoral sandy mud.**

Figure 12. Example images of benthic samples in the 0.1 m^2 Day Grab and on the sieve assigned to the ‘A5.4 Subtidal mixed sediments’ feature within the Dover to Folkestone MCZ (© Environment Agency and Natural England 2016).

The low within-BSH similarity is reflected in the clustering and associated SIMPROF analysis, with samples from both inside and outside the MCZ assigned to the 'A5.4 Subtidal mixed sediments' BSH split into 11 distinct communities (Table 6, Figure 8) and a range of associated MNCR biotopes (Figure 13). These communities are predominately defined by varying abundances of *S. spinulosa, Lumbrineris* sp. and *Mediomastus fragilis*, as well as the polychaete *Ampharete lindstroemi* ascidian *Molgula manhattensis*. Notably one community of two samples from the northeast of the MCZ (cluster *g,* Table 6) were characterised by the high abundance of the amphipod *Ampelisca diadema* (>3000 individuals per sample) forming biogenic structures with closely packed tubes, and assigned the *Ampelisca*-dominated sediment biotope (SS.SMU.ISaMu.AmpPlon, A5.335, Figure 12).

There was no significant difference in the infauna community composition between the stations assigned to 'A5.4 Subtidal mixed sediments' inside and outside the MCZ (ANOSIM Global R = 0.03, $p > 0.05$; Figure 8). Similarly, there was no difference in univariate metrics between 'A5.4 Subtidal mixed sediments' stations sampled inside and outside the MCZ. There were, however, two stations (DVFK67 and DVFK68) that were assigned as mussel reef biotopes and these had correspondingly high biomass values (see Table 4).

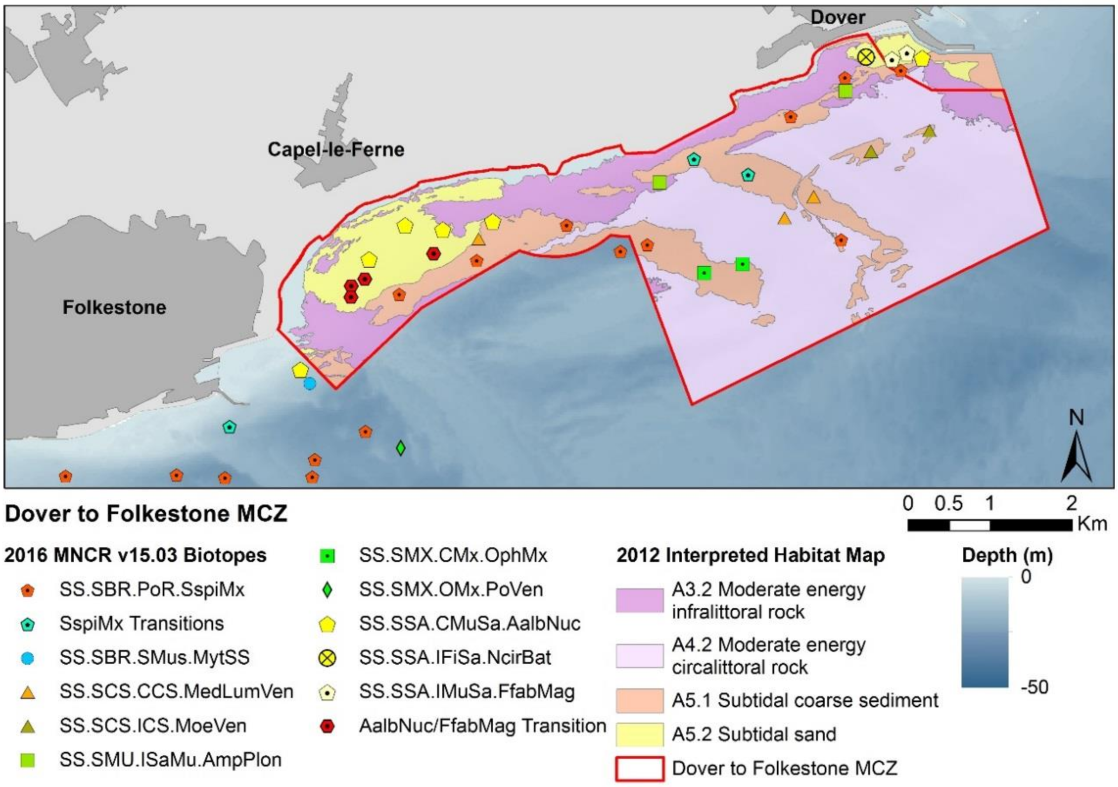

Figure 13. Distribution of *Sabellaria* and other biotopes at stations sampled inside and outside the Dover to Folkestone MCZ (© Natural England and Environment Agency 2022).

3.3 Habitat Features of Conservation Importance (FOCI)

3.3.1 Other Habitat FOCI

To fulfil Objective 2 of the report, two potential Habitat FOCI were recorded during the survey. Dover to Folkestone MCZ is not designated for 'Ross Worm (*Sabellaria spinulosa*) Reefs' but many of the infaunal samples from 'A5.4 Subtidal mixed sediments', both inside and outside the Dover to Folkestone MCZ, were assigned to a *Sabellaria spinulosa* reef biotope (Figures 13 and 14). The average abundance per grab of *Sabellaria spinulosa* in stations assigned to reef biotopes, inside and outside the site, was 657 worms, compared to 11 worms in non-reef biotopes.

Sabellaria spinulosa reefs are spatially extensive structures, that are distinctly raised above the surrounding seabed, and which can persist for a number of years (UK Biodiversity Action Plan Priority Habitat Descriptions. *Sabellaria* Reefs. 2008). *Sabellaria spinulosa* is actually widely distributed around the UK, often forming spatially localised and temporary crusts or aggregations that are not considered to be true reefs (UK Biodiversity Action Plan Priority Habitat Descriptions. *Sabellaria* Reefs 2008). The grab photographs do not indicate any obvious reef structures.

The site is also not designated for 'Blue Mussel Beds (including intertidal beds on mixed and sandy sediments)' but two stations (DVFK67 and DVFK68) west of the site boundary were assigned to the 'Blue Mussel Beds' biotope, with 517 and 175 *Mytilus edulis* enumerated respectively (SS.SBR.SMus.MytSS, A5.625). A smaller number of adult Blue mussels (39) were also found at station DVFK 20 inside the MCZ, but this was not characterised as a 'Blue Mussel Beds' biotope.

3.4 Species Features of Conservation Importance (FOCI)

3.4.1 Native Oyster (*Ostrea edulis*)

The Native Oyster (*Ostrea edulis*) was not observed during the survey or recorded in the infaunal data.

3.4.2 Other Species FOCI

The surveys reported here were not designed to specifically monitor (or identify the presence of) species FOCI. As such, this should not be interpreted as an absence of these species FOCI from the site.

3.5 Non-indigenous species (NIS)

Both the Slipper limpet *Crepidula fornicata* and the Australian barnacle *Austrominius modestus* were present in low numbers at one station within the MCZ (the coarse sediment station DVFK12). Four *A. modestus* individuals were also identified in a sample collected for confirming the presence of *Sabellaria spinulosa* from station DVFK47. Outside the MCZ, these species were present in low numbers at a total of seven mixed sediment stations (Figure 15).

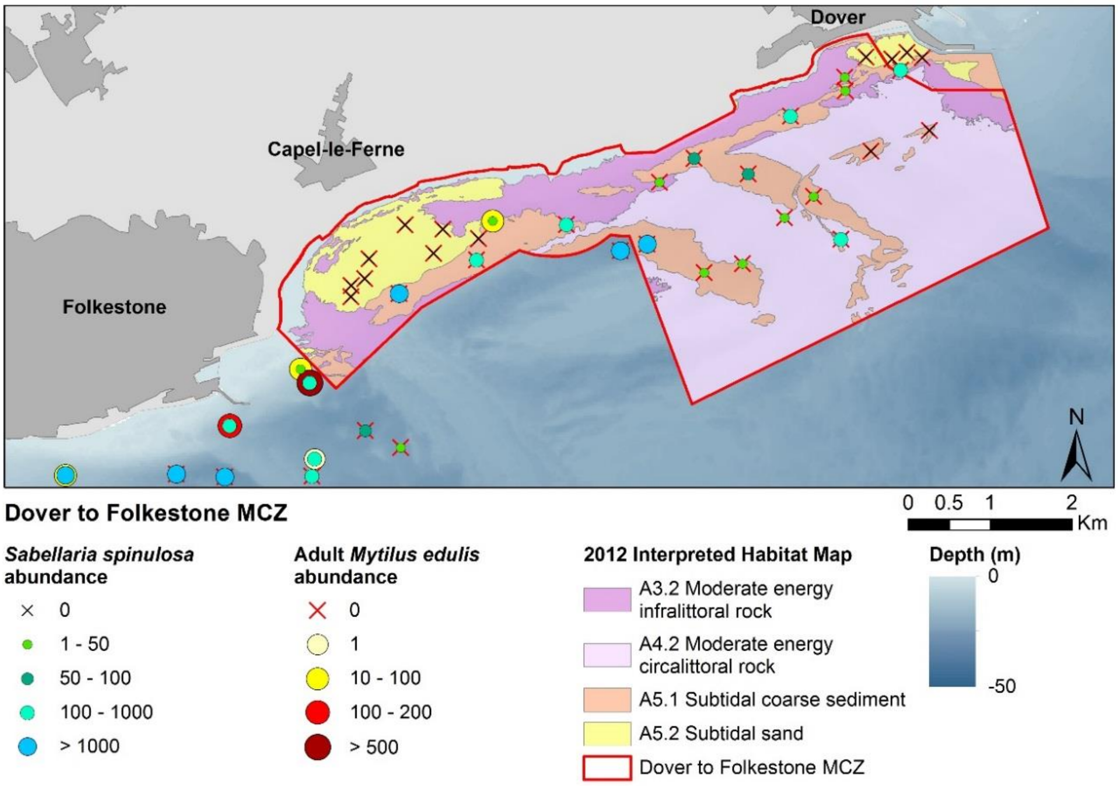

Figure 14. Abundance of the Ross Worm *Sabellaria spinulosa* and Blue Mussel *Mytilus edulis* at stations inside and outside the Dover to Folkestone MCZ (© Natural England and Environment Agency 2022).

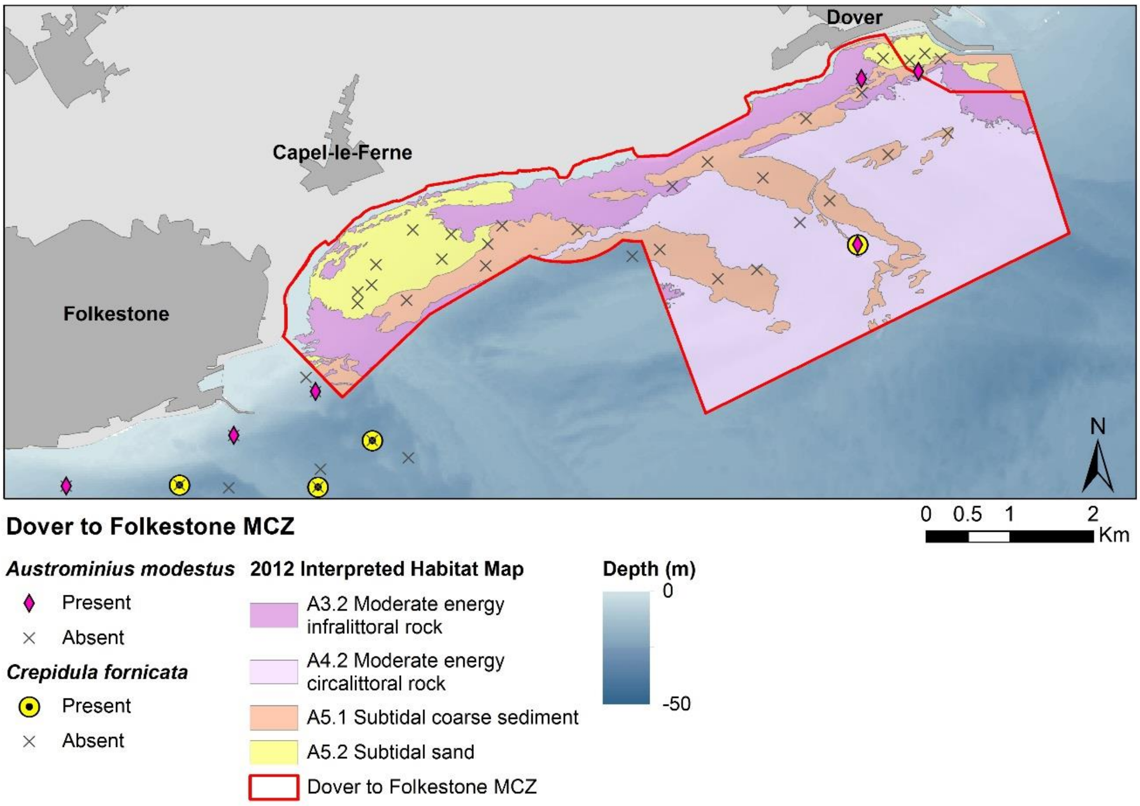

Figure 15. Abundance of non-native species at stations inside and outside the Dover to Folkestone MCZ (© Natural England and Environment Agency 2022).

3.6 Supporting processes

3.6.1 Sediment quality parameters

Four sediment contaminant samples were collected to provide information for the 'Sediment contaminants' supporting processes attribute (Report Objective 1) for the designated sediment features (Table 2). Station DVFK31 just outside Dover harbour exceeded the OSPAR EAC/ERLs for mercury, chromium, lead, and seven polycyclic aromatic hydrocarbons (PAHs) and two polychlorinated biphenyls (PCBs). The IQI score at this station was the third lowest recorded, although the sample was still classified as "Good". It is possible that shipping, the activities of the port or associated dredging may have led to contamination of nearby sediments but this study is not designed to assess the source of contamination and there are too few samples to carry out a robust spatial analysis of sediment contamination.

Levels of contaminants at the other three stations were generally less than the EAC/ERL threshold, with the exception of Chromium which exceeded the threshold at all stations (the OSPAR background level EAC and elevated ERL thresholds are set as the same concentration for chromium).

The recorded exceedances of EAC thresholds for organic compounds should also be treated with caution as the multiplication factors used for normalisation were large (6.7 to 50 for PAHs and PCBs) because some stations contained little organic carbon (Annex 7).

3.7 Additional monitoring requirements

3.7.1 Marine litter

Plastic fragments greater than 1 mm were recorded in 18 out of 48 infaunal samples, with nylon, wire (and plastic coating) and twine noted (Objective 3 of the survey). The number of pieces per sample ranged from 1 to 4 pieces and they corresponded to "A14. Other" sub-category of the OSPAR/ICES/IBTS marine litter classification system.

4 Discussion

This discussion presents evidence for future assessment and monitoring of designated features of the Dover to Folkestone MCZ, as required to achieve the report objectives stated in Section 1.2.3.

4.1 Subtidal sedimentary BSH

All four designated sediment BSHs were identified in the grab survey: ‘A5.1 Subtidal coarse sediment’, ‘A5.2 Subtidal sand’, ‘A5.3 Subtidal mud’ and ‘A5.4 Subtidal mixed sediments’.

4.1.1 Extent and distribution

The proportion and location of samples assigned to ‘A5.2 Subtidal sand’ (mostly restricted to East Wear Bay) was broadly in line with the acoustically derived habitat map used to plan the survey. This suggests that sediment composition across this area has been stable in the short term (Figure 3).

Stations DVFK31, DVFK59 and DVFK60 that targeted a small area of sand outside of Dover harbour were determined to be sand, mud and coarse sediment respectively. This could suggest that this area is more dynamic in terms of sediment stability or there is some finer scale patchiness not adequately represented by the habitat map.

The distribution of ‘A5.1 Subtidal coarse sediment’ (5 samples) and ‘A5.4 Subtidal mixed sediments’ (12 samples) differs from acoustically-derived habitat map generated from the 2012 verification survey (Colenutt *et al.,* 2015, Figure 3). Certain approaches to interpreting acoustic data struggle to differentiate between areas of mixed and coarse sediment (Diesing *et al.,* 2014) and such areas could be better defined as a mosaic of the two BSHs. Additionally, the south and west areas mapped as ‘A5.1 Subtidal coarse sediment’ were only groundtruthed with still images in the 2012 verification survey, and therefore would have only assessed the surface of the sediment. These two factors combined may have led to the underrepresentation of the ‘A5.4 Subtidal mixed sediments’ BSH in that area of the MCZ, rather than any change in feature extent.

‘A5.3 Subtidal mud’ was not mapped on the 2012 acoustically-derived habitat map, and was only recorded as a point sample. This station was resampled in 2016 and was recorded as ‘A5.4 Subtidal mixed sediments’ (DVFK56). Similarly, only one station was recorded as ‘A5.3 Subtidal mud’ inside the MCZ in this survey, in an area expected to be ‘A5.2 Subtidal sand’. This suggests that the BSH is present only in small patches, which could be subject dynamic temporal change.

4.1.2 Structure and function: Biological communities

The stations inside and outside at Dover to Folkestone MCZ were assigned to 18 infauna communities (Figure 8, Table 6), many of which were primarily composed of stations assigned of the ‘A5.4 Subtidal mixed sediments’ BSH. A number of species were recorded at high densities across the site and have characteristics that will be important for altering habitat structure; *Ampelisca diadema*, *Mytilus edulis* and *Sabellaria spinulosa*.

Two stations within the 'A5.4 Subtidal mixed sediments' BSH were assigned to the SS.SMU.ISaMu.AmpPlon biotope (*Ampelisca* spp., *Photis longicaudata* and other tube building amphipods and polychaetes in infralittoral sandy mud). An example of this biotope is shown in the bottom row of Figure 10 where the flattened, flexible mud-covered tubes are clearly visible due to the high density of *Ampelisca diadema*. The tubes have the potential to alter the sedimentary and biogeochemical properties of the seabed surface due to modification of seabed to water column exchange processes, as well as through the presence of faecal pellets produced by the amphipods (see Rigolet *et al.,* 2014, Woodin *et al.,* 2010 and references therein).

Ampelisca diadema dominates both the abundance and the biomass of these samples and they form distinct assemblages from the rest of the mixed sediment samples (Figure 8, Table 6). When this species was removed from the dataset, multivariate community analysis still identified the community at these two stations as being distinct from the other mixed sediments stations. Although far from conclusive, this suggests that the presence of the *Ampelisca* tubes affects which other taxa are found locally (Rigolet *et al.,* 2014).

The Blue Mussel *Mytilus edulis* is thought to be an important species in modifying the environment. Mussel beds alter the benthic-water exchange, provide increased structural complexity and associated niches, and provide hard surfaces for the attachment of other organisms. The production of a layer faeces and pseudo-faeces below the mussels also provides habitat and nutrients for other species (Mainwaring *et al.,* 2014, McLeod *et al.,* 2014 and references therein).

Two adjacent nearshore stations to the south-west of the Dover to Folkestone MCZ were assigned to the 'Mytilus edulis beds on sublittoral sediment' (SS.SBR.SMus.MytSS, A5.625) biotope (one definitively, and one as a transition biotope). Another adjacent station was also high in numbers and biomass of mussels suggesting that there may be a sizeable mussel bed just outside the western boundary of the MCZ.

There were eight stations within the MCZ, and nine outside, which were assigned to *Sabellaria* biotopes. There is good evidence that, irrespective of the presence of *Sabellaria*, there are communities here distinct from non-*Sabellaria* biotopes in the 'A5.4 Subtidal mixed sediments' BSH. Moreover, when excluding *Sabellaria* from the data, diversity (species richness, Simpson's Evenness 1-λ') was greatest in the samples from the *Sabellaria* reef biotopes compared to the non-reef mixed sediment samples (one-way ANOVA, transformed data, $p < 0.05$). There was no evidence, however, of a relationship between infaunal diversity and *Sabellaria* abundance.

Within the 'A5.3 Subtidal sand' BSH, *Nucula nitidosa* was a key species. This shallow-burrowing deposit feeding bivalve is not noted for being a habitat engineer (see

Sabatini and Ballerstedt, 2008) but individuals will obviously contribute to bioturbation and energy transfer among trophic levels.

The sampling in this survey was sufficient to detect differences in abundance, species richness, IQI and multivariate species composition between different BSH within the MCZ (Annex 4), with the exception of ‘A5.3 Subtidal Mud’ where only one sample was collected, although only the IQI achieved. As only four infauna samples were collected in the 2012 verification survey (one from each BSH), this inhibited the process of estimating the number of samples required to detect change (following the recommendations of Marubini (2014)). *A posteriori* power analysis indicated that sampling effort within the ‘A5.4 Subtidal mixed sediments’ BSH should be increased in future surveys if a BACI approach is to be applied to univariate indices of community structure (Annex 4).

The mean Infaunal Quality Index across the site indicated that ‘A5.1 Subtidal Coarse Sediment’, ‘A5.3 Subtidal mud’ and ‘A5.4 Subtidal mixed sediments’, were at ‘High’ ecological status (IQI of > 0.75) for equivalent Water Framework Directive classifications, and ‘A5.2 Subtidal sand’ at ‘Good’ status (IQI of 0.64-0.75). This suggests that there is limited anthropogenic pressures impacting the site. It should be noted, that the IQI has not been shown to consistently respond to the presence of non-indigenous species (notably *Crepidula fornicata*) or abrasion pressure from fishing activities on faunal communities (Phillips and Green, in prep). At the time of writing, the site is still open to fishing using bottom-towed gear, so this cannot be ruled out as potentially impacting the sediment features of the site.

4.2 Species Features of Conservation Importance (FOCI)

The Dover to Folkestone MCZ is designated to protect the Native Oyster (*Ostrea edulis*) feature.

4.2.1 Presence and distribution of species

Although the survey was not designed to specifically assess the species features of this site, no evidence was found of Native Oysters in the Dover to Folkestone MCZ. Oysters were not recorded in the verification survey in 2012 and the Selection Assessment Document (SAD) only cites four records. However, this should not be interpreted as the species is absent from the site. Future monitoring should involve a survey tailored to baseline Native Oyster populations in the site, possibly through the use of a dive survey for close examination of seabed habitats.

4.3 Non-indigenous species (NIS)

Both non-native species recorded, *Crepidula fornicata* and *Austrominius modestus*, are often found on coarse sediments on the south coast of England. The survey generated no evidence that *Crepidula fornicata* was acting as a habitat structure by forming extensive beds, as the species was only found at one station within the MCZ. Similarly, numbers of the native *Balanus crenatus* were greater than *Austrominius* *modestus* by an order of magnitude. Neither species were recorded in the 2012 verification survey of the MCZ, although only four samples were collected for infauna analysis (Colenutt *et al*., 2015), so it is likely they were present but not sampled.

4.4 Supporting processes

4.4.1 Sediment BSH; Supporting processes: Sediment contaminants

Sediment contamination is monitored under the OSPAR Convention for the contaminants of greatest concern, either in terms of toxicity, persistence, or bioaccumulation. The contaminants found within the Dover to Folkestone MCZ were assessed using the same BACs, ERL/EACs as the OSPAR assessments (OSPAR Commission 2012).

Trends in sediment contamination by cadmium, mercury and lead are generally downwards within the OSPAR regions but levels of mercury and lead still occur above the ERL in the North Sea and English Channel, particularly around the estuaries of large rivers (OSPAR Commission, 2012). Both cadmium and lead were above the ERL at a station in close proximity to the port of Dover, suggesting a possible source of these metals.

Within the OSPAR regions, both PCBs and PAHs are generally above their EACs (OSPAR Commission, 2012). At the station in close proximity to the port, seven out of ten PAHs and two out of seven PCBs were above the EACs. PAHs and PCBs at the other stations were generally below the EACs, again suggesting that the port and associated activities may be a source of contaminants.

Although the contaminants near the port are above the EAC/ERLs, which indicates potential for an ecological impact, and the multivariate faunal composition at this station is distinct from other sandy stations, there isn't strong evidence of an impact. Species composition may simply be different because the area of sand is geographically distinct from the main sandy area in East Wear Bay. The sample consisted mainly of the small amphipod *Bathyporeia guilliamsoniana* and amphipods are generally considered to be sensitive to pollutants (De la Ossa-Carretero *et al.,* 2012).

PCB 118 is the most toxic congener (OSPAR Commission, 2012) and was above the EAC at three of the four stations. There was however no evidence of an adverse ecological impact inside the site, as the mean Infaunal Quality Index (IQI) score for each BSH (coarse, mud and mixed) was > 0.75, which is equivalent to 'High Ecological Status' for the Water Framework Directive (Table 4) and above 0.64 (Good Ecological Status) for the sand BSH.

4.5 Additional monitoring requirements

4.5.1 Marine litter

Trawl surveys of the seabed have found that the English Channel is one of the most contaminated areas within the OSPAR region in terms of plastic waste (OSPAR Commission, 2017). The number of plastic fragments found within grab samples was generally low (0 to 4 items per grab) compared to other recent surveys in south east England such as The Swale Estuary MCZ baseline survey, where samples with over 50 fragments per grab were recorded (Miller and Green, 2018), although it is difficult to put that number into context. There are no large rivers in the vicinity which might be a source of plastic contamination and the site may be subject to the movement and redistribution of existing plastic particles within the English Channel by currents. If a standardised counting and recording protocol is in place then monitoring at the site could provide evidence of the effectiveness of better terrestrial waste handling although this may take some time if grab sampling only detects smaller breakdown products of larger items that are already in the environment.

5 Recommendations for future monitoring

To fulfil Objective 4, various recommendations have been made in the following two sections for future Dover to Folkestone MCZ monitoring.

5.1 Operational and survey strategy recommendations

- The 'A5.4 Subtidal mixed sediments' would be a potential BSH to use for a future 'sentinel' monitoring programme, as it is has a wide diversity of communities, is present both inside and outside the MCZ, and the BSH is spatially distributed across the site. A combination of BACI (before-after, control-impact) and temporal change sampling designs could then be implemented to assess the efficacy of the current GMA.
- To detect a 20 % change in species richness of 'A5.4 Subtidal mixed sediments' at 80 % power, 45 samples would be required (Annex 4), that could be hard to achieve. However, differences in community structure will be detected with far fewer samples, and metrics such as the IQI can detect 20 % change at a similar power with fewer samples.
- Any future acoustic surveys should ensure that sufficient groundtruthing is undertaken in areas originally predicted to be 'A5.1 Subtidal coarse sediment' in the south and west of the site, in order to increase the resolution of the Broadscale Habitat mapping in areas that have had different BSH recorded since the 2012 verification survey.
- This survey had a separate aim to use a drop-down camera to monitor the rock features of the MCZ. Due to high turbidity levels, it was not possible to utilise the resulting still images for any meaningful analysis. The 2012 verification survey was able to obtain still images with good visibility (Colenutt *et al.,* 2015), but there may not be many periods where weather and tidal conditions allow for such images to be collected. An alternative approach would be to use a camera with a freshwater lens and surveying at slack water which should minimise the turbidity enough to be able to at least identify the biotope present.
- The IQI and contaminants results suggested that outside eastern boundary of the site, close to Dover Harbour, could be subject to higher anthropogenic pressures than the west and south of the site. A targeted spatial assessment of sediment contaminants, combined with infauna and PSA sampling, could be undertaken to investigate this in more detail. Repeat sampling the same stations as used in this study will enable trends in concentrations to be identified.
- The acoustic data used to create the habitat map was collected by the UKHO in 2010 (Colenutt *et al.,* 2015). Due to the ephemeral nature of *Sabellaria* biotopes, it is likely that the acoustic data is too old now to confidently assess the extent of the biogenic reef across the site using the 2016 Day Grab data,

although the 2010 acoustic backscatter could be examined to see if Sabellaria reef signals could be present. Should further information be required on the presence of *Sabellaria* reef, a joint acoustic (potentially sidescan sonar) and groundtruthing survey should be performed, following the approach used by Jenkins *et al.* (2018). A similar approach could be used to assess the extent and density of potential Blue Mussel Beds at the western end of the MCZ.

- Future monitoring should involve a survey tailored to baseline Native Oyster populations in the site, possibly through the use of a dive survey for close examination of seabed habitats.
- NIS should continue to be monitored within the site given the high density of shipping activity in the area acting as a major vector for the introduction of new species to UK marine waters (Stebbing *et al*., 2014). *Crepidula fornicata* numbers should also be monitored as this species is included on the list of NIS selected for the assessment of Good Environmental Status in GB waters under MSFD Descriptor 2 (Stebbing *et al.,* 2014).

5.2 Analysis and interpretation recommendations

- Multivariate analysis methods generally offer the greatest power in detecting spatial differences and temporal changes and should be used alongside statistical analysis of univariate summary statistics.
- The adjacent Dover to Deal MCZ is also designated for ‘A5.4 Subtidal mixed sediments, and subjected to similar pressures as Dover to Folkestone MCZ. Future analyses could consider merging the datasets from the two MCZs (if sampled simultaneously) to undertake an assessment of change across the Dover area.
- Consider mapping future distributions of ‘A5.1 Subtidal coarse sediment’ and ‘A5.4 Subtidal mixed sediments’ as a matrix of BSH. A similar approach was used for surveying The Manacles MCZ, where the same BSH could not be easily differentiated due to their interwoven distribution (Brown and Mitchell, 2018).

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Annex 1. Abbreviations

BSH	Broadscale Habitats
Cefas	Centre for Environment, Fisheries and Aquaculture Science
CP2	Charting Progress 2
CHP	Civil Hydrography Programme
Defra	Department for Environment, Food and Rural Affairs
EA	Environment Agency
EUNIS	European Nature Information System
FOCI	Feature of Conservation Interest
GES	Good Environmental Status
GMA	General Management Approach
IFCA	Inshore Fisheries and Conservation Authority
JNCC	Joint Nature Conservation Committee
NMBAQC	North East Atlantic Marine Biological Analytical Quality Control Scheme
MBES	Multibeam echosounder
MCZ	Marine Conservation Zone
MPA	Marine Protected Area
MPAG	Marine Protected Areas Group
MSFD	Marine Strategy Framework Directive
NE	Natural England
NIS	Non-Indigenous Species
OSPAR	Oslo-Paris Convention for the Protection of the Marine Environment of the North-East Atlantic
PSA	Particle Size Analysis
PSD	Particle Size Distribution
RV	Research Vessel
SAC	Special Area of Conservation
SAD	Selection Assessment Document
SNCB	Statutory Nature Conservation Body
SOCI	Species of Conservation Interest

Annex 2. Glossary

Definitions signified by an asterisk (*) have been sourced from Natural England and JNCC Ecological Network Guidance (NE and JNCC, 2010).

Activity	A human action which may have an effect on the marine environment; e.g. fishing, energy production (Robinson, Rogers and Frid, 2008).*
Annex I Habitats	Habitats of conservation importance listed in Annex I of the EC Habitats Directive, for which Special Areas of Conservation (SAC) are designated.
Anthropogenic	Caused by humans or human activities; usually used in reference to environmental degradation.*
Assemblage	A collection of plants and/or animals characteristically associated with a particular environment that can be used as an indicator of that environment. The term has a neutral connotation, and does not imply any specific relationship between the component organisms, whereas terms such as ‘community’ imply interactions (Allaby, 2015).
Benthic	A description for animals, plants and habitats associated with the seabed. All plants and animals that live in, on or near the seabed are benthos (e.g. sponges, crabs, seagrass beds).*
Biotope	The physical habitat with its associated, distinctive biological communities. A biotope is the smallest unit of a habitat that can be delineated conveniently and is characterised by the community of plants and animals living there.*
Broadscale Habitats	Habitats which have been broadly categorised based on a shared set of ecological requirements, aligning with level 3 of the EUNIS habitat classification. Examples of Broadscale Habitats are protected across the MCZ network.
Community	A general term applied to any grouping of populations of different organisms found living together in a particular environment; essentially the biotic component of an ecosystem. The organisms interact and give the community a structure (Allaby, 2015).
Conservation Objective	A statement of the nature conservation aspirations for the feature(s) of interest within a site, and an assessment of those human pressures likely to affect the feature(s).*
EC Habitats	EC Habitats Directive (Council Directive 92/43/EEC on the

Directive	Conservation of natural habitats and of wild fauna and flora) requires Member States to take measures to maintain natural habitats and wild species of European importance at, or restore them to, favourable conservation status.
Epifauna	Fauna living on the seabed surface.
EUNIS	A European habitat classification system, covering all types of habitats from natural to artificial, terrestrial to freshwater and marine.*
Favourable Condition	When the ecological condition of a species or habitat is in line with the conservation objectives for that feature. The term ‘favourable’ encompasses a range of ecological conditions depending on the objectives for individual features.*
Feature	A species, habitat, geological or geomorphological entity for which an MPA is identified and managed.*
Feature Attributes	Ecological characteristics defined for each feature within site-specific Supplementary Advice on Conservation Objectives (SACO). Feature Attributes are monitored to determine whether condition is favourable.
Features of Conservation Importance (FOCI)	Habitats and species that are rare, threatened or declining in Secretary of State waters.*
General Management Approach (GMA)	The management approach required to achieve favourable condition at the site level; either maintain in, or recover to favourable condition.
Habitats of Conservation Importance (HOCI)	Habitats that are rare, threatened, or declining in Secretary of State waters.*
Impact	The consequence of pressures (e.g. habitat degradation) where a change occurs that is different to that expected under natural conditions (Robinson, Rogers and Frid, 2008).*
Infauna	Fauna living within the seabed sediment.
Joint Nature Conservation Committee (JNCC)	The statutory advisor to Government on UK and international nature conservation. Its specific remit in the marine environment ranges from 12 - 200 nautical miles offshore.
Marine Strategy Framework Directive (MSFD)	The MSFD (EC Directive 2008/56/EC) aims to achieve Good Environmental Status (GES) of EU marine waters and to protect the resource base upon which marine-related economic and social activities depend.

Marine Conservation Zone (MCZ)	MPAs designated under the Marine and Coastal Access Act (2009). MCZs protect nationally important marine wildlife, habitats, geology and geomorphology, and can be designated anywhere in English and Welsh inshore and UK offshore waters.*
Marine Protected Area (MPA)	A generic term to cover all marine areas that are 'A clearly defined geographical space, recognised, dedicated and managed, through legal or other effective means, to achieve the long-term conservation of nature with associated ecosystem services and cultural values' (Dudley, 2008).*
Natura 2000	The EU network of nature protection areas (classified as Special Areas of Conservation and Special Protection Areas), established under the 1992 EC Habitats Directive.*
Natural England	The statutory conservation advisor to Government, with a remit for England out to 12 nautical miles offshore.
Non-indigenous Species	A species that has been introduced directly or indirectly by human agency (deliberately or otherwise) to an area where it has not occurred in historical times and which is separate from and lies outside the area where natural range extension could be expected (Eno *et al.,* 1997).*
Pressure	The mechanism through which an activity has an effect on any part of the ecosystem (e.g. physical abrasion caused by trawling). Pressures can be physical, chemical or biological, and the same pressure can be caused by a number of different activities (Robinson, Rogers and Frid, 2008).*
Special Areas of Conservation	Protected sites designated under the European Habitats Directive for species and habitats of European importance, as listed in Annex I and II of the Directive.*
Species of Conservation Importance (SOCI)	Habitats and species that are rare, threatened or declining in Secretary of State waters.*
Supplementary Advice on Conservation Objectives (SACO)	Site-specific advice providing more detailed information on the ecological characteristics or 'attributes' of the site's designated feature(s). This advice is issued by Natural England and/or JNCC.

Annex 3. Infauna data truncation protocol.

Raw taxon abundance and biomass matrices can often contain entries that include the same taxa recorded differently, erroneously or differentiated according to unorthodox, subjective criteria. Therefore, ahead of analysis, data should be checked and truncated to ensure that each row represents a legitimate taxon and that they are consistently recorded within the dataset. An artificially inflated taxon list (i.e., one that has not had spurious entries removed) risks distorting the interpretation of pattern contained within the sampled assemblage.

It is often the case that some taxa have to be merged to a level in the taxonomic hierarchy that is higher than the level at which they were identified. In such situations, a compromise must be reached between the level of information lost by discarding recorded detail on a taxon's identity and the potential for error in analyses, results, and interpretation if that detail is retained.

Details of the data preparation and truncation protocols applied to the infaunal datasets acquired at Dover to Folkestone MCZ ahead of the analyses reported here are provided below:

- Where there are records of one named species together with records of members of the same genus (but the latter not identified to species level) the entries are merged and the resulting entry retains only the name of the genus.
- Taxa are often assigned as 'juveniles' during the identification stage with little evidence for their actual reproductive natural history (with the exception of some well-studied molluscs and commercial species). Many truncation methods involve the removal of all 'juveniles'. However, a decision must be made on whether removal of all juveniles from the dataset is appropriate or whether they should be combined with the adults of the same species where present. For the infaunal data collected at Dover to Folkestone MCZ: where a species level identification was labelled 'juvenile', the record was combined with the associated species level identification, when present or the 'juvenile' label removed where no adults of the same species had been recorded.
- Records of meiofauna (i.e., nematodes) were removed.
- Records of fish species were removed.

Annex 4. Assessment of sampling sufficiency

The ability to detect change depends on what sort of change we are interested in (the question being asked), the magnitude of change, the magnitude and the scale of temporal and spatial variability in the environment, the rate of Type I errors deemed permissible, and sampling effort (Wilding *et al.,* 2015), as well as the statistical method of data analysis (non-parametric procedures are often considered to be less powerful than parametric ones).

A typical set of criteria used when considering power analysis in ecological contexts is to determine the level of sample effort needed to detect a change of 20% with a power of 80% and a Type I error rate of 5%. However, a wider range of options is considered in Table 5.

Whilst there have been suggestions to use alternative Type I error rates to boost power without the associated costs of sampling (The UK Marine Biodiversity Monitoring Strategy, 2016; Marubini, 2014), this depends on being able to determine the costs of making incorrect decisions, which is beyond the scope of this work.

Methods of power analysis do not exist for the non-parametric permutation approaches to the multivariate community analyses used here (Wilding *et al.,* 2015, p4). Consequently, only univariate summary statistics of community composition (species richness, Shannon index, Simpson's Evenness, Hill's N1) and health (IQI) will be considered.

Although the statistical analysis used in this report used ANOVA, and any future analysis is likely to do the same to account for the BACI design, power analysis was carried out in Minitab using the independent two sample t-test option. It therefore doesn't consider increased power gained from repeated sampling over time, nor does it consider any issues arising from heteroscedasticity in the data. Estimates of standard deviation came from the untransformed sample data collected from 'A5.4 Subtidal mixed sediments' inside the site.

The results of the power analysis (Table 7) agree with the outcome of the analyses described in section 2. Due to the some BSHs having more samples collected in the survey than others (Table 4), there were differences in the power analysis results between habitat types for some metrics (e.g. species richness and the IQI), but the derived diversity indices were generally not responsive. It should be noted that multivariate analysis of community composition is likely to be more powerful than analysis of univariate metrics.

Additional insights about effective monitoring strategies can also be obtained through spatial autocorrelation analysis. Spatial autocorrelation is a natural phenomenon in which observations from nearby locations are likely to have values more similar than would be expected due to chance alone (Fortin *et al.,* 2002). Positive autocorrelation occurs when taxa are distributed in clumps or patches, or form aggregations. For example, *Sabellaria spinulosa* reefs are colonised by gregarious settlement, with existing aggregations of *Sabellaria spinulosa* encouraging settlement of larvae (Wilson 1970), therefore two sampling units taken in close proximity are likely to be highly spatially autocorrelated.

Table 7. Predicted number of samples needed to obtain a statistical power of 80% to detect a given level of change in each of the univariate metrics of community structure of 'A5.4 Subtidal mixed sediments' in the Dover to Folkestone MCZ (© Natural England and Environment Agency 2022). The number of samples required to detect change in the base statistics (abundance and species richness) is given as a percent change in the mean. Change is given in absolute values for the derived statistics. Power analysis based on a two-sample independent t-test, using untransformed data and a significance level of 5%

Metric	Mean	StDev	Number of samples needed to detect		
			10% change	20% change	50% change
Abundance	1208	1155	1432	359	59
Species Richness	90	30	176	45	9
			Number of samples needed to detect given magnitude of change		
H' Diversity	2.7	1.1	476 (d 0.2)	120 (d 0.4)	21 (d 1.0)
1-λ' Diversity	0.74	0.27	459 (d 0.05)	116 (d 0.1)	30 (d 0.2)
N1 Diversity	23	20	253 (d 5)	64 (d 10)	17 (d 20)
IQI	0.81	0.07	32 (d 0.05)	9 (d 0.1)	4 (d 0.2)

The randomised sampling design employed in this study was chosen to minimise the influence of positive spatial autocorrelation i.e. sample locations ensured spatial independence.

The degree of spatial dissimilarity is evaluated for the 2016 Dover to Folkestone MCZ survey data by quantifying the significance of spatial autocorrelation in the benthic communities sampled at each station within the 'A5.4 Subtidal mixed sediment' BSH. This analysis is performed by computing the Mantel's correlogram. Mantel's correlogram (Sokal, 1986; Oden and Sokal 1986) is a special case of a simple Mantel test (Mantel, 1967), a measure used to evaluate the resemblance between two matrices (e.g. ecological and geographic distance), where the analysis is partitioned into a series of discreet distance classes (analogous to the lag bins of a semivariogram). That is, a first distance matrix is evaluated by computing a standardised Mantel statistic (r_M) for all pairs of points within the first distance class; then a second matrix is scored for all pairs of points within the second distance interval, and so on. The result is analogous to an autocorrelation function or semivariogram but performed on a multivariate distance matrix (c.f. Legendre and Legendre, 1998, pp. 736-738) where no spatial correlation is $r_M = 0$ and a perfect positive autocorrelation is $r_M = 1$.

Before the correlogram was computed a Hellinger transformation was performed on the species data in order to reduce the influence (give lower weighting) to taxa with low counts or many zeros. The mantel correlogram for the Dover to Folkstone 2016 MCZ data was computed in the *Vegan R* package (Oksanen *et al.,* 2017). Here a multiple testing approach is employed to produce the correlogram where the Mantel test is performed for each distance class over multiple permutations (n=999) to compute the significance (*p* value) of spatial autocorrelation. There is an inherent increased risk of Type I error in the multiple testing approach so the Holm (1979) approach was used to correct *p* after permutation testing. Distance classes were assigned following Sturges' rule (Sturges, 1926): number of classes = 1 + (3.3219 x $\log_{10}n$) where *n* is the number of pairwise distances.

The correlogram for Dover to Folkestone (shown above) has a single significant distance class indicating a significant (*p* = 0.05) positive correlation (Mantel *r* = 0.095) at distance class 1 (0 – 1.07 km). Beyond this separation distance (>1.07 km) no significant autocorrelation is identified. This means that for practical purposes (i.e. future monitoring) measurements (samples) taken more than 1.07 km apart (or conservatively 2.7 km apart – the separation distance of distance class 2), can be considered as spatially independent with respect to infaunal community composition.

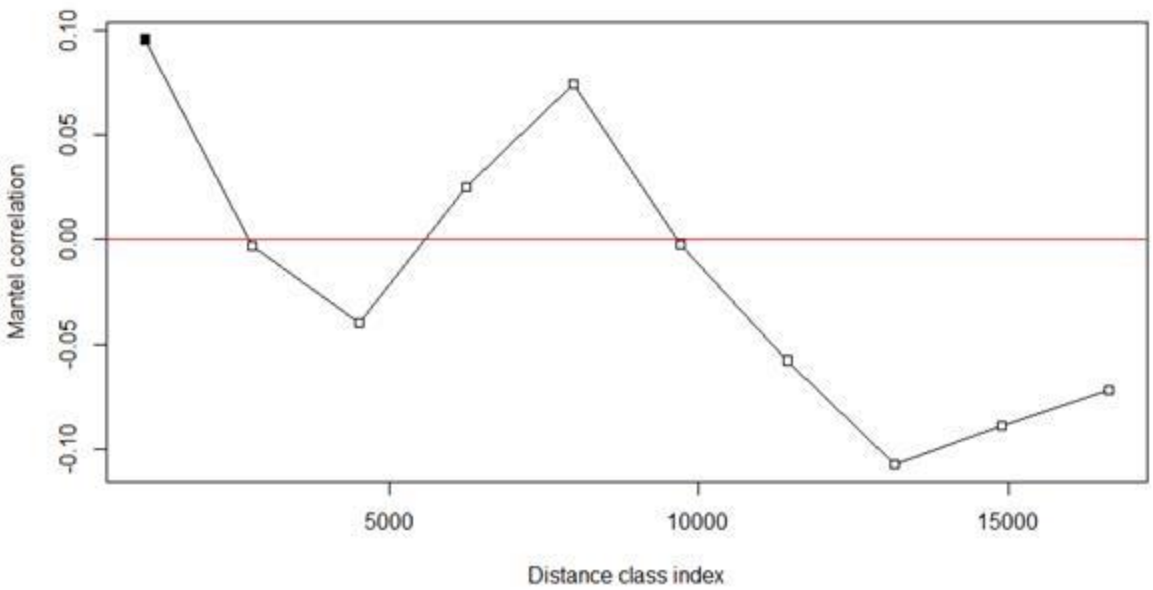

Figure 16. Mantel correlogram of the Hellinger transformed Dover to Folkestone faunal data. Filled black squares indicate significant multivariate spatial autorelation after Holm correction for multiple testing (n=999 permutations) (© Natural England and Environment Agency 2022).

Annex 5. Marine litter

Table 8. Standardised categories and sub-categories of litter items for Sea-Floor from the OSPAR/ICES/IBTS for North East Atlantic and Baltic. Guidance on Monitoring of Marine Litter in European Seas, a guidance document within the Common Implementation Strategy for the Marine Strategy Framework Directive, MSFD Technical Subgroup on Marine Litter, 2013.

A: Plastic	B: Metals	C: Rubber	D: Glass/ Ceramics	E: Natural products/ Clothes	F: Miscellaneous
A1. Bottle	B1. Cans (food)	C1. Boots	D1. Jar	E1. Clothing/ rags	F1. Wood (processed)
A2. Sheet	B2. Cans (beverage)	C2. Balloons	D2. Bottle	E2. Shoes	F2. Rope
A3. Bag	B3. Fishing related	C3. Bobbins (fishing)	D3. Piece	E3. Other	F3. Paper/ cardboard
A4. Caps/ lids	B4. Drums	C4. Tyre	D4. Other		F4. Pallets
A5. Fishing line (monofilament)	B5. Appliances	C5. Other			F5. Other
A6. Fishing line (entangled)	B6. Car parts				
A7. Synthetic rope	B7. Cables				
A8. Fishing net	B8. Other				
A9. Cable ties					
A10. Strapping band					
A11. Crates and containers					
A12. Plastic diapers					
A13. Sanitary towels/ tampons					
A14. Other					

Related size categories
A: ≤ 5*5 cm = 25 cm^2
B: ≤ 10*10 cm = 100 cm^2
C: ≤ 20*20 cm = 400 cm^2
D: ≤ 50*50 cm = 2500 cm^2
E: ≤ 100*100 cm = 10000 cm^2
F: ≥ 100*100 cm = 10000 cm^2

Annex 6. Non-indigenous species (NIS).

Table 9. Taxa listed as non-indigenous species (present and horizon) which have been selected for assessment of Good Environmental Status in GB waters under MSFD Descriptor 2 (Stebbing *et al.,* 2014).

Species name	List	Species name	List
Acartia (Acanthacartia) tonsa	Present	*Alexandrium catenella*	Horizon
Amphibalanus amphitrite	Present	*Amphibalanus reticulatus*	Horizon
Asterocarpa humilis	Present	*Asterias amurensis*	Horizon
Bonnemaisonia hamifera	Present	*Caulerpa racemosa*	Horizon
Caprella mutica	Present	*Caulerpa taxifolia*	Horizon
Crassostrea angulata	Present	*Celtodoryx ciocalyptoides*	Horizon
Crassostrea gigas	Present	*Chama sp.*	Horizon
Crepidula fornicata	Present	*Dendostrea frons*	Horizon
Diadumene lineata	Present	*Gracilaria vermiculophylla*	Horizon
Didemnum vexillum	Present	*Hemigrapsus penicillatus*	Horizon
Dyspanopeus sayi	Present	*Hemigrapsus sanguineus*	Horizon
Ensis directus	Present	*Hemigrapsus takanoi*	Horizon
Eriocheir sinensis	Present	*Megabalanus coccopoma*	Horizon
Ficopomatus enigmaticus	Present	*Megabalanus zebra*	Horizon
Grateloupia doryphora	Present	*Mizuhopecten yessoensis*	Horizon
Grateloupia turuturu	Present	*Mnemiopsis leidyi*	Horizon
Hesperibalanus fallax	Present	*Ocenebra inornata*	Horizon
Heterosigma akashiwo	Present	*Paralithodes camtschaticus*	Horizon
Homarus americanus	Present	*Polysiphonia subtilissima*	Horizon
Rapana venosa	Present	*Pseudochattonella verruculosa*	Horizon
Sargassum muticum	Present	*Rhopilema nomadica*	Horizon
Schizoporella japonica	Present	*Telmatogeton japonicus*	Horizon
Spartina townsendii var. anglica	Present		
Styela clava	Present		
Undaria pinnatifida	Present		
Urosalpinx cinerea	Present		
Watersipora subatra	Present		

Table 10. Additional taxa listed as non-indigenous species in the JNCC 'Non-native marine species in British waters: a review and directory' report by Eno *et al.* (1997) which have not been selected for assessment of Good Environmental Status in GB waters under MSFD Descriptor 2.

Species name (1997)	Updated name (2017)
Thalassiosira punctigera	
Thalassiosira tealata	
Coscinodiscus wailesii	
Odontella sinensis	
Pleurosigma simonsenii	
Grateloupia doryphora	
Grateloupia filicina var. luxurians	*Grateloupia subpectinata*
Pikea californica	
Agardhiella subulata	
Solieria chordalis	
Antithamnionella spirographidis	
Antithamnionella ternifolia	
Polysiphonia harveyi	*Neosiphonia harveyi*
Colpomenia peregrine	
Codium fragile subsp. atlanticum	
Codium fragile subsp. tomentosoides	*Codium fragile* subsp. *atlanticum*
Gonionemus vertens	
Clavopsella navis	*Pachycordyle navis*
Anguillicoloides crassus	
Goniadella gracilis	
Marenzelleria viridis	
Clymenella torquata	
Hydroides dianthus	
Hydroides ezoensis	
Janua brasiliensis	
Pileolaria berkeleyana	
Ammothea hilgendorfi	
Elminius modestus	*Austrominius modestus*
Eusarsiella zostericola	
Corophium sextonae	
Rhithropanopeus harrissii	
Potamopyrgus antipodarum	
Tiostrea lutaria	*Tiostrea chilensis*
Mercenaria mercenaria	
Petricola pholadiformis	
Mya arenaria	

Annex 7. Sediment contaminants

Table 11. Sediment contaminant results for the four stations sampled for contaminants analysis in the Dover to Folkestone 2016 survey (© Natural England and Environment Agency 2022). Heavy metal contaminants are normalised to 5% aluminium and organic contaminants are normalised to 2.5% carbon. Aluminium, organic carbon and nitrogen are presented as non-normalised values. OSPAR thresholds: < BAC = Background Assessment Concentrations shaded light blue, > EAC = Environmental Assessment Criteria shaded green and > ERL = Effects Range Low shaded red. Values reported as less than were taken as half the face value.

	Material (dry weight at 30ºC)	**Unit**	**BAC**	**EAC**	**ERL**	**DVFK01**	**DVFK20**	**DVFK30**	**DVFK31**
Heavy metals	Mercury	mg/kg	**0.07**		**0.15**	0.07	0.04	0.05	0.23
	Aluminium, HF Digest	mg/kg				18900	12200	11900	7580
	Iron, HF Digest	mg/kg				147884	80328	65966	106860
	Arsenic, HF Digest	mg/kg	**25**			109	74	54	65
	Cadmium, HF Digest	mg/kg	**0.31**		**1.2**	0.17	0.2	0.23	0.16
	Chromium, HF Digest	mg/kg	**81**		**81**	94	141	156	185
	Copper, HF Digest	mg/kg	**27**		**34**	18.1	17.1	14.3	16.7
	Lead, HF Digest	mg/kg	**38**		**47**	45	36.8	42.9	63.9
	Lithium, HF Digest	mg/kg				47.6	49.2	44.5	58.2
	Manganese, HF Digest	mg/kg				2778	1012	882	976
	Nickel, HF Digest	mg/kg	**36**			52.6	33.4	28.3	48.4
	Zinc : HF Digest	mg/kg	**122**		**150**	148	106	100	122
Chloro-carbons	Hexachlorobenzene	µg/kg				0.337	0.654	0.845	2.5
	Hexachlorobutadiene	µg/kg				0.337	0.654	0.845	2.5
	Tributyl Tin as Cation	µg/kg				5.3	8.2	8.4	13.2
	Carbon, Organic as C	%				0.371	0.191	0.148	<0.1
	Nitrogen as N	mg/kg				1090	<200	<200	<200

	Material (dry weight at 30ºC)	Unit	BAC	EAC	ERL	DVFK01	DVFK20	DVFK30	DVFK31
Polyaromatic hydrocarbons (PAHs)	Anthracene	µg/kg	5	85		29.1	49.5	41.4	75.5
	Benzo(a)anthracene	µg/kg	16	261		80	217	182	429
	Benzo(a)pyrene	µg/kg	30	430		73	232	194	565
	Benzo(ghi)perylene	µg/kg	80	85		43	141	118	305
	Chrysene + Triphenylene	µg/kg	20	384		83	229	191	486
	Fluoranthene	µg/kg	39	600		124	387	324	755
	Indeno(1,2,3-c,d)pyrene	µg/kg	103	240		52	162	136	322
	Naphthalene	µg/kg	8	160		17	52	42	125
	Phenanthrene	µg/kg	32	240		71	168	141	125
	Pyrene	µg/kg	24	665		113	364	306	750
Polybrominated diphenyl ethers (PBDEs)	2,2,4,4,5,5-Hexabromodiphenyl ether :- {PBDE 153}	µg/kg				0.067	0.209	0.253	0.5
	2,2,4,4,5,6-Hexabromodiphenyl ether :- {PBDE 154}	µg/kg				0.067	0.131	0.253	0.5
	2,2,4,4,5-Pentabromodiphenyl ether :- {PBDE 99}	µg/kg				0.168	0.327	0.507	1.25
	2,2,4,4,6-Pentabromodiphenyl ether :- {PBDE 100}	µg/kg				0.067	0.131	0.541	0.5
	2,2,4,4-Tetrabromodiphenyl ether :- {PBDE 47}	µg/kg				0.236	0.458	0.591	1.75
	2,4,4-Tribromodiphenyl ether :- {PBDE 28}	µg/kg				0.067	0.131	0.338	0.5
Polychlorinated biphenyls (PCBs)	PCB - 028	µg/kg	0.22	1.7		0.337	0.654	0.845	2.5
	PCB - 052	µg/kg	0.12	2.7		0.337	1.309	0.845	2.5
	PCB - 101	µg/kg	0.14	3		0.337	0.654	0.845	2.5
	PCB - 118	µg/kg	0.17	0.6		0.337	0.654	0.845	2.5
	PCB - 138	µg/kg	0.15	7.9		0.337	0.654	0.845	2.5
	PCB - 153	µg/kg	0.19	40		0.337	0.654	0.845	2.5
	PCB - 180	µg/kg	0.1	12		0.337	0.654	0.845	2.5

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