

JNCC/Cefas Partnership Report Series

Report No. 42

Farnes East Marine Conservation Zone (MCZ) Monitoring Report 2018

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September 2024

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ISSN 2051-6711

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ISSN 2051-6711

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This report should be cited as:

Downie, A., Clare, D. & McCabe, C. (2024). Farnes East Marine Conservation Zone (MCZ) Monitoring Report 2018. JNCC/Cefas Partnership Report 42. Peterborough, ISSN 2051- 6711.

<https://hub.jncc.gov.uk/assets/52696bf1-7eae-480d-8780-b25d788cf8c9>

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Funded by:

Department for Environment, Food and Rural Affairs (Defra) Marine and Fisheries Nobel House 17 Smith Square London SW1P 3JR

Please Note:

This work was delivered by Cefas and JNCC on behalf of the Marine Protected Areas Survey Coordination & Evidence Delivery Group (MPAG) and sponsored by Defra. MPAG was established in November 2012 and continued until March 2020. MPAG, was originally established to deliver evidence for Marine Conservation Zones (MCZs) recommended for designation. In 2016, the programme of work was refocused towards delivering the evolving requirements for Marine Protected Area (MPA) data and evidence gathering to inform the assessment of the condition of designated sites and features by SNCBs, to inform Secretary of State reporting to Parliament. MPAG was primarily comprised of members from Defra and its delivery bodies which have MPA evidence and monitoring budgets and/or survey capability. Members included representatives from Defra, JNCC, Natural England, Cefas, the Environment Agency, the Inshore Fisheries Conservation Authorities (IFCAs) and the Marine Management Organisation (MMO).

Since 2010, offshore MPA surveys and associated reporting have been delivered by JNCC and Cefas through a JNCC\Cefas Partnership Agreement (which remained the vehicle for delivering the offshore survey work funded by MPAG between 2012 and 2020).

Executive summary

Farnes East Marine Conservation Zone (MCZ) is a joint inshore and offshore site located off the northeast of England, approximately 11 km from the Northumberland coast. The site has a total area of 945 km² and ranges in depth from 30 m to 100 m. The site was designated due to the composition of the seabed, which predominantly comprises subtidal sediments with small patches of moderate energy circalittoral rock. Additionally, the site was designated for the presence of the 'Sea-pen and burrowing megafauna communities' (SPBMC) habitat Feature of Conservation Importance (FOCI) in the southeast corner of the site (a section of the Farne Deeps trench), and the presence of the species FOCI ocean quahog (*Arctica islandica*).

The primary aim of this report is to explore and describe the attributes of the designated features within Farnes East MCZ, based on a dedicated monitoring survey conducted between April and May 2018, to inform future assessment and monitoring of feature condition. The two main objectives of the 2018 survey were:

- 1) to further characterise and map the extent of the SPBMC habitat FOCI feature, which was under-represented in previously acquired data and consequently not well defined, and
- 2) to acquire quantitative data to enable sentinel (Type 1) monitoring of the designated Broadscale Habitat (BSH) features 'Subtidal coarse sediment', 'Subtidal sand', 'Subtidal mud', and 'Subtidal mixed sediments'.

Whilst 'Moderate energy circalittoral rock' is designated for the site, its spatial extent is limited rendering its characterisation beyond the scope of the 2018 survey. This report therefore focuses on the sediment BSHs and the SPBMC and ocean quahog FOCI.

A summary of report objectives and their outcomes and conclusions are provided below.

Table 1. Summary of report objectives and outcomes.

Objective Outcome

Provide a description of the extent, distribution, structure, and functional attributes of the designated sediment Broadscale Habitats (BSH): 'Subtidal coarse sediment', 'Subtidal sand', 'Subtidal mud', and 'Subtidal mixed sediments'

Sampling for the sediment BSH was conducted at 16 stations, covering the range of particle size distributions within each BSH to achieve a set of stations that are representative of the substrate conditions over the site, with 10 replicate Hamon grab samples collected at each station. The representative BSH stations all showed high within-station biological and sediment composition variability.

Up to three different BSH were observed at more than half of the representative multi-replicate stations. Large differences in the proportions of the gravel, sand, and mud sediment fractions were observed within stations, indicating high variability at fine (i.e. within-station) spatial scales. This was reflected in the associated macrofaunal communities, with the average withinstation similarity ranging from 34% to 54%.

Macrofaunal community composition also varied significantly between all stations, regardless of BSH. However, the same key structural taxa tended to characterise stations with the same BSH. This was reflected in the biotopes assigned to stations, namely 1) '*Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand' for 'Subtidal sand' BSH, 2) '*Owenia fusiformis* and *Amphiura filiformis* in offshore circalittoral sand or muddy sand' for 'Subtidal mud' BSH, and 3) '*Sabellaria spinulosa* on stable circalittoral mixed sediment' for 'Subtidal mixed sediments' BSH.

Ecological functioning similarly varied both within and across stations; relatively high macrofaunal community production estimates and bioturbation potentials were observed in 'Subtidal mixed sediments', particularly at two stations where *S. spinulosa* was a key structural and functional species, whilst *E. pusillus* had an important role in the sand-dominated 'Subtidal sand' and 'Subtidal coarse sediment' BSHs.

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1 Introduction

This report primarily explores data acquired from the first dedicated survey of Farnes East Marine Conservation Zone (MCZ) as part of the Marine Protected Area (MPA) monitoring programme in English waters. The specific aims of the report are discussed in more detail in Section 1.2.

This report **does not** aim to assess the condition of the designated features of the MPA. Statutory Nature Conservation Bodies (SNCBs) use evidence from MPA monitoring reports in conjunction with other available evidence (e.g. activities, pressures, historical data, survey data collected from other organisations or collected to address different drivers) to make holistic assessments of the condition of designated features within an MPA.

1.1 Site overview

Farnes East MCZ is a joint inshore and offshore site located off the northeast of England, approximately 11 km from the Northumberland coast, situated in the 'Northern North Sea' Charting Progress 2 (CP2) sea area (Area 4, Figure 1). The site has a total area of 945 km² and ranges in depth from 30 m to 100 m. The site was designated due to the composition of the seabed, which is predominantly subtidal sediments with small patches of moderate energy circalittoral rock. The western half of the site is dominated by subtidal coarse sediment in its shallower portions, with subtidal mixed sediments in deeper water. The eastern side largely consists of subtidal sand, with an increasing mud content towards the deeper southeast corner of the site where a section of the Farnes Deep glacial trench occurs within the site boundary. The trench, which is the deepest part of the MCZ, contains subtidal mud.

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1.2 Existing data and maps

During the site verification phase of the MPA programme, two surveys were undertaken at Farnes East MCZ (rMCZ at that time) aboard the RV *Cefas Endeavour*: one in 2012 and one in 2014*.* The main site verification survey was conducted in March 2012 (CEND0412; Whomersley *et al.* in press; Eggleton *et al.* 2015), to verify the presence and extent of Broadscale Habitats (BSH; 'Moderate energy circalittoral rock', 'Subtidal coarse sediment', 'Subtidal sand', 'Subtidal mud' and 'Subtidal mixed sediments') and habitat FOCI ('Peat and clay exposures') features proposed for designation within the site. The survey targeted 102 sampling stations placed in triangular lattice grids, according to the predicted extent of BSH derived from the UK SeaMap 2010 (v7) and the habitat map provided in the Site Assessment Document (SAD; Net Gain 2011). One hundred 0.1 m² mini-Hamon grab samples were collected and camera sledge videos and still images were collected from a further 36 stations (Whomersley *et al.* in press; Eggleton *et al.* 2015). The 2012 survey confirmed the presence of all four sediment BSH originally recommended for designation, but the 'Moderate energy circalittoral rock' BSH and 'Peat and clay exposures' habitat FOCI were not observed. However, the survey encountered the species FOCI ocean quahog (*Arctica islandica*) and habitat FOCI 'Sea-pen and burrowing megafauna communities' (SPBMC), with Norway lobster (*Nephrops norvegicus*) constructing burrows within the deep mud habitat. Two species of sea pen, the slender sea pen (*Virgularia mirabilis*) and phosphorescent sea pen (*Pennatula phosphorea*) were also observed. Neither FOCI were proposed for designation at the time of the survey. The SPBMC habitat FOCI was only observed at three stations in the 2012 survey and, as a result, its extent could not be delineated.

An updated habitat map of the site was produced in 2013 (Eggleton *et al.* 2015). This was based on the groundtruth data collected in 2012 and multibeam echosounder (MBES) bathymetry and backscatter data collected in that year by EGS (International) Ltd (EGS 2012) and in 2005 by the Maritime and Coastguard Agency under the Civil Hydrographic Programme (CHP). The updated BSH map indicated that 'Moderate energy circalittoral rock' is present at Farnes East, albeit covering a much smaller extent than suggested by UK Seamap.

The second survey in March 2014, conducted aboard the RV *Cefas Endeavour* (CEND0514; McIlwaine, in press; Eggleton *et al.* 2015), was specifically designed to target the newly mapped 'Moderate energy circalittoral rock' and the area with anecdotal evidence of 'Peat and clay exposures', with the aim of collecting additional evidence on the presence and extent of these two habitats. The survey collected drop camera video and still images from 56 tows, and three stations were sampled with a 0.1 $m²$ mini-Hamon grab. The presence of 'Moderate energy circalittoral rock' was confirmed but the habitat FOCI 'Peat and clay exposures' was still not encountered. The 2012 and 2014 data were used to further update the BSH map (Figure 2).

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Figure 2. Broadscale Habitat (BSH) map of Farnes East MCZ produced by Eggleton *et al*. (2015).

Because of the verification results, the site was designated for the sediment BSHs and circalittoral rock BSH, along with the SPBMC habitat FOCI and ocean quahog (*Arctica islandica*) species FOCI (Table 2).

Table 2. Designated features and General Management Approaches (GMAs) of Farnes East MCZ. Information from Robson (2014). GMA: Maintain in favourable condition / Recover to favourable condition).

Please note: the data used in this report were not specifically collected to target species FOCI.

1.3 Aims and objectives

Conservation objectives

Site-specific conservation objectives serve as benchmarks against which to monitor and assess the efficacy of management measures in maintaining a designated feature in, or restoring it to, 'favourable condition'.

As detailed in the [Farnes East MCZ Designation Order,](https://www.legislation.gov.uk/ukmo/2016/7/pdfs/ukmo_20160007_en.pdf) the conservation objectives for the site are that the designated features:

- 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.

Report aims and objectives

The primary aim of this report is to explore and describe the attributes of the designated features within Farnes East MCZ, to inform 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 report concentrates on the sediment BSH and the SPBMC Habitat FOCI. Whilst 'Moderate energy circalittoral rock' is designated for the site, it is not very prevalent. As a result, the rock habitat was given the lowest priority of all survey objectives, and it was not realistic to cover within the scope of the survey set by weather and the other survey priorities. For a full description of objectives and their prioritisation see Wood *et al*. (2020). The objectives of this report and the associated outputs are provided in Table 3.

To achieve Objective 1, selected Feature Attributes and supporting processes of the designated features are described as defined in Joint Nature Conservation Committee (JNCC) Conservation Advice for the site, from the [Site Information Centre](https://jncc.gov.uk/our-work/farnes-east-mpa/#conservation-advice) (SIC) (see Table 3).

Table 3. Report objectives and outputs.

* As defined in Supplementary Advice on Conservation Objectives (SACO) for the Farnes East MCZ.

2 Methods

2.1 Survey design

Between April and May 2018, a dedicated monitoring survey was conducted at the Farnes East MCZ onboard the RV *Cefas Endeavour*. The aim of the survey was to acquire sentinel (Type 1) monitoring data to contribute to the development of a monitoring time-series for the Farnes East MCZ. The two main objectives of the survey were:

- 1) to further characterise and map the extent of the SPBMC habitat FOCI feature, which was under-represented in previously acquired data and consequently not well defined, and
- 2) to acquire quantitative data to enable sentinel (Type 1) monitoring of the designated BSH features 'Subtidal coarse sediment', 'Subtidal sand', 'Subtidal mud', and 'Subtidal mixed sediments'.

Two different sampling approaches were required to meet the objectives (Figures 3 and 4).

A detailed description of the survey strategy, including descriptions of the statistical approaches used to determine the BSH subtypes and macrofauna community analysis, as well as all planned and completed sampling can be found in Wood *et al*. (2020). The two survey strategies are summarised below.

SPBMC habitat FOCI – grid of single samples

The SPBMC habitat FOCI had only been observed at three stations in the previous surveys of Farnes East MCZ, and its extent within the site could not be delineated based on such limited data. The previously mapped 'Subtidal mud' feature, consisting of two large basins and additional smaller patches of mud, was used as the basis of survey design for delineation and characterisation of the SPBMC feature (Figure 3). Existing data suggest the two largest basins differ in their particle size distribution; in that the southern basin has a higher mud content, whilst the northern basin is sandier. The southern basin is also currently fished for Norway lobster (*Nephrops norvegicus*) using otter trawls. This, together with a power analysis of macrofaunal metrics from the 2012 grab samples and the objective to confirm the extent of the SPBMC feature resulted in a survey design of 90 planned survey stations, across the two main mud basins (45 stations each) in an equidistant triangular lattice grid design with a minimum separation of 1 km.

Each station was sampled with a single 10-minute camera sledge tow, collecting video and stills imagery to confirm the presence of burrows and/or mounds, and record occurrence of sea pens. A single 0.1 m^2 mini-Hamon grab sample was collected per station to acquire particle size data, to determine sediment BSH type and investigate the macrofaunal community associated with burrowed mud habitats (JNCC 2014). Time limitations, arising partly from downtime with the camera sledge and partly an underestimation of the deployment time required for camera sledges (as opposed to drop cameras), reduced the number of stations visited to 52 (26 in each mud basin). Stations were prioritised for sampling based on maintaining geographical coverage and minimising long transits between stations. The locations of the planned and completed stations are presented in Figure 3. A subset of these stations ($n = 4$) also formed part of the sampling design for acquisition of sentinel monitoring data (Figure 4).

Sentinel monitoring of BSH – representative stations

There are four sediment BSHs present at Farnes East MCZ; 'Subtidal coarse sediment', 'Subtidal sand', 'Subtidal mud' and 'Subtidal mixed sediments' (Figure 3). Particle size analysis (PSA) and power analysis (using species richness and abundance metrics from macrofaunal data collected in 2012) both indicated high within-group variability for all BSH (Wood *et al.* 2020). This variability was too great to consider them as uniform units for sampling using a random approach. To capture the full range of particle size distributions and macrofaunal communities across the site, whilst at the same time reducing variability in sampling units used for future comparison, it was decided to approach the sentinel sampling via a smaller number of monitoring stations with increased replication across a range of particle size distributions within each BSH. Monitoring single stations with replication, instead of multiple, single sample stations across BSH classes, was selected to reduce variability in macrofaunal communities among replicate samples and to increase the power to detect temporal change within stations. Replication allows quantification of within-station variability, enabling robust time-series trend analysis for monitoring.

Multiple stations were included within each BSH, to give confidence that the stations collectively represented the variability in sediment composition and faunal communities across the whole site. Each of the four BSH were divided into two subtypes based on PSA data from 2012, and two sampling stations were in each subtype, giving a total of 16 stations (four BSH x 2 subtypes x 2 stations; Figure 4). Each were sampled with ten replicates inside a 50 m bullring around the station (as used for all grab sampling in the MPA programme). Analysis of the 2012 survey data was used to verify that the planned stations also captured the variation in macrofaunal community composition throughout the Farnes East MCZ.

Figure 3. Location of ground truth samples planned and collected at Farnes East MCZ in 2018. SPBMC Grid = grid of single camera sledge tow and Hamon grab stations for survey Objective 1; Representative BSH = representative stations in each Broadscale Habitat with 10 replicates each; Drop video = stations planned for drop video at known 'Moderate energy sublittoral rock' locations, not completed due to time restraints. Stations planned but not sampled are indicated with crosses.

Figure 4. Schematic of the sampling design for the two main objectives: 1) Map the extent of the SPBMC habitat FOCI in the two main mud basins (North/South) and 2) Sentinel (Type 1) monitoring of the designated BSH features. The four mud representative stations are also part of the SPBMC sampling grid. Type refers to the subcategory of the BSH identified from previous PSA; Samples refers to individual samples/replicates; CS = Camera Sledge, HG = Hamon Grab.

2.2 Data acquisition, preparation, and analysis

This section summarises how data were acquired, prepared, and analysed to achieve the report objectives (see Table 3). Sampling methods are described in detail in Annex 2. Data preparation and analytical methods are described in detail in Annex 3. The data derived from the two sampling strategies were treated separately in analysis. Consequently, in this report the grid of stations with camera sledge tows and a single grab sample targeting the mud habitats are referred to as the 'mud basin' stations and the 16 stations with ten replicate grabs are referred to as 'representative' stations.

Acquisition of grab samples and data preparation

Grab samples were successfully collected for sediment PSA and macrofauna from all 16 'representative' stations (n = 10 per station) and all 52 'mud basin' stations (n = 1 per station). Data were prepared as such:

- \triangleright The sediment particle size data were divided into 0.5 phi (ϕ) intervals using a combination of sieving (greater than 1 mm fraction) and laser diffraction (less than 1 mm fraction). Organisms were identified to the lowest possible taxonomic resolution, enumerated (abundance), and weighed (biomass).
- \triangleright Cobbles present in the grab samples were accounted for separately from the PSA samples as they were not considered to change the particle size of the sediment, but rather to provide additional hard substrata. Measurements (dimensions, rock type and volume) were recorded on board and the cobbles were then returned to each macrofauna sample. Cobbles content in samples was calculated as proportion (%) of the total sample volume rather than being included in the PSA. Incorporating the cobble proportion in the PSA would not change the BSH classes as it increases the proportion of gravel but does not change the sand:mud ratio.
- \triangleright Whilst benthic grabs typically extract infaunal taxa, some of the key taxa sampled by this method within the Farnes East MCZ are epifaunal (e.g. the tubicolous polychaete *Sabellaria spinulosa*). Therefore, the broader term 'macrofauna' is appropriate and is used throughout. Prior to analysis, macrofaunal data were truncated by removing

organisms that are not benthic invertebrates (e.g. fish) and organisms whose taxonomic resolution could potentially produce misleading estimates of community composition and diversity (e.g. an organism identified to the family level when there are also organisms from the same family identified to the genus or species level).

Acquisition of seabed imagery

High-definition video footage and digital still images were captured during single 11-minute camera sledge tows per station, travelling at \sim 0.3 knots and covering \sim 100 m distance over the seabed. Fan lasers were mounted on the camera sledge frame, giving a consistent gated view of an 80 cm wide transect. The camera set-up is described in more detail in Wood *et al*. (2020).

Analysis of seabed imagery

Poor visibility resulting from suspended sediment in the water column meant the still images collected were deemed to be of very poor quality (Turner *et al*. 2016). Consequently, only the video footage was analysed, by Seastar Survey Ltd.

The primary objective of video analysis was to verify and characterise occurrences of the SPBMC habitat FOCI. Burrow density is a defining characteristic of SPBMC, irrespective of the presence or absence of sea pens. Table 4 lists the density thresholds for combinations of small (1–3 cm diameter) and large (greater than 3 cm diameter) burrows for classifying a mud habitat into SPBMC, according to Robson *et al*. (2014).

Each video was segmented into sections considered to represent different seabed habitat types. Each video segment was further assigned into an image quality category (according to Turner *et al*. 2016) and the field of view of the gated area (between laser lines) was calculated. Each segment was assigned a BSH and Marine Habitat Classification for Britain and Ireland (MHCBI) biotope. Analysis of burrow density was completed in the [BIIGLE](https://biigle.de/) video annotation software. Burrows were annotated on the video using circles drawn to encompass the burrow opening. The circle diameter is considered representative of burrow size for further analysis. Burrow annotations were restricted to the gated area to allow calculation of burrow density from the known field of view. Burrows were annotated when they were fully inside the laser gates or intersected the right line laser. The circle diameter in pixels was converted to centimetres by relating it to the pixel distance between the lasers with a known real-world distance. Burrow counts by size (1–3 cm and greater than 3 cm) from annotations were further converted into densities, and where the thresholds shown in Table 4 were met, the segment was assigned to SPBMC.

Table 4. Burrow density thresholds (Robson *et al*. 2014) used for classifying mud habitats as 'Sea pen and burrowing megafauna communities' (SPBMC).

Poor visibility in a subset of the video tows prevented a full quantitative analysis of burrow densities. The extent and spatial distribution of SPBMC in the mud basins (Report Objective 1) was illustrated by plotting the density of large (greater than 3 cm diameter) and small (1– 3 cm diameter) burrows derived from video annotations, for those tows where visibility conditions allowed reliable counts. The density of large burrows from valid tows was also

plotted against depth and the proportion of gravel, sand, and mud in corresponding grab samples, to illustrate links between depth, substrate and burrow size and density.

To address the species composition of component communities and the presence of key structural and influential species in SPBMC (Report Objective 1), sea pen species and other clearly identifiable, conspicuous epifauna, including burrow-forming taxa (see Robson *et al.* 2014), were recorded to the highest taxonomic resolution possible. Taxa were annotated for the whole width of the video field of view to provide a comprehensive taxon list for each tow. The poor visibility limited the consistency of taxon observations, which were extracted as counts in the data but were only used as presence/absence observations in this report. None of the fauna were utilised in any quantitative analysis.

Occurrences of litter were further recorded using OSPAR/ICES/IBTS categories (see Annex 4) as were visible impacts or other modifiers, such as trawl marks or evidence of strong currents, to address Objective 4.

Estimation of bottom fishing activity

The spatial variability of bottom fishing activity at Farnes East MCZ was estimated using data from two sources. Mean annual subsurface abrasion Swept Area Ratio (SAR, see Church *et al*. 2016 for methods) from 2009 to 2018, available as raster layers gridded at 0.05 degrees resolution, was principally used to indicate the distribution of the intensity of fishing activity across the site. These data were supported by data derived by delineating areas with trawl marks visible in full coverage MBES backscatter data collected at the site in 2012 (Eggleton *et al.* 2015). The Vessel Monitoring System (VMS) derived SAR grid represents a longer-term estimate of pressure but is limited by its low resolution. The area with trawl marks derived from the MBES data has much higher resolution but is a snapshot in time. Both estimates of fishing activity are shown in Figure 5.

Figure 5. Fishing activity at Farnes East MCZ. Mean Annual Swept Area Ratio (SAR) (Church *et al*. 2016), over 2009 to 2018 with areas mapped as mud in the site verification habitat map (Eggleton *et al*. 2015) overlain in grey. The area where trawl marks are visible in MBES backscatter data collected in 2012 (Eggleton *et al*. 2015) is outlined in red. An example of the trawl marks is shown in a subset panel.

Statistical analysis

The following text provides a broad overview of the assessments used to address several report objectives. The specific analytical methods are detailed in Annex 3 'Numerical and statistical analyses'.

For Report Objective 1, the ten replicate grab samples from each of the 16 'representative' stations were used to assess the structural and functional attributes of macrofaunal communities. First, consistency (internal similarity) in macrofaunal community composition within each station and differences (external dissimilarity) in macrofaunal community composition between each pair of stations were assessed. Variation in macrofaunal community composition in relation to BSH was not formally analysed, because replicate sediment samples from the same station were often classified as different BSH. However, it was noted when there were similarities in community composition across stations with the same BSH. Univariate metrics of macrofaunal community structure – total number of taxa per sample, total abundance per sample, Shannon-Weiner Diversity Index (Shannon 1948; hereafter 'Shannon Index'), and Pielou's Evenness Index (Pielou 1966; hereafter 'evenness') – were calculated and plotted for each station (mean and 95% confidence interval), as were estimates of secondary production and bioturbation (sediment reworking) potential. The latter two metrics were considered relevant as they underpin the provision of food for demersal predators (e.g. benthivorous fish) and the regulation of biogeochemical cycling (e.g. decomposition and nutrient regeneration), respectively (Vander Zanden & Vadeboncoeur 2002; Solan *et al.* 2004). The methods used to estimate secondary production and bioturbation potential are described in Annex 3 'Ecological function estimates'.

Taxa that contributed the most to compositional similarity of the ten replicate samples at each of the 16 stations were considered the key structural taxa, whereas taxa that made the largest contributions to total secondary production and/or bioturbation potential were considered the key functional taxa. Any reef-forming taxa that occurred at sufficiently high densities were also considered key with respect to their habitat provision function. The key structural and functional taxa at each station were compared to those that characterise MHCBI biotopes (JNCC 2015) and were used, alongside formal statistical analysis of macrofaunal communities and information on the sediment habitat type, to assign a biotope to each station. The method for assigning biotopes is detailed in Annex 3 'Assigning biotopes'. The above procedures were also applied to macrofaunal community data from the 52 'mud basin' stations (each represented by a single sample), except here stations were grouped based on the results of a cluster analysis (i.e. rather than the ten replicate samples forming a station-level group, the stations with macrofaunal communities that clustered together were treated as replicates of the same cluster-level group).

Macrofaunal community data were also the focus of the analyses used to address Report Objectives 5, 6 and 7. To assess potential long-term changes in community composition (Report Objective 5), cluster analyses were used to compare data from the ten replicate samples at each of the 'representative' stations in 2018 to data from the single sample collected from each of the same stations in 2012. Univariate structural indices recorded in 2012 were then compared to the range of values obtained for the same station in 2018. The seasonal mismatch of the sampling between the two years (i.e. March in 2012; April/May in 2018) needed to be considered during these direct data comparisons. To calculate the number of replicates required per station for future monitoring (Report Objective 6), a power analysis was used to determine the sample size needed to have an 80% chance of detecting a 20% change in each metric. Finally, to address Report Objective 7, macrofaunal communities in areas exposed and not exposed to fishing (based on SAR and the presence of trawl marks on the seabed) were compared. The assessment focused on community composition and two metrics that appear to be negatively affected by trawl disturbance,

namely the Margalef Index (van Loon *et al.* 2018) and the *W*-statistic derived from ABC curves (Tuck *et al.* 1998; Kaiser *et al.* 2000). The former metric represents the total number of taxa relative to total abundance and is expected to decline due to a loss of sensitive taxa and/or an increase in the density of opportunists in response to trawling. The latter metric represents the relative biomass dominance of large *vs*. small organisms and is expected to decline due to a greater impact of trawling on large organisms.

3 Results

3.1 Particle size analysis (PSA)

Representative BSH stations

The PSA results from the 'representative' stations showed high variability in sediment content across the site and in the replicates at individual stations (Figure 6). Often variability was greater within than between stations. In line with predictions, the stations targeting the mixed sediments that were selected based on their coarser elements (numbers 13 and 14; Figure 7) were consistently of mixed sediment type with a high percentage of the coarser particle size fractions. The other two stations targeting mixed sediments (numbers 7 and 8; Figure 7) were, however, sandier and were classed as both 'Mixed' and 'Muddy sand' substrate types, based on the prominence of the larger gravel fraction. All the stations targeting coarse sediments (numbers 1–4; Figure 7) contained mixtures of sandy and coarse sediment fractions. One station was entirely sand (number 4; Figure 7) with a very uniform grain size distribution between replicates. The stations targeting 'Sand and muddy sand' maintained the characteristics used to select them. The two stations characterised by more prominent fine sand and mud fractions (numbers 5 and 6; Figure 7) were predominantly classified as 'Sand and muddy sand'. One of the stations selected to represent the coarser sand fractions (number 9 in Figure 7) showed a very uniform grain size distribution in the sand fraction between all replicates, and the other (number 10; Figure 7) contained a mixture of sand and gravel fractions, where some replicates were classified as 'Coarse'. The two stations located in the deeper southern mud basin (numbers 15 and 16; Figure 7) showed the most uniform particle size distributions with fine sand and mud fractions. The two northern mud basin replicate stations (numbers 11 and 12; Figure 7) contained mixtures of mud and coarser fractions, with replicates falling into both 'Mud and sandy mud' and 'Mixed' sediment types.

Figure 6. Classification of particle size distribution (half phi) information for each replicate sample at the 16 representative BSH monitoring stations at Farnes East MCZ (2018) into one of the sediment Broadscale Habitats (BSH; coloured areas) plotted on a true scale subdivision of the BGS-modified Folk triangle (Folk 1954; Long 2006). Point colour indicates the target BSH of the stations determined based on the 2012 survey samples (CEND0412; Whomersley *et al.* in press). Point shape corresponds to each of the four stations targeting within BSH variability (see Section 2.1).

(a)

(b)

Figure 7 a & b. Particle size distribution at the 16 stations with multiple replicates. The numbered plots show the percentage of the total PSA sample made up of each half phi fraction for the ten replicates at each station. Lines are coloured based on the BSH assigned to each sample based on PSA. Orange = Coarse sediment, green = Mixed sediments, yellow = Sand and brown = Mud. The dashed lines indicate boundaries between the gravel, sand and mud sediment fractions used for BSH. Corresponding numbers indicate the location of the stations on the map.

Mud basin stations

The majority (81%) of the 48 single grab samples taken in the mud basins fell into the 'Sandy mud' and 'Muddy sand' sediment types, confirming a 'Mud and sandy mud' substrate type, with a mud/sand ratio increasing with depth. Two of the 26 stations in the northern basin were 'Sand and muddy sand' and five were 'Mixed sediments'. In the southern mud basin, two of the 26 stations were 'Mixed sediments' (Figure 8).

Figure 8. Particle size distribution for each sample in the single grab sampling grid in the mud basins at Farnes East MCZ (2018) plotted on a true scale subdivision of the BGS-modified Folk triangle with the sediment Broadscale Habitats (BSHs) illustrated as coloured areas (Folk 1954; Long 2006). Point colour corresponds to samples collected in the northern and southern basins and shape indicates whether the accompanying video tow was classified as 'Not Habitat FOCI' (Not FOCI) or as 'Sea-pen and burrowing megafauna communities' (SPBMC) based on burrow density analysis or was excluded from analysis due to poor visibility.

Grab sample cobble content

Seven of the 16 representative BSH stations had cobbles in one to seven of the replicate samples (Figure 9). No cobbles were present in the single mud basin grab samples. Cobbles made up 3-50% of the total volume of the samples they were present in (Figure 9B). Cobbles make up a larger proportion of those samples with a smaller total volume (Figure 9C) potentially due to cobbly ground being less likely to yield large samples. The station with the highest number of replicate samples with cobbles, FRNEM096, also had the highest proportion by volume of cobble in the samples.

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Figure 9. Proportion of replicate grab samples at Farnes East MCZ (2018) with cobbles (A), proportion of sample volume made up of cobbles in each replicate at the representative stations (B) and relationship between cobble proportion and total sample volume (C).

3.2 Broadscale Habitats (BSH)

The survey has shown that whilst the BSH map produced by Eggleton *et al*. (2015), is a good approximation of the general BSH distribution across the site (Figure 9), it is not possible to capture the local fine scale variability using the BSH framework. Replicate sampling locations were designed to revisit sampling stations included in the generation of the habitat map and hence cannot be used to independently validate it.

Although the 'representative' station survey strategy targeted each BSH equally (four stations in each sediment BSH (see Section 2.1), 'Sublittoral sand' was the most prevalent BSH observed in the samples, whilst fewer 'Sublittoral coarse sediment' and 'Sublittoral mud' samples were acquired than expected (Table 5). At many stations replicate samples were classified into multiple BSH. 'Sublittoral sand' was present at the stations for all other BSH types. Two stations (FRNEM091 and FRNEM093) had replicates with three different BSHs ('Sublittoral sand', 'Sublittoral coarse sediment' and 'Sublittoral mixed sediments'). Only at five of the 16 replicated stations did all replicates consistently represent a single BSH. The two 'Sublittoral mud' stations in the southern mud basin and two of the 'Sublittoral sand' stations returned the expected BSH in all replicates. One of the targeted 'Sublittoral coarse sediment' stations also had a consistent BSH across all replicates, but contrary to expectations the BSH observed was 'Sublittoral sand' (Figure 9).

The single grab samples collected in 2018 across the mapped 'Sublittoral mud' area (which were independent of the data used to produce the habitat map) showed that both mud basins also included areas of 'Sublittoral mixed sediments'. In the northern basin, where the ratio of sand to mud fractions was higher, 'Sublittoral sand' was also observed. Without replication at these stations, however, it is not possible to say whether the sediment is predominantly mixed or sandy, or (as is more likely based on the replicate stations) a fine scale mixture of multiple BSHs. The habitat map shows narrow ridges of mixed sediments traversing the mud basins (Figure 9), where a small change in the bottom topography can lead to deposition of finer sediments or alternatively the exposure of underlying coarser fractions. Considering the within-station heterogeneity, spatial accuracy and the resolution and vintage of the acoustic data, the data collected on this survey cannot improve on the existing BSH map.

Table 5. Number of grab samples and camera sledge tows collected in each Broadscale Habitat (BSH) at Farnes East MCZ. Only the number of video tows is given; still images were collected but were of poor quality due to suspended sediment.

* Four of the 52 'mud basin' grab stations were also 'replicate' stations and are accounted for under that heading.

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Figure 10. Proportion of Broadscale Habitats (BSH) assigned to grab samples. The representative BSH stations with 10 replicates are indicated with a black circle.

3.3 Macrofaunal communities

This section presents the results of analyses of macrofaunal community data obtained through the collection of grab samples. Report Objectives 1, 5, 6, and 7 (see Table 3) are addressed: Section 3.3.1 uses data collected from the 16 'representative' stations in 2018 to address Report Objective 1; Section 3.3.2 uses the same data alongside data from the same stations in 2012 (n = 1 at each station) to address Report Objectives 5 and 6; and Section 3.3.3 uses data collected from the 52 'mud basin' stations in 2018 to address Objectives 1 and 7.

Variation across stations and BSHs

Community structure

Average within-station Bray-Curtis similarity of macrofaunal community composition ranged from 34% (FRNEM100) to 54% (FRNEM089) (Table 14; Annex 6). Stations with the most variability (i.e. the lowest internal similarity) were those that displayed multiple BSH based on their replicate PSA samples (FRNEM092, FRNEM093, FRNEM098 and FRNEM100), with 'Subtidal sand' recorded alongside 'Subtidal coarse sediment' and/or 'Subtidal mixed sediments'. Stations with relatively consistent macrofaunal community composition (i.e. high internal similarity) tended to be classified as just one BSH, either 'Subtidal mud' (FRNEM044), 'Subtidal sand' (FRNEM099 and FRNEM101) or 'Subtidal mixed sediments' (FRNEM096). However, in contrast to the overall pattern, the station that exhibited the most consistency in macrofaunal community composition (FRNEM089) had both 'Subtidal mud' and 'Subtidal mixed sediments'.

Macrofaunal community composition at each station differed significantly from all other stations (Analysis of Similarities (ANOSIM); *p* less than 0.05 for all pairwise comparisons), with the average between-station similarity ranging from 8% (FRNEM045 *vs.* FRNEM100) to 48% (FRNEM089 *vs.* FRNEM090). Moreover, variation in macrofaunal community composition across stations closely reflected variation in BSH type. That is, stations with the same BSHs tended to have relatively similar assemblages, whilst stations with different BSHs tended to have relatively dissimilar assemblages (Figure 10).

Figure 11. Non-metric multidimensional scaling (nMDS) ordination of macrofaunal community composition, based on $log_e(x+1)$ taxa abundances, at the 16 stations sampled with ten replicate grab samples. The colour of symbols indicates the Broadscale Habitats (BSH) recorded at the station: red = 'Subtidal coarse sediment', yellow = 'Subtidal sand', brown = 'Subtidal mud', green = 'Subtidal mixed sediments', orange = 'Subtidal sand' and 'Subtidal coarse sediment' or 'Subtidal mixed sediments', purple = 'Subtidal mud' and 'Subtidal mixed sediments'. The shape of the symbols distinguishes different stations with the same BSH. Gradients in the three sediment fractions that define BSHs (mud, sand and gravel) are shown as vectors. Two-dimensional stress = 0.16.

The macrofaunal taxa that characterised each station (i.e. key structural taxa) are shown in Table 14 (Annex 6). Generally, stations classified as 'Subtidal mud' (including those where some samples were classified as 'Subtidal mixed sediments') were characterised by the polychaetes *Diplocirrus glaucus*, *Galathowenia oculata*, and *Chaetozone setosa*, the cumacean *Diastylis lucifera*, the amphipod *Harpinia antennaria*, and ribbon worms (Nemertea). Stations classified as 'Subtidal coarse sediment' or 'Subtidal sand' were mainly characterised by the sea urchin *Echinocyamus pusillus* and a range of polychaetes, although the identities of the characteristic polychaete taxa varied from station to station. Stations classified largely or exclusively as 'Subtidal mixed sediments' were characterised by the reef building polychaete *Sabellaria spinulosa* and various taxa that live attached to hard surfaces, such as the barnacle *Verruca stroemia*, the chiton *Leptochiton asellus*, the hydroid *Sertularia* sp., the bryozoan *Escharella ventricosa*, the coral *Caryophyllia (C.) smithii* and a range of polychaetes.

Univariate indices of macrofaunal density and diversity varied across stations and BSHs. Total abundance was generally around 1,000 individuals per $m²$ in 'Subtidal coarse sediment', 'Subtidal sand' and 'Subtidal mud' but was as low as 600 individuals $m²$ at two stations where the seabed varied between the first two of these BSHs (FRNEM098 and FRNEM100; Figure 10A). Stations located on 'Subtidal mixed sediments' had relatively high macrofaunal densities, particularly at the station FRNEM096 where total abundance was > 5,000 individuals m-2 (Figure 10A). Notably, this station had the highest cobble content (Figure 9). Variation in the total number of taxa per sample (richness) showed a similar

pattern to total abundance, though the difference between 'Subtidal mixed sediments' and other BSHs was generally less pronounced (Figure 10B). Nevertheless, richness was notably high at FRNEM096 (\sim 80 taxa 0.1 m⁻² on average; Figure 10B), with one grab sample from this station containing 135 taxa. The evenness of taxa abundances was largely consistent across stations, ranging from 0.75 to 0.9 (where 1 is perfectly even) (Figure 10C). However, FRNEM096 was again the exception and had an average evenness of just 0.6 (Figure 10C). Shannon Index, which reflects both the richness and evenness of taxa, showed no clear patterns across stations or BSHs and ranged from around 2.5 and 3.5 (Figure 10D).

Figure 12. Bar graphs (mean and 95% confidence interval) for A) total abundance, B) total number of taxa, C) Pielou's evenness, D) Shannon Index, E) community production and F) community bioturbation potential of macrofauna at the 16 stations sampled with ten replicate grabs. The colour of bars indicates the BSH at the station: red = 'Subtidal coarse sediment', yellow = 'Subtidal sand', brown = 'Subtidal mud', green = 'Subtidal mixed sediments', orange = 'Subtidal coarse sediment / Subtidal mixed sediments' & 'Subtidal sand', purple = 'Subtidal mud' & 'Subtidal mixed sediments'.

Ecological function

Estimates of ecological function also varied across stations and BSHs. As with macrofaunal density and diversity, estimated macrofaunal community production and bioturbation potential were relatively high for 'Subtidal mixed sediments' (FRNEM096 and FRNEM097, not FRNEM091; Figure E-F). One station with multiple BSHs (based on its sediment PSA data; FRNEM093) had high but variable macrofaunal community production (Figure E), which is explained by unusually high values for two of the ten replicate samples from this station. Both community production and community bioturbation potential estimates were lowest at the two stations with particularly low total abundance and taxa richness (FRNEM098 and FRNEM100), where the seabed sediments were classified as 'Subtidal coarse sediment' and 'Subtidal sand' (Figure E-F).

The taxa that contributed most to estimated macrofaunal community production (i.e. key functional taxa) are shown in Table 16. In 'Subtidal mixed sediments', where production was relatively high (FRNEM096 and FRNEM097), the main contributors included the polychaetes *S. spinulosa*, *Lumbrineris cingulata* and *Notomastus* sp*.*, the crab *Ebalia tuberosa*, and Nemertea. Taxa that were major contributors to community production at just one of these stations included the crab *Atelecyclus rotundatus*, the chiton *Leptochiton asellus,* the bivalve *Timoclea ovata* at FRNEM096, and the brittle stars *Amphiura filiformis* and *Ophiactis balli* at FRNEM097. At the other station with high macrofaunal community production (FRNEM093), the main contributors were the bivalves *Modiolus* sp., *Arctica islandica* and sea anemones Actiniaria. Elsewhere within the MCZ, the major contributors to community production varied in relation to the habitat, with the polychaetes *D. glaucus* and *Streblosoma* sp. dominant in muddy sediments and the polychaete *Notomastus* sp., the brittle star *A. filiformis* and the sea urchin *Echinocardium cordatum* dominant in sand. Nemertea, the polychaete genus *Nephtys* spp., and the bivalve *Lucinoma borealis* were major contributors to macrofaunal community production irrespective of BSH.

The taxa that contributed most to community bioturbation potential (i.e. key functional taxa) are shown in Table 17 (Annex 7). The most productive taxa in 'Subtidal mixed sediments' (FRNEM096 and FRNEM097) tended to also have high bioturbation potential. However, the contribution of *S. spinulosa* to bioturbation potential was low compared to its secondary production, whereas the contributions of mobile burrowers such as the polychaetes *L. cingulata* and *Paramphinome jeffreysii* and the sea urchin *E. pusillus* to bioturbation potential were relatively high. Similarly, the most productive taxa at other stations (where the BSH was not 'Subtidal mixed sediments') tended to also be among the main contributors to bioturbation potential, but with *E. pusillus* in sandy sediments and acorn worms (Enteropneusta) in muddy sediments making relatively large contributions to bioturbation potential.

The capacity for *S. spinulosa* to form biogenic reefs also requires consideration from an ecological function perspective. These reefs add structural complexity to soft sediment habitats and can create new niches for species to occupy. Indeed, the dense aggregations of *S. spinulosa* in 'Subtidal mixed sediments' (FRNEM091, FRNEM096 and FRNEM097) were associated with relatively diverse assemblages and a range of taxa that live attached to the substrate, that were not characteristic of communities elsewhere in the MCZ. However, only at FRNEM096 did *S. spinulosa* reach a density that would be expected for a reef $(-1,500$ individuals m⁻²), which implies 'medium reefiness' (Gubbay 2007).

Bioturbation and habitat creation functions are also provided by burrowing megafauna, such as the Norway lobster (*Nephrops norvegicus*). However, these taxa are not effectively sampled by the mini-Hamon grab used to sample macrofauna. Burrowing megafauna form part of the SPBMC habitat FOCI within the Farnes East MCZ. Therefore, this feature is specifically addressed in Section 3.5.1.

Temporal comparisons

At stations surveyed in both 2012 and 2018, macrofaunal assemblages in replicate samples collected in 2018 tended to cluster together (*p* ≥ 0.05) and differ significantly (*p* less than 0.05) from the assemblages in the single samples collected in 2012 (Figure 12). However, univariate indices (total abundance, total number of taxa, evenness, and Shannon Index) in 2012 tended to fall within the range of values observed across the ten replicate samples at the same station in 2018 (Figure 13). Together these results indicate the possibility of temporal changes in macrofaunal community composition within the MCZ between 2012 and 2018, but there is no indication of broadscale changes to macrofaunal density and diversity.

The taxa that contributed most to compositional dissimilarity at each station between 2012 and 2018 are shown in Table 20 (Annex 8).The taxon lists vary from station to station, but some broad trends from 2012 to 2018 are apparent, including the appearance in 2018 of the cumacean *Diastylis lucifera* in 'Subtidal mud' and 'Subtidal mixed sediments' (five stations), an increase in the abundance of the polychaete *Sabellaria spinulosa* in 'Subtidal mixed sediments' (three stations), an increase in the abundance of the sea urchin *Echinocyamus pusillus* across all BSHs (eight stations), and a widespread reduction in the abundance of several polychaetes such as *Galathowenia oculata* (12 stations), *Hilbigneris gracilis* (six stations), and *Anobothrus gracilis* (five stations).

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Figure 13. Dendrogram of macrofaunal community composition, based on log_e(x+1) taxa abundances, of the 16 stations sampled with ten replicate grabs in 2018 (black circles) and a single grab in 2012 (orange circles). Distinct clusters (significantly different at *p* < 0.05) are separated by black branches.

Figure 14. A) total abundance, B) total number of taxa, C) Pielou's evenness, and D) Shannon Index of macrofauna at the 16 stations sampled with ten replicate grabs in 2018 (black circles) and a single grab in 2012 (orange circles).

Concerning future surveys of the 16 'representative' stations, the results of a power analysis to determine how many replicate samples are required to have an 80% chance of detecting a 20% change in univariate indices are shown in Table 6. The required number of samples ranges from 22 to 142 for total abundance and from 9 to 78 for total number of taxa. Therefore, the level of replication used for the 2018 survey ($n = 10$) is likely to be insufficient for monitoring temporal changes in these indices. Evenness and Shannon Index would require a much lower number of samples to detect a 20% change (generally greater than 10). However, it should be noted that the likelihood of an equivalent proportional change may differ between indices (e.g. a 20% change in the Shannon Index may be less likely than a 20% change in the total number of taxa).

Variation within mud basins

Community structure

Macrofaunal communities within the two mud basins group into seven significantly different clusters (SIMPROF: *p* less than 0.05), but only two of these clusters consist of communities from more than two stations (Figure 14). These two clusters (f and g) are therefore the focus of the following results. Cluster f is largely distributed within the northern basin and cluster g exclusively within the southern basin (Figure 15). Many of the taxa that characterise the two main clusters (i.e. the key structural taxa) are the same as those that characterise muddy sediments at the 'representative' stations where ten replicate grab samples were collected (see section 3.3.1). However, taxa that were characteristic only of cluster f include the Caudofoveatan mollusc *Chaetoderma nitidulum*, the brittle star *Amphiura filiformis* and the bivalve *Thyasira flexuosa*, whereas taxa that were characteristic only of cluster g include acorn worms (Enteropneusta), the polychaete *Heteromastus filiformis*, and the bivalve *Abra nitida* (Table 15; Annex 6). Mean total abundance and the total number of taxa were higher in cluster f than in cluster g (Figure 16A–B). Both clusters had an evenness of approximately 0.9 and a Shannon Index of approximately 3 (Figure 16C–D).

Figure 15. Non-metric MDS ordination of statistically significant (*p* less than 0.05) macrofaunal community clusters, based on log $e(x+1)$ taxa abundances, at the 52 stations within the two mud basins. $2D$ stress = 0.21.

Figure 16. Spatial distribution of macrofaunal community clusters within the two mud basins at Farnes East MCZ.

Ecological function

Estimated community production and bioturbation potential were higher in cluster f than in cluster g (Figure 16E–F). There were both similarities and differences in the key functional taxa of each cluster. Taxa that were major contributors to community production in both clusters include the polychaetes *Diplocirrus glaucus* and *Nephtys hombergii*, the bivalve *Lucinoma borealis*, and ribbon worms (Nemertea) (Table 18; Annex 7). Other major contributors to production include the bivalve *Arctica islandica*, the crab *Corystes cassivelaunus* and the sea urchin *Echinocardium cordatum* in cluster f, and the polychaetes *Streblosoma* sp., *Praxillella affinis* and the sea urchin *Brissopsis lyrifera* in cluster g (Table 18; Annex 7). Community bioturbation potential was driven mainly by the same taxa that drove community production, but the brittle star *Amphiura filiformis* and acorn worms

(Enteropneusta) made relatively large contributions to bioturbation, whereas the contribution of *A. islandica* (cluster f) was relatively small (Table 19 in Annex 7).

Figure 17. Bar graphs (mean and 95% confidence interval) for A) total abundance, B) total number of taxa, C) Pielou's evenness, D) Shannon Index, E) community production and F) community bioturbation potential of macrofaunal community clusters at the 52 stations where the BSH 'Subtidal mud' was targeted.

Potential role of trawling activity

Within the southern mud basin, macrofaunal community composition in areas with a high swept area ratio (seabed is trawled greater than 3 times per year on average) differed significantly from areas with a low swept area ratio (seabed is trawled \sim 1 time per year on average) (ANOSIM: R = 0.384, *p* = 0.007; Figure 17A). Moreover, community composition in the unfished northern mud basin differed significantly from areas of the southern mud basin with a high swept area ratio (ANOSIM: $R = 0.415$, $p = 0.001$; Figure 17A) but did not differ significantly from areas with a low swept area ratio (ANOSIM: $R = 0.03$, $p = 0.537$; Figure17A). When fishing activity was inferred from the presence of trawl marks on the seabed, macrofaunal community composition differed significantly between southern areas with and without trawl marks (ANOSIM: $R = 0.243$; $p = 0.003$; Figure 17B); however, the lower R value indicates that the magnitude of the difference was small compared to the difference between areas distinguished by the swept area ratio. Community composition in the northern area differed significantly from community composition in southern areas with and without trawl marks (ANOSIM: R = 0.476, *p* = 0.001 and R = 0.189, *p* = 0.048, respectively: Figure 17B).

Despite the differences in macrofaunal community composition in relation to trawling activity, inspection of two metrics that are expected to be responsive to disturbance (the Margalef Index and the W-statistic) showed little indication of trawl impacts (Figure 18). When trawling activity was inferred from the swept area ratio, the southern area with high activity had the lowest mean Margalef Index value (Figure 18A), suggesting a relatively disturbed benthic assemblage. However, the low W-statistic in areas of the southern basin with low activity, albeit with relatively high variability around the mean, possibly contradicts this suggestion (Figure 18C). When trawling activity was inferred from the presence of trawl marks, the mean Margalef Index was slightly lower in trawled areas (Figure 18B); however, the Wstatistic was stable across all three areas, thus suggesting consistent levels of disturbance to the macrofaunal communities.

Sediment composition within the MCZ varied in relation to fishing activity, with relatively heavily fished southern areas being somewhat muddier than areas with low fishing activity (Figure 19). This indicates that the physical attributes of the trawled habitat, rather than trawling *per se*, may drive the observed variation in macrofaunal community composition in relation to trawling activity. Indeed, the relationship between macrofaunal community composition and mud content is significant (RELATE: R = 0.327, *p* less than 0.05).

Figure 18. Non-metric MDS ordinations of macrofaunal community composition, based on log_e(x+1) taxa abundances, at the 52 stations within a northern mud basin and southern mud basin with different levels of fishing activity. Fishing activity is inferred from A) the swept area ratio and B) the presence of trawl marks on the seabed. Two-dimensional stress = 0.21.

Figure 19. Bar graphs (mean and 95% confidence interval) for the Margalef Index of macrofauna in relation to A) Swept Area Ratio (SAR) and B) the presence of trawl marks on the seabed, and the Wstatistic of macrofauna (from ABC curves) in relation to C) SAR and D) the presence of trawl marks on the seabed. 'North' indicates the northern mud basin, where there is no activity, and 'South' indicates the southern mud basin, where fishing activity is variable.

Figure 20. Bar graph (mean and 95% confidence interval) for % mud content in relation to A) Swept Area Ratio (SAR) and B) the presence of trawl marks on the seabed. 'North' indicates the northern mud basin, where there is no activity, and 'South' indicates the southern mud basin, where fishing activity is variable.

3.4 Biotopes

This section assigns biotopes to the areas surveyed within the Farnes East MCZ to contribute to the completion of Report Objective 1 (see Table 3). The closest matches of Marine Habitat Classification of Britain and Ireland (MHCBI; v 15.03) biotopes to macrofaunal communities from grab samples at each 'representative' station and each of the main community clusters in the mud basins are shown in Table 7. It should be noted that whilst a statistical approach was used to help assign biotopes to stations and clusters (see 'Assigning biotopes' in Annex 3). The results of statistical analyses used to help assign biotopes to each station/cluster are presented in Annex 9.

Station / Cluster Biotope code Biotope description 044, 045, 089, 090, 099, 102, cluster f SS.Ssa.Osa.OfusAfil *Owenia fusiformis* and *Amphiura filiformis* in offshore circalittoral sand or muddy sand 091, 096 SS.SBR.PoR.SspiMx *Sabellaria spinulosa* on stable circalittoral mixed sediment 092, 093, 094, 095, 097, 098, 100, 101 SS.Ssa.CFiSa.EpusOborApri *Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand cluster g SS.Ssa.Osa.OfusAfil / SS.SMxCMx.MysThyMx Transitional biotope between '*Owenia fusiformis* and *Amphiura filiformis* in offshore circalittoral sand or muddy sand' and '*Mysella bidentata* and *Thyasira* spp. in circalittoral muddy mixed sediment'

Table 7. Marine Habitat Classification of Britain and Ireland biotopes matched to 'representative' stations (n = 16) and the main community clusters (f and g) in the two mud basins within the Farnes East MCZ.

In contrast to biotopes assigned to grab samples from the mud basins (clusters f and g, Table 7), the two main biotopes (or habitats if biotopes could not be assigned using the available data) observed from video tows in the two mud basins were 'Seapens and burrowing megafauna in circalittoral fine mud' (SS.SMu.CFiMu.SpnMeg) and 'Circalittoral muddy sand' (SS.SSa.CMuSa). The habitats 'Circalittoral fine mud' (SS.SMu.CFiMu), 'Circalittoral sandy mud' (SS.SMu.CSaMu) and 'Circalittoral mixed sediment' (SS.SMx.CMx) were also found, primarily in the northern basin but also the western edge of the southern basin (Figure 20). The 'Seapens and burrowing megafauna in circalittoral fine mud' biotope was observed throughout both mud basins. However, complex burrows were observed only in the deeper, muddier southern basin whilst sparser simple burrows were present in the northern basin and edges of the southern basin. Figure 21 shows screen capture examples from video tows in three of the main habitats identified. 'Circalittoral fine mud' (SS.SMu.CFiMu) and 'Circalittoral sandy mud' (SS.SMu.CSaMu) are not shown due to poor visibility. Examples are given of both types of burrowed mud classified as the 'Seapens and burrowing megafauna in circalittoral fine mud' (SS.SMu.CFiMu.SpnMeg). Table 8 shows the number of video segments in each habitat or biotope, as assigned visually by the video analyst. The video analysts included segments with sandier and coarser sediments in their corresponding grab samples as the 'Seapens and burrowing megafauna in circalittoral fine mud' biotope than those exceeding the burrow density threshold for SPBMC derived from annotated burrow counts (see Section 3.5.1), suggesting their visual estimation of burrows may be an overestimation.

Figure 21. Distribution of Marine Habitat Classification of Britain & Ireland habitats and biotopes assigned to stations using video footage and grab samples across Farnes East MCZ.

Figure 22. Examples of the main Marine Habitat Classification of Britain & Ireland habitats and biotopes assigned to video segments. 'Circalittoral fine mud' (SS.SMu.CFiMu) is excluded due to poor visibility.

Table 8. Marine Habitat Classification of Britain & Ireland. Habitats and biotopes assigned visually by analyst to video segments in the two mud basins within the Farnes East MCZ.

3.5 Habitat Features of Conservation Importance (FOCI)

'Sea-pen and burrowing megafauna communities'

Camera tows and grabs placed in the two mud basins to establish the extent of the SPBMC habitat FOCI indicated that not all the area mapped as mud basins in the site verification map is composed of 'Sublittoral mud'. Coarser sediment fractions were present at the edges and on narrow ridges traversing the mud basins, with associated grab samples from eight stations classed as mixed sediments, and two as sand. Fine scale local variability can be observed in the video tows. Four of the stations with grab samples classified as 'Sublittoral mixed sediments' and one as 'Sublittoral sand' include parts of the video tow still categorised as 'Sublittoral mud' by the video analysts. Of the 52 camera tows, 42 were at least partially visually classified as 'Sublittoral mud'.

Two distinct types of burrowed mud were observed in the video tows classified as 'Sublittoral mud'. The difference is illustrated by the burrow densities calculated from the BIIGLE annotations of burrow diameter, where large complex burrows with densities above the threshold values for FOCI assignation were confined to the deeper southern mud basin (Figure 22). Whilst both types of burrowed mud have been assigned to the 'Seapens and burrowing megafauna in circalittoral fine mud' biotope (SS.SMu.CFiMu.SpnMeg), on visual evaluation by the video analyst, the annotated burrow size data do not support the classification of all of these tows as the SPBMC habitat FOCI. Of the 39 video tows classified as SS.SMu.CFiMu.SpnMeg by the analyst, seven had visibility too poor for quantitative analysis, 18 exceeded the threshold for SPBMC densities of small and/or large burrows and 14 were below the threshold. All tows that exceeded the FOCI threshold based on the annotated burrow size data were in the southern mud basin. However, the visibility of the tows assigned to SS.SMu.CFiMu.SpnMeg in the western part of the northern basin was too poor for consistent counts.

Figure 23. Density of large (greater than 3 cm diameter) and small (1–3 cm diameter) burrows in the video tows covering the basins mapped as mud in the site verification habitat map (Eggleton *et al.* 2015). Tows where visibility was too poor to consistently enumerate burrows are indicated in grey. Thresholds for SPBMC are greater than 0.1 large burrows per m^2 , greater than 1 small burrow per m^2 or greater than 1 small and/or large burrows per m2.

There is a low percentage of gravel in grab samples associated with camera tows where SPBMC is present, and the mud percentage is higher than in the tows where the SPBMC burrow density is not met, whilst the sand fraction remains the same in both (Figure 23). The density of large burrows increases with depth and the proportion of mud in associated grab samples. High densities of large burrows are only seen where gravel content is close to zero (Figure 24). SPBMC corresponds closely to grab cluster group g. Of the 14 stations in group g that have burrow counts, 13 are assigned as SPBMC, whilst the corresponding number for grab cluster group f is only 3 in 21.

Figure 24. (a) Distribution of the 'Sea-pen and burrowing megafauna communities' (SPBMC) FOCI in the video tows covering the basins mapped as mud in the site verification habitat map (Eggleton *et al.* 2015). Pie charts show the proportion of gravel, sand and mud in associated grab samples; (b) The range of gravel, sand and mud percentage values in SPBMC and other tows.

Figure 25. Relationship between density of large burrows and (a) depth, (b) percentage gravel, (c) percentage sand, and (d) percentage mud. The grey lines show a loess smooth fit to the data.

Taxa recorded in video tows with burrow counts are given in Table 9. Sea pens occur in low numbers (1–3 individuals per camera tow) in muddy, sandy, and mixed substrata over the whole area investigated (Figure 25). *Pennatula phosphorea* sea pens were present at 18 stations, eight of which were assigned to SPBMC. *Virgularia mirabilis* was present at one station, which was muddy but did not exceed the burrow count threshold. The SPBMC stations share many of their associated species with the other mud habitats. *Nephrops norvegicus*, the hagfish (*Myxine glutinosa*) and sea mouse (*Aphrodita aculeata*) were more commonly observed at the SPBMC stations, whilst bryozoans, true anemones, true crabs, squat lobsters, sea stars, gobies, and dragonets (*Callionymus* sp.), present in other mud habitats, were less common or absent. The only taxa observed in association with the SPBMC stations were spider crabs and fish, such as the snake blenny (*Lumpenus lampretaeformis*) and gurnards (Triglidae).

Table 9. Taxa observed in video tows with good visibility (N = 39). The percentage of stations where each taxon is present is given for the 'Sea-pen and burrowing megafauna communities' (SPBMC) habitat FOCI, other mud habitats and other substrata. (* and shading represents Burrowing taxa and sea pens).

Figure 26. Distribution of sea pens (*Pennatula phosphorea* and *Virgularia mirabilis*) in the video tows covering the basins mapped as mud in the site verification habitat map (Eggleton *et al.* 2015).

Undesignated habitat FOCI

The reef building habitat FOCI taxon, Ross worm (*Sabellaria spinulosa*) was present at two stations (FRNEM091 and FRNEM096) (Figure 26). At FRNEM096, the density of *S. spinulosa* (1439 ± 283 individuals m-2), indicates 'medium reefiness' as defined by Gubbay (2007). The lower density (275 \pm 49 individuals m⁻²), at FRNEM091 is below the threshold for reef. However, it should be noted that *S. spinulosa* density is not one of the main characteristics proposed by Gubbay (2007) to define reef presence and quality, which includes elevation, extent, and patchiness. Data on these characteristics are not available for the Farnes East MCZ.

Figure 27. Undesignated habitat FOCI species found in grab samples.

3.6 Species FOCI

The ocean quahog (*Arctica islandica*) was observed at 24 of the 64 stations sampled with a mini-Hamon grab in 2018. Most specimens were of a small size: Only two individuals longer than 5 cm in length were recorded, one at FRNEM052 and one at FRNEM093 (Figure 27). Figure 28 (a) shows the locations where *A. islandica* was observed in 2018 and in the 2012 survey (CEND0412; Whomersley *et al.* in press). At the replicated stations, when present, it was observed in 1–8 of the replicates. Between one and two individuals were retained per grab replicate. Densities at the replicated stations (where present) ranged from 1– 10 individuals m⁻². At the station with the highest density (10 individuals m⁻²), a single replicate would only have an 80% chance of detection, meaning single samples are inefficient at estimating its presence or density at a site. Figure 28 (b) shows the number of individuals retained per replicate for the 16 replicated stations.

A. islandica is a major contributor to macrofaunal community production and bioturbation potential at FRNEM93 and FRNEM95, where it was present at a density of six and five $intividuals$ m⁻², respectively (see Annex 7).

Figure 28. Images of ocean quahog (*Arctica islandica*) specimens larger than 5 cm in length.

Figure 29. Observations of the ocean quahog (*Arctica islandica*) at Farnes East MCZ: a) presence at grab stations from 2018 and 2012, and b) number of individuals in each replicate sample in 2018 replicated stations.

3.7 Non-indigenous species (NIS)

The only non-indigenous species (see Annex 5), observed at Farnes East MCZ was the polychaete *Goniadella* sp. The locations of samples with *Goniadella* sp. occurrence are shown in Figure 29. It must be noted that each of the stations where *Goniadella* sp. were observed were those with multiple replicates, with the species being present in 1–5 of the replicates. Densities ranged between 1–10 individuals m-2.

Figure 30. Locations of observed non-indigenous species at Farnes East MCZ.

3.8 Marine litter

Litter was observed at three stations sampled in 2018. Pieces of plastic sheet (Plastic: A2, see Annex 4) were seen on the video at FRNEM004 and in one replicate grab sample at FRNEM093. A grab sample at FRNEM090 contained a rubber seal with plastic binding (Rubber: C5. Other, and Plastic: A14. Other; see Annex 4).

3.9 Observed anthropogenic activities and pressures

No signs of human activities were observed on the survey.

4 Discussion

The analyses in this report provide an overview of the extent and distribution, along with structural and functional attributes of the designated features within the Farnes East MCZ. Additionally, whilst the specific metrics for indicating whether the condition of the features has been maintained, is improving, or is in decline are not yet available, a provisional comparison was made between communities present at the time of the monitoring survey and those observed in single-replicate samples collected from the same stations in 2012. Macrofauna communities in fished and non-fished mud areas were further investigated through a descriptive comparison, to afford insight to the potential effects of bottom trawling in the mud basins, which have a General Management Approach (GMA) of 'Recover'. The results relevant to decisions on next steps in monitoring the site are discussed below, before presenting specific recommendations for future monitoring.

4.1 Community composition in relation to physical habitat

Macrofaunal community composition varied significantly from station to station (Figure 10). However, the same group of key structural taxa tended to characterise areas of 'Subtidal coarse sediment' and/or 'Subtidal sand', whilst another group characterised areas where 'Subtidal mud' was dominant, and another, relatively diverse, group characterised areas where the substrate was mainly or exclusively 'Subtidal mixed sediments' (Table 14; Annex 6). These groupings are reflected in the biotopes that were assigned to stations, namely 1) '*Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand', 2) '*Owenia fusiformis* and *Amphiura filiformis* in offshore circalittoral sand or muddy sand', and 3) '*Sabellaria spinulosa* on stable circalittoral mixed sediment', respectively (Table 7). It should be noted, however, that the stations assigned to the first biotope often had coarser substrates than indicated by the biotope description. This is not unexpected, as biotopes in The Marine Habitat Classification of Britain and Ireland are defined by the macrofauna present rather than the BSH present. Indeed, biotopes are known to often cover more BSH than the one featured in the biotope description. Furthermore, whilst *Amphiura filiformis* and *Owenia* sp. were consistently present at stations assigned to the second biotope, they were generally not among the key structural taxa (Table 14; Annex 6). A notable feature of 'Subtidal mixed sediments' at the site is that they often contained cobbles (Figure 9A). Station FRNEM096 had particularly high cobble content (Figure 9B), which possibly explains the prevalence of the reef forming S. spinulosa (section 3.5.2) and associated high macrofaunal abundance and diversity (Figure 12).

Cluster analysis of macrofaunal data collected throughout the two mud basins reaffirmed the presence of the biotope '*Owenia fusiformis* and *Amphiura filiformis* in offshore circalittoral sand or muddy sand' within the northern basin (community cluster f), but also indicated that it forms a transitionary biotope with '*Mysella bidentata* and *Thyasira* spp. in circalittoral muddy mixed sediment' in the southern basin (community cluster g) (Table 7). In contrast to the 'Mixed sediment' habitat description of the matched biotope, community cluster g occurred in the deepest part of the southern basin where mud content is the highest and gravel content the lowest (24 of the 25 grab samples in cluster g were classified as 'Sublittoral mud'). The video tows at stations with grabs in cluster g were likewise all classified as 'Seapens and burrowing megafauna in circalittoral fine mud', indicating a mismatch between biotopes assigned to the same locations based on the different sampling methods. A similar discrepancy was seen for cluster f, where 35 of the 48 grab samples in the cluster group were classified as 'Sublittoral mud', 12 as 'Mixed sediment' and only one as 'Sand'. Biotopes or habitats assigned visually from video included 19 instances of 'Seapens and burrowing megafauna in circalittoral fine mud' and eight instances of 'Circalittoral muddy sand'.

Although in some cases the visual assignations to the burrowed mud biotope may have been unwarranted (as noted by the burrow counts based the size cut-offs applied to burrows with diameters calculated from sized annotations) the mismatch between biotopes is not surprising. The source of core records used to derive the biotope description and representative species lists for the two biotopes matched to the grab samples are based on fauna from grab sampling, whilst the presence of burrows and characterising epifauna for 'Seapens and burrowing megafauna in circalittoral fine mud' are derived from video and images. The large fauna creating the burrows (such as *Nephrops norvegicus*, mobile epifauna and sparse sessile epifauna such as burrowing anemones) that form the biotope description for 'Seapens and burrowing megafauna in circalittoral fine mud' are usually not collected by grab sampling. Similarly, although traces of many of the larger polychaetes that characterise the infaunal biotope descriptions can be seen on video and still images, taxonomic identification is not possible. Most of the identifying taxa for the infauna-based biotope descriptions are not observable by video approaches. Improved holistic sampling with both grab and camera approaches would help to match the descriptions based on taxa they do share and allow for these biotopes to be identified more accurately and consistently.

Comparing macrofauna from the replicate grab samples collected in 2018 to those from the single grab samples collected from the same stations in 2012 suggests there have been compositional changes over time (Figure 12) but no broadscale changes in macrofaunal density or diversity (Figure 13). It is important to note here that a true statistical comparison is precluded by the lack of replication in 2012. As such, these results should be interpreted with caution. It is also worth noting that the 2018 survey was conducted later in the year than the 2012 survey (April/May *vs*. March), meaning that differences in communities may partly or fully reflect a seasonal cycle. The effects of any changes to faunal identification between years (e.g. the taxonomic resolution that could be achieved) also require consideration. As the data for each year were combined and truncated as a single dataset prior to analysis (following the procedure outlined in 'Macrofaunal data preparation' in Annex 3), such effects will have been mitigated. However, some apparent temporal changes to communities (e.g. a replacement of *Chaetozone setosa* with *C. zetlandica* at station 44; see Table 20 in Annex 8) suggest that some identification inconsistencies may have influenced the results. It is, however, also possible that there has been a genuine turnover of species from the same genus.

The decision to collect replicate grab samples ($n = 10$) from 'representative' stations for the 2018 survey was informed by the high variability in macrofaunal communities across stations with the same BSH in 2012 (Wood *et al.* 2020). It was postulated during planning of the 2018 survey that within-station variability would be relatively low, thus providing the statistical power necessary to detect temporal changes in macrofaunal communities associated with each designated BSH, using a 'manageable' sample size. However, multiple (up to three) different BSH were observed in replicates at more than half of the representative stations, with large differences in the proportions of the gravel, sand, and mud sediment fractions. Whilst some of the 'Sublittoral mud' and 'Sublittoral sand' stations had more uniform sediment distributions, others were very variable at a fine spatial scale. Macrofaunal communities similarly exhibited a high level of variability even at the station level, with average within-station compositional similarity ranging from 34% (FRNEM100) to 54% (FRNEM089) (Table 14; Annex 6). This biological variability appears to reflect the underlying within-station variability in sediment composition, as stations with multiple BSHs (based on PSA of grab samples) tended to have the greatest variability in macrofaunal community composition. The stations with high variability in sediment composition and BSH (e.g. FRNEM089, FRNEM092, FRNEM098 and FRNEM 100) are probably unsuitable for monitoring temporal changes to biological communities associated with the designated BSHs at the site. Moreover, the biological variability means that the number of replicate grab samples required to have an 80% chance of detecting a 20% change in common community metrics, such as total abundance and taxa richness, ranges from 20 to over 100 for most

stations, including those where only a single BSH was recorded (Table 6). This is not practicable for a cost-effective monitoring programme unless a smaller subset of stations were selected, such as those that require relatively few samples. However, this would reduce the representativity of the survey, as stations that require fewer samples tend to be from muddy areas of the MCZ (FRNEM044, FRNEM045, FRNEM089, and FRNEM090).

4.2 Ecological function

As with structural attributes of macrofaunal communities, there was variability in ecological functioning both within and across stations. However, at the BSH level, relatively high macrofaunal community production and bioturbation potential were observed in 'Subtidal mixed sediments', particularly at stations 96 and 97 (Figure 10E-F). A key structural and functional species at these stations is the tubicolous polychaete *Sabellaria spinulosa* (see Annex 6 and Annex 7). Aside from its role in productivity and sediment reworking, this species can also perform a habitat provisioning function by forming reefs that support other fauna (Pearce *et al.* 2011). This is possibly reflected by the high macrofaunal total abundance and diversity observed in 'Subtidal mixed sediments' (Figure 10A-B), particularly at station 96, where the density of *S. spinulosa* is high enough to constitute 'medium' reef (~ 1,500 individuals m-2; Gubbay 2007). S*. spinulosa* is an ephemeral species and the spatial distribution of the reefs it forms can change on a short timescale. Therefore, any habitat provision function provided by *S. spinulosa* at specific locations within the MCZ may not be persistent over time.

Sandy and muddy sediments within the MCZ each had their own set of key functional species, most of which were influential both in terms of community production and bioturbation potential (see Annex 7). Like *S. spinulosa* in mixed sediments, *E. pusillus* was key with respect to both structure and function in sandy sediments ('Subtidal sand' and 'Subtidal coarse sediment'; see Annex 6 and Annex 7) and was characteristic of the assigned biotope (Table 7). As the abundance of both *S. spinulosa* and *E. pusillus* appears to have substantially increased between 2012 and 2018 (see Table 20 in Annex 8), the longterm temporal dynamics of these species is likely to have important implications for the structure and function of designated BSHs.

4.3 Habitat FOCI

The Marine Habitat Classification of Britain and Ireland biotope 'Seapens and burrowing megafauna in circalittoral fine mud' was recorded across both mud basins in the visual estimates made by the video analyst (Figure 20). However, the burrows seen in the northern basin were smaller and sparser, and large complex burrows were concentrated in the southern basin (Figure 22). When the visual classifications were compared to burrow density estimates made from annotations in BIIGLE (using circles to measure burrow openings), none of the 11 stations assigned to the biotope in the northern basin by the analyst (where visibility was sufficient for comparison), exceeded the burrow density thresholds used. Although the definition of the Marine Habitat Classification of Britain and Ireland biotope is not the same as for SPBMC, and the burrow count thresholds are not applied, the analyst's tendency to assign the 'Seapens and burrowing megafauna in circalittoral fine mud' biotope more liberally may stem from an issue with estimating burrow size visually. Most burrows that were annotated measured smaller than 1 cm diameter. On the other hand, the oblique angle of the camera and the location in the video frame, where annotations are made, will cause errors in sizing. Still, the sizes derived from annotation are more precise and repeatable, making annotating burrows a quantifiable measure that can be developed for future use to monitor habitat quality.
The conspicuous epifauna observed in SPBMC were largely similar to those observed in the more sandy and mixed sediments. Sea pens were observed across both mud basins in consistently low densities and, although slightly more common at the muddy stations, they were also seen on sand and mixed sediments. Low numbers of sea pens on varying substrata are common, and occur in biotopes and habitats other than SPBMC, which are defined by the presence of burrows. Most of the other taxa observed in video tows also occur both in SPBMC and other habitats, excluding the Norway lobster (*N. norvegicus*), burrowing anemones and spider crabs, as well as snake blennies and gurnards seen exclusively in conjunction with SPBMC. The density and size of burrows at Farnes East MCZ increased with depth, which is also correlated with increasing mud content, whilst the number of taxa reduced. The SPBMC stations in the northern mud basin and stations in the southern basin with lower burrow counts are associated with macrofaunal cluster group f, whereas the deepest part of the southern mud basin where the highest counts of the large (greater than 3 cm) complex burrows are concentrated is associated with cluster group g (Figure 30).

Both the high burrow count video tows and grab samples in cluster group g had the lowest diversity (number of taxa). The reason for the lower diversity at these stations cannot be determined due to multiple confounding factors. The part of the southern mud basin where high burrow count video tows and cluster group g are located is trawled more than three times per year, on average, with trawl marks providing evidence of impact at a finer spatial scale (Figure 30). Whilst they spatially overlap with the area where bottom contact fisheries are occurring, the resolution of the fishing effort data is too coarse to link individual stations to impact. The comparison must be made at the level of a group of stations with a higher or lower likelihood of impact. Furthermore, it is not possible to infer that lower diversity is a result of fishing impact due to the confounding environmental gradients of depth and mud content. The colocation of the highest counts of large complex burrows and bottom fishing is not surprising given the fishery is targeting *N. norvegicus*, which is the species that contributes most to the construction of large burrows. It is possible the lower diversity is a result of repeated impact by bottom gears, but it is equally possible the lower diversity is a result of the higher mud content or depth, and the association with the fished area is due to the habitat preference of *N. norvegicus* for muddier ground. Inspection of community metrics that are considered sensitive to trawling revealed no clear impact of this activity to the macrofauna in grab samples (Figure 18). This may suggest that trawling at current levels within the Farnes East MCZ has had little influence on macrofaunal communities associated with 'Subtidal mud'. However, it should also be noted that the survey was not specifically designed to test the impacts of trawling, and the differences in environmental conditions between the areas exposed to different levels of trawling activity (e.g. mean mud content; Figure 19) may have influenced the results.

Figure 31. Location of SPBMC confirmed by measured burrow counts in relation to grab cluster groups f and g and fishing activity.

The high density of the Ross worm (*Sabellaria spinulosa*) at FRNEM096, a mainly mixed sediment station in the southern part of the site, indicated the presence of the habitat FOCI 'Ross Worm (*Sabellaria spinulosa*) reefs' which is not a designated feature of the site. The density of *S. spinulosa* observed (~1500 individuals m-2) is expected to correspond to 'medium reefiness' according to the criteria set by an inter-agency workshop on defining and managing *S. spinulosa* reefs (Gubbay 2007). Elevated diversity at the station in comparison to other locations is potentially a further indication of reef presence. A drop camera would need to be deployed to confirm the presence and extent of potential *S. spinulosa* reef. One other station also showed elevated numbers of *S. spinulosa* but did not meet the density threshold criteria for reef.

4.4 Species FOCI

Farnes East MCZ is also designated for the species FOCI ocean quahog (*Arctica islandica*). The survey was not specifically targeted to detect or monitor *A. islandica*, which is difficult to monitor using grab samples because of its low rate of detection. The species was, however, found across the site on all substrata. Most individuals caught were small. The replicate samples from the representative stations confirmed that multiple samples were required to

reliably establish presence of *A. islandica* at a specific location, even at the stations where it was most numerous. This highlights the care that must be taken if comparing occurrence at the site over time. Comparisons should not be made between revisits to single sample stations. Given the wide spatial distribution and lack of habitat preference shown by ocean quahog at Farnes East MCZ, assuming equal likelihood of catchability, some inference could be drawn from comparing occurrence rates over the whole site.

5 Recommendations

It was determined prior to the 2018 survey that it is not practical to monitor the Farnes East MCZ via grab sampling throughout the extent of each designated BSH as the spatial variability of substrate and macrofaunal communities mean that an unfeasible number of stations would need to be sampled to detect change with sufficient confidence. The 2018 survey, instead, collected replicate grab samples from a relatively small number of stations that are representative of each BSH present at the site. However, both sediment composition (including BSH type) and macrofaunal communities were variable at many of these stations, indicating that the ecology of the designated BSHs is heterogenous at both broad and fine spatial scales. This poses a challenge for future monitoring.

The two mud basins have relatively consistent macrofaunal communities compared to the rest of the site. Monitoring the SPBMC habitat FOCI and the associated 'Sublittoral mud' BSH, which are the only designated features at the site with a 'Recover to favourable status' management objective, may be undertaken using a combination of grab sampling and camera sledge tows. Should fishing regulations be implemented for the site, monitoring the effectiveness of these regulations in the recovery of the SPBMC should account for depth and relative mud content variations across the southern mud basin (where the SPBMC occurs) and use comparative stations outside the site. Comparison of the currently fished and unfished areas within the site is not recommended due to the differences in these environmental conditions.

Application of the BIIGLE annotation software to estimate burrow size and density has been adopted here for the first time in the MPA monitoring programme. Lessons learned from this exercise have been taken forward and the method is currently being expanded for use at other sites and in the development of monitoring strategies. The key analytical messages from its application to the Farnes East MCZ data are summarised below in sections 5.1 and 5.2.

5.1 Operational and survey strategy

- Comprehensive monitoring of all Broadscale Habitats (BSH) via grab sampling is not recommended at Farnes East MCZ, because of the exceptionally high variability observed both at the site and local scales resulting in a cost-prohibitive number of stations required to detect change with confidence. Any grab sampling of designated BSH other than 'Subtidal mud' within the Farnes East MCZ should, where possible, focus on specific stations where sediment composition and macrofaunal communities have relatively low within-station variability. This will reduce the number of replicates required to detect temporal change associated with a specific BSH to a high level of confidence (see Table 6).
- 'Subtidal mud' has relatively low spatial variability in macrofaunal communities, thus requiring a relatively small number of samples for effective monitoring. It is associated with the only designated habitat FOCI at the site (SPBMC) and it is the habitat on which fishing within the MCZ largely occurs. The 'Subtidal mud' BSH is therefore a good candidate for being the primary focus of future monitoring of the Farnes East MCZ.
- The likelihood of encountering poor visibility limits the success of quantitative analysis from video tows on mud habitats in this region. The time of year for successive surveys should be chosen to optimise conditions for video acquisition.
- Successive surveys should be scheduled for the same time of year to minimise the influence of seasonality in biological communities. Conducting surveys at different

times of the year can make it impossible to distinguish long-term change from seasonal cycles.

- The long-running programme of *Nephrops* underwater television surveys by Cefas under the auspices of an ICES expert group (ICES 2020) in Functional Unit 6 (Farne Deeps) consists of 110 stations with annually repeated camera sledge tows in the southern mud basin outside of Farnes East MCZ. The survey covers a fishing intensity gradient and a range of sea pen densities. Video footage from these surveys can provide additional material, which will be especially useful as a control area with continued fishing if fishing regulations are put into place for the site and should be considered when planning any future monitoring surveys.
- The possibility of extending the annual *Nephrops* surveys at Farne Deeps to include stations inside the Farnes East MCZ should be explored for its potential as a costeffective source of monitoring data with regular sampling of a control area with continued fishing activity and a multidecadal timeseries on variability under different levels of fishing.
- A drop-frame camera tow is recommended to confirm the presence of 'Ross worm (*Sabellaria spinulosa*) reef'.
- Investigatory drop-frame camera tows, or imagery from a camera integrated into the grab frame, are recommended at stations where grab sampling of coarse or mixed sediments is planned. This would help to determine if the cobble content would limit grab samples collection and provide additional information to assign a BSH class to the station.

5.2 Analysis and interpretation

- Camera sledge video footage is recommended as the primary source of data for monitoring SPBMC. Still images were of poor quality due to light from the flash being reflected from the high load of suspended sediment in the water column, which is likely to be the case if conditions at the time of survey are not optimal.
- Poor visibility from occasional sediment plumes obstructed the view of the seafloor for parts of some video tows. The exclusion of poor visibility segments in future should be managed by dividing the tows into distance-based segments, deciding on a minimum number of segments required and selecting a set of adequate quality segments from each tow to reach an equal sample size. This would be beneficial to excluding tows with stretches of poor quality.
- Burrow density is recommended as a quantitative metric for monitoring SPBMC. Whilst a burrowed mud habitat is very unlikely to change to 'not-burrowed' unless the sediment type changes, quantifiable burrow sizes and counts may give an indication of changes in habitat quality over time.
- The BIIGLE annotation method was found to be useful in terms of its consistency and its ability to extract quantitative estimates of burrow size and density.
- For improved repeatability of density estimates, better specification of what constitutes a burrow would help with consistency between video analysts. For consistent sizing when using an oblique camera angle, burrow annotations should be done only in a specified part of the frame, together with annotation of the line lasers.
- A habitat/site-specific Epifauna Identification Protocol (EIP) should be developed for the SPBMC at the site, considering the primary source of imagery (video) and the potential for poor visibility. Consistently identifiable taxa should be annotated in the same quantitative video segments as burrows for detection of change in associated epifaunal communities.
- More detailed annotation of faunal traces, such as siphons, feeding traces and worm tubes and casts should be included in video analysis for better comparison with associated grab samples.
- Cobbles are very unlikely to affect the classification of coarse and mixed sediments, but high cobble content may indicate the presence of hard substrata and hence impact on the type of fauna present in the sample by including attached taxa. For this reason, where cobbles are present in grab samples they should be treated as hard substrata rather than being included in the PSA.
- It is also recommended that cobble content is considered as a proportion of the sample (% total volume) made up of hard substrata.
- Visual verification of broader cobble content at the station using seabed imagery is also recommended.

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Annex 1. Glossary

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

Annex 2. Data acquisition

Grab sampling

Sediment samples for particle size distribution (PSD) and macrofauna analyses were collected using a 0.1 $m²$ mini-Hamon Grab. This grabbing device was selected due to its versatility in sampling the different sediment habitats present within the Farnes East MCZ.

A 500 ml sub-sample was taken from each grab sample and stored at -20 °C prior to determining the particle size distribution. Sediment samples were processed following the recommended method 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 sediment Broadscale Habitats (BSH).

The faunal fraction was sieved over a 1 mm mesh, photographed, then fixed in buffered 4% formaldehyde. Faunal samples were processed to extract all fauna present in each sample. Fauna were identified to the lowest taxonomic level possible, enumerated and weighed (blotted wet weight) to the nearest 0.0001 g following the recommendations of the NMBAQC scheme (Worsfold *et al.* 2010).

Seabed imagery

Seabed imagery data were acquired using a camera sledge. High-definition video footage and digital still images were captured along 11-minute tows. Fan lasers (80 cm apart) and point lasers (19 cm apart) were mounted on the camera sledge to standardise the image field of view for quantitative analysis.

Sensors logging bottom temperature, altitude, bearing, and depth recorded data for the duration of the tow.

The full camera specification is available in Wood *et al*. (2020).

Annex 3. Data preparation and analysis

Hydrodynamic modelling

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 sediment Broadscale Habitats (BSH) using a modified version of the classification model produced during the Mapping European Seabed Habitats (MESH) project (Long 2006).

Macrofaunal data preparation

Macrofaunal taxa recorded in mini-Hamon grab samples were checked for the application of consistent and up-to-date nomenclature using the [WoRMS match taxa tool](http://www.marinespecies.org/aphia.php?p=match) [accessed 10/08/20]. The following truncation steps were then applied to the up-to-date taxa list:

- Where a species was recorded alongside members of the same genus (but the latter not identified to species level), the entries were merged, and the resulting entry retained only the name of the genus.
- Taxa recorded above the genus level were removed from the dataset when lower taxonomic levels of the same group were recorded to avoid having to reduce the taxonomic resolution of records.
- Taxa are often assigned as 'juveniles' during the identification stage with little evidence for their actual reproductive natural history (except for some wellstudied 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 macrofaunal data collected at the Farnes East MCZ, if 'juvenile' records were recorded at the same taxonomic level as 'adult' records then the two records were combined, whereas if juveniles were recorded at a higher taxonomic level than adults then the 'juvenile' records were removed to avoid having to reduce the taxonomic resolution of the 'adult' records.
- Records of taxa that are not benthic macrofauna (e.g. fish) were removed.

The retained taxa were assigned the abundance recorded in grab samples. In cases where it was not possible to determine whether one or more individuals of a taxon were present (e.g. with small colonial taxa), an abundance of '1' was assigned (*sensu* Callaway *et al.* 2018; Downie *et al.* 2018).

For analyses focused on the 2018 data, the truncation process was applied to this dataset only. For analyses of differences in macrofaunal communities between 2012 and 2018, data from the two years were combined and the truncation process was applied to this integrated dataset.

Ecological function estimates

Secondary production

Macrofaunal community production (kJ m⁻² year⁻¹) was estimated indirectly using abundance and biomass data from grab samples collected during the 2018 survey. First, any taxa that could not be both enumerated and weighed were removed from the datasets. Measured (wet) biomass values were then converted to energy values, using published conversion factors (Brey *et al.* 2010), and converted to annual production values using a multi-parameter empirical model (Brey 2001). This method unifies previous habitat-specific approaches into a multiple regression model and is one of the most reliable and robust methods for estimating secondary production (Cusson & Bourget 2005; Dolbeth *et al.* 2005).

To produce macrofaunal community production estimates for each grab sample, the mean biomass (kJ m^{−2}), mean abundance (individuals m^{−2}) and individual body mass (kJ) of each taxon were entered into the empirical model along with station specific depths (recorded during the survey) and mean annual bottom water temperatures for the year preceding sample collection (extracted from a 7 km gridded model for the [European north-west shelf](http://marine.copernicus.eu/) [accessed 24/08/20]). The broad taxonomic group of each taxon was specified, along with whether it is subtidal or intertidal (all subtidal at this site), infaunal or epifaunal, and motile or sessile. With this information, production by each taxon was calculated and these values were summed to estimate faunal community production.

As the prediction error associated with community level production values is unknown, caution must be applied when interpreting model results. That said, the large prediction errors typically associated with population-level estimates are greatly reduced when pooled to the community level (Brey 2001). It should also be noted that the model requires mean annual abundance and biomass data for each taxon, whereas the available community data in 2018 are from a single survey conducted in April/May. As the abundance and biomass of taxa typically varies throughout the year, an under- or overestimation of total secondary production is possible. The degree to which this influences results will depend on how closely abundance and biomass in April/May resemble annual values.

Bioturbation potential

The bioturbation potential (i.e. the capacity of organisms to rework sediment) of macrofauna found in grab samples collected during the 2018 survey was quantified using a widely applied index (Solan *et al*. 2004, 2012; Birchenough *et al.* 2012; Morys *et al*. 2017). To derive this index, the per capita contribution of each taxon to bioturbation was calculated by multiplying the square root of its mean body mass (g) by a mobility score and then by a score derived from its sediment reworking mode. Scores ranged from 1 to 5, reflecting an increasingly positive effect on bioturbation (Table 10). This information was sourced from a comprehensive bioturbation classification of marine benthic invertebrates (Queirós *et al.* 2013), supplemented by additional literature review where required. The per capita bioturbation potential of each taxon was then multiplied by its abundance within a grab sample (converted to individuals $m²$) to give the population bioturbation potential. Finally, the population bioturbation potentials of all taxa within a grab sample were summed to give community bioturbation potential.

Table 10. Biological traits used to calculate the bioturbation potential of taxa in grab samples collected from the Farnes East MCZ in 2018. The scores associated with each mobility category and sediment reworking mode are shown (a higher score indicates a larger positive effect on bioturbation potential).

Habitat provision

The potential for habitat provision by the tubicolous polychaete *Sabellaria spinulosa* was inspected by comparing its density at sampling stations (individuals $m⁻²$) to those typical of reefs of low, medium, and high quality (Gubbay 2007).

Non-indigenous species (NIS)

The taxon lists generated from the grab samples and seabed imagery data were crossreferenced against lists of non-indigenous target species which have been selected for assessment of Good Environmental Status in GB waters under MSFD Descriptor 2 and identified as significant by the GB Non-Native Species Secretariat. These taxa are listed in Annex 5.

Numerical and statistical analyses

Multivariate analyses of macrofaunal communities were conducted in PRIMER (version 7; Clarke & Gorley 2015). Taxa abundances were transformed by $log_e(x+1)$ to downweigh the influence of numerically dominant taxa and allow variation in less abundant taxa to be detected. For each macrofaunal dataset analysed, a resemblance matrix was created from the Bray-Curtis similarities of each pair of grab samples. All multivariate analyses were performed on these resemblance matrices.

Two types of multivariate analysis were used to test whether macrofaunal communities differed significantly. Analysis of similarities (ANOSIM) was used to test for differences between pre-defined groups of samples (i.e. those from different stations or exposed to different levels of trawling activity), whereas hierarchical cluster analysis and associated similarity profile analysis (SIMPROF) were used to systematically divide samples into groups such that samples from the same group are not significantly different and samples from different groups are significantly different. Variation in macrofaunal community composition with respect to pre-defined groups was illustrated using non-metric MDS ordinations, whereas the hierarchical clustering of communities was illustrated using dendrograms. RELATE was used to test whether there was a significant correlation between macrofaunal

communities and the environment (e.g. mud content). For all analyses, a significant difference between groups or a significant correlation with the environment was concluded when *p* less than 0.05.

The similarity percentage (SIMPER) routine was used to determine the average similarity of macrofaunal communities in the same group (either pre-defined or derived from cluster analysis) and in different groups. SIMPER was also used to identify the taxa that contributed most to within-group similarity (i.e. the taxa that characterise a group) and between-group dissimilarity (i.e. the taxa that distinguish groups). The taxa that characterised groups were interpreted as the key structural taxa.

To assess variation in univariate community indices within and between groups, bar charts were plotted showing the mean and 95% confidence interval for all samples within each group. The same approach was used to assess variation in ecological function indices. Taxa within each group were then ordered by their mean contribution to each ecological function (i.e. from highest to lowest population-level secondary production or bioturbation potential), and the taxa that made the largest contributions were inferred to be the key functional taxa.

To inform potential future surveys, power analyses were used to determine how many samples would be required to have an 80% chance of detecting a 20% change in univariate community indices at each of the 16 stations where replicate samples were collected in 2018. The power analyses were conducted using the *power.t.test* function in R (R Core Team 2020).

Assigning biotopes

Biotopes were assigned to the areas surveyed within the Farnes East MCZ.

A selection of potential Marine Habitat Classification of Britain and Ireland (v15.03) biotopes were chosen based on the sediment and habitat information. The taxa associated with these biotopes were compared to the taxa that characterise the 'representative' stations and the main mud basin community clusters (see Annex 6) to determine how closely the potential biotopes and the sampled macrofauna communities match up.

In PRIMER, the abundance matrices for macrofauna samples from the Farnes East MCZ were merged with abundance matrices for the following biotopes for statistical analysis:

- SS.SCS.CCS.MedLumVen
- SS.SSa.CFiSa.EpusOborApri
- SS.SSa.OSa.OfusAfil
- SS.SMx.CMx.MysThyMx
- SS.SBR.PoR.SspiMx.

For the 'mud basin' clusters, characteristic taxa were first compared to those of SS.SCS.ICS.CumCset, SS.SSa.OSa.MalEdef, SS.Ssa.Osa.OfusAfil, SS.SMx.Omx.PoVen, SS.SMx.CMx.MysThyMx, SS.SSa.CFiSa.ApriBatPo, and SS.SSa.CFiSa.EpusOborApri using expert judgement. This informed the decision to merge the abundance matrices from the mud basins with the matrices for a subset of these biotopes (SS.SSa.CFiSa.EpusOborApri, SS.SMx.Omx.PoVen, SS.Ssa.Osa.OfusAfil, and SS.SMx.CMx.MysThMx) for statistical analysis.

For both 'representative' stations and 'mud basin' clusters, the merged macrofauna datasets were transformed by $log_e(x+1)$ in accordance with the approach used to identify

characteristic taxa within the Farnes East MCZ. Bray-Curtis similarity resemblance matrices were produced from these data and nMDS plots were created to illustrate the association between macrofauna samples and biotopes (see Figure 31 in Annex 9). As the assignment of biotopes provided *a priori* conditions for testing, ANOSIM was used to statistically determine the similarity between the communities of the stations/clusters and those associated with the biotopes. The R values that resulted from these tests are presented in Table 21 (stations) and Table 22 (clusters) in Annex 9. Communities are increasingly different as the R value increases and, therefore, a lower R values indicates a closer match between the station/cluster and the biotope.

The results of these statistical analyses were considered alongside information on the key taxa (for community samples and biotopes) and the sedimentary habitat before a final recommendation was made on which biotope should be assigned to each station/cluster. In instances where one biotope had a better statistical fit, but another had a more reasonable descriptor fit (for example if the sedimentary habitats were significantly different), a closer biotope has been recommended. In instances where community clusters were like two or more biotopes, a 'transitional' biotope was recommended, consisting of primary, secondary, and (if required) tertiary biotopes, starting with the most statistically similar.

All recommendations were reviewed by a JNCC specialist and any changes in the recommended biotope were discussed and agreed before being assigned.

Annex 4. Marine litter categories

Table 11. Categories and sub-categories of litter items for Sea-Floor (European Commission 2013).

Annex 5. Non-indigenous species lists

Table 12. 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).

Table 13. 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.

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Annex 6. Key structural taxa

Table 14. Taxa that contributed to a cumulative 70% of the internal Bray-Curtis similarity for macrofaunal communities, based on log_e(x+1) taxa abundances, at the 16 stations sampled with ten replicate grabs. The average community similarity and the Broadscale Habitats (BSHs) at each station are shown, with the number of samples in brackets.

Table 15. Taxa that contributed to a cumulative 70% of the internal Bray-Curtis similarity for the two main macrofaunal community clusters, based on loge(x+1) taxa abundances, in the northern and southern mud basins. The average community similarity and the Broadscale Habitats (BSHs) for each cluster are shown, with the number of samples in brackets.

Annex 7. Key functional taxa

Table 16. Taxa that contributed to a cumulative 70% of macrofaunal community production at the 16 stations sampled with ten replicate grabs. The average community similarity and the Broadscale Habitats (BSHs) at each station are shown, with the number of samples in brackets.

Table 17. Taxa that contributed to a cumulative 70% of macrofaunal community bioturbation potential at the 16 stations sampled with ten replicate samples. The average community similarity and the Broadscale Habitats (BSHs) at each station are shown, with the number of samples in brackets.

Annex 8. Macrofaunal communities in 2012 *vs.* **2018**

Table 20. Taxa that make up the top ten contributors to dissimilarity in macrofaunal communities, based on log_e(x+1) taxa abundances, between 2012 and 2018 at the 16 'representative' stations (n = 1 for 2012; n = 10 for 2018). The average community similarity in 2012 and 2018 and the Broadscale Habitats (BSHs) at each station are shown, with the number of samples in brackets. Taxa abundances in 2012 and 2018 are presented using untransformed data.

Annex 9. Biotopes

Figure 32. Non-metric MDS ordinations of macrofaunal community composition, based on loge(x+1) taxa abundances, at A) the 16 stations sampled with ten replicate grabs (2D stress = 0.25) and B) the 52 stations within the two mud basins (2D stress = 0.24). Macrofaunal community composition of potential MHCBI biotopes are also shown in both A) and B).

Table 21. ANOSIM results comparing macrofaunal community composition, based on log_e(x+1) taxa abundances, at the 16 stations sampled with ten replicate grabs to the taxa associated with potential biotopes. A closer statistical match is indicated by a lower R value (the lowest R value is in bold). The biotope assigned to each station (based on statistical analysis and inspection of characteristic taxa and sedimentary habitat) are shown alongside any relevant notes relating to biotope assignment.

Table 22. ANOSIM results comparing macrofaunal community composition, based on log_e(x+1) taxa abundances, of the two main clusters within the mud basins. A closer statistical match is indicated by a lower R value (the lowest R value is in bold). The biotope assigned to each community clusters (based on statistical analysis and inspection of characteristic taxa and sedimentary habitat) are shown alongside any relevant notes relating to biotope assignment.

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