Offshore Wind Power Limited

West of Orkney Offshore EIA Report

Volume 2, Supporting Study 5: Benthic Environmental Baseline Report

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Benthic and Environmental Survey

West of Orkney Windfarm

Volume 2 - Environmental Baseline Report

WEST OF ORKNEY WINDFARM

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West of Orkney Windfarm – List of Survey Reports

Offshore Geophysical Site Investigation Report

Volume	Title	OI Document No.	TEPUK Document No.
Vol. 1	Offshore Agreement Area (OAA) Results Report	103836-TOT-OI-SUR-REP-OOAREP	GB-GEN-00-MMT-000007
Vol. 2a	ECC Whiten Head Bank to Crosskirk Results Report (Including Greeny Geo Spur)	103836-TOT-OI-SUR-REP-ECCWHBCREP	GB-GEN-00-MMT-000008
Vol. 2b	ECC Stormy Bank to Crosskirk Results Report	103836-TOT-OI-SUR-REP-ECCSBCREP	GB-GEN-00-MMT-000009
Vol. 2c	ECC Route Centre Lines	103836-TOT-OI-SUR-REP-ECCRCLREP	GB-GEN-00-MMT-000010
Vol. 3	MMO/PAM Report	103836-TOT-OI-SUR-REP-MMOPAMREP	GB-GEN-00-MMT-000011
Vol. 4	Operations Report	103836-TOT-OI-SUR-REP-OPEREP	GB-GEN-00-MMT-000012
Vol. 5	UHRS Interpretation Report	103836-TOT-OI-SUR-REP-INTREPP1	GB-GEN-00-MMT-000013

Benthic and Environmental Report

Volume	Title	OI Document No.	TEPUK Document No.
Vol. 1	Habitat Assessment Report	104164-TOT-OI-SUR-REP-HABASRE	GB-GEN-00-MMT-000029
Vol. 2	**Environmental Baseline Report**	**104164-TOT-OI-SUR-REP-ENVBASRE**	**GB-GEN-00-MMT-000030**
Vol. 3	MMO Report	104164-TOT-OI-SUR-REP-MMOREP	GB-GEN-00-MMT-000031
Vol. 4	Operations Report	104164-TOT-OI-SUR-REP-OPERATRE	GB-GEN-00-MMT-000032

Geotechnical Investigation Report

Volume	Title	OI Document No.	TEPUK Document No.
Vol. 1	Shallow Geotechnical Results Report	103836-TOT-OI-SUR-REP-SHGEOTER	GB-GEN-00-MMT-000036

Project Field Report

Volume	Title	OI Document No.	TEPUK Document No.
N/A	Geophysical Survey Field Report Phase I	103836-TOT-OI-SUR-REP-FIEREPP1	GB-GEN-00-MMT-000020
N/A	Geophysical Survey Field Report Phase II	103836-TOT-OI-SUR-REP-FIEREPP2	GB-GEN-00-MMT-000018
N/A	Geophysical Survey Field Report Phase III	103836-TOT-OI-SUR-REP-FIEREPP3	GB-GEN-00-MMT-000019
N/A	Environmental Survey Field Report	104164-TOT-OI-SUR-REP-ENVFIELDREP	GB-GEN-00-MMT-000033
N/A	Shallow Geotechnical Investigation Field Report	103836-TOT-OI-FIE-REP-SHGEOTEI	GB-GEN-00-MMT-000035

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Abbreviations and Definitions

ΣEPA 16 PAH	Sum of the 16 EPA PAHs
ΣPCB7	Sum of the ICES seven ICES PCBs
AL1	Action Level 1
AL2	Action Level 2
BAC	Background Assessment Concentrations
CCME	Canadian Council of Ministers of the Environment
CEFAS	Centre for Environment, Fisheries and Aquaculture Science
DBT	Dibutyltin
DDV	Drop Down Video
DNA	Deoxyribonucleic Acid
DP	Designated Person
DPR	Daily Progress Report
DVV	Dual Van Veen grab
EAC	Environmental Assessment Criteria
EC	European Commission
ECC	Export Cable Corridor
eDNA	Environmental DNA
EIA	Environmental Impact Assessment
ERL	Effect Range-Low
EUNIS	European Nature Information System
ICES	International Council for the Exploration of the Sea
GIS	Geographic Information System
GNSS	Global Navigation Satellite System
HG	Hamon Grab
ISQG	Interim Sediment Quality Guidelines
JNCC	Joint Nature Conservation Committee
LAT	Lowest Astronomical Tide (vertical datum)
LoD	Limit of Detection
MAC	Mobilisation and Calibration
MAG	Magnetometer
MBT	Monobutyltin
MBES	Multibeam Echo Sounder
MDS	Multi-Dimensional Scaling
MESH	Mapping European Seabed Habitats
M/V	Motor Vessel
nMDS	Non-Metric Multi-Dimensional Scaling
NEA	Norwegian Environment Agency
NMBAQC	National Marine Biological Analytical Quality Control
OAA	Option Agreement Area

OCP.....Organochlorine Pesticides
OI.....Ocean Infinity Group Holding (Sweden) AB
OSPAR .....The Oslo and Paris Conventions for the protection of the marine environment of the North-East Atlantic
PAH.....Polycyclic Aromatic Hydrocarbons
PCA .....Principal Component Analysis
PCB .....Polychlorinated Biphenyls
PEL.....Probable Effect Level
PBDE.....Polybrominated Flame Retardants
PMF .....Priority Marine Feature
PPS .....Pulser Per Second
PRIMER.....Plymouth Routines in Multivariate Ecological Research
PSA .....Particle Size Analysis
PSU .....Practical Salinity Unit
REDOX .....Reduction-Oxidation
SBL.....Scottish Biodiversity List
SBP .....Sub-bottom Profiler
SIMPER .....Similarity Percentage Analysis
SIMPROF.....Similarity Profiles
SOW .....Scope of Work
SSS.....Side Scan Sonar
TBT .....Tributyltin
THC.....Total Hydrocarbons
TOC.....Total Organic Carbon
TOM .....Total Organic Matter
TSS.....Total Suspended Solids
UK.....United Kingdom
USBL .....Ultra Short Base Line
UTC.....Coordinated Universal Time
UTM.....Universal Transverse Mercator
VORF.....Vertical Offshore Reference Frame
VRM.....Vector Ruggedness Measure

Executive Summary

This report details the results of the Environmental Baseline Survey for the West of Orkney offshore windfarm, located 28 km west of Hoy, Orkney, Scotland.

The offshore benthic and environmental survey data acquisition included sediment sampling and imagery, with continuous video, water sampling and Conductivity, Temperature, and Depth profiling to establish a baseline for the habitats and faunal communities within the survey area. The benthic and environmental survey was carried out from the survey vessel M/V Geo Ranger between the 15th of August and the 13th of September 2022.

Seabed imagery was undertaken at all of the 82 planned grab samples sites, as well as 17 standalone drop down video transects. Samples were collected at 73 of the planned 82 grab sample sites. All of the 20 planned water sample sites, including the 4 sites selected for sampling at two different tidal cycles, were completed.

A nearshore survey was carried out between the 16th and the 26th of October by Spectrum Geosurvey Limited and Ocean Ecology Limited from the survey vessel M/V Spectrum 1. Based on geophysical data and the completion of nine drop down video (DDV) transects, four nearshore grab sample sites were selected. Out of the four selected grab sample sites, three were successfully sampled. All five planned water sampling sites were successfully sampled.

Geophysical data were used to determine water depths, surficial geology, seabed features, shallow geology, and objects present within the survey area. Instruments used during the geophysical survey included Multibeam Echo Sounder, Side Scan Sonar Sub-Bottom Profiler, Sparker and Magnetometer. During the nearshore geophysical survey, vessels acquired Multibeam Echo Sounder, Side Scan Sonar, Magnetometer and Sub-Bottom Profiler data.

The geophysical interpretation combined with the environmental data was used as the basis for the European Nature Information System habitat classifications and assessments of potential areas and species of conservation importance.

A total of seven European Nature Information System habitats including two habitat complexes, as well as four habitats of conservation importance, were identified within the survey area. The taxonomic assemblages from the acquired grab sample data indicate the presence of 15 sample-specific habitats across the survey area, including 6 transitional habitat complexes.

One Habitats Directive Annex I habitat was interpreted to be present, 1170 Reefs with its two subtypes “Bedrock Reefs”, and “Stony Reefs”. Areas of “Stony Reefs” were assessed and delineated based on current guidance on resemblance. This resulted in the division of areas of hard substrata into four categories of “Stony Reefs”: Potential Reef, Low Resemblance, Low to Medium Resemblance and Medium Resemblance. The Potential Reefs are mainly located in the western Option Agreement Area and Export Cable Corridors whereas the Low to Medium Reefs are primarily located in the Option Agreement Area.

The results of the particle size analysis showed limited variation in sediment composition throughout the survey area, with sand and gravel being the main sediment fractions.

Metal concentrations were generally low, but levels of arsenic and nickel exceeded thresholds at several grab sample sites. Hydrocarbon concentrations were low and variable with generally higher concentrations in the nearshore samples, with a few sites exceeding threshold values for polycyclic aromatic hydrocarbons. Polychlorinated biphenyl and organochloride concentrations were low and only exceeded the limit of detection at a few sites. Organotin concentrations were below the limit of detection for all analysed samples. Concentrations of brominated flame retardants were overall low and exceeded threshold values in all the samples which were above the limit of detection.

The faunal analyses of the grab sample showed that the phyletic composition was dominated by annelids. The two most abundant taxa were the annelid *Owenia* and the roundworm Nematoda. Pielou's Evenness index, Shannon-Wiener index, and Simpson's Index of Dominance had limited variation, were as Margalef's Richness Index presented a slightly higher variation, with the Similarity Profile Routine test identifying 21 faunal groups. Mollusca comprised most of the biomass. The colonial fauna was dominated by Bryozoa.

The most abundant non-colonial fauna identified in stills was Mollusca, followed by Arthropoda, and the colonial fauna with the highest coverage was Bryozoa.

Density measurements showed no indication of strong pycnoclines in the survey area. Temperature measurements exhibited limited variation, appearing most variable offshore and declining with depth. The lowest salinity measurements were all obtained from the surface water and the average salinity of 34.8 PSU is in line with measurements in Scottish waters.

Turbidity sensor measurements were generally low, with higher concentrations noted in the Export Cable Corridors when compared to the Option Agreement Area. The levels of Total Suspended Solids were also generally low, typically below the threshold of <5 mg/l at most sites but presented higher at sites W15 – W20 and W24 in the Export Cable Corridors.

Environmental DNA, conducted on the water samples, from the Vertebrate assay detected 42 taxa and the sequence was dominated by *Gadidae* sp. while the Fish assay detected 34 taxa and the sequence was dominated by Atlantic mackerel *Scomber scombrus*. The Invertebrate assay detected 407 taxa and was dominated by a sequence of *Siphonophorae* sp. The Invertebrate assay detected taxa from Animalia, Chromista, Fungi, Plantae and Protozoa. The Marine Mammal assay detected 7 taxa and the sequence was dominated by common bottlenose dolphin *Tursiops truncates.*

Fourteen species of conservation importance and one non-native species were identified within the survey area.

The majority of the delineated habitat polygons had ground-truthed data collected resulting in 66 % of the survey area having a MESH confidence score of ≥76. Additionally, 38 % of the area had a confidence score of ≥90. The overall high scores indicate a robust assessment and confidence in the habitats identified within the Option Agreement Area and Export Cable Corridors.

1. Introduction

1.1 Project Information

Ocean Infinity (OI) have been contracted by TotalEnergies E&P UK (TEPUK) to perform a benthic and environmental survey and a geophysical survey for the West of Orkney offshore windfarm Option Agreement Area (OAA) and the preferred Export Cable Corridors (ECC) to the Scottish mainland. The OAA extends over 657 km² and is located approximately 28 km west of Hoy, Orkney, Scotland (Figure 1).

A nearshore benthic and environmental survey and a geophysical survey were also performed by Spectrum Geosurvey Limited and Ocean Ecology Limited for the West of Orkney offshore windfarm. The nearshore scope of work covered Crosskirk Bay and the surrounding area on the Caithness coast, Scotland.

Project specific details are presented in Table 1.

Table 1 Project details.

Client:	TotalEnergies E&P UK Ltd
Project:	West of Orkney Windfarm
OI Sweden AB Project Number:	104164
Survey Type:	Benthic and Environmental Survey
Area:	North-East Atlantic - West of Orkney, UK
Survey Period:	August - September 2022 October 2022 (Nearshore Survey)
Survey Vessel:	M/V Geo Ranger M/V Spectrum 1
OI Project Manager:	Sara Andersson
Client Project Manager:	Mark Grove Smith

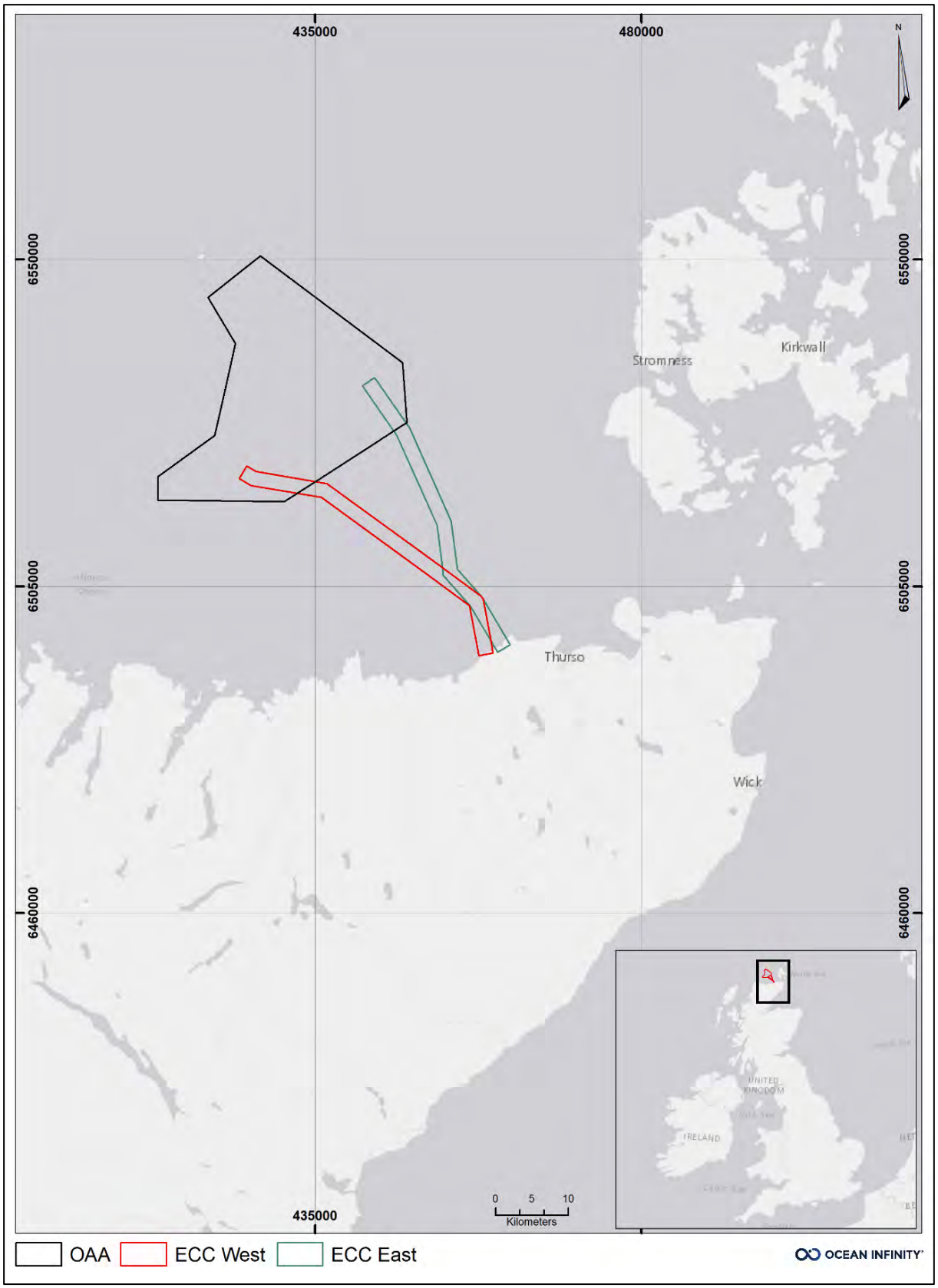


Figure 1 Overview of the survey area.

1.2 Scope of Work – Benthic and Environmental Survey

The aim of the Benthic and Environmental Survey was to collect data for Habitat Assessment and to provide an Environmental Baseline to allow the future determination of possible environmental impacts as a result of windfarm development in the area. This consisted of both an offshore and nearshore sampling campaign.

The following summarises the Environmental Survey Scope of Work:

- Drop down video (DDV) for identifying epifauna and habitats
- Grab sampling for faunal taxonomy, biomass, particle size analysis (PSA) and contaminants
- Water sampling together with CTD profiling for contaminants and Environmental DNA (eDNA)
- Identify and delineate habitats and protected features, such as Annex I habitats and PMFs
- Provide an Environmental Baseline to support the Environmental Impact Assessment (EIA) process

1.3 Scope of Work – Geophysical Survey

The aim of the Geophysical Survey was to acquire data to evaluate the seabed and sub-seabed conditions, including potential associated hazards (geohazards or man-made hazards), affecting the future installation of wind turbines and subsea cables.

The Geophysical Survey scope included the acquisition of closely spaced (62.5 m survey line separation) multibeam echo sounder (MBES), side scan sonar (SSS), magnetometer (MAG) and sub-bottom profiler (SBP) data in the Option Agreement Area (OAA) and Export Cable Corridors (ECCs).

1.4 Purpose of Document

The purpose of this Report is to present the Benthic and Environmental Baseline survey methodology and results within the survey area. This report, together with overview charts and Geographic Information System (GIS) database, presents the baseline environmental conditions from the West of Orkney Windfarm.

Areas of specific interest within the survey area are presented in this report as well as in the GIS overview habitat charts. All existing OI data from the Geophysical and, Benthic and Environmental Survey are correlated to each other and compared against the existing background information and the publicly available environmental data, to strengthen the accuracy of the interpretations.

This Report comprises findings from the Habitat Assessment Report (104164-TOT-OI-SUR-REP-HABASRE) and further includes the result of the subsequent Environmental Baseline Survey's laboratory analyses. Where results from the laboratory analyses have led to a re-assessment of the habitat classifications these have been included in this Report, as well as updated in the Habitat Assessment Report.

2. Survey Parameters

2.1 Geodetic Datum and Grid Coordinate System

2.1.1 Geodetic Datum

The geodetic datum parameters used during acquisition during the project are presented in Table 2.

Table 2 Geodetic Datum.

Horizontal Datum: (WGS84 EPSG4326)	
Datum	World Geodetic System 1984 (6326)
Ellipsoid	World Geodetic System 1984 (7030)
Prime Meridian	Greenwich (8901)
Semi-major axis	6 378 137.000 m
Semi-minor axis	6 356 752.3142 m
Inverse Flattening (1/f)	298.257223563
Unit	International metre

2.1.2 Projection Parameters

The projection parameters used during the project are presented in Table 3.

Table 3 Projection parameters.

Projection Parameters	
Projection	UTM
Zone	30 N
Central Meridian	03° 00’ 00’’ W
Latitude origin	0
False Northing	0 m
False Easting	500 000 m
Central Scale Factor	0.9996
Units	metres

2.2 Vertical Reference Parameters

The vertical reference parameters used during the project are presented in Table 4.

Table 4 Vertical reference parameters.

Vertical Reference Parameters	
Vertical reference	LAT
Height model	VORF

2.3 Time Datum

Coordinated universal time (UTC) was used on all survey systems on board all vessels. The synchronisation of the vessel's onboard system is governed by the pulse per second (PPS) issued by the primary positioning system. All displays, overlays and logbooks are annotated in UTC as well as the daily progress report (DPR).

3. Survey Performance

3.1 Survey Tasks

The survey tasks are presented in Table 5.

Table 5 Environmental survey tasks.

Task	Date	Description
OAA and ECCs Offshore		
Mobilisation and calibration	2022-08-15 2022-08-21	Mobilisation and calibration in Scrabster
Environmental survey	2022-08-21 2022-09-11 2022-08-21 2022-09-11	First grab site completed Last grab site completed First water/eDNA site completed Last water/eDNA site completed
Crew changes	2022-08-31 2022-09-07	First crew change Second crew change
Demobilisation	2022-09-12 2022-09-13	Personnel and equipment demobilisation in Scrabster
ECCs Nearshore		
Mobilisation and calibration	2022-10-16 2022-10-21	Mobilisation and calibration in Scrabster
Environmental survey	2022-10-22 2022-10-25 2022-10-23 2022-10-25 2022-10-25	First water site completed Last water site completed DDV began and completed First grab site completed Last grab site completed
Crew changes	2022-10-16	First crew change
Demobilisation	2022-10-26	Personnel and equipment demobilisation in Scrabster

3.2 Mobilisation and Calibration Test

Mobilisation and Calibration (MAC) commenced on the 15th of August 2022 in Scrabster and was completed on the 21st of August 2022. Equipment was trialled alongside the jetty and outside the harbour area.

For a detailed description of the calibration performance and results please refer to the MAC report 104164-TOT-OI-MAC-REP-GEORANGER. Further information about the equipment set-up and performance can be found in the Benthic and Environmental Survey Operations Report.

Mobilisation and Calibration for the environmental nearshore survey commenced on the 16th of October in Scrabster and was completed on the 21st of October following the completion of the nearshore geophysical survey. Equipment was trialled alongside the jetty and outside the harbour area. For further information about the environmental survey set up and performance, please refer to OEL_SPEWES0522_MOB_V01.

3.3 Vessels and Equipment

M/V Geo Ranger

The Environmental survey operation was conducted by the M/V Geo Ranger, shown in Figure 2.

The vessel M/V Geo Ranger is equipped with navigation and positioning systems as stated in Table 6 and Table 7. The vessel was equipped with a stern A-frame and two (2) winches for the deployment and recovery of equipment.

Further information about the equipment set-up and performance can be found in the Environmental Operations Report 104164-TOT-OI-SUR-REP-OPERATRE.

Figure 2 M/V Geo Ranger.

Table 6 Vessel Equipment M/V Geo Ranger.

Equipment	Model	Quantity
Primary positioning system	C-nav 3050 with c-navc2 corrections on the sf2 service	1
Secondary positioning system	Trimble SPS855 with Fugro G4+ corrections	1
Heading/motion sensor	Ixblue Hydrins 3	1
USBL	Kongsberg Hipap 502	1
Sound velocity profiler	Valeport SVX2	2

Table 7 M/V Geo Ranger Environmental Survey Equipment.

Equipment	Model
Benthic Grab	Dual Van Veen (2 x 0.1 m^2), Day Grab (0.1 m^2) Hamon Grab (0.1 m^2)
Video system and photographic camera	STR SeaSpyder
Water Sampler	Rosette sampler & 5 L Niskin Bottles
eDNA Sampler	Vampire Sampler

M/V Spectrum 1

The nearshore environmental survey operations were conducted by the M/V Spectrum 1, shown in Figure 3. The vessel M/V Spectrum 1 is equipped with navigation and positioning systems as stated in Table 8 and Table 9. The vessel is equipped with an aft deck crane for the deployment and recovery of equipment.

Figure 3 M/V Spectrum 1 (Image Source: Mobilisation & Calibration report – Spectrum 1, 02.22.16.MOB.S1).

Table 8 Vessel Equipment M/V Spectrum 1.

Equipment	Model	Quantity
Primary positioning system	Applanix POS MV GNSS antenna	1
Secondary positioning system	Applanix POS MV secondary GNSS antenna	1
Heading/motion sensor	Applanix POS MV IMU	1
USBL	Sonardyne Ranger 2	1
Sound velocity profiler	Valeport SWiFT miniSVP	1

Table 9 M/V Spectrum 1 Environmental Survey Equipment.

Equipment	Name
Benthic Grabs	Shipek Grab (0.05 m^2) Hamon Grab (0.1 m^2)
Video system and photographic camera	SubC Imaging Rayfin BPE
Water Sampler	5 L Niskin Bottles
Water Profiler	YSI EXO 3 Sonde

4. Methodology

4.1 Survey Design

The number and location of environmental sample sites were decided based on depth variation, sediment, and habitat changes, as delineated during the geophysical survey, to provide benthic data for all habitats interpreted across the survey area. The survey plan was in line with the OWPL Benthic Survey Sampling Strategy (100-WOW-CON-G-GA-0001-01).

A Senior Benthic Ecologist reviewed the geophysical data and planned the benthic and environmental survey. To ensure that the different habitats as interpreted from the Side Scan Sonar (SSS), Multibeam Echo Sounder (MBES), including normalised backscatter values, were ground-truthed a total of 95 sites (82 dedicated for grab sampling and 13 for standalone video transects) were selected for sampling. A detailed account of selected sites, including an overview of the geophysical data at each sample site, is presented in Appendix A.

Final sampling sites were agreed upon in consultation with the Client prior to the commencement of the sample collection, and the site selection was validated through a rationale submitted to the Client. Water sampling locations were provided by the Client to OI.

Grab sampling was planned at a total of 82 sites. One (1) sample for taxonomic analyses (including biomass), one (1) sample for Particle Size Analysis (PSA), and one (1) sample for contaminants analyses were planned at each site. Samples from 31 of these sites were also analysed for pesticide and flame-retardant contaminants (Figure 4).

Replicate grab samples for fauna, PSA and contaminants analyses were planned at a total of 16 sites. The replicate samples were collected as a backup and not included in the analyses.

Before conducting grab sampling the Drop Down Video camera system (DDV) was deployed at each grab sample site. A minimum of 11 still images, with continuous video, were acquired at each grab sample site to connect epifaunal and faunal assemblages. Standalone video transects were planned at 13 sites, with a minimum of seven (7) images acquired.

Water sampling together with CTD profiling was planned at a total of 20 sites widely distributed across the OAA and ECCs. At four (4) of these sites, water sampling was to be carried out during spring tides and then sampled again during neap tides. Water samples were collected at three (3) depths (Bottom, Middle and Top). Further details about the sampling strategy can be found in the West of Orkney Windfarm Benthic Survey: Sampling Strategy Plan Doc No. 100-WOW-CON-G-GA-0001-01.

The environmental nearshore survey was conducted by Ocean Ecology Limited following the completion of the nearshore geophysical survey. Nine (9) DDV transects were planned and were conducted prior to grab sampling, with a minimum of six (6) images with continuous video acquired per transect. Grab sampling was planned at six (6) sites for taxonomic and biomass analyses, PSA, and contaminant analyses. Planned grab site suitability was assessed following a review of collected DDV data. Water sampling and profiling were planned at five (5) sites. Water samples were collected at three (3) depths (Bottom, Middle and Top).

The boundary between the offshore and nearshore surveys is based on the offshore vessels' operational safety due to the steep incline to shore and is located approximately 1.5 km out from the shoreline. The Benthic and Environmental survey sampling overlaps within this area.

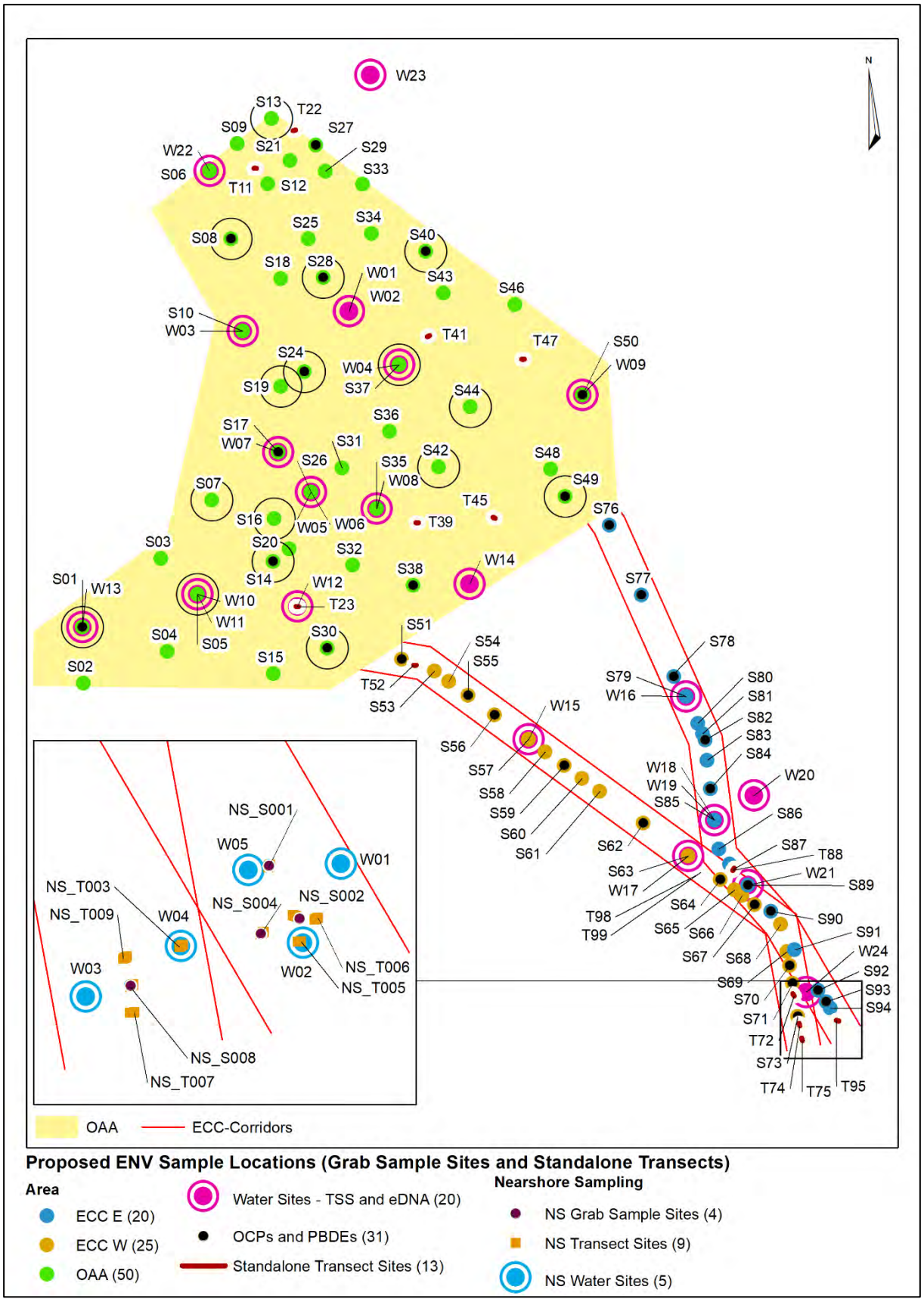


Figure 4 Overview of the proposed sampling design including the Nearshore survey.

4.2 Offshore Field Methods

4.2.1 Drop Down Video

An STR SeaSpyder Drop Down Video system was used to investigate grab sample sites as well for the standalone video transects (Figure 5 and Figure 6).

Figure 5 SeaSpyder DDV System.

Figure 6 SeaSpyder DDV example still photo.

A minimum of two (2) still photos were acquired every 5 m at each grab sample transect with continuous video recordings. The length of each transect at the planned grab sample sites was 50 m. The grab transects were nominally extended by 100 m if potential Annex I habitats were detected, to delineate the feature of interest.

The length of the standalone video transects was planned to be 150 m and performed in areas identified to potentially host habitats and/or features of conservation value i.e., potential Annex I habitats. Still photos were taken at the start of the transect and approximately every 25 m, with video being recorded continuously. Stills were taken more frequently if the seabed exhibited features of interest i.e., reefs and/or evidence of increased diversity.

The camera was positioned as close as possible to the pre-selected starting point using the vessel's dynamic positioning system during the survey. The camera frame was lowered onto the seabed to adjust the camera focus. When the camera focus was set, an initial photo was taken, before the video recording was initiated.

The camera frame was eased off the seabed and towed slowly at approximately 0.3 – 0.5 knots. It was positioned as close to the seabed as possible with an approximate altitude of 0.5 – 1 m. Altitude was determined by seabed topography and weather conditions.

A field log was maintained of the stills acquired for habitat assessments, at each grab sample with preliminary descriptions of findings. Anthropogenic impacts that were visible were recorded including evidence of fishing activity, existing infrastructure (e.g. pipes, cables), marine debris (ropes, chains, equipment) and litter.

Prior to grab sampling, an experienced marine biologist reviewed all video transect data onboard to confirm the presence/absence of any potentially sensitive habitats or features of conservation interest.

4.2.2 Faunal Grab Sampling

The primary grab sampler utilised for the faunal sampling was the Dual Van Veen (DVV; 2 x 0.1 m^2) and the secondary grab sampler, e.g. in areas of coarse sediment, was the Hamon grab (HG; 0.1 m^2) (Figure 7 and Figure 8). Fifty-four (54) faunal samples were acquired using the DVV, while 16 samples were acquired using the HG.

Upon retrieval, samples were checked for adequate sample volume and samples covering less than 0.1 m^2 of bottom surface sediment were deemed unacceptable. No samples of less than 5 cm (7 cm in fine sediments) of penetration depth for the DVV or 7 litres for HG were considered acceptable samples.

If an acceptable sample was not achieved within three (3) attempts at the grab sample site (e.g., in areas of coarse sediment) then this was recorded, and the survey continued with the next grab sample site. Samples that were not accepted were not included in any statistical analyses.

If an acceptable sample was not achieved for the replicate samples at the original location within the three (3) attempts (e.g., in areas of coarse sediment), then a fourth attempt was conducted. The fourth attempt was repositioned slightly, up to 50 m, to obtain a representative sample. Observations of existing geophysical data were undertaken onboard by the experienced marine biologist in order to determine the closest area of suitable substrate.

Acquired samples were carefully sieved using seawater in a 5 mm mesh sieve over a 0.5 mm mesh sieve. Faunal samples were preserved on board in 96 % ethanol directly after the sieving was completed. The 5 mm and 0.5 mm fractions were kept in separate jars, labelled with a unique label containing the grab sample site ID and replicate number.

A field log of sampling equipment, sample positions, including time, sediment type, and water depth, was kept for later reference. Samples were photo-documented in situ. For further information regarding sample volume and the number of attempts, see the Environmental Survey Field Report (104164-TOT-OI-SUR-REP-ENVFIELDREP).

Figure 7 Dual Van Veen grab sampler.

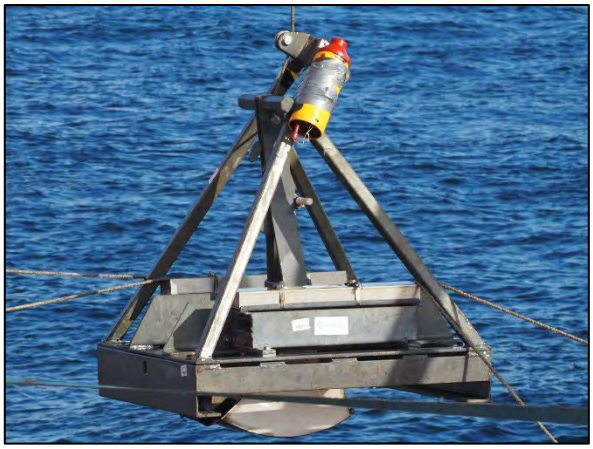


Figure 8 Hamon grab sampler.

4.2.3 Particle Size and Contaminants Grab Sampling

Samples were taken to investigate the sediment's physical and chemical properties, including the presence of any contaminants. The primary grab sampler utilised for the PSA and contaminants sampling was the Dual Van Veen (DVV). The Hamon Grab (HG) was used as a secondary grab to sample PSA in areas of coarse sediment, however, the HG could not be used for contaminant samples. Sixty (60) PSA samples were acquired using the DVV, while seven (7) samples were acquired using the HG. All 57 contaminant samples were acquired using the DVV.

Upon retrieval, samples were checked for adequate sample volume and samples covering less than 0.1 m^2 of bottom surface sediment were deemed unacceptable. No samples of less than 5 cm (7 cm in fine sediments) of penetration depth for the DVV or 2.7 litres for HG were considered acceptable samples (Worsfold, Hall, & O'Reilly, 2010; Davies, et al., 2001).

Sample re-attempts follow the same procedures as outlined in 4.2.2.

Samples for metals, organics (Total Organic Matter (TOM) and Total Organic Carbon (TOC)), hydrocarbons (Total Hydrocarbons (THC) and Polycyclic Aromatic Hydrocarbon (PAH)), Polychlorinated Biphenyls (PCB), organotins (Monobutyltin (MBT), Dibutyltin (DBT) and Tributyltin (TBT)), pesticides and flame-retardants were sampled from an undisturbed surface. The sediments were collected with a plastic spoon for metals and a metal spoon for organics, hydrocarbons, PCB, organotins, pesticides and flame-retardant to ensure minimal contamination risk. The grab sampler was cleaned between samples and sample sites.

A one (1) litre plastic container was used for the metal samples as well as PSA samples. For the contaminant analyses of organics, hydrocarbons, PCBs, organotins, pesticides and flame-retardants, a 250 ml tin container was used for storage. The different containers ensured that there was no outside contamination of the samples.

The sample containers were labelled with a unique sample site ID. The contaminants samples were stored frozen (-21°C) according to the analysing laboratory's recommendations before and during shipment.

REDOX potential was measured at each grab sample site, either from the faunal sample or the PSA sample, using a probe. Measurements were taken from 4 cm deep in the collected sample. The readings on the probe were recorded in the field protocols.

A field log of sampling equipment, sample positions including time, sediment type, and water depth was kept for later reference. Samples were photo-documented in situ. For further information regarding sample volume and the number of attempts, see the Environmental Survey Field Report (104164-TOT-OI-SUR-REP-ENVFIELDREP).

4.2.4 Water Sampling and CTD

Water sampling was performed using 5 L Niskin bottles attached to a Rosette sampler (Figure 9). The open bottles were lowered into the water and closed at pre-assigned depths. A CTD and external turbidity sensor was fitted to the Rosette sampler. There were five (5) Niskin bottles attached to the Rosette, two (2) for bottom water samples, one (1) for the midwater sample, and two (2) for topwater samples. The bottles were labelled according to the depth they triggered (Bottom, Middle and Top). Samples from the three (3) depths were collected from a single cast, where water was collected within 1-2 m below the sea surface, at mid-depth and 1-2 m above the seabed. Where grab sampling and water sampling coincided, the water sampling was acquired prior to the deployment of the DDV or grab sampler.

As the Rosette sampler was winched down to the bottom, the CTD and the external turbidity sensor recorded depth, temperature, conductivity/salinity, Chlorophyll-a, dissolved oxygen, REDOX potential, and turbidity in the water column. When close to the seabed, a position fix was taken, and the rosette sampler was then winched upwards for recovery. During the upcast, the bottles closed at their pre-assigned depths to collect the water samples.

Once the sampler was recovered to the vessel, the water for TSS analysis was collected from the Bottom, Middle and Top bottles into pre-labelled 1 L plastic containers and stored in the onboard freezer at -21°C.

Retrieved samples were assigned a sample number and their UTM coordinates, date and time of collection, and water depth were documented. The data from the CTD and turbidity sensor was downloaded and saved to a hard drive separately upon recovery for each cast at each sample site.

Figure 9 Rosette sampler equipped with Niskin bottles, CTD and turbidity sensor.

4.2.5 eDNA

Upon recovery of the rosette sampler, two (2) Niskin bottles for Top and Bottom water were filtered for eDNA using the Vampire sampling pump and following the guidance specifications provided by NatureMetrics. Water sampling for eDNA was carried out to determine the presence of fish, vertebrates, marine mammals, and invertebrates.

Care was taken to minimize contamination by performing the eDNA sampling in a dedicated area on the back deck. Separate eDNA sampling kits, consisting of Nitrile gloves, enclosed filters, a syringe filled with preservative solution, silicone hose, specimen bag, datasheet and disinfectant wipe were used for each sample/water body to avoid cross-contamination.

The silicone hose was attached to the outlet of the Niskin bottle with an enclosed filter (0.8 µm pore size, polyethersulfone) attached to the hose adapter. Once the entire 5 L Niskin bottle was filtered through, the filter was carefully detached from the hose. A syringe filled with 1.5 mL DNA preservative solution was twisted onto the filter and the entire preservative solution was slowly added into the filter. The filter was then detached from the syringe and sealed with a separate Luer Lock cap.

Filters were stored in a resealable specimen bag, which in turn were placed into the eDNA kit bags alongside the datasheet noting the Sample ID and depth from which it was taken. Samples were then stored in a chest freezer at -21 °C.

4.3 Nearshore Field Methods

4.3.1 Drop Down Video

An Ocean Ecology (OEL) freshwater housing with a SubC Rayfin Camera System was used to investigate each proposed grab sampling site as well as for standalone video transects (Figure 10 and Figure 11).

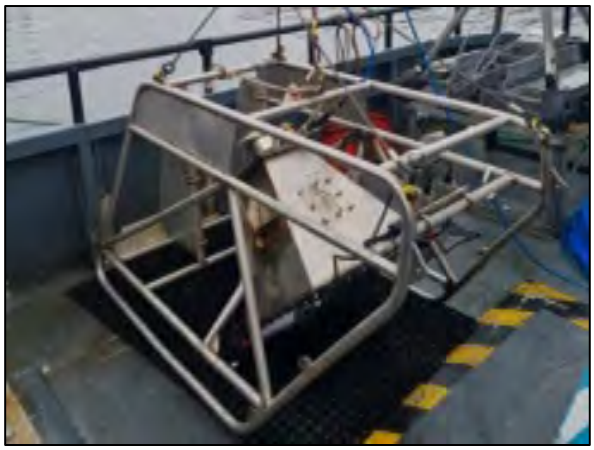


Figure 10 OEL SubC Rayfin Camera System (Image Source: Survey Operational Report – 02.22.16.OPS).

Figure 11 SubC Rayfin Camera System example still photo (Image Source: Survey Operational Report – 02.22.16.OPS).

A minimum of 1 still image was acquired every 5 m at each transect with continuous video recordings. The length at each planned grab sample site was 50 m. The camera was deployed to the seabed and slowly towed just above the seabed whilst recording continuous video footage. The footage was reviewed in real-time by the onboard marine biologist.

Prior to grab sampling, an experienced marine biologist reviewed all video transect data onboard to confirm the presence/absence of any potentially sensitive habitats or features of conservation interest.

4.3.2 Faunal Grab Sampling

Grab sampling was conducted following the completion of the camera scope and review of video transect data. The grab sampler utilised for the faunal sampling was the Hamon grab (HG; 0.1 m^2). The HG is suitable for sampling in areas of coarse sediments, such as gravel, pebbles, and small cobbles.

Upon retrieval, samples were checked for adequate sample volume and samples covering less than 0.1 m^2 of bottom surface sediment were deemed unacceptable. No samples of less than 7 litres for HG were considered acceptable samples.

If an acceptable sample was not achieved within three (3) attempts at the grab sample site (e.g., in areas of coarse sediment) then this was recorded, and the survey continued with the next grab sample site. Samples that were not accepted were not included in any statistical analyses. Acquired samples were carefully sieved using seawater in a 5 mm mesh sieve over a 0.5 mm mesh sieve. Faunal samples were processed and stored in 10 L plastic buckets.

A field log of sample positions, including time, sediment type, and water depth, was kept for later reference.

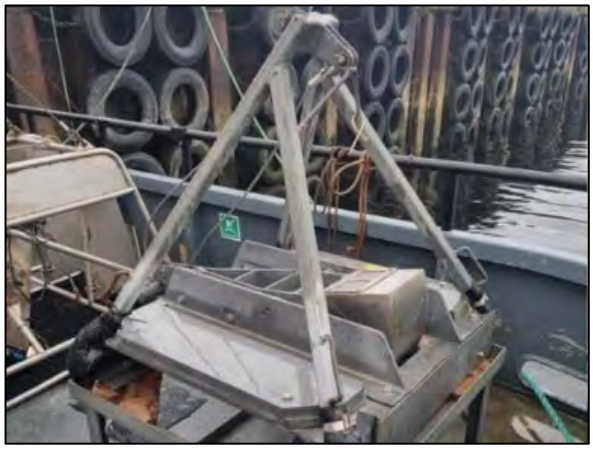


Figure 12 Hamon grab onboard the aft deck (Image Source: Survey Operational Report –, 02.22.16.OPS).

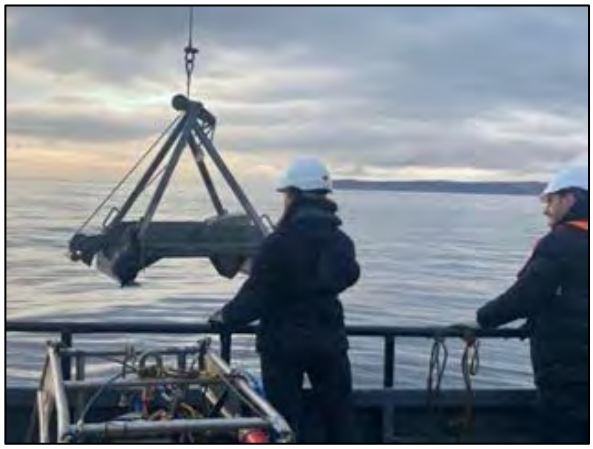


Figure 13 Hamon grab wet test (Image Source: Survey Operational Report –, 02.22.16.OPS).

4.3.3 Particle Size and Contaminants Grab Sampling

Samples were taken to investigate the sediment's physical and chemical properties, including the presence of any contaminants. The primary grab sampler utilised for PSA and contaminants grab sampling was a Shipek grab sampler (0.05 m^2). The Shipek grab sampler was deployed and recovered using the deck crane on the starboard side.

Upon retrieval, samples were checked for adequate sample volume. Should a sufficient sample not be acquired after 3 attempts with the Shipek grab, the site was abandoned.

Samples for metals, organics (TOM and TOC), hydrocarbons (THC and PAH), PCB, organotins (MBT, DBT and TBT), pesticides and flame-retardants were sampled from an undisturbed surface. Amber glass containers were used for the contaminant samples. PSA samples were stored in plastic containers.

A field log of sample positions, including time, sediment type, and water depth, was kept for later reference.

4.3.4 Water Sampling and Water Profiling

Water sampling was performed using a 5L Niskin bottle. The open bottle was lowered into the water via the winch on the vessel crane. The bottle was closed by a surface-deployed messenger weight. Water samples were taken from three (3) depths (Top, Middle and Bottom) in the water column. At each sampling location, a back-up water sample was taken for each depth.

Collected water samples were stored in 1 L plastic bottles. Processed water samples were stored in the onboard freezer. A YSI EXO 3 Sonde water profiler was used to collect water column profile data from the Top to the Bottom. The profiler was hand-deployed over the port side of the vessel. As the profiler was lowered, it recorded depth, conductivity, dissolved oxygen, turbidity, pH, and temperature. The collected profile data was downloaded, and quality checked on-site following retrieval of the YSI EXO 3 Sonde profiler.

4.4 Laboratory Methods

4.4.1 Particle Size Analyses

The Particle Size Analysis (PSA) was conducted by UK-based company Kenneth Pye Associates Limited. (KPAL).

Up to one litre of sediment from each sample site was analysed to detail the different particle fraction components with a combination of sieving and sedimentation methods.

PSA samples were analysed in accordance with the National Marine Biological Analytical Quality Control (NMBAQC) Guidelines for Particle Size Analysis (PSA) for Supporting Biological Analysis (Mason, 2022) to provide data over the complete particle size range allowing determination of the gravel to sand plus mud ratio. KPAL also hold MMO accreditation for particle size analysis.

Samples were wet separated at 2.0 mm. The >2.0 mm fraction, where present, was analysed using nested British Standard sieves at 'half' phi intervals. The sub-2.0 mm fraction was analysed via laser diffraction (size range 0.04 µm to 2.0 mm). The laser and sieve data were mathematically merged and calculations of particle size summary parameters (percentages of mud, sand, and gravel, silt/clay ratio, sand/mud ratio, median, mean, d10, d90, etc.) were calculated using GRADISTAT software (Blott & Pye, 2001).

The particle sizes were grouped into five large textural groups for description purposes (Table 10). The samples were described according to British standard 1377 (British Standard, 2010) and BGS-modified Folk classification (Long, 2006).

Table 10 British standard (2010) sieve sizes.

Classification	Particle Size Intervals (Diameter mm)	Grouped Classification
Boulder	>75	Boulders/cobbles
Cobble	75 – 64	
Coarse Gravel	64 – 20	Gravel
Medium Gravel	20 – 6	
Fine Gravel	6 – 2	
Coarse Sand	2 – 0.6	Sand
Medium Sand	0.6 – 0.2	
Fine Sand	0.2 – 0.063	
Coarse Silt	0.063 – 0.02	Silt
Medium Silt	0.02 – 0.006	
Fine Silt	0.006 – 0.002	
Clay	<0.002	Clay

4.4.2 Sediment Chemical and Contaminant Analyses

The sediment chemical and contaminant analyses were conducted by the UK-based company SOCOTEC. The different compounds that were analysed along with detection limits are stated in Table 11. The analyses included concentrations/contents of Total Organic Carbon (TOC), Total Organic Matter (TOM), metals, organotins (MBT, DBT, TBT), Total Hydrocarbon Content (THC), Polyaromatic Hydrocarbons (PAH), Polychlorinated Biphenyls (PCB), Organochlorine Pesticides (OCP), and Brominated Flame Retardants (PBDE).

Table 11 Marine sediment chemical and contaminant analyses.

Test Marine Sediment Contaminant Analyses	Method	Accreditation	Method Reporting Limit, PPM Unless Stated Otherwise
Particles Size Analysis and Distribution (PSA, PSD)	NMBAQC	NMBAQC	N/A
Total Organic Carbon	Sulphurous acid/combustion at 1600°`C/NDIR	UKAS 17025	0.02 %
Total Organic Matter by LOI	Combustion at 450°C	Not accredited	0.20 %
Metals suite: As(0.5), Cd(0.04), Cr(0.5), Cu(0.5), Hg(0.01), Ni(0.5), Pb(0.5), Zn(2)	Microwave assisted HF/Boric extraction & ICPMS	UKAS 17025	Limits of detection within parentheses.
Organotins: MBT(0.001), DBT(0.001), TBT(0.001)	ASC/SOP/301	UKAS/MMO	Limits of detection within parentheses.
THC (inc. saturates)	Solvent extraction & GC-FID	Not accredited	100 µg/kg (Total) 1 µg/kg (Individual alkanes)
PAH	Solvent extraction & GC-MS	UKAS 17025	1 µg/kg
PCB	Solvent extraction & GC-MS	UKAS/MMO	0.00008 mg/kg
OCP	Solvent extraction & GC-MS	UKAS/MMO	0.0001 mg/kg
PBDE	Solvent extraction & GC-MS	Not accredited	0.05 µg/kg (BDE209 0.1 µg/kg)

Table 12 Water sample analyses.

Test Marine Sediment Contaminant Analyses	Method	Accreditation	Method Reporting Limit, PPM Unless Stated Otherwise
Total Suspended Solids	WSLM10	Not accredited	5 mg/l

4.4.3 Biological Analyses

The macrofaunal analyses were conducted by the UK-based company APEM Ltd. Analyses were conducted in accordance with the NMBAQC scheme (Worsfold, Hall, & O'Reilly, 2010), and all the samples were quality controlled.

The macrofaunal grab samples were sorted from sediment residue, and the fauna was identified to the lowest taxonomic level possible, mainly species, counted and weighted. When the species could not be identified, the specimen was grouped into the nearest identifiable taxon of a higher rank (i.e. genus, family, or order etc). If the species remained unknown but clearly separated from any other found specimen within the same genus, it was assigned a “Type” denomination, i.e. Type A or Type B. Juveniles were marked with the qualifier “juvenile”, and later excluded from further statistical analyses. Biomass was measured for each taxon within each sample, to the nearest 0.1 mg. All macrofaunal analyses followed the NMBAQC scheme.

4.5 Data Analysis

4.5.1 Visual Data Analysis

The stills were analysed to identify species and species densities, including seabed substrate. The video recordings were used to aid in the assessment of features and extent of habitats. Particular attention was paid to the elevation of habitats above ambient seabed level, together with their spatial extent, percentage biogenic cover, and patchiness, as these are key criteria for evaluating areas of conservation interest.

Quantitative methods were used for the identification of biota in still photographs, with all the data presented as individuals per square metre and percentage cover of colonial species. Stills were analysed in AutoCAD Map 3D, where visual epibenthic fauna was counted, and results were summarised in a log containing scientific name, position, date, time, and stills ID. Qualitative methods were used for the identification of biota in the video recordings. For a more detailed description of identified species, see Appendix E.

4.5.2 Acoustic Data Analysis

Multibeam echo sounder (MBES) and side scan sonar (SSS) data from the geophysical survey together with the epifaunal composition from visual ground truthing data was used to determine the extent of habitats (Figure 14 and Figure 15). Video recordings, as well as field descriptions of grab samples, were also used to assign habitat classifications and delineate habitat boundaries.

The classification of the seabed sediments has been derived from the acoustic character of the SSS data and correlated with the general seabed morphology, MBES DTM and multibeam backscatter (only available in offshore data). The SSS data is presented as a grey scale image mosaic where the darker grey to black colours indicate higher intensity sonar returns corresponding to coarser sediments, and lighter grey colours indicate lower intensity sonar returns corresponding to finer sediments.

Once all data has been reviewed, the different data sets are aligned, and classifications are extrapolated based on textural similarity, reflectivity, and topographical features. The different datasets are combined to strengthen the accuracy of interpretations. Extrapolating a large area based on a low number of samples may lead to a lower hierarchic biotope level for that area, compared to the actual biotope level for the samples within the habitat. If two different habitats are classified at two different sites/transects in what appears to be a similar habitat, based on the geophysical interpretation, this may lead to the assignment of a matrix of the two habitats.

These compromises are reviewed individually. A smaller homogenous and distinctive area can be assigned to a higher hierarchic level compared to a larger and more variable area containing several different biotopes. The result of the habitat classification is presented in the results section and GIS charts.

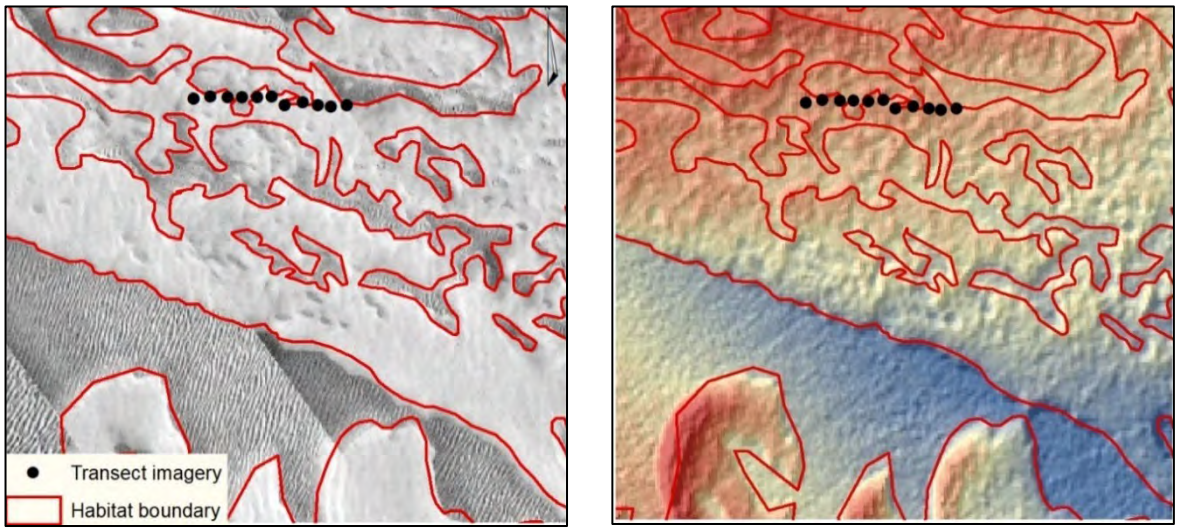


Figure 14 Example of side scan sonar image with rippled scour depression and areas of finer sediments.

Figure 15 Corresponding bathymetric image with rippled scour depression and areas of finer sediments.

4.5.3 Backscatter

The use of backscatter data to assist habitat interpretations and mapping is a methodology under development and is increasingly used in these types of analyses (Lurton and Lamarche, 2015).

Backscatter Normalised Values is a measurement of the MBES echo that is scattered in the direction of the transducer. This data records the intensity, in decibels (dB), of the echo that returns to the transducer after the emitted pulse interacts with the seabed. The backscatter amplitude varies with several factors such as frequency, beam pattern, range and losses due to absorption and spreading, angle with the seabed as well as sediment type and other factors.

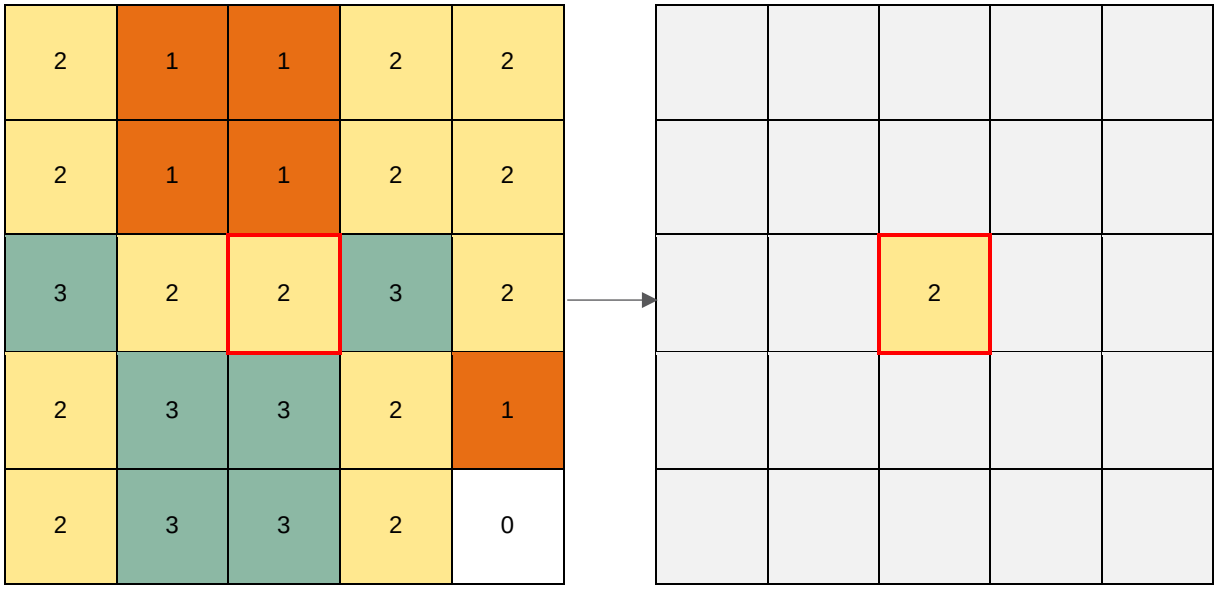
The offshore raw data were processed with the Fledermaus (FMGT) software, which applied various standard normalisations to the data to compensate for how the intensity varied across the swath producing a grayscale floating-point raster image gridded at 1 m, where each gridded cell contains a measured intensity value.

The intensity decibel value interval varied between the datasets acquired within the OAA and along the ECC due to their varying directionality. The range was -1 dB (hard seabed) to -44/-30 dB (soft seabed) for the exported raster data. Backscatter values varied across a small spatial scale, making interpretations on a larger scale challenging due to the small-scale variation.

To mitigate this, the Focal Statistics tool in ArcGIS was used to reduce the variation in the values. The backscatter raster data was imported into ArcGIS and a raster image was created based on the measured intensity values for each cell and plotted.

Within ArcGIS, a secondary raster image was created through the calculation of the cell value with the Focal Statistics tool. The tool calculates a new value for each input cell based on the neighbouring cell values. The new value output was based on the average value of the neighbouring cells in a 5x5 m (5x5 cells) square area with the target cell included (Table 13). The new cells maintained the original cell size of 1x1 m.

Table 13 Focal Statistics settings.



Ground-truthing data, imagery and filed descriptions of sediment, together with geophysical data, SSS and MBES, were used to align the backscatter reflectivity intervals based on the trends interpreted, with regards to substrate and habitats (Lurton and Lamarche, 2015). However, there were limiting factors due to the numerous and morphologically different ripple features.

The difficulties, that features such as ripples, impose on backscatter data are due to changes in elevation and angle of the seabed. These affect the amount of reflected sound, resulting in values indicating too hard or too soft a substrate. These potential errors are partially mitigated by using the Focal Statistics tool in ArcGIS, as the interpolation used in the tool averages out the overestimated and underestimated values from the backscatter.

Outlier values from the outermost ranges from the data sets were naturally excluded as the grouping of the intervals was set and these are detailed in Table 14. Nearshore backscatter data was subject to operational and technical difficulties resulting in noise disturbances in the dataset and the backscatter data could not be aligned with the offshore data to a satisfactory level. Nearshore backscatter data was excluded from this report.

The absence of Nearshore backscatter data does not affect the overall interpretation of the benthic conditions within the nearshore area due to the full coverage of primary geophysical data such as SSS, and MBES but also due to the existence of ground truthing data i.e., grab sampling and DDV sampling. Dataset from which mapping and assessments are primarily delineated.

Table 14 Backscatter Intensity colour schema for each area (intensity is presented in dB).

Datasets	Colour Bars & Classes (dB)	Outliers (dB)
RAW OAA	-46.38 to -20.9 -20.89 to -17.98 -17.97 to -15.25 -15.24 to -12.92 -12.91 to 3.227	N/A
Focal Statistics OAA	-26.96 to -18.62 -18.61 to -17.65 -17.64 to -14.05 -14.04 to -12.91 -12.90 to -7.12	-44 to -27; -7 to -1
RAW ECC W and E	-29.50 to -19.56 -19.55 to -17.41 -17.40 to -15.26 -15.25 to -12.98 -12.97 to 4.75	N/A
Focal Statistics ECCs	-24.90 to -18.95 -18.94 to -17.95 -17.94 to -14.96 -14.95 to -13.06 -13.05 to -8.05	-30 to -25; -8 to -1

4.5.4 Rugosity

The geophysical survey of the OAA and ECC areas produced a bathymetric dataset which has been used to characterise the seabed morphology and interpret seabed features within the survey area.

The bathymetric data was used to derive seabed roughness, an MBES derivative to provide further information on the seabed topography and enhance the identification of seabed features in particular areas of interest.

An ArcGIS toolbox, Benthic Terrain Modeler, and tool Vector Terrain Ruggedness (VRM) were used to produce a model of the surface rugosity to better illustrate trends within one area of interest in the OAA related to Stony Reef features (Walbridge, Slocum, Pobuda, & Wright, 2018).

MBES depth data was exported in a 1-metre mosaic which was clipped to the extent of the "Low to Medium Stony Reef" Resemblance delineated area of interest prior to the Rugosity model being produced.

Different resolutions of the MBES data were reviewed, to select the best resolution that minimizes the impact of noise while maintaining a good representation of the variability. As the algorithm looks at neighbouring cells, it was deemed appropriate to clip the dataset before any computing in order to minimize influence and focus on the variation within the polygon of interest.

The VRM measures seabed rugosity as the variation in the three-dimensional orientation of grid cells within a small-scale neighbourhood (5 x 5 m) taking slope and aspect into account. In this instance, a 5 x 5 m cell neighbourhood was elected to reflect the minimum criteria of Stony Reefs as well as to illustrate variability on a small spatial scale. The VRM intervals were adjusted manually (by reviewing the statics and spread of values within different areas) based on the heterogeneity, or lack thereof, as interpreted from the ground-truthing data, visual and acoustic. No outlier values were removed (Table 14).

Table 15 Rugosity colour scheme for the "Low – Medium" Resemblance Stony Reefs within the OAA.

Datasets	Colour Bars & Classes	Outliers (dB)
VRM (SD=1)	0.000001073 - 0.000079538 0.000079538 - 0.000228187 0.000228187 - 0.000376835 0.000376835 - 0.172429562	N/A
VRM (Values aligned with ground- truthing data)	0.000001073 - 0.000045 0.000045 - 0.00023 0.00023 - 0.0027	N/A

4.5.5 Particle Size Analyses

Sediment particle size distribution statistics for each sample were calculated from the raw data by the laboratory. Main sediment fractions and percentages were plotted to examine sediment composition changes across the survey area and used to aid the habitat assessment. Multivariate analyses were undertaken on the PSA data set, to identify patterns in the sediment distribution. Analyses included hierarchical clustering employing the Euclidean distance resemblance matrix, SIMPROF analysis and principal component analysis (PCA). The dataset was normalised prior to the analyses being performed.

PSA results were analysed using the Plymouth Routines in Multivariate Ecological Research (PRIMER) v7.0 statistical package (Clarke & Gorley, PRIMER v7: User Manual/Tutorial. Plymouth: PRIMER-E., 2015) and normalised before being included in any statistical analysis. Data for the percentage composition was analysed in a cluster analysis using the Euclidean distance. A Principal Component Analysis (PCA) was undertaken on the sediment data set to identify spatial patterns and relationships between variables.

Detailed results for each grab sample site are provided in Appendix F.

4.5.6 Sediment Chemical and Contaminant Analyses

Environmental Quality Standards (EQS) for metals and hydrocarbons in sediments are not yet developed for UK waters.

Assessment criteria developed by the Canadian Council of Ministers of the Environment (CCME) together with the Centre for Environment, Fisheries and Aquaculture Science (CEFAS) guideline action levels for disposal of dredged material have been considered common practice to use.

The Oslo and Paris Conventions for the protection of the marine environment of the North-East Atlantic (OSPAR) Environmental Assessment Criteria (EAC) have also been used as guidelines for metal, PAH, and PCB concentrations, when applicable, within this report. The Canadian sediment quality guidelines include two values as assessment criteria, the Interim Sediment Quality Guidelines (ISQG) and Probable Effect Level (PEL).

The ISQG are threshold levels that are set to protect all aquatic life during an indefinite period of exposure, and for values above PEL, adverse effects are expected to occur frequently (CCME, 1995; CCME, 2001). For concentrations between the ISQG and PEL, adverse effects occur occasionally.

CEFAS Action Levels are used as a part of assessing the contamination status in dredged material, where material below Action Level 1 (AL1) generally indicates that contaminant levels are of no concern, while contaminant levels above Action Level 2 (AL2) generally are considered unsuitable for disposal in the sea (MMO, 2015).

OSPAR's Environmental Assessment Criteria (EACs) are under development, and OSPAR uses "Effect range-low" (ERL) values for sediment assessment of metals and PAH, where EACs are not available. The ERL value indicates a concentration below which adverse effects on organisms are rarely observed (OSPAR, CEMP 2011 assessment report, 2011). Further, Background Assessment Concentrations (BAC) are used to indicate if concentrations of man-made substances exceed expected background levels (OSPAR, 2020).

Condition classes established by the Norwegian Environmental Agency (NEA) for contamination in coastal sediments (NEA, 2016, revised 2020) for metals, PAH and other organic compounds were also used. This system uses five classes, class 1 – Background levels, class 2 – Good, with no known toxic effects, class 3 – Moderate, with chronic effects at long-term exposure, class 4 – Poor, with acute toxic effects at short-term exposure and class 5 – Very Poor, with extensive toxic effects.

There are no OSPAR or UK contamination threshold values regarding THC for marine sediments. In the absence of such guidelines, Dutch intervention levels for aquatic sediments can offer a useful comparison. Concentrations above the Dutch intervention values represent a serious level of contamination, where functional properties of the sediment are seriously impaired or threatened (Hin, Osté, & Schmidt, 2010). Detailed results are presented in Appendix G.

4.5.7 Univariate Statistical Analyses

Univariate analyses were undertaken using PRIMER v7.0 statistical package (Clarke & Gorley, PRIMER v7: User Manual/Tutorial. Plymouth: PRIMER-E., 2015).
Univariate analyses included the primary variables, the number of taxa (S) and abundance (N) together with Margalef's index of Richness (D), Pielou's index of Evenness (J), Shannon- Wiener index of Diversity (H') and the Simpson's index of Dominance (1-λ) which are summarised in Table 16.

Table 16 Univariate statistical analyses.

Analyses	Parameters	Formula	Description
No. of Taxa (S)	Species richness	S	The number of species (taxa) in each sample.
No. of Individuals (N)	Abundance	N	The number of individuals in each sample.
Margalef's Index of Richness (D)	Richness	D = (S-1) / ln(N)	A measure of the number of species present for a given number of individuals
Shannon-Wiener Index of Diversity (H')	Diversity	H' = Σi Pi ln(Pi)	The diversity index incorporates both species richness and equitability, where Pi is the proportion of the total count arising from the/th species. A lower value equals a high chance that all abundance is concentrated to one species.

Analyses	Parameters	Formula	Description
Pielou's Index of Evenness (J)	Evenness	J = H' / ln (s)	Measures how evenly individuals are distributed between species. Gives a value between 0 to 1, where a higher value equals a more even community.
Simpson's Index of Dominance (1-λ)	Dominance	λ = (Σ pi2)	Dominance index between 0 – 1 where 0 corresponds to assemblages whose total abundance is dominated by one or very few of the species present and 1 represents a more evenly species distribution.

4.5.8 Multivariate Statistical Analyses

Multivariate analysis was undertaken using PRIMER v7.0 statistical package (Clarke & Gorley, PRIMER v7: User Manual/Tutorial. Plymouth: PRIMER-E., 2015).
The statistical analyses were based on macrofaunal data derived from the taxonomic analyses of a single replicate from each grab sample site (only one (1) replicate was analysed if two (2) replicates were collected at a given site). Grab samples with insufficient sample volume were excluded from the statistical analyses. Abundances were expressed as the number of individuals per 0.1 m^2.

The macrofaunal organisms were separated into non-colonial and sessile colonial fauna. Colonial fauna was not quantified in the laboratory analysis and was treated separately in the statistical analyses.

All colonial fauna was also considered epifauna. Juvenile (JUV) taxa were excluded from the dataset. Foraminiferans were excluded from the datasets. The faunal composition was linked to physical variables such as depth and sediment composition.

Square root transformation was applied to the non-colonial enumerated fauna datasets before calculating the Bray-Curtis similarity measures. This transformation was made to prevent abundant species from influencing the Bray-Curtis similarity index measures, excessively and to take the rarer species into account (Clarke & Warwick, 2001).

The macrofaunal laboratory results were compared for faunal composition between sampling sites. Site-related differences in community structure were examined in a clustering analysis using Euclidean distance and the Bray-Curtis similarity coefficient. This method is common when measuring ecological distance in biological sample data.

Multi-Dimensional Scaling (MDS) analysis was undertaken in conjunction with the cluster analysis. The MDS analysis is based on the same similarity matrix as that of the cluster analysis and produces a multidimensional ordination of samples.

The number of restarts was set to 999 with a minimum stress of 0.01. The MDS plot visualises the relative (dis)similarities between samples; the closer they are the more similar the species composition between the samples. The degree to which these relations can be satisfactorily represented is expressed as the stress coefficient statistic, low values (<0.1) indicate a good ordination with low probabilities of misleading interpretation. Generally, the higher the stress, the greater the likelihood of non-optimal solutions (Clarke & Warwick, 2001).

A Similarity profiling algorithm (SIMPROF) test was run in conjunction with the cluster analysis, which was used to identify significantly different naturally occurring groups among grab samples.

The results are presented in the cluster dendrogram as black lines indicating significant statistical differences. Red lines represent samples that are not statistically different. The SIMPROF is based on taxa, and the abundance of each taxon in each sample, thus different SIMPROF groups may host similar fauna which differ in abundance.

A Similarity Percentage Analysis (SIMPER) was undertaken following the cluster analysis. SIMPER examines variable relations to each other and presents the species' contributions and similarities within and among groups.

PSA data was analysed in PRIMER and normalised before being included in any statistical analysis. Data for the percentage composition was analysed in a cluster analysis using the Euclidean distance.

A Principal Component Analysis (PCA) was undertaken on the sediment data set in order to identify spatial patterns and relationships between variables.

The relationship between the physical and biological data was tested using the BIOENV method, with Spearman rank correlations, in the BEST procedure in PRIMER v.7. This analysis identifies variables that exert the greatest influence on the spatial distribution of the input datasets. Prior to the BEST analyses species abundance data were square root transformed and the physical variables were normalised.

4.6 Habitat Classification

Habitats were classified to the lowest hierarchic level possible and based on interpretations that combine biotope descriptions of species abundance, diversity, depth and seabed features from grab samples, video and photos acquired at each sample site.

The classification of the communities of the different habitat types was based on physical characteristics such as benthic geology, wave exposure, tidal currents, temperature, and salinity together with key species present in the area. In addition, normalized backscatter data from the offshore MBES was used to delineate habitats in areas of homogenous sediments.

The EUNIS classification (EEA, 2022) is divided into six hierarchic levels, Figure 16. At Level 1, the habitats are divided into marine, coastal and terrestrial habitats. The marine habitats are further divided into three separate categories: benthic, pelagic and ice-associated habitats.

At Level 2, the biological zone and presence/absence of rock is a classification criterion, and at Level 3, the classifications are separated into marine regions.

Level 4 gives references to specific taxa. For rocky substrates, the major epifauna is used, and for softer substrates, the classification relies on both zonation and physical attributes. Further, at Level 5, the classification is based on both the physical and biological characteristics of the habitats, and classes are defined with both infauna and epifauna on different substrates. At the highest level, level 6, the different characterising taxa are associated with different environmental characteristics of the habitat.

If two different habitat classifications within what appears to be a similar habitat are identified, without any apparent differences in the interpreted geophysical data, a low number of transects may lead to the assignment of a matrix of two habitats. Extrapolating a large area based on a low number of samples may lead to a lower hierarchic biotope level for that area, than the actual biotope level for a singular sample within the habitat.

These compromises are reviewed individually. A smaller homogenous and distinctive area can be assigned to a higher hierarchic level compared to a larger and more variable area containing several different biotopes. The result of the habitat classification is presented in the results section and GIS as well as AutoCAD charts.

L1	(M) Marine Habitats
L2	(MC4) Circalittoral mixed sediment
L3	(MC42) Atlantic circalittoral mixed sediment
L4	(MC421) Faunal communities of Atlantic circalittoral mixed sediment
L5	(MC4211) *Cerianthus lloydii* and other burrowing anemones in circalittoral muddy mixed sediment
L6	(MC42111) *Cerianthus lloydii* with *Nemertesia* spp. and other hydroids in circalittoral muddy mixed sediment

Figure 16 Example of 2022 EUNIS Hierarchy.

4.7 Protected Habitats and Species Assessments

For the assessment and classification of potential areas and/or species of conservation importance, the following legislation and guidelines have been applied when relevant.

The European Commission (EC) Habitat Directive specifies the European nature conservation policy (EUR 28, 2013). Species and habitats of special interest for conservation are specified in the different annexes to the directive.

Annex I states the habitats of special conservation interest and Annex II states the species of special conservation interest. Among the habitats specified in Annex I are the "Reefs" (code 1170). Reefs can be of biogenic, e.g. mussel beds or corals, or geogenic origin, e.g. stony areas with epifauna.

The Oslo and Paris Conventions for the protection of the marine environment of the North-East Atlantic (OSPAR), list protected species and habitats, as well as sensitive habitats and species in need of protection in the North-East Atlantic (OSPAR, 2008). This serves also as a complement to the EC Habitats Directive.

The species and habitats found in this survey were compared to the list of Scottish Priority Marine Features (PMF) (Tyler-Walters H. , et al., 2016) that further defines the habitats and species which are considered to be marine nature conservation priorities in Scottish waters.

In addition to the above-mentioned policies and guidelines the Scottish Biodiversity List (SBL) identifying the species and habitats which are the highest priority for biodiversity conservation in Scotland was also consulted (Scottish Biodiversity Forum, 2012).

In the Habitat Directive's interpretation manual (EUR 28, 2013) reefs are explained as follows:

"Reefs can be either biogenic concretions or of geogenic origin. They are hard compact substrata on solid and soft bottoms, which arise from the sea floor in the sublittoral and littoral zone. Reefs may support a zonation of benthic communities of algae and animal species as well as concretions and corallogenic concretions."

The distinction between what *is* and what *is not* a "reef" is not so precise and is generally referred to as "reefiness". This is particularly relevant in the case of the tube-building polychaete, *Sabellaria spinulosa* and areas of cobbles and boulders (Stony Reefs).

If for example *S. spinulosa* or the horse mussel, *Modiolus,* is found in an area it does not automatically qualify as a "reef", Annex I habitat or a potential Annex I habitat. Therefore, a scoring/assessment system based on a series of physical, biological and spatial characteristics is used to assess the degree of "reefiness".

A method to assess "reefiness" was presented by Gubbay (2007), and involves the quantification of three separate criteria: elevation (average tube height in cm), Area (m^2) and patchiness (percentage cover), as presented in Table 17 and Table 18. A similar assessment matrix for stony reefs by Irving (2009), is presented in Table 19.

Table 17 Proposed chart for Sabellaria spinulosa reef identification (Gubbay, 2007).

Characteristic	Not A Reef	"Reefiness"		
		Low	Medium	High
Elevation (cm) (Average tube height)	<2	2 – 5	5 – 10	>10
Extent (m^2)	<25	25 – 10 000	10 000 – 1 000 000	>1 000 000
Patchiness (% cover)	<10	10 – 20	20 – 30	>30

The general definition of biogenic reefs is made by (Holt, Rees, Hawkings, & Seed, 1998) as;

"Solid, massive structures which are created by accumulations of organisms, usually arising from the seabed or at least clearly forming a substantial, discrete community or habitat which is very different from the surrounding seabed. The structure of the reef may be composed almost entirely of the reef-building organism and its tubes or shells, or it may to some degree be composed of sediments, stones and shells bound together by the organism."

To assess the overall 'reefiness' the Collins (2010) method of combining the three separate criteria (elevation, extent and patchiness) as established by Gubbay (2007) was implemented. The reef structure is assessed in Step 1 followed by Step 2 aimed to categorise the final 'reefiness' (Table 18).

The patchiness of *S. spinulosa* was derived from the visual data analysis and the per cent coverage was calculated from each still image taken along the transect. Elevation of the *S. spinulosa* tubes was estimated from each still image taken along the transect. The area was calculated from boundaries (polygons) drawn in GIS and based on the interpreted geophysical and bathymetrical data.

Table 18 Sabellaria spinulosa Reef Structure Matrix (Step 1) and S. spinulosa Reef. *Structure Matrix vs Area Matrix (Step 2) to determine final "Reefiness" (Collins, 2010).*

Step 1						
Reef Structure Matrix			**Elevation (cm)**			
			<2	**2 – 5**	**5 – 10**	**>10**
			Not a reef	Low	Medium	High
Patchiness (%)	<10	Not a reef	Not a reef	Not a reef	Not a reef	Not a reef
	10 – 20	Low	Not a reef	Low	Low	Low
	20 – 30	Medium	Not a reef	Low	Medium	Medium
	>30	High	Not a reef	Low	Medium	High

Step 2					
Reef Structure VS Area		**Area (m^2)**			
		<25	**25 – 10 000**	**10 000 – 1 000 000**	**>1 000 000**
		Not a reef	Low	Medium	High
Reef Structure	Not a reef	Not a reef	Not a reef	Not a reef	Not a reef
	Low	Not a reef	Low	Low	Low
	Medium	Not a reef	Low	Medium	Medium
	High	Not a reef	Medium	High	High

Table 19 Guidelines used to categorise 'reefiness' for stony reefs (Irving, 2009).

Measure of Resemblance	**Not a Stony Reef**	**Low**	**Medium**	**High**
Composition	<10 %	10-40 % Matrix supported	40-95 %	>95 % Clast supported
Notes: Diameter of cobbles/boulders being greater than 64 mm. Percentage cover relates to a minimum area of 25 m^2. This 'composition' characteristic also includes 'patchiness'.				
Elevation	Flat Seabed	<0.064 m	0.064 m-5 m	>5 m
Notes: Minimum height (64 mm) relates to minimum size of constituent cobbles. This characteristic could also include 'distinctness' from the surrounding seabed.				
Extent	<25 m^2	>25 m^2		
Biota	Dominated by infaunal species			>80 % of species present composed of epifaunal species.

This scoring system indicates that stony reefs should be elevated by at least 0.064 m and with a composition of at least 10 % stones, covering an area of at least 25 m^2 and having an associated community of largely epifaunal species.

For Stony Reefs with a Low resemblance, the methodologies proposed by Brazier and Golding *et al.* (2020; 2020) were consulted to assess whether or not an area would meet the criteria for inclusion in Annex I (1170) – Reefs, Stony Reefs. The methodology is still under review and development and is therefore not fully implemented but contains guidance on classifying and enumerating reef habitat "Key Species" as well as "Reef-Species" often present in Stony Reef habitats (Table 20).

Table 20 Guidelines used to categorise low resemblance stony reefs (Brazier, 2020).

	Key Species Count	Reef-Species Count
Reef	≥3	>20
Possible Reef	>1 and <3	>5 and <20
Not Reef	0	<5

For "Bedrock Reefs" no similar scoring system exists. In areas where the geophysical data cannot provide information on the degree of bedrock exposure, these areas will be delineated as "Potential Bedrock Reefs". The qualifying criteria for the classification "Bedrock Reefs" is the presence of bedrock that could support an epifaunal community.

4.8 MESH Confidence Assessment

The MESH (Mapping European Seabed Habitats) confidence assessment is a tool for assessing the confidence and/or accuracy of seabed habitat maps (MESH, 2010). This evaluation addresses three main questions:

- How good is the remote sensing;
- How good is the ground-truthing;
- How good is the data interpretation?

Each of these questions is further divided into four to six sub-questions, in total 15 questions were included in the original assessment tool. The weight of each question was left at the default settings. The map is scored based on these criteria and the scores are combined to produce an overall confidence score. To achieve the top score of 100, both physical and biological survey data are required. It is possible to score different sections of a map separately if the data/interpretation varied across the area.

An overall MESH value was initially assigned to the entire survey area, based solely on remote sensing and data interpretation. These values were then revised and increased for polygons that have ground-truthing data (grab samples, seabed imagery, and geotechnical samples), depending on the quality of the ground-truthing data. If multiple sample sites with ground-truthed data were located within a single polygon, the sample site with the highest MESH score represents the entire polygon.

5. Results

5.1 Field Operations

5.1.1 Offshore

DDV transects were undertaken at all of the planned 82 grab sample sites but, due to the identification of potential Annex I habitats on the DDV, grab samples were taken at only 73 of the planned 82 grab sample sites (Table 21, Figure 17).

Standalone DDV transects were performed at 17 locations, 13 were planned DDV transects and 4 were additional DDV transects (T96-T99). Contaminant analyses for flame retardants and pesticides were planned at 31 of the 82 grab sample sites but due to coarse sediment samples could only be acquired at 26 of these sites (Table 22, samples acquired highlighted green). Water sampling and CTD profiling were performed at all 20 sites including the 4 sites selected for sampling at two different tidal cycles. A geophysical overview of each site and transect is available in Appendix A with further information regarding sampled sites in Appendix B.

Table 21 Numbers of performed sample sites and transects.

Number of Sampled Sites	Video Transects (Inc at Grab Sites)	Sites Grab Sampled	Water Sample/CTD Sites
	99	73	20*

**Including the 4 sites selected for sampling at two different tidal cycles.*

Table 22 Acquired samples at offshore sampling sites.

Sample ID	Fauna	PSA	Metals	Organics	PBDE/OCP	TSS	eDNA
OAA_S01							
OAA_S03							
OAA_S05							
OAA_S06							
OAA_S07							
OAA_S08							
OAA_S09							
OAA_S12							
OAA_S13							
OAA_S14							
OAA_S16							
OAA_S17							
OAA_S19							
OAA_S20							
OAA_S21							
OAA_S24							
OAA_S25							
OAA_S26							
OAA_S27							
OAA_S28							
OAA_S29							
OAA_S30							
OAA_S31							

Sample ID	Fauna	PSA	Metals	Organics	PBDE/OCP	TSS	eDNA
OAA_S32							
OAA_S33							
OAA_S34							
OAA_S35							
OAA_S36							
OAA_S37							
OAA_S38							
OAA_S40							
OAA_S42							
OAA_S43							
OAA_S44							
OAA_S46							
OAA_S49							
ECE_S51							
ECE_S54							
ECE_S55							
ECE_S56							
ECE_S57							
ECE_S58							
ECE_S59							
ECE_S60							
ECE_S61							
ECE_S62							
ECE_S63							
ECE_S64							
ECE_S66							
ECE_S67							
ECE_S68							
ECE_S69							
ECE_S70							
ECE_S71							
ECE_S73							
ECW_S76							
ECW_S77							
ECW_S78							
ECW_S79							
ECW_S80							
ECW_S81							
ECW_S82							
ECW_S83							
ECW_S84							
ECW_S85							
ECW_S86							
ECW_S87							

Sample ID	Fauna	PSA	Metals	Organics	PBDE/OCP	TSS	eDNA
ECW_S89							
ECW_S90							
ECW_S91							
ECW_S93							
ECW_S94							
OAA_W01							
OAA_W02							
OAA_W03							
OAA_W04							
OAA_W05							
OAA_W06							
OAA_W07							
OAA_W08							
OAA_W09							
OAA_W10							
OAA_W11							
OAA_W12							
OAA_W13							
OAA_W14							
ECW_W15							
ECE_W16							
ECW_W17							
ECE_W18							
ECE_W19							
ECE_W20							
ECE_W21							
OAA_W22							
OAA_W23							
ECE_W24							

At the attempted grab sample sites, no fauna samples were acquired at three (3) sites (S04, S32 and S61) and fauna samples comprised low volume and/or washed-out samples at eight (8) sites (S19, S24, S25, S36, S54, S59, S81 and S87). At the attempted grab sample sites, no PSA samples were acquired at six (6) sites (S04, S08, S24, S59, S81 and S87) and PSA samples comprised low volume and/or washed-out samples at four (4) sites (S26, S36, S54 and S61).

At the attempted grab sample sites, no samples were acquired for analyses of organics or metals at 16 sites (S05, S08, S09, S14, S21, S24, S26, S29, S32, S36, S37, S59, S61, S66, S81 and S87). Samples for Organochlorine Pesticides and Brominated Flame Retardants were not acquired at six (6) sites (S08, S14, S24, S50, S59 and S92).

Replicates were acquired at 16 sites (S01, S05, S07, S08, S13, S14, S16, S19, S24, S28, S30, S37, S40, S42, S44 and S49). Out of these replicate sites, no fauna samples were acquired at two (2) sites (S05 and S37), fauna samples comprised low volume at two (2) sites (S19 and S24), no PSA samples were acquired at three (3) sites (S08, S24 and S37), and no chemical samples were acquired at five (5) sites (S05, S08, S14, S24 and S37).

5.1.2 Nearshore

Out of the nine (9) planned DDV sites, all were successfully investigated using DDV. Following the completion of the DDV scope, four (4) of the Client proposed grab sample sites were deemed suitable for sampling. Of these, three (3) were successfully sampled for particle size, chemistry, and fauna (Table 23).

All five (5) water samples and CTD sites were successfully sampled and profiled. Grab sample site NS_S002 was attempted but failed to acquire an acceptable sample volume for faunal, PSA and chemistry analyses (Table 26 samples acquired highlighted green).). Replicates were acquired at two (2) sites (NS_S001 and NS_S008) for chemicals and contaminants. These samples were collected as backup and not analysed.

Table 23 Numbers of performed nearshore sample sites and transects.

Number of Sampled Sites	Video Transects (Inc at Grab Sample Sites)	Sites Grab Sampled	Water Sample/CTD Sites
	9	3	5

Table 24 Acquired samples at nearshore sampling sites.

Sample ID	Fauna	PSA	Metals	Organics	PBDE/OCP	CTD/TSS	eDNA
NS_W01							
NS_W02							
NS_W03							
NS_W04							
NS_W05							
NS_S001							
NS_S002							
NS_S003							
NS_S004							
NS_S005							
NS_S006							
NS_S007							
NS_S008							

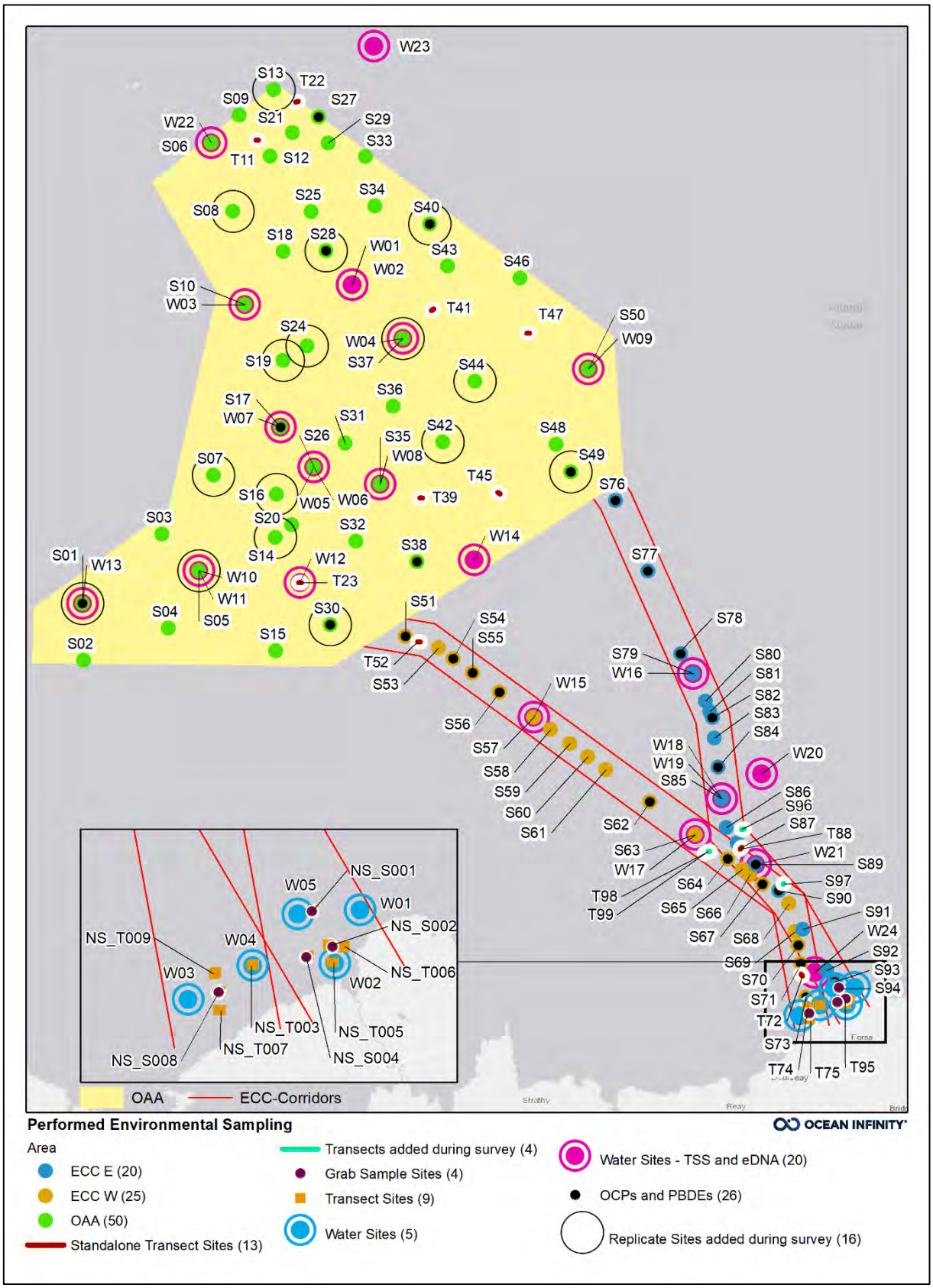


Figure 17 Overview of the performed environmental sampling.

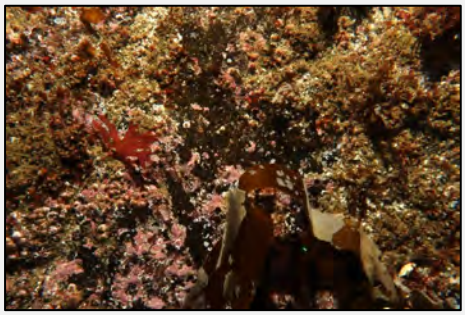
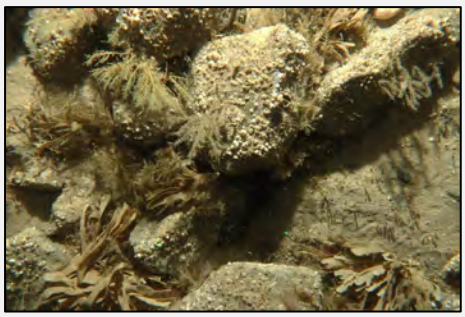
5.2 Summary of Identified Habitats

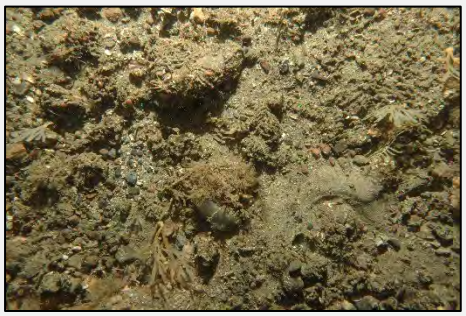
A total of seven (7) EUNIS habitats including two (2) habitat complexes were identified and delineated within the survey area. The identified fauna further indicates the presence of two (2) additional site-specific habitats; **MB121** – Kelp and seaweed communities on Atlantic infralittoral rock and **MC128** – *Sabellaria* on Atlantic circalittoral rock.

The classification codes at four (4) of the grab sample sites (S20, S55, S71 and S80) have changed from those stated in the first draft of the Habitat Assessment report 104164-TOT-OI-SUR-REP-HABASRE due to the results of the laboratory PSA analyses. Grab sample sites S20 and S71 changed from **MC52** – Atlantic circalittoral sand to **MC32**- Atlantic circalittoral coarse sediment/**MC52** Atlantic circalittoral sand, while S55 and S80 changed from **MC32** – Atlantic circalittoral coarse sediment/**MC52** – Atlantic circalittoral sand to **MC52** – Atlantic circalittoral sand. An overview of the identified habitats and sample sites is presented in Table 25 and further illustrated in Figure 19 to Figure 22.

The ID column in Table 25 defines the colour in the charts for the specific habitat type.

Table 25 Identified habitats within the surveyed area.

Habitat Image	ID	Habitat Classification	EUNIS Habitat Code	Site ID
		Atlantic infralittoral rock	MB12	T74, T75 and T95 NS_T005, NS_T006, NS_T007 and NS_T009
		Kelp and seaweed communities on Atlantic infralittoral rock	MB121	T75 NS_T005 and NS_T006
		Atlantic circalittoral rock	MC12	S02, S05, S10, S15, S32, S48, S53, S65 and S84 T23, T45, T47, T52, T72, T88, T96, T97 and T98 NS_T003

Habitat Image	ID	Habitat Classification	EUNIS Habitat Code	Site ID
		Sabellaria on Atlantic circalittoral rock	MC128	S53 and S54 T52
		Atlantic circalittoral coarse sediment	MC32	S07, S13, S17, S19, S21, S30, S31, S33, S34, S35, S44, S46, S58, S79, S82, S84 and S92 T22 and T72
		Atlantic circalittoral coarse sediment/ Atlantic circalittoral sand	MC32/ MC52	S01, S03, S06, S09, S12, S14, S19, S20, S28, S33, S55, S56, S58, S71, S73, S86, S91 and S93 T22
		Atlantic circalittoral mixed sediment	MC42	S04, S08, S16, S18, S19, S24, S25, S26, S29, S36, S37, S43, S50, S54, S57, S59, S61, S65, S68, S78, S81, S83, S84, S85, S87 and S89 T11, T22, T39, T41, T97 and T99
		Atlantic circalittoral sand	MC52	S27, S30, S38, S40, S42, S49, S51, S60, S62, S63, S64, S66, S67, S68, S69, S70, S76, S77, S80, S90 and S94 T72 and T99 NS_S001, NS_S002, NS_S004, NS_T007 and NS_S008

Habitat Image	ID	Habitat Classification	EUNIS Habitat Code	Site ID
		Atlantic circalittoral sand/Atlantic circalittoral rock	MC52/ MC12	NS_S004

5.2.1 Sample Specific Habitats

The taxonomic assemblages from the acquired grab sample data further indicate the presence of 15 sample-specific habitats across the survey area, including 6 transitional habitat complexes, that are presented in Table 26.

For the sample-specific habitats classification, a bottom-up approach was implemented to identify community patterns. Many of the infralittoral assignments are for communities that were a sufficient match with a qualifying descriptor, mainly fauna, while others are listed as 'cf.', where they deviated. As these had similarities to the better-described examples, often forming a continuum, it was deemed appropriate to assign them as comparable.

The Infralittoral and Circalittoral divide mainly relates to hard substrata, as it was defined to correlate with depth limits for algae. Groups which fitted no biotope well with regards to species descriptors were assigned based on the closest fauna and particle size analysis results.

Table 26 Sample-specific habitats within the surveyed area.

Notes	Habitat Classification	EUNIS Habitat Code	JNCC Biotope Code	Sample ID
Variant with low abundance of *Asbjornsenia pygmaea.*	*Moerella spp*. with venerid bivalves in Atlantic infralittoral gravelly sand	cf. MB3233	cf. SS.SCS.ICS.MoeVen	S05, S07, S13, S17, S21, S34, S44, S46, S58, S82 and S91
-	*Moerella spp*. with venerid bivalves in Atlantic infralittoral gravelly sand	MB3233	SS.SCS.ICS.MoeVen	S01, S03, S06, S12, S14, S20, S28, S35, S69,S70, S71 and S73
-	*Hesionura elongata* and *Microphthalmus similis* with other interstitial polychaetes in Atlantic infralittoral mobile coarse sand	MB3234	SS.SCS.ICS.HeloMsim	S93, S94, NS_S001, NS_S004 and NS_S008
Transition between two biotopes.	*Flustra foliacea* and *Hydrallmania falcata* on tide-swept circalittoral mixed sediment/*Sabellaria spinulosa* on stable Atlantic circalittoral mixed sediment	MC4214/ MC2211	SS.SMx.CMx.FluHyd/ SS.SBR.PoR.SspiMx	S57 and S59

Notes	Habitat Classification	EUNIS Habitat Code	JNCC Biotope Code	Sample ID
Transition between two biotopes.	*Flustra foliacea* and *Hydrallmania falcata* on tide-swept circalittoral mixed sediment/*Branchiostoma lanceolatum* in Atlantic circalittoral coarse sand with shell gravel	MC4214/ MC3215	SS.SMx.CMx.FluHyd/ SS.SCS.ICS.MoeVen	S09, S24, S31 and S37
Transition between two biotopes.	*Flustra foliacea* and *Hydrallmania falcata* on tide-swept circalittoral mixed sediment/*Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand	MC4214/ MC5211	SS.SMx.CMx.FluHyd/ SS.Ssa.CfiSa.EpusOborApri	S27, S29, S33, S43, S49, S67, S80 and S90
Transition between two biotopes. Infauna does not sufficiently match for a standard biotope description.	*Flustra foliacea* and *Hydrallmania falcata* on tide-swept circalittoral mixed sediment/Atlantic offshore circalittoral mixed sediment	MC4214/ MD42	SS.SMx.CMx.FluHyd/ SS.SMx.Omx.#	S08, S16 and S26
Infauna does not sufficiently match for a standard biotope description.	Faunal communities of Atlantic circalittoral sand	MC521	No suitable matching habitat	S83 and S86
Transition between two biotopes. Infauna does not sufficiently match for a standard biotope description.	Faunal communities of Atlantic circalittoral sand/Atlantic offshore circalittoral mixed sediment	MC521/ MC42	No suitable matching habitat/ SS.SMx.Omx.#	S56, S60, S78, S79, S81, S89 and S87
Variant with low abundance of *Ophelia borealis.*	*Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand	MC5211	SS.Ssa.CfiSa.EpusOborApri	S55
Variant without *Ophelia borealis*.	*Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand	cf. MC5211	cf. S.Ssa.CfiSa.EpusOborApri	S64, S66, S84 and S85
Transition between two biotopes.	*Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand/*Moerella spp*. with venerid bivalves in Atlantic infralittoral gravelly sand	MC5211/ MB3233	SS.Ssa.CfiSa.EpusOborApri/ SS.SCS.ICS.MoeVen	S30, S38, S40, S42 and S51

Notes	Habitat Classification	EUNIS Habitat Code	JNCC Biotope Code	Sample ID
Variant of biotope.	*Abra prismatica*, *Bathyporeia elegans* and polychaetes in circalittoral fine sand	MC5212	cf. SS.Ssa.CfiSa.ApriBatPo	S68
Variant with low abundance of *Lagis koreni.*	*Lagis koreni* and *Phaxas pellucidus* in Atlantic circalittoral sandy mud	MC6215	SS.Smu.CsaMu.LkorPpel	S62, S76 and S77
Variant without *Lagis koreni.*	*Lagis koreni* and *Phaxas pellucidus* in Atlantic circalittoral sandy mud	cf. MC6215	cf. SS.Smu.CsaMu.LkorPpel	S63

5.3 Area Descriptions

The following area description is sub-divided into a description of the OAA survey area and a description of ECC survey area consisting of ECC West and ECC East.

The survey area is located in a high-energy environment with strong tidal- and wave-current transport, resulting in a complex and dynamic substrate composition.

Habitats **MC32** – Atlantic circalittoral coarse sediment, **MC52** – Atlantic circalittoral sand, and complex of **MC32/MC52** were generally devoid of any conspicuous epibenthic fauna and any epibenthic fauna noted present was in sparse abundance.

Habitats **MC42** – Atlantic circalittoral mixed sediment and **MC12** – Atlantic circalittoral rock both comprised a wider range of species and were in higher abundance, compared to **MC32**, **MC52** and **MC32/MC52**. The most frequently noted colonial species were bryozoans such as *Flustra foliacea*, *Securiflustra securifrons*, *Alcyonidium diaphanum* and *Pentapora foliacea*, cnidarian *Alcyonium digitatum*, *Abietinaria abietina*, *Nemertesia antennina* and *Tubularia indivisa*. In addition, poriferans such as *Polymastia boletiformis*, *Sycon ciliatum*, *Stelligera stuposa*, *Axinella infundibuliformis* and *Hymedesmia paupertas* were identified. Epibenthic fauna identified in the imagery was generally diverse and presented a similar composition, with regards to abundance and diversity, for **MC42** and **MC12**.

Macroalgae were identified in the nearshore sections and generally comprised Rhodophyta, Corallinales, Phaeophyceae and *Laminaria* sp.

Detailed listings of all identified taxa are presented in Appendix C, Appendix D and Appendix E.

5.3.1 Bathymetrical Overview

Water depths within the OAA range from a minimum of 45 m on the Stormy Bank to a maximum of 99 m in the far east of the OAA. Two shoals are evident, one to the northeast at Stormy Bank, and the other to the southwest at Whiten Head Bank; separated by a central area where the water is deeper (Figure 18).

Although the seabed has a relatively high topographic variability, slope angles are typically very gentle (<1°) to gentle (<5°). Higher slope angles are associated with seabed features such as ridges, rippled scour depressions and megaripples.

Along the southern slopes of Stormy Bank predominant E-W currents have formed elongated linear to curvilinear N-S trending depressions with depths on the order of 1 m. Boulders with associated scour marks are evident on top of the bank and to the north with varying sizes but are typically on the order of 1.5 m in diameter extending 0.3 m to 0.5 m above the surrounding seabed.

Along the SE edge of the Whiten Head Bank, two prominent and well-defined ridges rise approx. 5 m above the surrounding seabed. To the southeast of these features, currents have formed NNE-SSW to N-S trending linear to curvilinear rippled scour depressions with depths in the order of 1 m. On the top of the bank, the

seabed appears rugged in patches and boulders are apparent. The boulders, typically associated with scour marks, measure on the order of 1.5 m in diameter.

The Central Area has parts of the Whiten Head Bank and Stormy Bank protruding into it. The top of these banks is defined by rougher sediments with a scattering of boulders with associated scouring. The boulders measure around 0.5 m. Between the higher banks, we see more elongated linear to curvilinear sediment bedforms running in a N-S trending direction with depths on the order of 0.5 m. Their orientation and shape suggest a strong current flow within the area.

On the NW flank of Whiten Head Bank, broad E-W trending rippled scour depressions are overprinted by N-S trending depressions forming a jagged, saw tooth pattern. Low profile, poorly developed bedforms also occur in this area suggesting the presence of variable currents on and around the bank.

The maximum depth along the ECC East, as surveyed during the offshore survey, is 99.34 m whilst the minimum depth is 33.95 m approaching the inshore extent of the survey. From the maximum to minimum depth, the seabed rises gently with an average slope of 0.4° before steepening again with an average slope of 0.9° for the final 5 km. Slope values are generally very gentle to gentle with the occasional moderate gradient found over individual small-scale bedforms.

Boulders, often with associated scour, are evident throughout the corridor. The direction of the scour suggests a dominant W-E flow. An area of ridges and linear to curvilinear rippled scour depressions extend across the corridor in a WSW- ENE orientation.

Sand waves and megaripples are evident with average heights of 2.5 m. Bedforms orientated N-S and E-W are also evident suggesting complex flow conditions within the area.

The maximum depth along the ECC West, as surveyed during the offshore survey, is 111.41 m whilst the minimum depth is 38.80 m approaching the inshore extent of the survey. The route descends gently with an average slope of 0.5° from the OAA border before rising with an average slope of 0.8° towards the inshore extent of the survey. Slope values are generally gentle with the occasional moderate or steep gradient found over various bedforms.

North-south trending linear to curvilinear rippled scour depressions with depths in the order of 1 m are visible, and undulations in the seabed are visible between with further east-west orientated linear to curvilinear rippled scour depressions evident. Areas of megaripples associated with rippled scour depressions are located to the west of the centreline. Bedforms with north-south and east-west orientations are also evident suggesting complex flow conditions in the area.

Water depths within the Nearshore areas of the ECC East and West range from a maximum depth of approximately 51 m furthest from shore, 1.5 km, to a minimum of approximately 1 m in the very inshore areas comprising bedrock along the coastline. The depth decline is rapid from the shallow bedrock to the coarse sediment comprising cobbles and boulders bordering the bedrock at an approximate depth of 28 m. Mega ripples are present as well as dense boulder fields as well as areas where the underlying rock deposits have been covered with sand.

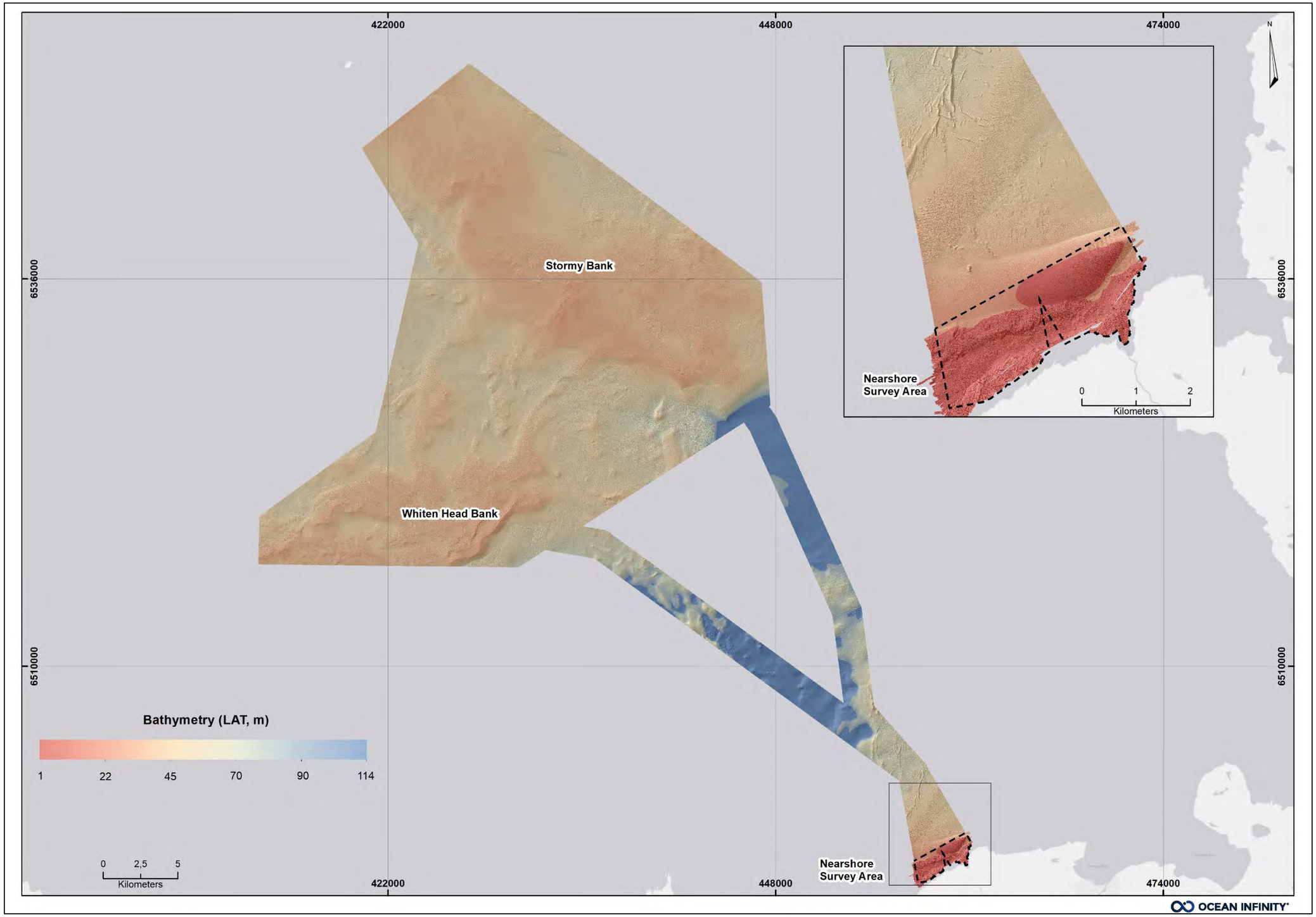


Figure 18 Bathymetric overview of the survey area.

5.3.2 OAA

The majority of the OAA, including Whiten Head Bank, Central and Stormy Bank area, comprises coarse sediments (Figure 19).

Whiten Head Bank

Water depths within the Whiten Head Bank range from 47 m to 82 m and the seabed mainly comprises mixed and rocky sediments, characterised by dense cobbles and boulders classified as **MC42** – Atlantic circalittoral mixed sediment and **MC12** – Atlantic circalittoral rock.

The **MC42** and **MC12** habitats dominate Whiten Head Bank together with areas of rippled scour depressions, classified as **MC32** – Atlantic circalittoral coarse sediment. The **MC32** habitat comprises densely packed coarse sand, gravel, and pebbles with a coarse homogenous surface.

To illustrate the complexity of the substrate composition within the OAA, scattered areas of rippled coarse sands and gravel have been interpreted as a habitat complex of **MC32** – Atlantic circalittoral coarse sediment and **MC52** – Atlantic circalittoral sand (Figure 20). The delineation of the **MC32/MC52** complex, as a separate habitat to **MC32**, was implemented to further show the variation of the coarse sediment composition and subsequently increase the resolution of the mapping.

Areas of finer sediments, surrounded by the rippled scour depressions, have been classified as **MC52** – Atlantic circalittoral sand. These areas are predominantly present in the southernmost section of Whiten Head Bank extending into the ECC West corridor.

Central Area

Water depths within the Central area range between 77 m in the northern sections and 50 m in the southwest which includes a section of the Whiten Head Bank. Across the Central sections of the OAA, habitats **MC42** and **MC12** habitats together with areas of **MC32**, remain prevalent. Areas of **MC32/MC52** are present to a large extent throughout the Central area. The **MC52** habitat is present in the mid and southeastern sections of the Central area, surrounded by rippled scour depressions classified as **MC32**.

Stormy Bank

Water depths within the Stormy Bank range from 45 m, at the top of the bank, to 100 m, in the southeastern section bordering the ECC East corridor.

The majority of the Stormy Bank is interpreted to comprise **MC42** with areas of ripple features and rippled scour depressions, **MC32**. These habitats are most prominent in the northern and central sections of the Stormy Bank. Areas of dense cobbles and boulders, **MC12**, are predominantly interpreted to be present in the northernmost section, with a few isolated areas along the easternmost boundary. The **MC52** habitat is the most prevalent in the deeper parts, south of the Bank, extending into the ECC East corridor.

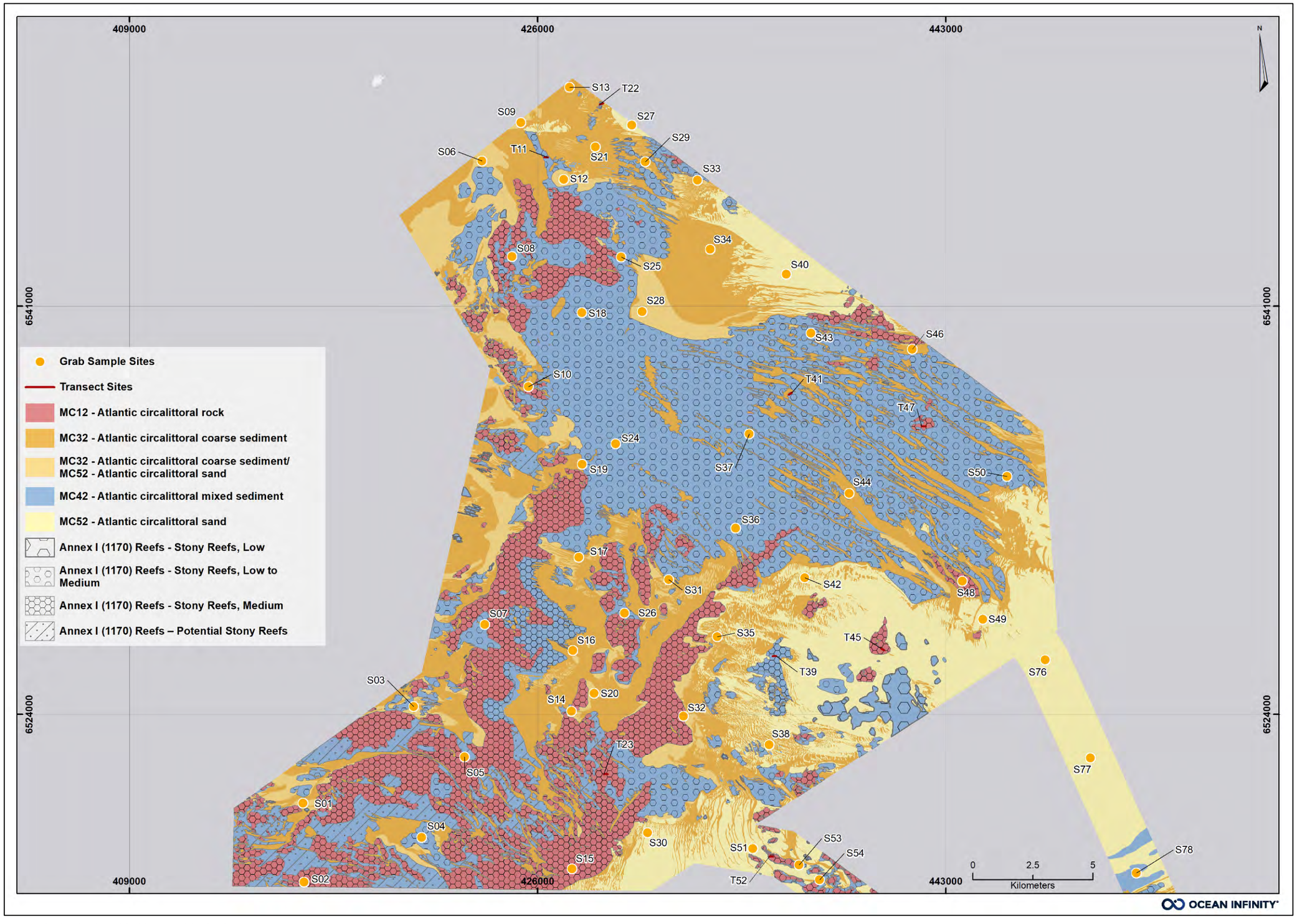


Figure 19 Distribution of habitats within the OAA survey area.

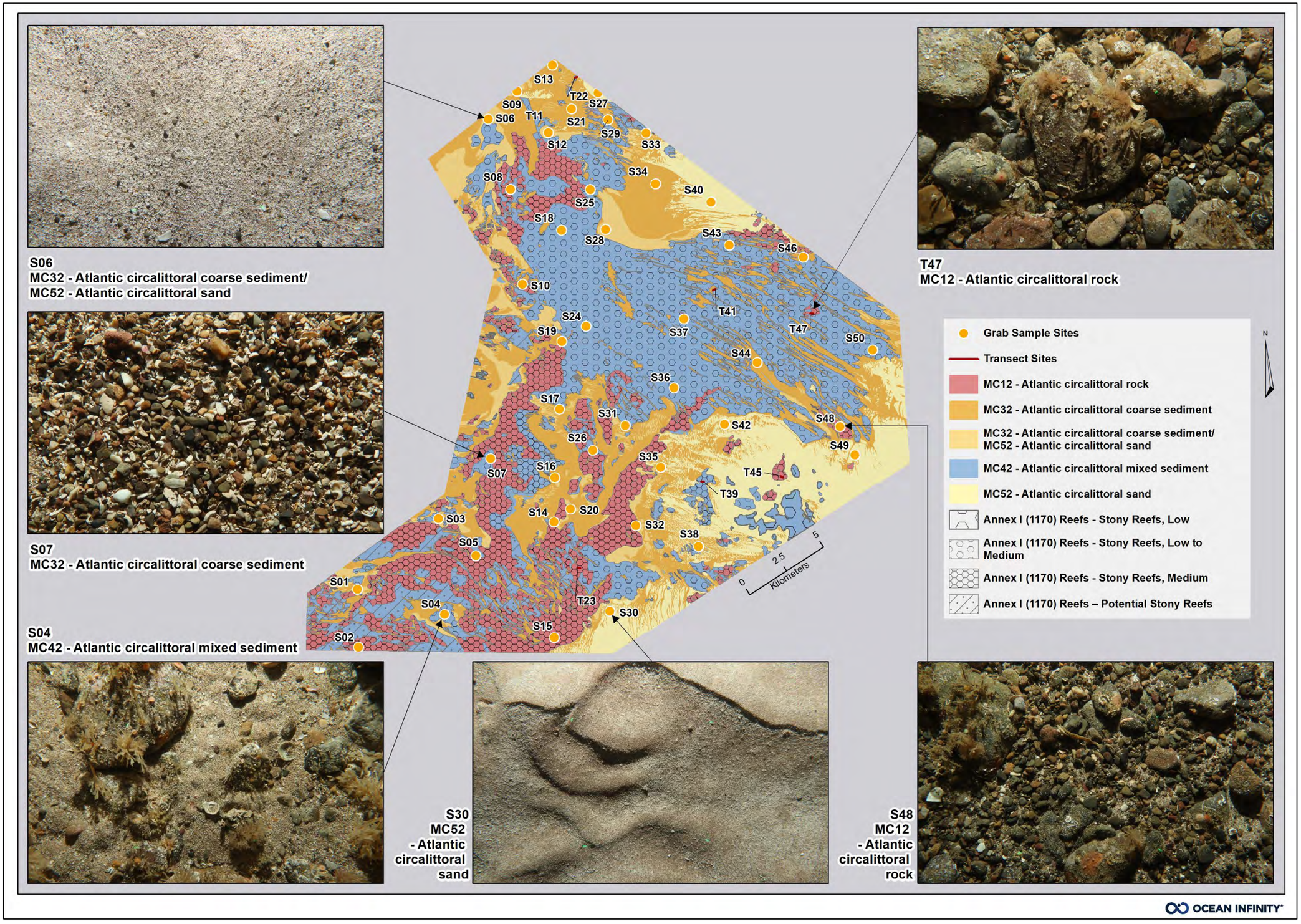


Figure 20 Classified habitats within the OAA, exemplified by imagery data.

5.3.3 ECC West and ECC East

The ECC West and East route corridors partially overlap each other, and the depths range from 115 m to 33 m, and 106 m to 33 m, respectively.

ECC West (Whiten Head Bank to Crosskirk)

Habitat **MC52** – Atlantic circalittoral sand is interpreted along the entire ECC West, with increased presence southwards. In the northernmost section, **MC52** is intermixed with mound features comprising dense cobbles and boulders. These areas are classified in parts as **MC12** – Atlantic circalittoral rock, and where cobble/boulder density was interpreted to be sparser as **MC42** – Atlantic circalittoral mixed sediment. Progressing southwards, the **MC12** and **MC42** habitats are scattered across the route corridor and occur predominantly at the outer edges (Figure 21).

Areas associated with rippled scour depressions, classified as **MC32** – Atlantic circalittoral coarse sediment, are present throughout the ECC West corridor. Transitional areas, at the boundaries between **MC42** and **MC52** habitats, are classified as **MC32** – Atlantic circalittoral coarse sediment/**MC52** – Atlantic circalittoral sand (Figure 22).

ECC East (Stormy Bank to Crosskirk)

The **MC52** habitat is also interpreted along the entire ECC East, dominating the northernmost sections, and decreasing in presence southwards. The mid-sections of the ECC East's corridor are dominated by coarse sediments, cobbles, and boulders. The seabed alternates between areas of **MC42**, rippled scour depressions classified as **MC32** with intermediary patches of coarse sands, and **MC32/MC52**.

Areas of isolated **MC12** are present at the easternmost edge of the ECC East, at the intersection with ECC West, with the features extending across both route corridors. The intersection between ECC West and East comprises a very dynamic seabed with most sedimentary fractions present. Here the **MC42** habitat extends across both route corridors, with the intermediary sediments classified as predominantly **MC52**, as well as **MC32** and **MC32/MC52**.

Towards the landfalls, spanning both route corridors, are large areas comprising flat rather featureless seabed, **MC52**, as well as areas of prominent ripple features, **MC32**.

ECC West and ECC East Nearshore

Two large areas of **MC12** – Atlantic circalittoral rock and **MB12** – Atlantic infralittoral rock characterise the width of both the ECC West and ECC East route corridors. The bedrock comprises exposed bedrock with Kelp as well as patches comprising sand and gravel. The presence of large boulders is noted predominantly at the lower edge of **MB12** – Atlantic infralittoral rock bordering **MC52** – Atlantic circalittoral sand (Figure 23).

Between these areas of bedrock, a channel of predominantly **MC52** – Atlantic circalittoral sand is interpreted to be present. Several areas of **MC42** – Atlantic circalittoral mixed sediment as well as areas of habitat complex **MC52** – Atlantic circalittoral sand/**MC12** – Atlantic circalittoral rock. Areas classified as **MC52 –** Atlantic circalittoral sand/**MC12 –** Atlantic circalittoral rock comprise areas where it is notable rock presence underneath a veneer of sand, and where the cobbles and boulders are protruding (Figure 24).

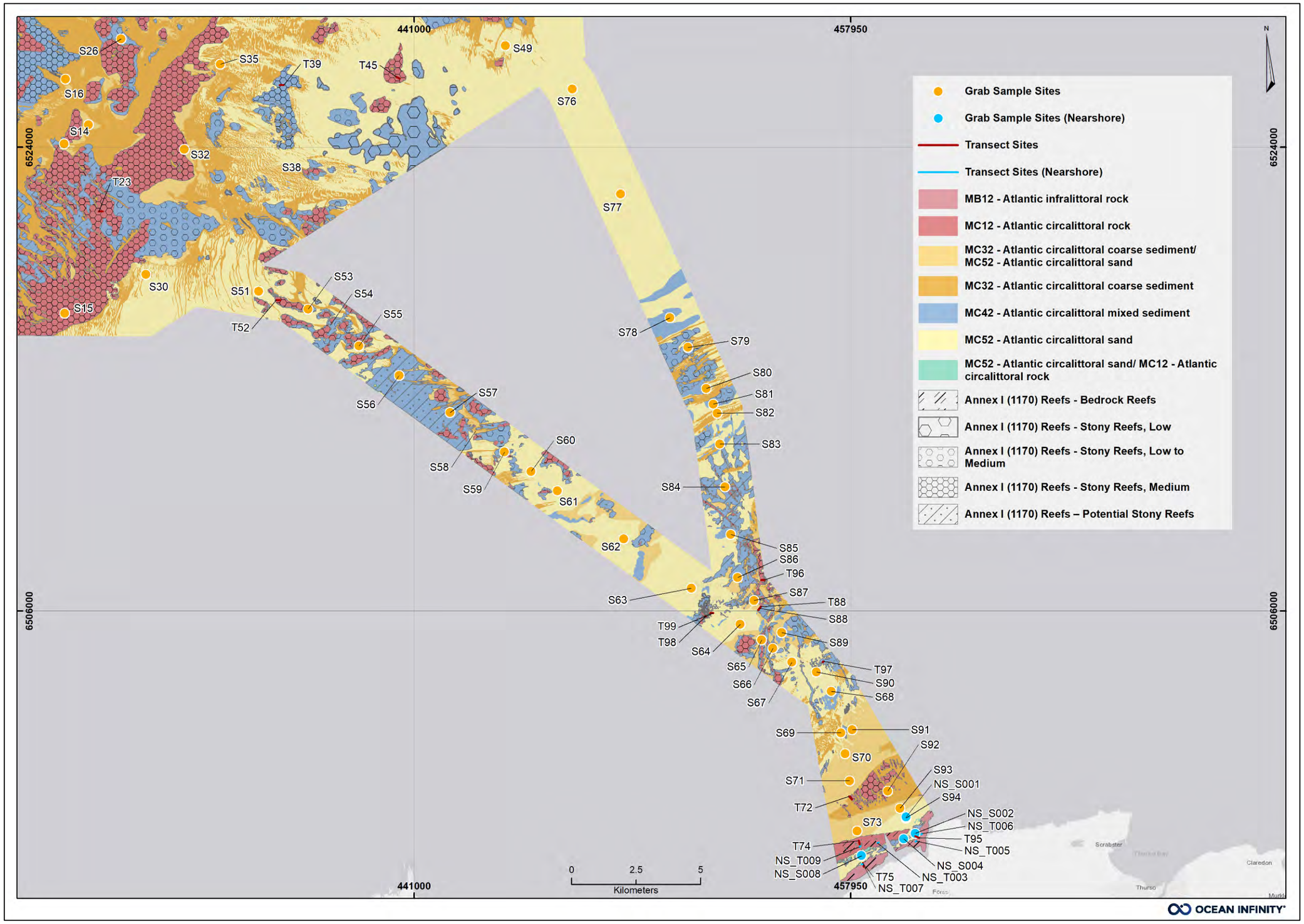


Figure 21 Distribution of habitats within the ECC West and East survey areas.

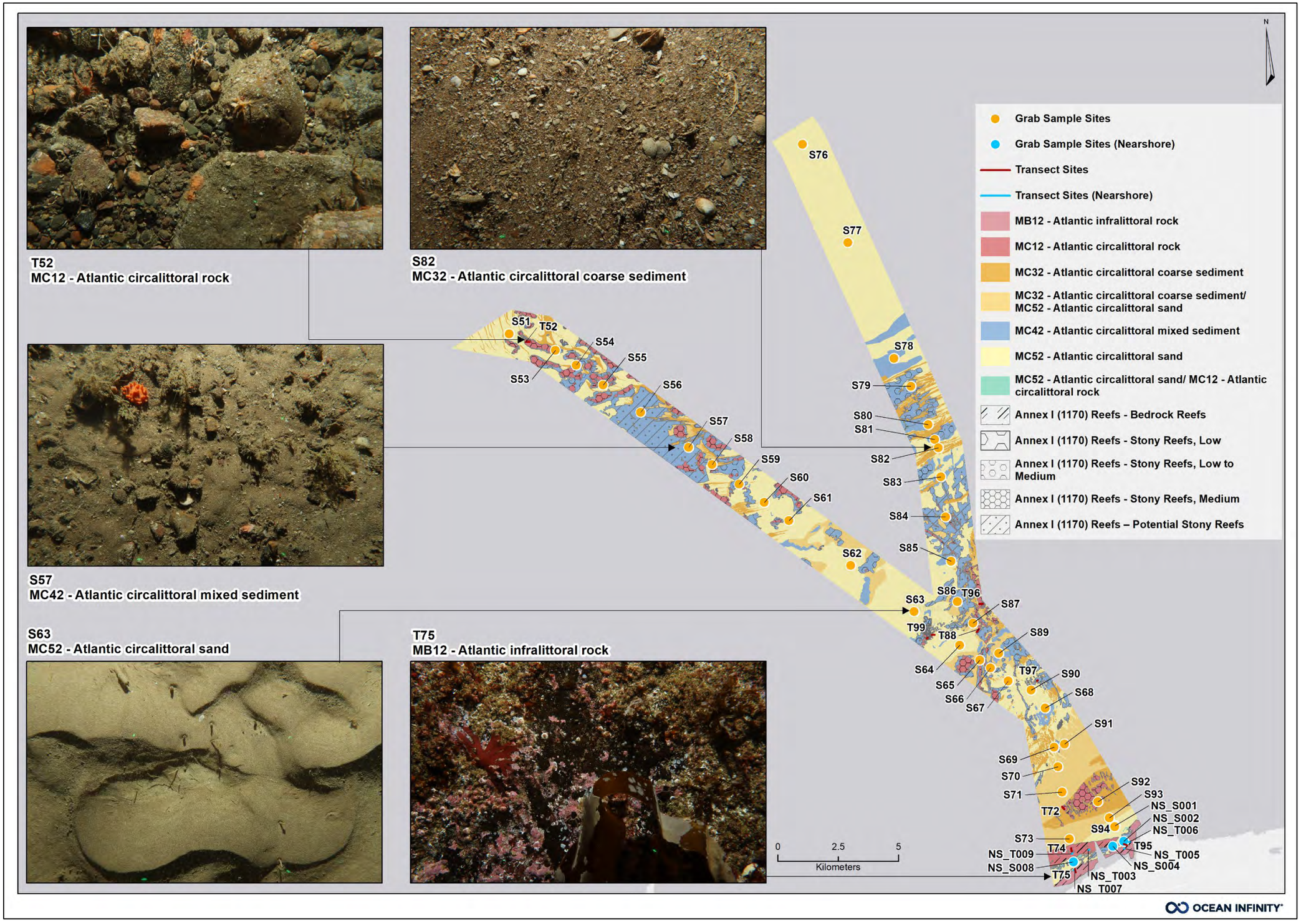


Figure 22 Classified habitats within the ECC West and East, exemplified by imagery data.

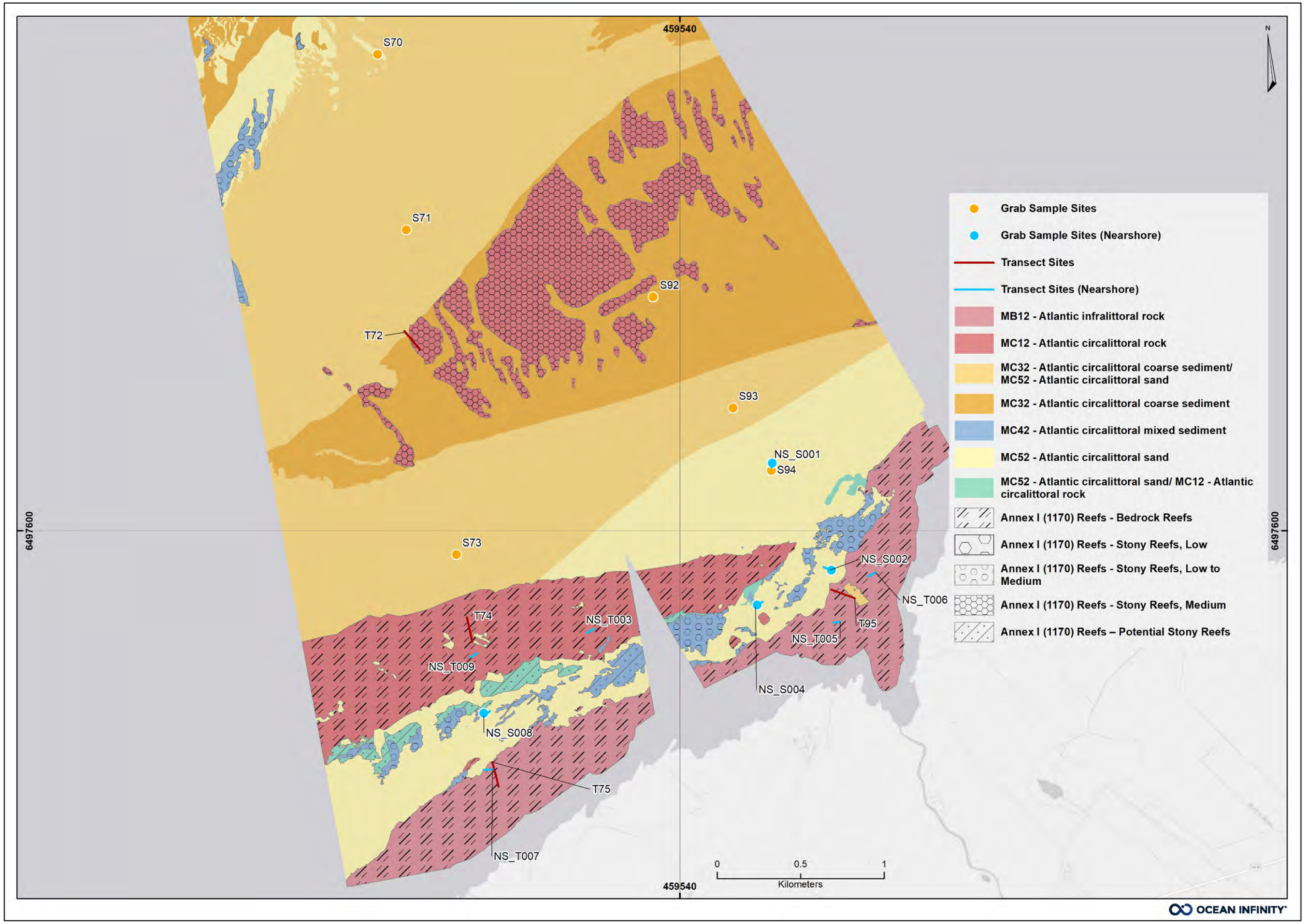


Figure 23 Distribution of habitats within the ECC West and East nearshore survey areas.

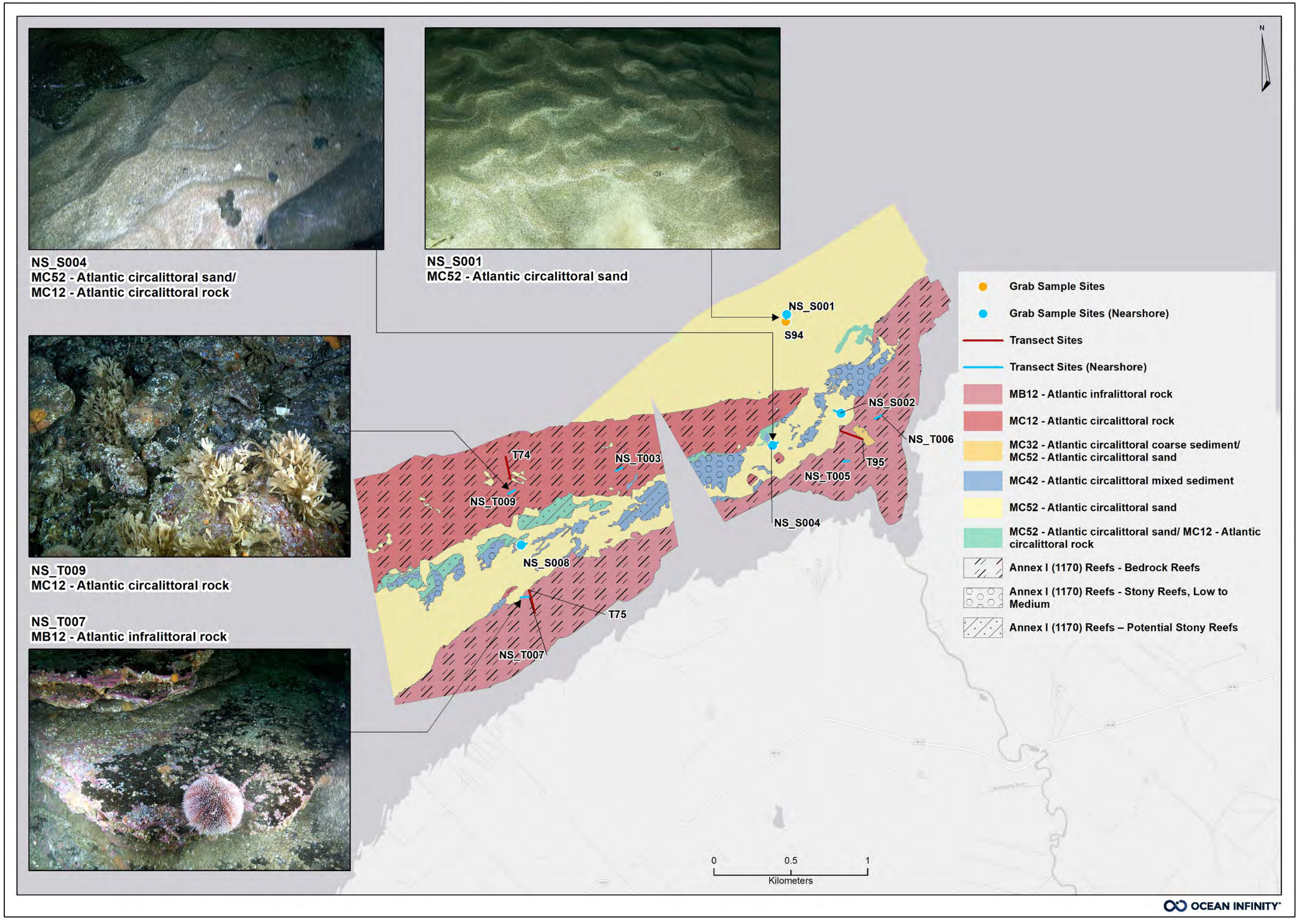


Figure 24 Classified habitats within the ECC West and East nearshore survey area, exemplified by imagery data.

5.4 Particle Size Distribution

In the Offshore area, 82 sites were selected for PSA sampling, out of which 67 samples were successfully acquired. In the Nearshore area, four (4) sites were selected for PSA sampling, out of which three (3) samples were successfully acquired.

Detailed results from the PSA are stated in Appendix F.

Sand was the dominating sediment fraction, with a mean content of 78.7 % (SD=24.6), followed by Gravel which had a mean content of 19.9 % (SD=25.0). The Mud content was low with a mean content of 1.5 % (SD=1.7), comprising 1.2 % (SD=1.5) Silt and 0.3 % (SD=0.3) Clay (Table 27).

The results from the PSA analyses showed a limited variation in the sediment composition. The OAA area primarily comprised Sand and Gravel, with variable ratios, with Sand dominating the ECC (including ECC West, ECC East and Nearshore) (Figure 25). These trends from the PSA results are further supported by the Backscatter data (Figure 26).

Note: PSA samples from grab sample sites OAA_S26, OAA_S36, ECW_S54 and ECW_ S61 comprised low sample volumes and are thus potentially unrepresentative.

Table 27 Summary of PSA results.

Sample ID	Depth (m)	Sediment Fraction (%)				Mud (%) (Silt + Clay)	BGS (1982) Classification (modified from Folk, 1954)
		Gravel	Sand	Silt	Clay		
OAA_S01	67	0.9	97.5	1.3	0.3	1.6	Sand
OAA_S03	67	11.5	87.9	0.4	0.2	0.6	Gravelly Sand
OAA_S05	56	55.6	43.0	1.1	0.4	1.5	Sandy Gravel
OAA_S06	55	7.8	91.8	0.3	0.1	0.4	Gravelly Sand
OAA_S07	64	54.7	44.9	0.4	0.1	0.5	Sandy Gravel
OAA_S09	56	48.5	51.1	0.3	0.2	0.4	Sandy Gravel
OAA_S12	57	4.9	94.8	0.2	0.1	0.3	Slightly Gravelly Sand
OAA_S13	63	51.2	48.5	0.2	0.0	0.3	Sandy Gravel
OAA_S14	61	62.3	37.5	0.1	0.1	0.2	Sandy Gravel
OAA_S16	67	43.9	54.0	1.8	0.3	2.2	Sandy Gravel
OAA_S17	69	19.2	80.3	0.3	0.1	0.4	Gravelly Sand
OAA_S19	60	52.7	45.9	1.3	0.2	1.5	Sandy Gravel
OAA_S20	65	1.1	98.4	0.3	0.2	0.5	Slightly Gravelly Sand
OAA_S21	60	48.0	51.5	0.3	0.1	0.4	Sandy Gravel
OAA_S25	53	64.6	35.1	0.2	0.1	0.3	Sandy Gravel
OAA_S26*	64	71.1	28.5	0.3	0.1	0.4	Sandy Gravel
OAA_S27	63	0.2	99.8	0.0	0.0	0.0	Sand
OAA_S28	56	1.8	97.8	0.2	0.2	0.3	Slightly Gravelly Sand
OAA_S29	56	11.9	88.1	0.0	0.0	0.0	Gravelly Sand
OAA_S30	68	0.4	99.0	0.4	0.3	0.7	Sand

Sample ID	Depth (m)	Sediment Fraction (%)				Mud (%) (Silt + Clay)	BGS (1982) Classification (modified from Folk, 1954)
		Gravel	Sand	Silt	Clay		
OAA_S31	68	34.1	65.3	0.4	0.2	0.6	Sandy Gravel
OAA_S32	56	3.8	95.5	0.5	0.3	0.7	Slightly Gravelly Sand
OAA_S33	65	0.2	99.8	0.0	0.0	0.0	Sand
OAA_S34	66	38.4	60.5	1.0	0.2	1.1	Sandy Gravel
OAA_S35	68	9.2	90.1	0.5	0.2	0.7	Gravelly Sand
OAA_S36*	53	79.1	20.7	0.1	0.0	0.1	Sandy Gravel
OAA_S37	50	48.2	51.5	0.3	0.0	0.3	Sandy Gravel
OAA_S38	67	0.2	98.9	0.5	0.4	0.9	Sand
OAA_S40	70	0.1	99.9	0.0	0.0	0.0	Sand
OAA_S42	66	0.1	99.4	0.4	0.1	0.5	Sand
OAA_S43	61	39.4	60.1	0.3	0.2	0.5	Sandy Gravel
OAA_S44	56	68.5	31.3	0.2	0.1	0.2	Sandy Gravel
OAA_S46	62	71.9	27.8	0.2	0.1	0.3	Sandy Gravel
OAA_S49	67	0.1	99.0	0.5	0.4	0.9	Sand
ECW_S51	78	0.2	98.9	0.6	0.4	1.0	Sand
ECW_S54*	79	72.9	26.0	0.9	0.1	1.1	Sandy Gravel
ECW_S55	85	4.3	93.4	1.6	0.7	2.3	Slightly Gravelly Sand
ECW_S56	88	44.2	53.0	2.4	0.4	2.8	Sandy Gravel
ECW_S57	85	40.1	56.2	3.2	0.5	3.7	Sandy Gravel
ECW_S58	93	10.1	87.1	2.4	0.3	2.7	Gravelly Sand
ECW_S60	97	0.2	94.8	4.3	0.8	5.1	Sand
ECW_S61*	102	13.4	81.5	4.3	0.8	5.1	Gravelly Sand
ECW_S62	110	0.1	93.3	5.5	1.1	6.6	Sand
ECW_S63	108	0.1	95.8	3.3	0.8	4.2	Sand
ECW_S64	96	0.3	97.0	2.1	0.7	2.8	Sand
ECW_S66	78	6.0	92.0	1.5	0.5	1.9	Gravelly Sand
ECW_S67	82	1.0	97.5	1.0	0.6	1.6	Sand
ECW_S68	76	0.7	98.0	0.8	0.5	1.4	Sand
ECW_S69	63	0.0	100.0	0.0	0.0	0.0	Sand
ECW_S70	60	0.3	98.3	0.8	0.5	1.3	Sand
ECW_S71	57	22.2	77.3	0.3	0.1	0.5	Gravelly Sand
ECW_S73	42	5.2	94.5	0.2	0.1	0.3	Gravelly Sand
ECE_S76	97	0.4	91.4	7.1	1.1	8.2	Sand

Sample ID	Depth (m)	Sediment Fraction (%)				Mud (%) (Silt + Clay)	BGS (1982) Classification (modified from Folk, 1954)
		Gravel	Sand	Silt	Clay		
ECE_S77	90	0.1	92.7	6.2	1.0	7.2	Sand
ECE_S78	93	4.2	92.1	3.0	0.7	3.7	Slightly Gravelly Sand
ECE_S79	85	57.8	41.0	1.0	0.2	1.2	Sandy Gravel
ECE_S80	85	0.8	97.3	1.2	0.7	1.9	Sand
ECE_S82	89	21.3	76.6	1.8	0.2	2.1	Gravelly Sand
ECE_S83	88	7.5	89.5	2.6	0.5	3.0	Gravelly Sand
ECE_S84	85	1.6	96.3	1.5	0.7	2.1	Slightly Gravelly Sand
ECE_S85	88	2.0	96.2	1.1	0.7	1.8	Slightly Gravelly Sand
ECE_S86	84	10.3	86.7	2.2	0.7	2.9	Gravelly Sand
ECE_S89	79	8.7	89.4	1.4	0.5	1.9	Gravelly Sand
ECE_S90	81	0.7	97.8	1.0	0.5	1.5	Sand
ECE_S91	67	46.8	52.1	0.9	0.2	1.1	Sandy Gravel
ECE_S93	41	0.1	99.9	0.1	0.0	0.1	Sand
ECE_S94	35	0.0	100.0	0.0	0.0	0.0	Sand
NS_S001	32	0.0	100.0	0.0	0.0	0.0	Sand
NS_S004	36	2.3	97.2	0.2	0.3	0.5	Slightly Gravelly Sand
NS_S008	34	0.0	100.0	0.0	0.0	0.0	Sand
Mean		**19.9**	**78.7**	**1.2**	**0.3**	**1.5**	
SD		**25.0**	**24.6**	**1.5**	**0.3**	**1.7**	
Min		**0.0**	**20.7**	**0.0**	**0.0**	**0.0**	
Max		**79.1**	**100.0**	**7.1**	**1.1**	**8.2**	
Median		**5.6**	**91.9**	**0.5**	**0.2**	**0.8**	

**Sample comprised low volume.*

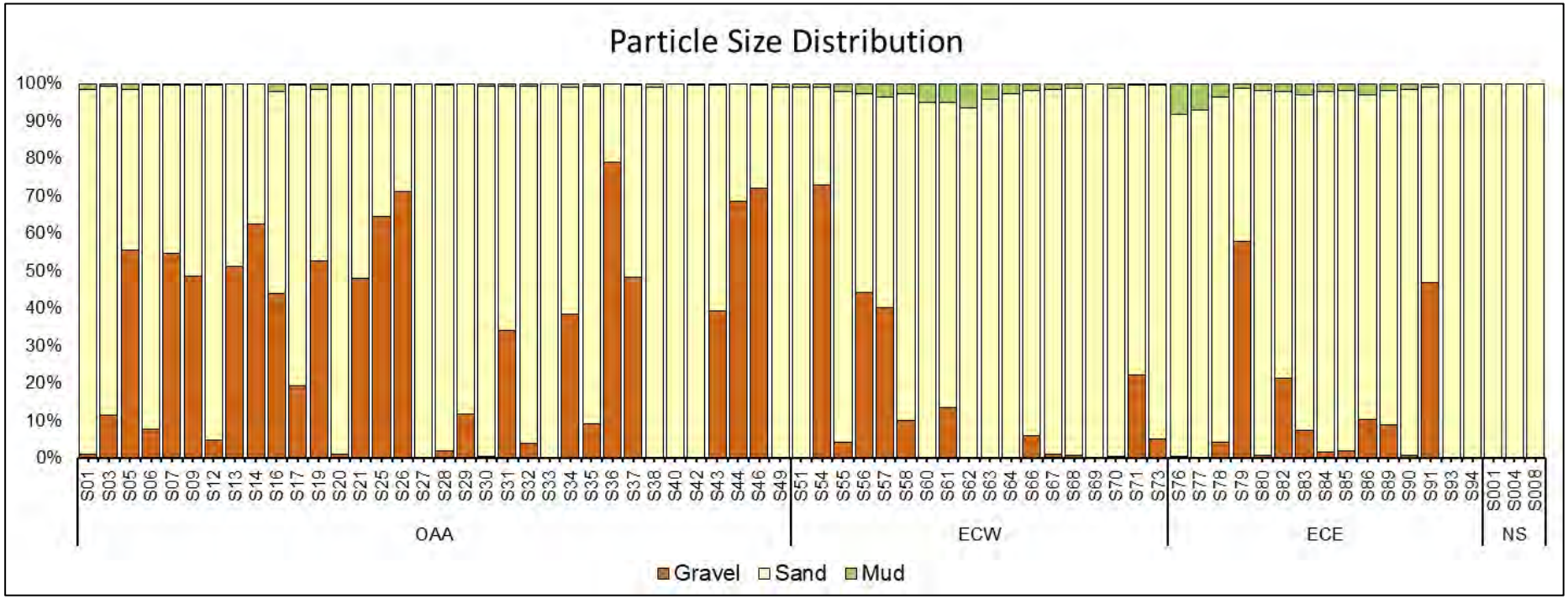


Figure 25 Particle Size Distribution from grab samples.

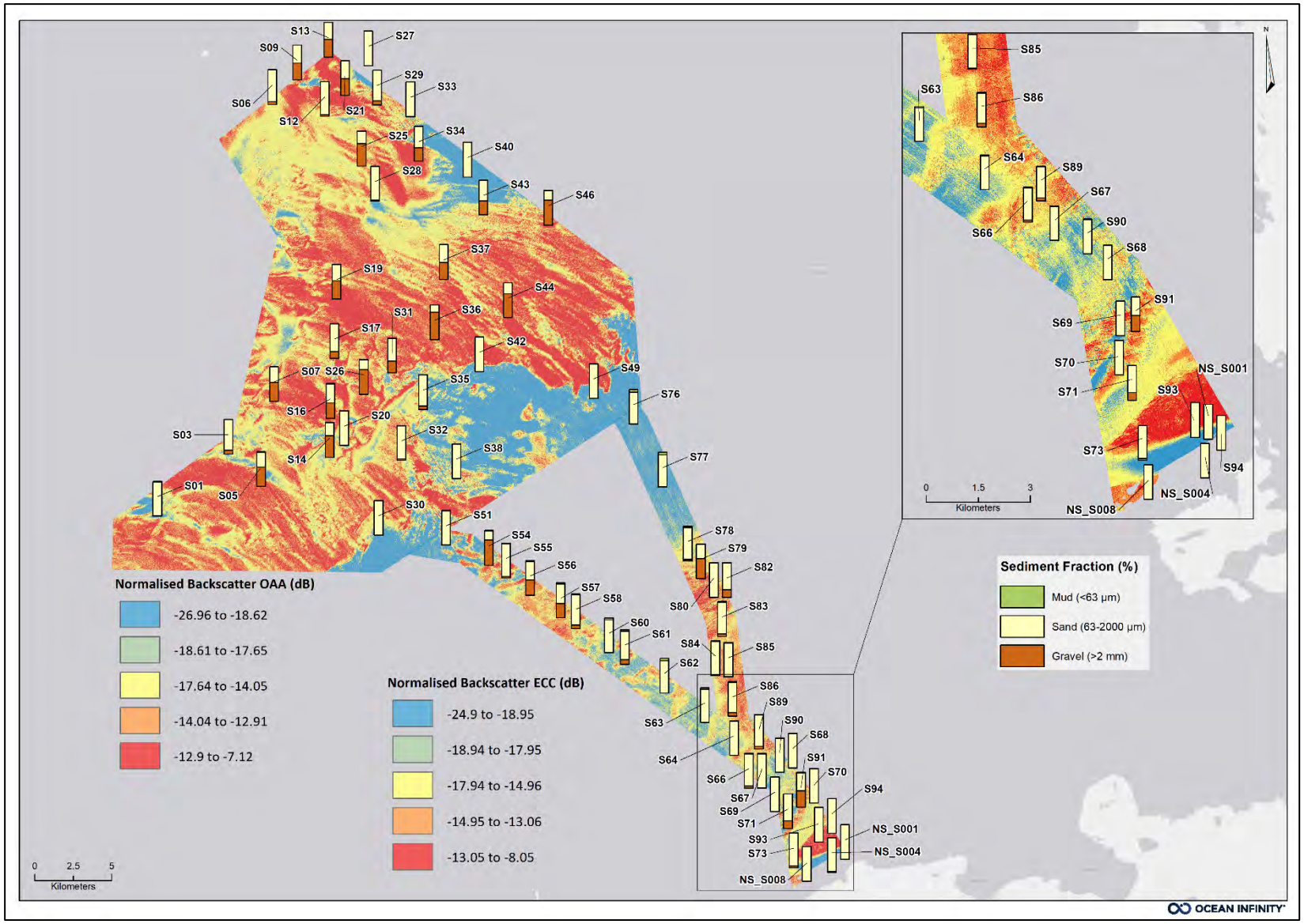


Figure 26 Overview of Particle Size Distribution superimposed with the offshore backscatter data.

5.4.1 Multivariate Analyses for Sediment

Multivariate analyses were undertaken on the PSA data set, to identify patterns in the sediment distribution. Analyses included hierarchical clustering employing the Euclidean distance resemblance matrix, SIMPROF analysis and principal component analysis (PCA). The datasets were normalised prior to the analyses being performed.

The SIMPROF analysis of the sediment composition produced 22 distinct groups separating the 66 grab sample sites with accepted sample volume (Figure 27).

Principal component 1 (PC1), explaining 68.5 % of the variation, separated the sites based on the gravel-to-sand ratio. Principal component 2 (PC2), explaining 31.5 % of the variation, separated the sites based on the mud content (Figure 28).

SIMPROF Groups **g**, **h**, **I**, **j**, **k**, **l**, **m**, **n**, **o**, **p**, **q**, **r**, **s**, **t**, **u**, and **v** comprise clean sand and sand with some gravel, corresponding to the Folk classes Sand, Slightly Gravelly Sand, and Gravelly Sand; Groups **a**, **b**, **c**, **d**, **e**, and **f** comprise coarse sediments, corresponding to the Folk class Sandy Gravel.

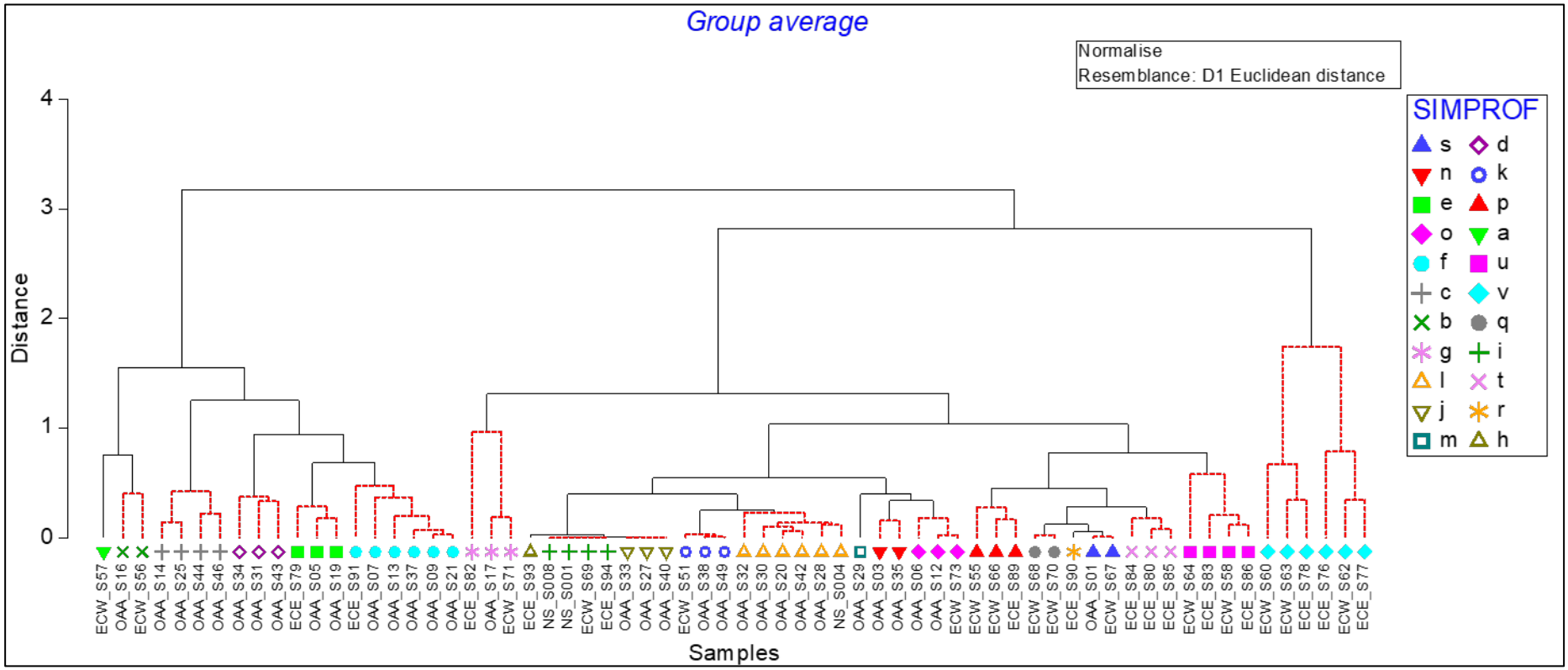


Figure 27 Dendrogram based on Euclidian distance for the sediment composition for accepted samples, showing SIMPROF groups with a 5 % significance level.

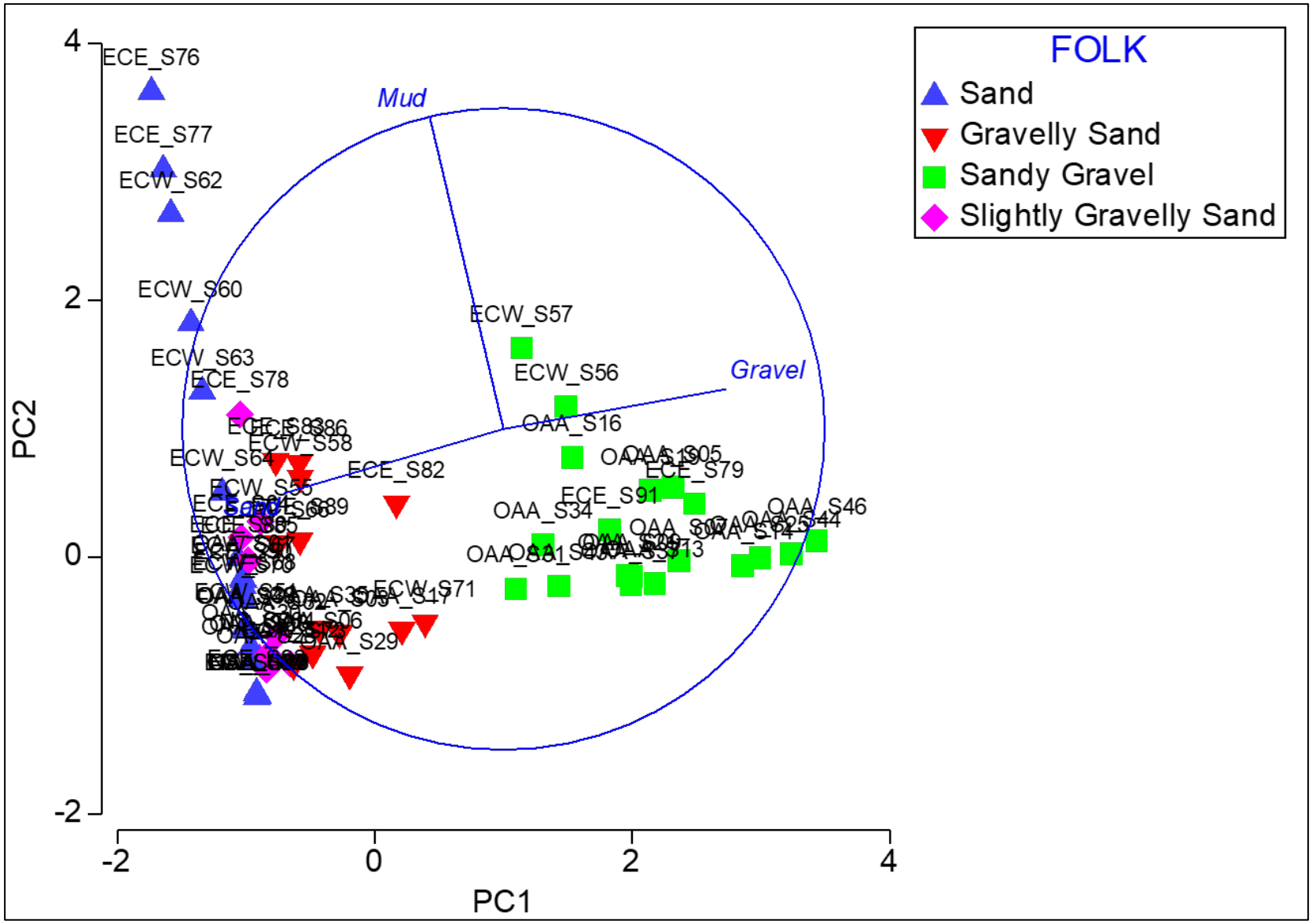


Figure 28 PCA plot of sediment composition for accepted samples, showing groups based on the FOLK classifications.

5.5 Sediment Chemical and Contaminant Analyses

Offshore sampling

A total of 82 sites were selected for chemical and contaminant sampling. Samples were successfully acquired at 57 sites, with one (1) site (OAA_S09) not having samples acquired for metal and organics analyses. Out of the 82 sites, 31 sites were selected for additional analyses of pesticide and flame-retardants, out of which 26 were successfully sampled.

Nearshore sampling

A total of four (4) sites were selected for chemical and contaminant sampling. Samples were successfully acquired at three (3) sites. All four (4) sites were selected for additional analyses of pesticide and flame-retardants, out of which three (3) were successfully sampled.

Detailed results from the chemical analyses are stated in Appendix G.

5.5.1 Metals

Metal concentrations were overall low throughout the survey area. Threshold values were exceeded at 13 grab sample sites (Table 28).

The threshold value for arsenic (As) according to the Canadian Council of Ministers of the Environment's (CCME) ISQG (7.24 mg/kg) was exceeded at seven (7) sites (Figure 29). Exceeding the ISQG threshold indicates "the possible effect range within which adverse effects occasionally occur" (CCME, 2001). Furthermore, one (1) of these samples exceeded the lower threshold of the Norwegian Environment Agency's (NEA) class 2- Good (15 mg/kg), thereby exceeding the expected natural background levels (class 1 – Background), according to NEA (NEA, 2016, revised 2020).

The threshold value for nickel (Ni) according to CEFAS' AL1 (20 mg/kg) was exceeded at eight (8) sites (Figure 30). Furthermore, two (2) of these samples exceeded the lower threshold of the NEA class 2 Good (30 mg/kg), thereby exceeding the expected natural background levels (class 1 – Background).

The arsenic and nickel concentrations do not display any clear geographical trends, but the samples from the ECC West are notably lower compared to the samples from the OAA and ECC East, with one (1) sample exceeding a threshold value for nickel (Figure 31; Figure 32). Both metals have their highest recorded levels at site OAA_S046, located in an area comprising coarse sediment just northeast of a larger area comprising mixed sediment.

Table 28 Metal concentrations (mg/kg dry weight) in samples with threshold values. Highlighted cells indicate where threshold values have been exceeded.

Analytes	As	Cr	Cu	Ni	Pb	Zn	Cd	Hg
Method	ICPMSS	ICPMSS	ICPMSS	ICPMSS	ICPMSS	ICPMSS	ICPMSS	ICPMSS
Limit of Detection	0.5	0.5	0.5	0.5	0.5	2	0.04	0.01
NEA 1 Background	0	0	0	0	0	0	0	0
NEA 2 Good	15	60	20	30	25	90	0.2	0.05
NEA 3 Moderate	18	620	-	42	150	139	2.5	0.52
NEA 4 Poor	71	6000	48	271	1480	750	16	0.75
NEA 5 Very Poor	580	15500	147	533	2000	6690	147	1.45
OSPAR ERL	81	34	-	47	150	150	1.2	0.15
CEFAS AL1	20	40	40	20	50	130	0.4	0.3
CEFAS AL2	100	400	400	200	500	800	5	3
CCME ISQG	7.24	52.3	18.7	-	30.2	124	0.7	0.13
CCME PEL	41.6	160	108	-	112	271	4.2	0.7
Dutch RIVM	85	380	190	210	580	2000	14	10
Units	**mg/kg**	**mg/kg**	**mg/kg**	**mg/kg**	**mg/kg**	**mg/kg**	**mg/kg**	**mg/kg**
OAA_S01	4.6	5.0	3.5	5.7	4.2	11.7	<0.04*	<0.01*
OAA_S03	6.0	7.0	4.1	9.0	4.8	9.8	0.05	<0.01*
OAA_S06	2.0	5.2	1.9	5.3	1.4	7.0	0.10	<0.01*
OAA_S07	10.2	18.0	6.7	18.1	6.4	33.3	<0.04*	<0.01*
OAA_S12	3.2	3.9	2.9	4.6	2.1	7.5	0.06	<0.01*
OAA_S13	6.3	6.5	3.6	8.2	7.7	15.6	0.09	<0.01*
OAA_S16	4.8	6.0	3.5	5.7	4.1	10.5	0.12	<0.01*
OAA_S17	6.6	6.4	2.5	8.0	4.3	10.8	<0.04*	<0.01*
OAA_S19	6.1	8.9	5.4	8.0	4.7	15.5	<0.04*	<0.01*
OAA_S20	3.6	5.0	2.5	7.3	3.0	7.6	<0.04*	<0.01*
OAA_S25	5.1	8.8	5.0	10.1	5.0	14.1	<0.04*	<0.01*
OAA_S27	2.4	3.7	2.1	3.9	1.5	7.9	<0.04*	<0.01*
OAA_S28	1.8	5.1	2.7	5.9	1.8	11.6	<0.04*	<0.01*
OAA_S30	3.0	4.2	2.6	6.2	2.1	8.5	<0.04*	<0.01*
OAA_S31	3.3	3.7	2.1	3.9	2.7	6.4	<0.04*	<0.01*
OAA_S33	1.8	2.9	2.1	3.3	1.4	6.0	<0.04*	<0.01*
OAA_S34	8.8	10.2	5.1	12.3	5.4	17.9	<0.04*	<0.01*
OAA_S35	3.8	4.6	2.4	3.7	1.9	5.7	<0.04*	<0.01*
OAA_S38	5.6	5.3	3.3	6.1	2.7	9.4	<0.04*	<0.01*
OAA_S40	2.6	6.0	3.0	13.4	1.8	8.6	<0.04*	<0.01*
OAA_S42	2.3	5.4	3.7	13.1	2.0	15.6	<0.04*	<0.01*
OAA_S43	6.9	11.1	5.3	21.5	4.1	19.0	<0.04*	<0.01*
OAA_S44	11.1	20.5	8.6	23.7	15.5	34.7	<0.04*	<0.01*
OAA_S46	17.8	18.9	8.5	39.3	15.1	36.0	<0.04*	<0.01*
OAA_S49	2.2	7.2	3.6	29.5	1.9	12.3	<0.04*	<0.01*
ECW_S51	3.6	6.2	2.9	14.2	2.4	10.0	<0.04*	<0.01*
ECW_S54	3.1	6.8	4.0	21.3	1.7	14.1	<0.04*	<0.01*
ECW_S55	<0.5	1.0	<0.5	4.9	<0.5	2.7	<0.04*	<0.01*
ECW_S56	<0.5	0.7	1.0	6.4	<0.5	2.8	<0.04*	<0.01*
ECW_S57	6.6	11.7	3.9	9.8	5.3	18.2	<0.04*	<0.01*
ECW_S58	5.2	7.5	2.8	6.0	5.5	12.0	<0.04*	<0.01*
ECW_S60	6.0	8.6	3.9	5.3	6.7	13.3	<0.04*	<0.01*
ECW_S62	2.7	9.8	3.0	6.2	4.9	15.3	<0.04*	<0.01*
ECW_S63	2.8	8.2	2.6	5.3	4.6	13.0	<0.04*	<0.01*
ECW_S64	7.2	29.4	3.0	6.7	6.7	13.0	<0.04*	<0.01*
ECW_S67	6.9	16.8	3.4	11.2	4.8	16.4	<0.04*	<0.01*
ECW_S68	5.8	7.2	2.1	4.9	3.8	11.0	<0.04*	<0.01*

Analytes	As	Cr	Cu	Ni	Pb	Zn	Cd	Hg
ECW_S69	5.8	5.4	2.1	4.4	2.3	8.7	<0.04*	<0.01*
ECW_S70	4.1	5.8	2.3	4.6	2.4	11.2	<0.04*	<0.01*
ECW_S71	4.7	3.5	2.7	10.7	3.9	30.9	<0.04*	<0.01*
ECW_S73	4.2	4.9	3.5	13.0	3.6	9.4	<0.04*	<0.01*
ECE_S76	1.8	8.6	4.1	22.4	4.2	14.0	<0.04*	<0.01*
ECE_S77	3.3	5.9	3.1	6.8	4.9	11.8	<0.04*	<0.01*
ECE_S78	3.2	8.0	3.3	7.9	4.9	11.8	<0.04*	<0.01*
ECE_S79	6.3	12.3	5.2	15.9	5.4	22.1	<0.04*	<0.01*
ECE_S80	4.5	11.8	4.1	34.8	5.4	15.2	<0.04*	<0.01*
ECE_S82	13.3	8.3	4.1	12.9	8.3	15.8	<0.04*	<0.01*
ECE_S83	8.9	10.5	3.5	9.3	6.9	14.3	<0.04*	<0.01*
ECE_S84	4.0	6.9	3.8	8.1	4.5	9.4	<0.04*	<0.01*
ECE_S85	4.5	8.9	7.0	11.3	5.6	12.9	<0.04*	<0.01*
ECE_S86	5.7	13.0	3.4	15.4	4.8	12.6	<0.04*	<0.01*
ECE_S89	6.1	7.6	3.5	6.7	4.5	9.2	<0.04*	<0.01*
ECE_S90	10.6	11.0	4.1	12.5	5.5	15.4	<0.04*	<0.01*
ECE_S91	5.1	5.2	2.9	20.1	6.9	13.0	<0.04*	<0.01*
ECE_S93	7.1	7.7	2.8	14.1	2.4	16.4	<0.04*	<0.01*
ECE_S94	4.3	5.7	2.3	6.4	1.6	8.8	<0.04*	<0.01*
NS_S001	5.7	6.3	4.9	6.8	2.6	12.1	<0.04*	0.02
NS_S004	5.0	6.8	3.5	5.8	2.5	11.4	<0.04*	0.02
NS_S008	3.8	8.1	3.4	5.1	2.1	10.4	<0.04*	0.01
Mean	**5.3**	**8.1**	**3.6**	**10.7**	**4.4**	**13.2**	**0.08**	**0.02**
SD	**3.0**	**4.8**	**1.5**	**7.6**	**2.7**	**6.7**	**0.03**	**0.01**
Min	**1.8**	**0.7**	**1.0**	**3.3**	**1.4**	**2.7**	**0.05**	**0.01**
Max	**17.8**	**29.4**	**8.6**	**39.3**	**15.5**	**36.0**	**0.12**	**0.02**
Median	**4.8**	**6.9**	**3.4**	**8.0**	**4.2**	**12.0**	**0.09**	**0.02**

**Not included in statistical analyses of Mean, SD, Min, Max and Median.*

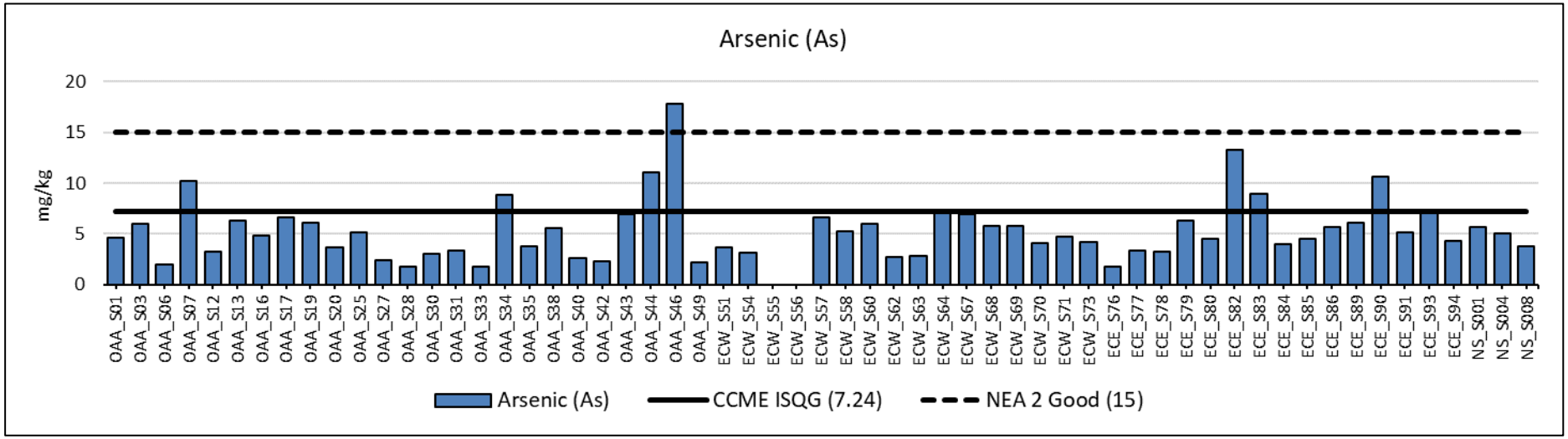


Figure 29 Arsenic (As) levels (mg/kg dry weight) from grab samples with threshold values.

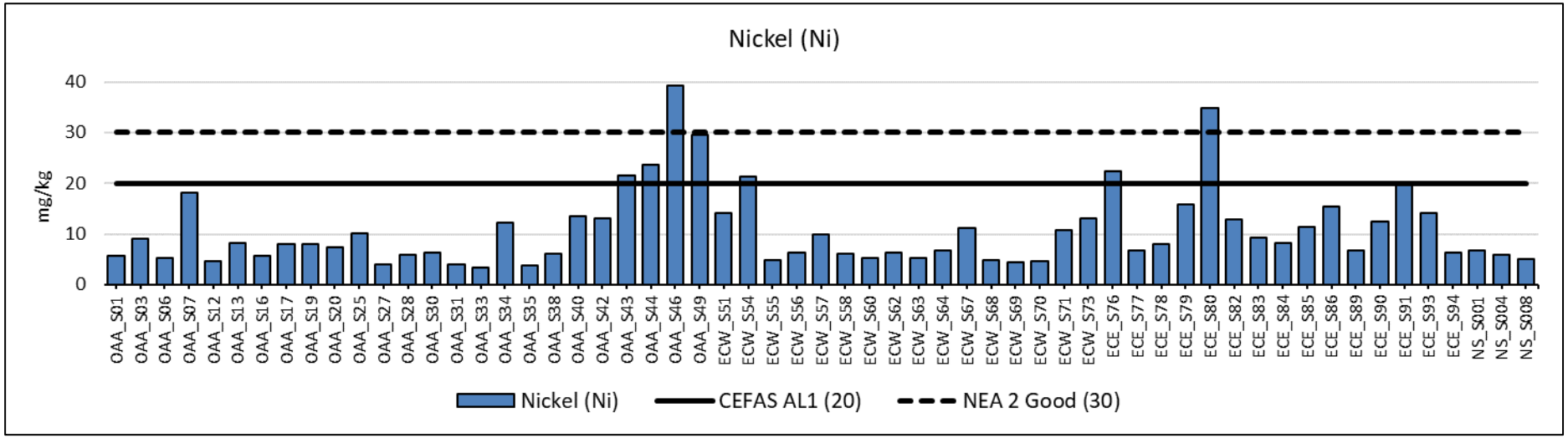


Figure 30 Nickel (Ni) levels (mg/kg dry weight) from grab samples with threshold values.

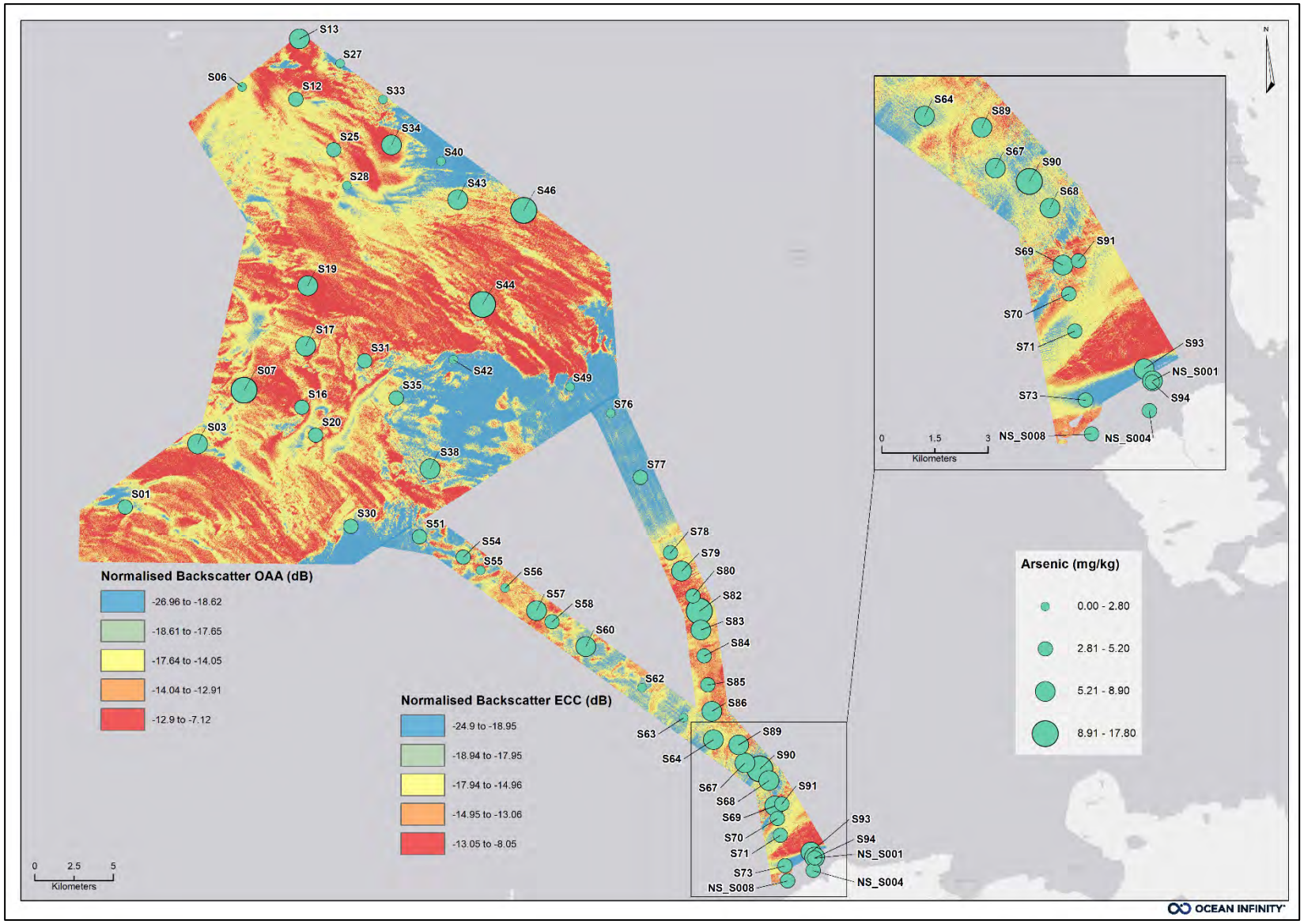


Figure 31 Arsenic (As) concentrations superimposed on the offshore backscatter data.

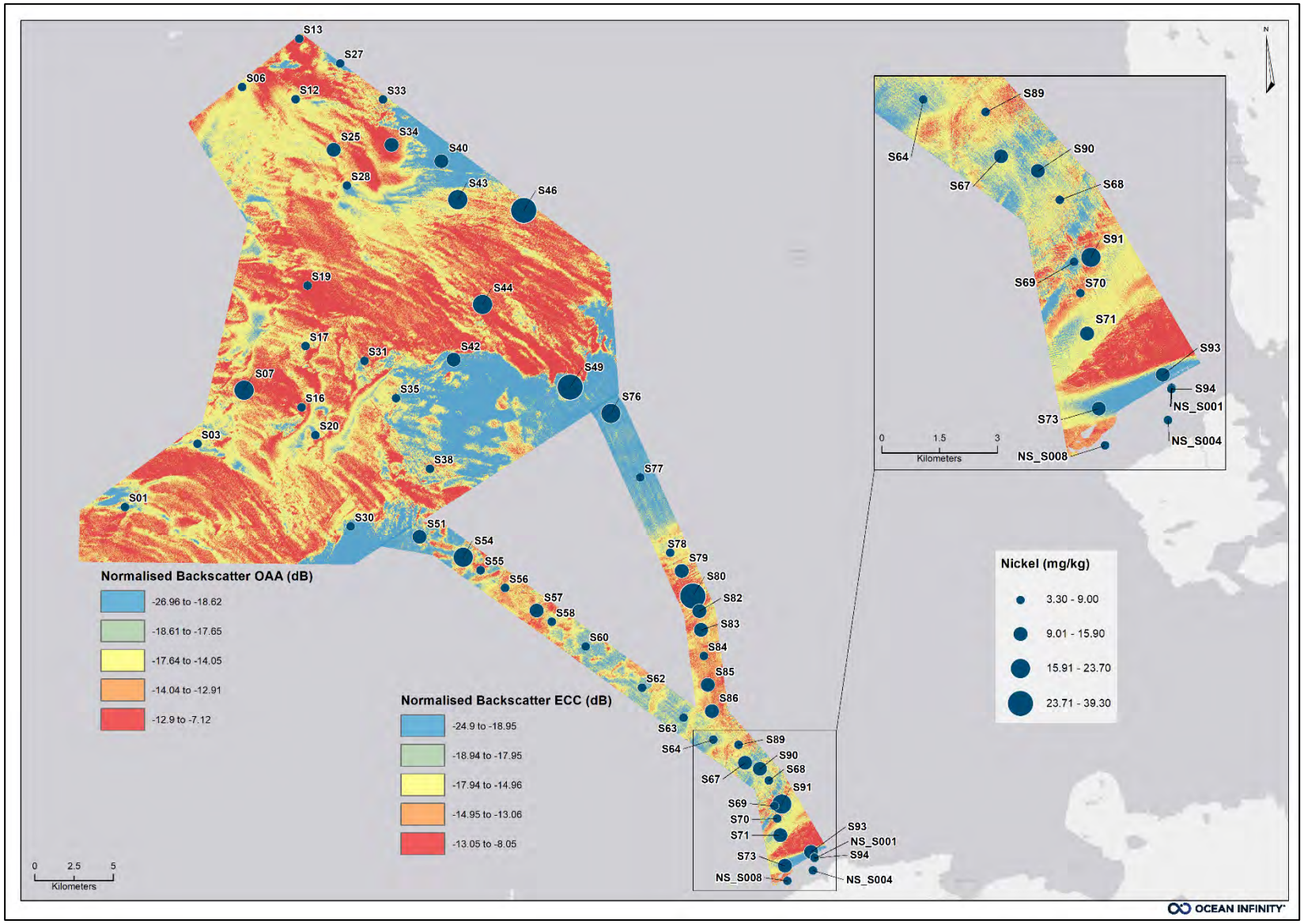


Figure 32 Nickel (Ni) concentrations superimposed on the offshore backscatter data.

5.5.2 Organics

Total Organic Matter (TOM) and Total Organic Carbon (TOC) both varied throughout the survey area, with a mean content of 1.5 % (SD=0.4) and 0.20 % (SD=0.09), respectively (Table 29).

Table 29 Total organic matter and total organic carbon in samples.

Analytes	Total Organic Matter	Total Organic Carbon
Method	Loss On Ignition (LOI)	WSLM59
Limit of Detection	0.2	0.02
Units	**%**	**%**
OAA_S01	1.1	0.19
OAA_S03	1.5	0.25
OAA_S06	1.7	0.29
OAA_S07	1.0	0.21
OAA_S12	1.5	0.25
OAA_S13	1.8	0.29
OAA_S16	1.6	0.25
OAA_S17	1.8	0.21
OAA_S19	1.2	0.19
OAA_S20	1.6	0.16
OAA_S25	1.4	0.21
OAA_S27	0.8	0.06
OAA_S28	1.6	0.14
OAA_S30	1.2	0.12
OAA_S31	1.7	0.17
OAA_S33	0.9	0.08
OAA_S34	1.6	0.15
OAA_S35	1.7	0.14
OAA_S38	1.4	0.10
OAA_S40	1.2	0.09
OAA_S42	2.0	0.12
OAA_S43	1.2	0.11
OAA_S44	1.0	0.13
OAA_S46	1.1	0.11
OAA_S49	1.1	0.12
ECW_S51	1.4	0.14
ECW_S54	1.7	0.55

Analytes	Total Organic Matter	Total Organic Carbon
Method	Loss On Ignition (LOI)	WSLM59
Limit of Detection	0.2	0.02
Units	**%**	**%**
ECW_S55	1.2	0.35
ECW_S56	1.7	0.56
ECW_S57	1.6	0.17
ECW_S58	1.9	0.24
ECW_S60	1.7	0.29
ECW_S62	1.7	0.29
ECW_S63	1.9	0.29
ECW_S64	1.7	0.22
ECW_S67	1.5	0.20
ECW_S68	1.2	0.14
ECW_S69	0.9	0.11
ECW_S70	1.4	0.16
ECW_S71	1.8	0.20
ECW_S73	2.3	0.20
ECE_S76	2.4	0.32
ECE_S77	2.1	0.31
ECE_S78	1.0	0.23
ECE_S79	1.5	0.22
ECE_S80	1.2	0.10
ECE_S82	1.5	0.21
ECE_S83	1.5	0.23
ECE_S84	1.2	0.11
ECE_S85	1.2	0.14
ECE_S86	1.6	0.20
ECE_S89	1.6	0.21
ECE_S90	1.4	0.19
ECE_S91	2.2	0.25
ECE_S93	1.9	0.18
ECE_S94	1.4	0.16
NS_S001	1.5	0.16

Analytes	Total Organic Matter	Total Organic Carbon
Method	Loss On Ignition (LOI)	WSLM59
Limit of Detection	0.2	0.02
Units	**%**	**%**
NS_S004	1.5	0.23
NS_S008	0.9	0.13
Mean	**1.5**	**0.20**
SD	**0.4**	**0.09**
Min	**0.8**	**0.06**
Max	**2.4**	**0.56**
Median	**1.5**	**0.19**

5.5.3 Hydrocarbons

Total Hydrocarbon Content (THC)

Total Hydrocarbon Content (THC) concentrations were low but variable throughout the survey area and did not exceed the Dutch RIVM intervention values at any of the grab sample sites (Table 30). The THC concentration from the sample from site ECE_S89 was notably higher compared to the rest of the sites in the Offshore area, with generally higher concentration found in the samples from the Nearshore area.

Polycyclic Aromatic Hydrocarbon (PAH)

Polycyclic Aromatic Hydrocarbon (PAH) concentrations were variable throughout the survey area, with threshold values being exceeded at five (5) sites (Table 30; Figure 33). The PAH concentrations were notably higher in the samples from the Nearshore area, being most evident in the sum of the 16 EPA PAHs (ΣEPA 16 PAH).

The lower threshold of NEA's class 2 – Good was exceeded for three (3) individual congeners and EPA 16 PAH in the sample from site ECW_S68.

The sample from site ECE_S89 exceeded the lower threshold of NEA's class 2 – Good for eight (8) individual congener and ΣEPA 16 PAH, as well as the lower threshold of NEA's class 3 – Moderate for one (1) individual congener.

The lower threshold of NEA's class 2 – Good was exceeded for 11 individual congeners and ΣEPA 16 PAH in the sample from site NS_S001, as well as one (1) individual congener exceeding the lower threshold of NEA's class 3 – Moderate and the CCME ISQG threshold, respectively.

The sample form site NS_S004 exceeded the lower threshold of NEA's class 2 – Good for nine (9) individual congeners and ΣEPA 16 PAH, as well as the CCME ISQG threshold for two (2) individual congeners and the lower threshold of NEA's class 3 – Moderate for one (1) individual congener.

The highest concentration of ΣEPA 16 PAH was recorded in the sample from site NS_S008 which exceeded the lower threshold of NEA's class 2 – Good, which was also exceeded for nine (9) individual congeners, as well as exceeding the CCME ISQG threshold for two (2) individual congeners and the lower threshold of NEA's class 3 – Moderate for one (1) individual congener.

Table 30 PAH and THC concentrations (µg/kg dry weight) in samples with threshold values. Highlighted cells indicate where threshold values have been exceeded.

Analytes	Acenaphthene	Acenaphthylene	Anthracene	Benzo[a]anthracene	Benzo[a]pyrene	Benzo[b] fluoranthene	Benzo[ghi]perylene	Benzo[e]pyrene*	Benzo[k] fluoranthene	Chrysene	Dibenzo[a,h] anthracene	Fluoranthene	Fluorene	Indeno[123, cd]pyrene	Naphthalene	Perylene *	Phenanthrene	Pyrene	ΣEPA 16 PAH	THC
Limit of Detection	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	-	1000
NEA 1 Background	0	0	0	0	0	0	0	-	0	0	0	0	0	0	0	-	0	0	0	
NEA2 Good	2.4	1.6	1.2	3.6	6	90	18	-	90	4.4	12	8	6.8	20	2	-	6.8	5.2	30	-
NEA 3 Moderate	96	33	4.8	60	183	-	-	-	-	-	27	-	150	-	27	-	780	84	2000	-
NEA 4 Poor	195	85	30	501	230	140	84	-	135	280	273	400	694	63	1754	-	2500	840	6000	-
NEA 5 Very Poor	19500	8500	295	50100	13100	10600	1400	-	7400	2800	2730	2000	34700	2300	8769	-	25000	8400	20000	-
OSPAR ERL	-	-	85	-	430	-	85	-	-	384	-	600	-	240	160	-	240	665	-	-
CEFAS AL1	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	-	-
CCME ISQG	6.71	5.87	46.9	74.8	88.8	-	-	-	-	108	6.22	113	21.2	-	34.6	-	86.7	153	-	-
CCME PEL	88.9	128	245	693	763	-	-	-	-	846	135	1494	144	-	391	-	544	1398	-	-
Dutch RIVM	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5000000
Unit	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**
OAA_S01	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S03	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1360
OAA_S06	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	3040
OAA_S07	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	13600
OAA_S09	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S12	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1180
OAA_S13	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	5410
OAA_S16	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S17	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	3270
OAA_S19	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S20	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S25	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1090
OAA_S27	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1050
OAA_S28	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1010
OAA_S30	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1240
OAA_S31	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S33	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S34	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	7040
OAA_S35	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	2740
OAA_S38	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000

Analytes	Acenaphthene	Acenaphthylene	Anthracene	Benzo[a]anthracene	Benzo[a]pyrene	Benzo[b] fluoranthene	Benzo[ghi]perylene	Benzo[e]pyrene*	Benzo[k] fluoranthene	Chrysene	Dibenzo[a,h] anthracene	Fluoranthene	Fluorene	Indeno[123, cd]pyrene	Naphthalene	Perylene *	Phenanthrene	Pyrene	ΣEPA 16 PAH	THC
Limit of Detection	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	-	1000
NEA 1 Background	0	0	0	0	0	0	0	-	0	0	0	0	0	0	0	-	0	0	0	
NEA2 Good	2.4	1.6	1.2	3.6	6	90	18	-	90	4.4	12	8	6.8	20	2	-	6.8	5.2	30	-
NEA 3 Moderate	96	33	4.8	60	183	-	-	-	-	-	27	-	150	-	27	-	780	84	2000	-
NEA 4 Poor	195	85	30	501	230	140	84	-	135	280	273	400	694	63	1754	-	2500	840	6000	-
NEA 5 Very Poor	19500	8500	295	50100	13100	10600	1400	-	7400	2800	2730	2000	34700	2300	8769	-	25000	8400	20000	-
OSPAR ERL	-	-	85	-	430	-	85	-	-	384	-	600	-	240	160	-	240	665	-	-
CEFAS AL1	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	-	-
CCME ISQG	6.71	5.87	46.9	74.8	88.8	-	-	-	-	108	6.22	113	21.2	-	34.6	-	86.7	153	-	-
CCME PEL	88.9	128	245	693	763	-	-	-	-	846	135	1494	144	-	391	-	544	1398	-	-
Dutch RIVM	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5000000
Unit	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**
OAA_S40	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S42	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S43	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
OAA_S44	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1010
OAA_S46	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	5280
OAA_S49	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
ECW_S51	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
ECW_S54	<1	<1	<1	<1	<1	1.32	<1	1.34	<1	<1	<1	<1	<1	1.69	1.35	<1	1.24	<1	5.60	3270
ECW_S55	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	1.04	<1	<1	<1	<1	1.04	<1000
ECW_S56	<1	<1	<1	<1	<1	1.35	<1	1.10	<1	<1	<1	<1	<1	1.42	<1	<1	<1	<1	2.77	<1000
ECW_S57	<1	<1	<1	<1	<1	1.42	1.03	1.53	1.03	<1	<1	<1	<1	1.88	1.33	<1	1.17	<1	7.86	<1000
ECW_S58	<1	<1	<1	<1	<1	2.00	1.21	1.44	1.13	1.06	<1	<1	<1	1.59	<1	<1	1.58	<1	8.57	4280
ECW_S60	<1	<1	<1	<1	<1	2.32	1.47	2.37	1.63	1.11	<1	1.02	<1	3.05	1.06	<1	1.78	<1	13.44	<1000
ECW_S62	<1	<1	<1	<1	<1	1.27	<1	1.41	1.02	<1	<1	<1	<1	1.64	1.29	<1	<1	<1	5.22	<1000
ECW_S63	<1	<1	<1	<1	<1	1.87	1.14	1.62	1.57	<1	<1	<1	<1	2.07	<1	<1	<1	<1	6.65	1030
ECW_S64	<1	<1	<1	<1	<1	1.96	1.23	1.52	1.61	<1	<1	<1	<1	2.08	<1	<1	<1	<1	6.88	1120
ECW_S67	<1	<1	<1	<1	<1	1.95	1.15	1.15	1.34	<1	<1	<1	<1	1.45	<1	<1	<1	<1	5.89	<1000
ECW_S68	<1	<1	<1	4.68	5.95	6.03	4.12	4.54	5.13	5.45	<1	6.51	<1	4.59	<1	1.51	1.26	6.30	50.02	1490
ECW_S69	<1	<1	<1	<1	<1	1.05	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	1.05	1500
ECW_S70	<1	<1	<1	<1	<1	1.72	<1	1.18	1.16	<1	<1	<1	<1	1.01	<1	<1	<1	<1	3.89	1750
ECW_S71	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000

Analytes	Acenaphthene	Acenaphthylene	Anthracene	Benzo[a]anthracene	Benzo[a]pyrene	Benzo[b] fluoranthene	Benzo[ghi]perylene	Benzo[e]pyrene*	Benzo[k] fluoranthene	Chrysene	Dibenzo[a,h] anthracene	Fluoranthene	Fluorene	Indeno[123, cd]pyrene	Naphthalene	Perylene *	Phenanthrene	Pyrene	ΣEPA 16 PAH	THC
Limit of Detection	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	-	1000
NEA 1 Background	0	0	0	0	0	0	0	-	0	0	0	0	0	0	0	-	0	0	0	
NEA2 Good	2.4	1.6	1.2	3.6	6	90	18	-	90	4.4	12	8	6.8	20	2	-	6.8	5.2	30	-
NEA 3 Moderate	96	33	4.8	60	183	-	-	-	-	-	27	-	150	-	27	-	780	84	2000	-
NEA 4 Poor	195	85	30	501	230	140	84	-	135	280	273	400	694	63	1754	-	2500	840	6000	-
NEA 5 Very Poor	19500	8500	295	50100	13100	10600	1400	-	7400	2800	2730	2000	34700	2300	8769	-	25000	8400	20000	-
OSPAR ERL	-	-	85	-	430	-	85	-	-	384	-	600	-	240	160	-	240	665	-	-
CEFAS AL1	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	-	-
CCME ISQG	6.71	5.87	46.9	74.8	88.8	-	-	-	-	108	6.22	113	21.2	-	34.6	-	86.7	153	-	-
CCME PEL	88.9	128	245	693	763	-	-	-	-	846	135	1494	144	-	391	-	544	1398	-	-
Dutch RIVM	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5000000
Unit	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**
ECW_S73	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	2820
ECE_S76	<1	<1	<1	<1	<1	2.80	3.44	2.05	2.12	1.22	<1	1.35	<1	3.94	<1	<1	1.83	<1	16.70	1310
ECE_S77	<1	<1	<1	<1	<1	2.32	2.35	1.40	1.38	<1	<1	<1	<1	2.82	1.35	<1	1.33	<1	11.55	1650
ECE_S78	<1	<1	<1	<1	<1	1.40	1.46	<1	1.22	<1	<1	<1	<1	1.76	<1	<1	<1	<1	5.84	<1000
ECE_S79	<1	<1	<1	<1	<1	1.33	<1	1.09	<1	<1	<1	<1	<1	1.31	<1	<1	<1	<1	2.64	<1000
ECE_S80	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
ECE_S82	<1	<1	<1	<1	<1	1.54	1.19	1.25	<1	1.23	<1	<1	<1	1.35	<1	<1	1.71	<1	7.02	2420
ECE_S83	<1	<1	<1	<1	<1	1.50	1.30	1.13	<1	<1	<1	<1	<1	1.58	<1	<1	1.40	<1	5.78	<1000
ECE_S84	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	1.13	<1	<1	<1	<1	1.13	<1000
ECE_S85	<1	<1	<1	<1	<1	1.08	<1	<1	<1	<1	<1	<1	<1	1.09	<1	<1	<1	<1	2.17	<1000
ECE_S86	<1	<1	<1	<1	<1	1.78	1.41	1.18	<1	<1	<1	<1	<1	1.90	<1	<1	<1	<1	5.09	<1000
ECE_S89	3.38	<1	9.19	14.0	13.1	11.2	7.35	8.14	10.8	13.8	1.69	30.4	3.07	8.61	5.13	3.11	28.6	23.8	184.12	27600
ECE_S90	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	<1000
ECE_S91	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	<1	0.00	1750
ECE_S93	<1	<1	<1	<1	<1	1.66	<1	1.27	1.44	1.58	<1	<1	<1	<1	<1	<1	<1	<1	4.68	17900
ECE_S94	<1	<1	<1	<1	<1	2.42	<1	1.58	1.60	1.62	<1	1.02	<1	1.03	<1	<1	<1	<1	7.69	11500
NS_S001	2.49	5.66	6.93	18.5	29.0	33.3	35.2	20.2	33.1	21.9	6.44	32.7	3.25	40.0	7.83	12.5	16.9	27.3	320.50	26600
NS_S004	1.89	7.54	6.03	19.5	33.2	37.3	37.8	21.2	33.9	23.3	7.17	28.2	3.02	42.8	5.38	12.0	15.2	23.4	325.63	19900
NS_S008	3.20	8.40	11.6	36.2	46.9	51.7	53.2	30.2	51.6	40.4	10.50	57.6	4.62	59.9	10.8	17.6	25.2	48.2	520.02	3230

**Not included in the EPA 16 PAHs.*

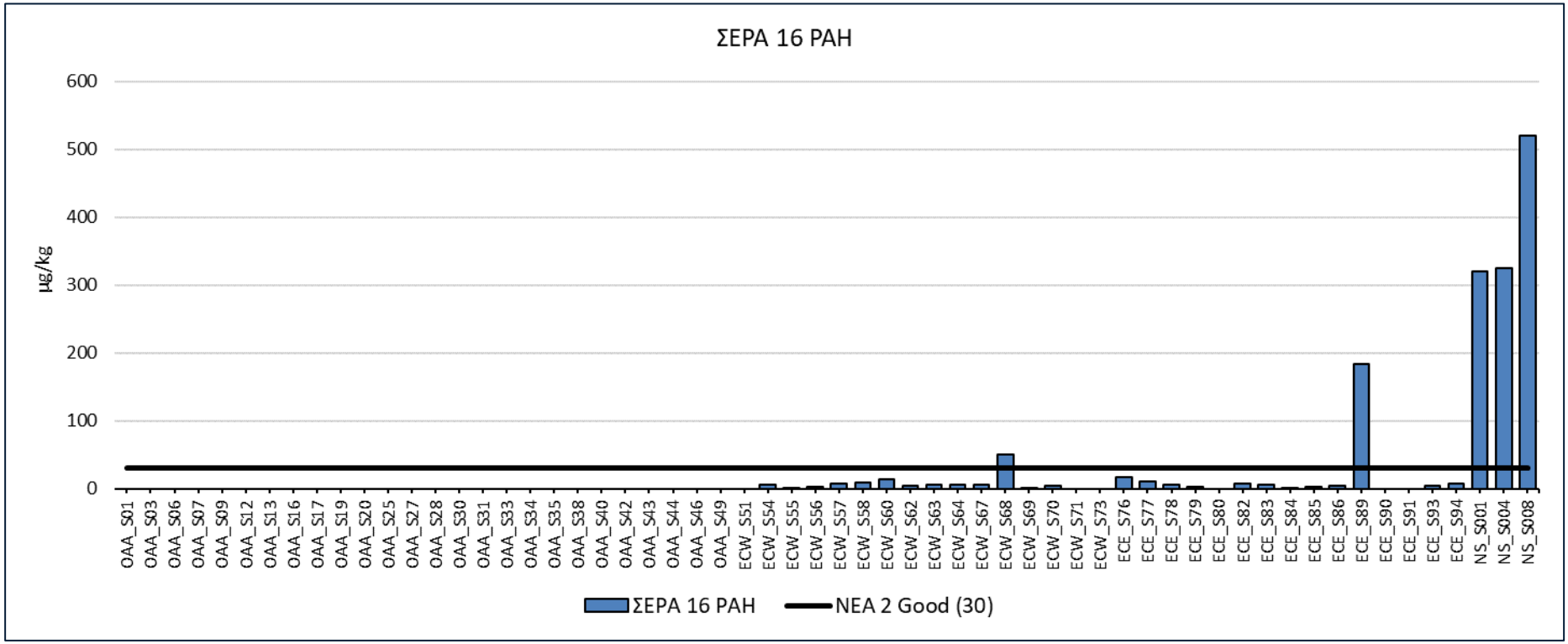


Figure 33 Levels of EPA 16 PAHs summarized (µg/kg) in samples with threshold values.

5.5.4 Polychlorinated Biphenyls (PCB)

Concentrations of Polychlorinated Biphenyls (PCB) were overall low throughout the survey area and exceeded the limit of detection at seven (7) of the 60 sampled sites (Table 31), all of which exceeded multiple threshold values. The lower threshold of NEA's class 2 – Good for the sum of the seven ICES PCBs (ΣPCB7) was exceeded by default for these sites, being set at 0.00 µg/kg.

5.5.5 Organotins (MBT, DBT & TBT)

None of the 60 acquired samples exceeded the limit of detection for any of the analytes (MBT, DBT & TBT).

5.5.6 Pesticides (OCP)

Concentrations of Organochlorine Pesticides (OCP) was low throughout the survey area and exceeded the limit of detection at three (3) of the 29 sampled sites (Table 32), with no threshold values being exceeded.

5.5.7 Brominated Flame Retardants (PBDE)

Concentrations of Brominated Flame Retardants (PBDE) were overall low throughout the survey area (Table 33). Out of the twelve analysed congeners, congener BDE209 had notably higher concentrations.

The lower threshold of NEA's class 2 – Good, as well as the OSPAR BAC threshold values, were exceeded for one or multiple congeners at 18 of the 29 sampled sites, corresponding to all sites with levels above the limit of detection.

Table 31 PCB concentrations (µg/kg dry weight) in samples exceeding LoDs, with threshold values. Highlighted cells indicate where threshold values have been exceeded.

Analytes	PCB 18	PCB 28	PCB 31	PCB 44	PCB 47	PCB 49	PCB 52	PCB 66	PCB 101	PCB 105	PCB 110	PCB 118	PCB 128	PCB 138	PCB 141	PCB 149	PCB 151	PCB 153	PCB 156	PCB 158	PCB 170	PCB 180	PCB 183	PCB 187	PCB 194	ΣPCB7	ΣPCB25
Limit of Detection	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	-	-
NEA 1 Background	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
NEA 2 Good	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0	-
NEA 3 Moderate	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4.1	-
NEA 4 Poor	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	43	-
NEA 5 Very Poor	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	430	-
OSPAR BAC	-	0.22	-	-	-	-	0.12	-	0.14	-	-	0.17	-	0.15	-	-	-	0.19	-	-	-	0.1	-	-	-	-	-
OSPAR EAC	-	1.7	-	-	-	-	2.7	-	3	-	-	0.6	-	7.9	-	-	-	40	-	-	-	12	-	-	-	-	-
CEFAS AL1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	10	20
CEFAS AL2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	200
CCME ISQG	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	21.5
CCME PEL	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	189
Unit	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**
OAA_S06	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	0.12	0.10	0.34	0.20	0.28	0.39	0.23	0.29	0.21	0.16	0.22	0.30	0.25	0.31	0.25	0.27	0.20	0.27	1.08	4.39
OAA_S28	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	0.14	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	0.14	0.14
OAA_S33	<0.08	<0.08	<0.08	0.17	<0.08	0.11	0.27	0.12	0.21	<0.08	0.23	0.09	<0.08	<0.08	<0.08	0.12	<0.08	0.09	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	0.66	1.41
OAA_S34	<0.08	0.09	0.11	0.14	0.13	0.12	0.14	0.16	0.17	0.13	0.17	0.18	0.10	0.13	0.21	0.16	0.15	0.18	0.13	0.16	0.14	0.16	0.16	0.14	0.15	1.05	3.51
ECW_S68	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	0.09	0.14	<0.08	0.13	0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	0.22	0.44
ECW_S69	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	<0.08	0.09	0.09	0.14	0.12	0.15	0.12	0.13	0.09	0.12	0.13	0.15	0.16	0.13	0.13	0.16	0.11	0.15	0.14	0.68	2.31
NS_S004	0.15	0.15	0.25	0.29	0.22	0.19	0.17	0.25	0.15	0.17	0.25	0.17	0.16	<0.08	0.14	0.18	0.12	0.12	<0.08	<0.08	0.09	0.08	<0.08	<0.08	0.17	0.84	3.47

Table 32 OCP concentrations (µg/kg dry weight) in grab samples exceeding LoDs, with threshold values.

Analytes	alpha-Hexachlorocyclohexane	beta-Hexachlorocyclohexane	gamma-Hexachlorocyclohexane	Dieldrin	Hexachlorobenzene	p,p' -Dichlorodiphenyl dichloroethane	p,p' -Dichloro diphenyldichloroethylene	p,p' -Dichloro diphenyltrichloroethane
Limit of Detection	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
NEA 1 Background	-	-	-	-	-	-	-	-
NEA 2 Good	-	-	-	-	-	-	-	0
NEA 3 Moderate		-	-	-	-	-	-	16
NEA 4 Poor	-	-	-	-	-	-	-	165
NEA 5 Very Poor	-	-	-	-	-	-	-	1647
OSPAR ERL	-	-	3	2	20	-	2.2	-
CEFAS AL1	-	-	-	5	-	-	-	1
CEFAS AL2	-	-	-	-	-	-	-	-
CCME ISQG	-	-	-	-	-	1.22	2.07	1.19
CCME PEL	-	-	-	-	-	7.81	374	4.77
Unit	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**
OAA_S27	<0.1	<0.1	<0.1	0.1	<0.1	<0.1	<0.1	<0.1
ECW_S71	<0.1	<0.1	<0.1	0.2	<0.1	<0.1	<0.1	<0.1
NS_S004	<0.1	<0.1	<0.1	0.4	<0.1	0.3	0.2	<0.1

Table 33 PBDE concentrations (µg/Kg dry weight) in grab samples, with threshold values. Highlighted cells indicate where threshold values have been exceeded.

Analytes	BDE17	BDE28	BDE47	BDE66	BDE85	BDE99	BDE100	BDE138	BDE153	BDE154	BDE183	BDE209	ΣPBDE
Limit of Detection	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.1	-
NEA 1 Background	-	-	-	-	-	-	-	-	-	-	-	-	-
NEA 2 Good	-	-	-	-	-	-	-	-	-	-	-	-	0
NEA 3 Moderate	-	-	-	-	-	-	-	-	-	-	-	-	62
NEA 4 Poor	-	-	-	-	-	-	-	-	-	-	-	-	79
NEA 5 Very Poor	-	-	-	-	-	-	-	-	-	-	-	-	1580
OSPAR BAC	-	0.05	0.05	0.05	0.05	0.05	0.05	-	0.05	0.05	0.05	0.05	-
Unit	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**	**µg/kg**
OAA_S01	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
OAA_S17	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
OAA_S27	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
OAA_S28	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.15	0.15
OAA_S30	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.32	0.32
OAA_S38	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.18	0.18
OAA_S40	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
OAA_S49	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.19	0.19
ECW_S51	<0.05	<0.05	<0.05	0.06	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	2.30	2.36
ECW_S54	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
ECW_S55	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.19	0.19
ECW_S56	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.55	0.55
ECW_S62	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	1.20	1.20
ECW_S64	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.42	0.42
ECW_S67	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.11	0.11
ECW_S70	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.33	0.33
ECW_S71	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.54	0.54
ECW_S73	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.08	8.75	8.83
ECE_S76	<0.05	<0.05	0.05	0.08	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	1.91	2.04
ECE_S77	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
ECE_S78	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
ECE_S82	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	1.40	1.40
ECE_S84	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
ECE_S89	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.18	0.18
ECE_S90	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
ECE_S93	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.1	0.00
NS_S001	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.47	0.47
NS_S004	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	0.53	0.53
NS_S008	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.2	0.00

5.6 Statistical Analyses from Grab Samples

5.6.1 Non-Colonial Fauna

The non-colonial epifauna was identified to the lowest taxonomic level possible and the individuals were enumerated. The infauna and non-colonial epifauna were combined and analysed together. When analysing phyletic composition, the following phyla: Chordata, Cnidaria, Hemichordata, Phoronida, Nemertea and Platyhelminthes were combined into the group "Others".

Offshore and nearshore grab sample sites were analysed together in Section 5.6. Grab sample sites OAA_S19, OAA_S24, OAA_S25, OAA_S36, ECW_S54, ECW_S59, ECE_S81, ECE_S87 and NS_S002 comprised insufficient sample volume and were excluded from all statistical analyses.

The colonial epifauna was identified to the lowest taxonomic level possible. The colonial epifauna was recorded as absent/present and analysed separately. The results are presented in 5.6.10.

A full list of species from the grab samples is presented in Appendix E.

5.6.2 Phyletic Composition

The phyletic composition of the non-colonial fauna identified from the grab samples is illustrated in Figure 34, and Figure 35, and summarised in Table 34. Annelida had the highest abundance, followed by Mollusca and Nematoda. These three phyla contributed to 78 % of the recorded individuals. Annelida had the highest diversity, followed by Arthropoda and Mollusca. These three phyla contributed to 94 % of the recorded taxa.

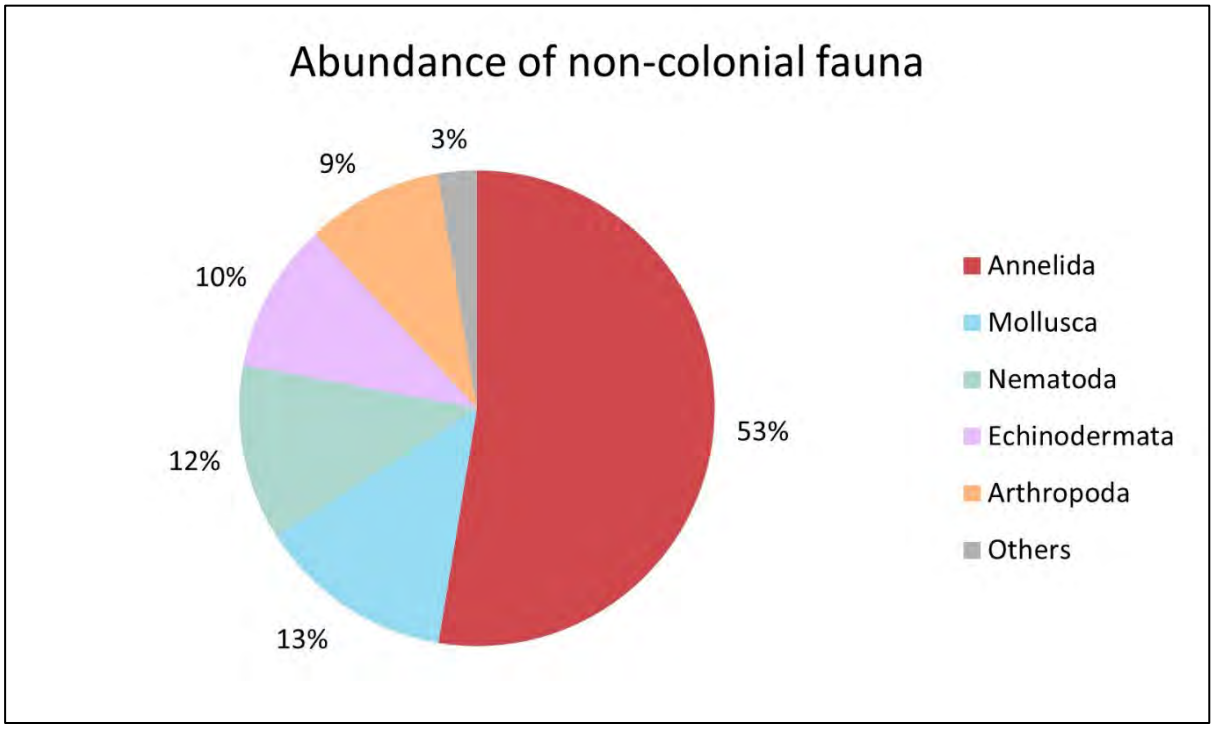


Figure 34 Abundance of non-colonial fauna from grab samples.

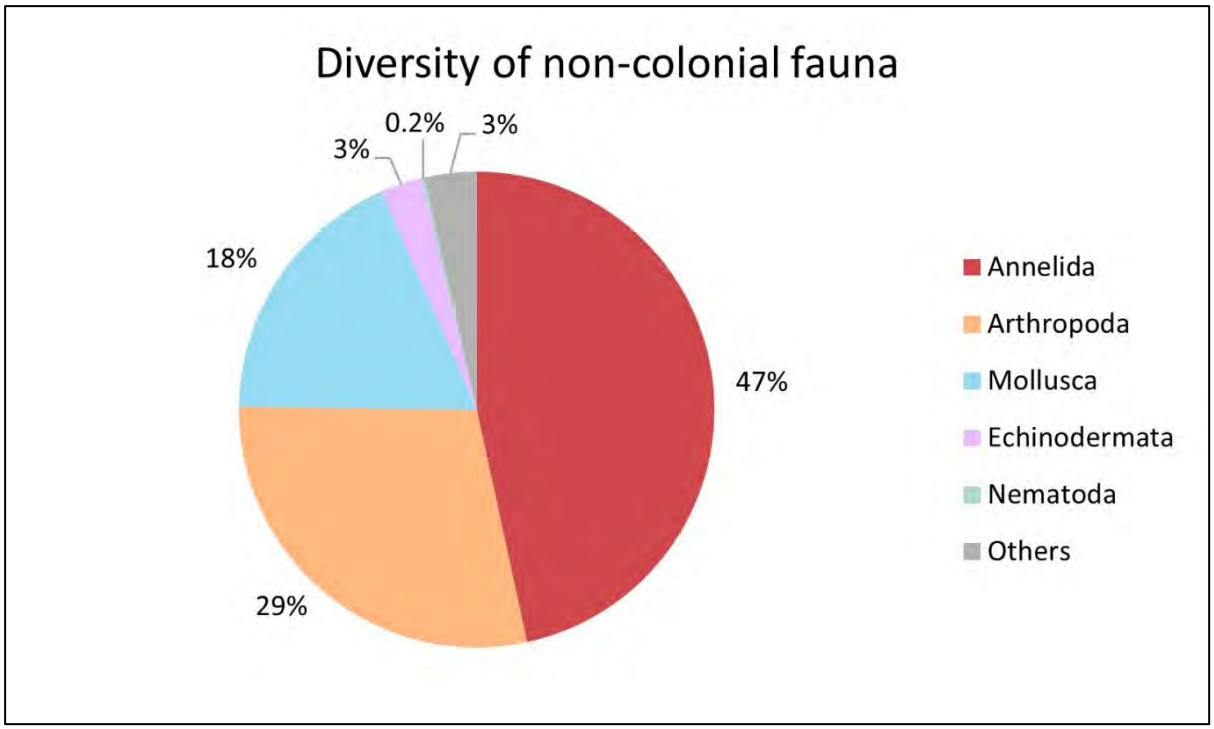


Figure 35 Diversity of non-colonial fauna from grab samples.

Table 34 Phyletic composition of non-colonial fauna from grab samples.

Phylum	Number of Taxa	Abundance (Total Number of Individuals)
Annelida	199	8178
Mollusca	78	2081
Nematoda	1	1856
Echinodermata	12	1584
Arthropoda	122	1439
Others	15	411
Total	**427**	**15 549**

A list of the ten most abundant taxa, with total abundance and frequency of occurrence, is presented in Table 35 and the distribution within the survey area is illustrated in Figure 36. The most abundant taxon is the annelid *Owenia*, with a total of 2332 individuals recorded, and the species occurred in 77 % of the grab samples.

Table 35 The ten most abundant taxa from grab samples, together with the frequency of occurrence.

Phylum	Taxa	Total Abundance	Mean Abundance	SD	Frequency of Occurrence (%)
Annelida	*Owenia*	2332	35.88	109.91	77
Nematoda	Nematoda	1856	28.55	41.19	92
Echinodermata	*Echinocyamus pusillus*	1500	23.08	17.96	95
Annelida	*Pisione remota*	490	7.54	16.28	38
Mollusca	*Asbjornsenia pygmaea*	480	7.38	15.30	68
Annelida	*Polygordius*	449	6.91	16.86	40
Annelida	*Glycera lapidum* (aggregate)	335	5.15	7.16	72
Annelida	*Spiophanes kroyeri*	268	4.12	8.49	52
Mollusca	*Goodallia triangularis*	266	4.09	6.39	55
Nemertea	Nemertea	254	3.91	3.28	83

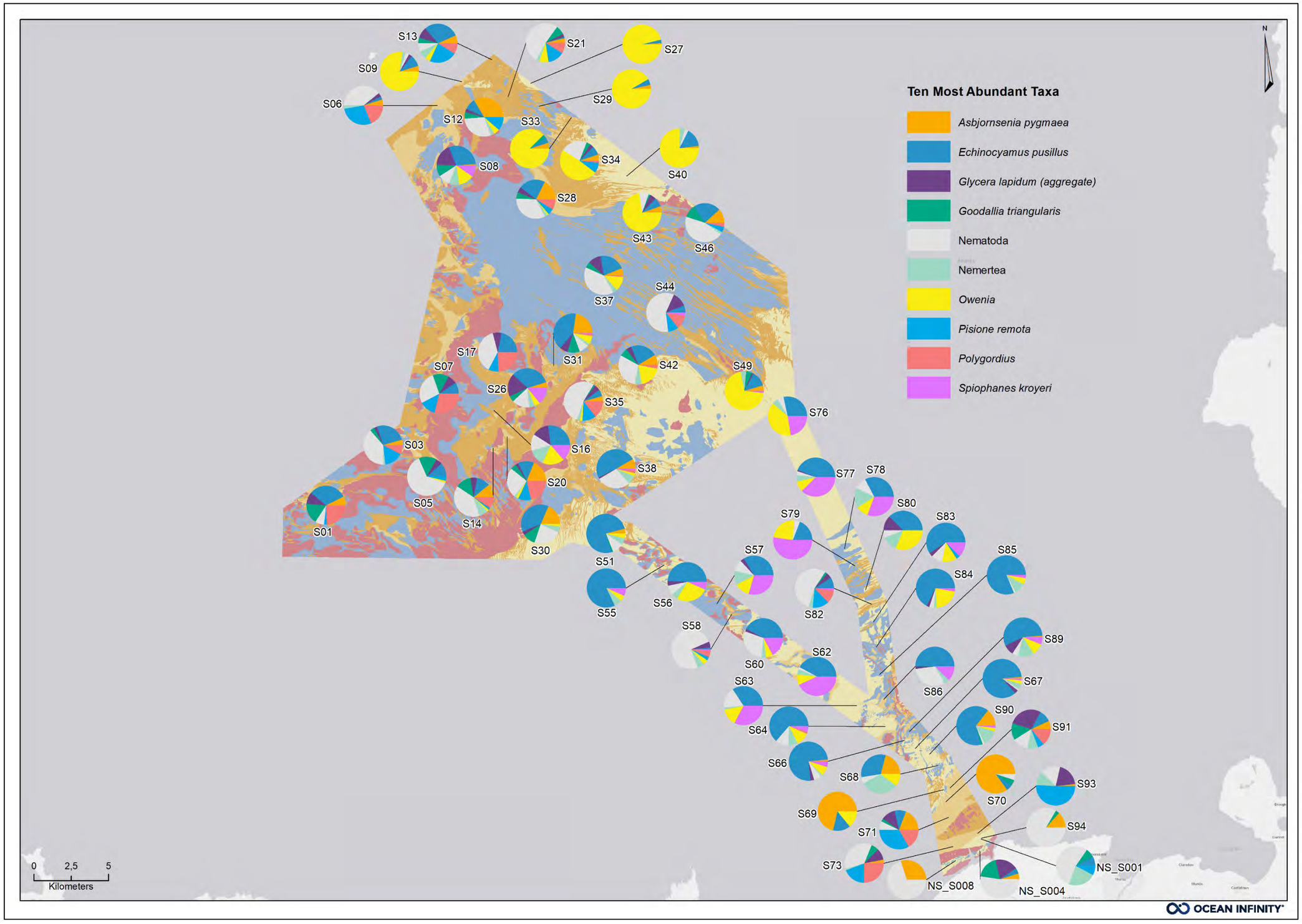


Figure 36 Overview of the ten most abundant taxa from grab samples.

A list of the ten most frequently occurring taxa, with total abundance, is presented in Table 36. The most frequently occurring taxon was the sea urchin *Echinocyamus pusillus*, which occurred in 95 % of the grab samples, with a total abundance of 1500 individuals.

Table 36 The ten most frequently occurring taxa from grab samples, together with total abundance.

Phylum	Taxa	Frequency of Occurrence (%)	Total Abundance
Echinodermata	*Echinocyamus pusillus*	95	1500
Nematoda	Nematoda	92	1856
Nemertea	Nemertea	83	254
Annelida	*Owenia*	77	2332
Annelida	*Glycera lapidum* (aggregate)	72	335
Mollusca	*Asbjornsenia pygmaea*	68	480
Annelida	*Aonides paucibranchiata*	68	183
Mollusca	*Goodallia triangularis*	55	266
Annelida	*Parexogone hebes*	54	131
Annelida	*Spiophanes kroyeri*	52	268

5.6.3 Univariate Statistical Analyses

Univariate analyses were performed to assess the non-colonial faunal richness, diversity, evenness and dominance. The results of the univariate analyses are presented in Table 37.

The number of Taxa (S) per site varied with a mean of 44 (SD= 20) where OAA_S16 contained the highest number of Taxa (99 different taxa) and NS_S001 and NS_S008 the lowest (9 different taxa). An overview of the number of Taxa (S) identified per grab sampling site in the survey area is presented in Figure 37.

The number of individuals (N) per site (expressed per 0.1 m^2) varied with a mean of 239 (SD= 164) where OAA_S27 contained the highest number of individuals (835 individuals) and ECW_S69 the lowest with 25 individuals. An overview of the number of individuals (N) identified per grab sampling site in the survey area is presented in Figure 38.

The species richness measured with Margalef's diversity index (D) varied between 1.80 and 15.87, with OAA_S16 having the highest value of 15.87. Pielou's evenness index (J') ranged from 0.19 to 0.93, with OAA_S26 and ECW_S69 having the highest value of 0.93.

The Shannon-Wiener index (H') ranged from 0.67 to 4.09, with OAA_S16 having the highest value of 4.09.

Simpson's index of dominance (1-λ) varied from 0.20 to 0.98, with OAA_S16 and OAA_S26 having the highest value of 0.98. An overview of the Shannon-Wiener index (H') identified per grab sampling site in the survey area is presented in Figure 39.

Table 37 Univariate indices of species richness, diversity and evenness for fauna in a single grab sample per site.

Sample ID	Number Of Taxa (S)	Number Of Individuals (N)	Margalef's Richness Index (D)	Pielou's Evenness Index (J')	Shannon-Wiener Index (H')	Simpson's Index of Dominance (1-λ)
OAA_S01	27	70	6.12	0.87	2.88	0.93
OAA_S03	40	303	6.83	0.69	2.56	0.86
OAA_S05	54	254	9.57	0.77	3.06	0.90
OAA_S06	25	312	4.18	0.73	2.35	0.86
OAA_S07	51	378	8.42	0.79	3.10	0.93
OAA_S08	77	343	13.02	0.81	3.52	0.94
OAA_S09	33	144	6.44	0.59	2.06	0.68
OAA_S12	31	424	4.96	0.71	2.44	0.86
OAA_S13	44	123	8.94	0.91	3.46	0.97
OAA_S14	28	86	6.06	0.85	2.82	0.92
OAA_S16	99	481	15.87	0.89	4.09	0.98
OAA_S17	38	210	6.92	0.79	2.87	0.91
OAA_S20	30	126	6.00	0.82	2.80	0.92
OAA_S21	42	149	8.19	0.81	3.04	0.91
OAA_S26	51	94	11.01	0.93	3.67	0.98
OAA_S27	36	835	5.20	0.19	0.67	0.20
OAA_S28	34	276	5.87	0.73	2.57	0.88
OAA_S29	29	393	4.69	0.29	0.99	0.33
OAA_S30	34	162	6.49	0.76	2.69	0.89
OAA_S31	61	201	11.31	0.84	3.47	0.94
OAA_S33	26	345	4.28	0.38	1.25	0.44
OAA_S34	61	788	9.00	0.68	2.79	0.86
OAA_S35	56	438	9.04	0.70	2.82	0.86
OAA_S37	42	155	8.13	0.78	2.93	0.91
OAA_S38	40	151	7.77	0.78	2.87	0.89
OAA_S40	55	323	9.35	0.60	2.40	0.75
OAA_S42	44	232	7.89	0.81	3.05	0.93
OAA_S43	30	282	5.14	0.51	1.72	0.61
OAA_S44	52	225	9.42	0.79	3.12	0.91
OAA_S46	69	481	11.01	0.81	3.44	0.95
OAA_S49	26	105	5.37	0.69	2.24	0.79

Sample ID	Number Of Taxa (S)	Number Of Individuals (N)	Margalef's Richness Index (D)	Pielou's Evenness Index (J')	Shannon-Wiener Index (H')	Simpson's Index of Dominance (1-λ)
ECW_S51	41	156	7.92	0.75	2.80	0.86
ECW_S55	47	168	8.98	0.80	3.08	0.90
ECW_S56	57	227	10.32	0.84	3.39	0.94
ECW_S57	90	407	14.81	0.79	3.54	0.95
ECW_S58	54	365	8.98	0.67	2.68	0.82
ECW_S60	92	480	14.74	0.83	3.76	0.96
ECW_S62	72	300	12.45	0.84	3.57	0.95
ECW_S63	57	261	10.06	0.86	3.47	0.96
ECW_S64	36	171	6.81	0.82	2.94	0.91
ECW_S66	31	106	6.43	0.71	2.44	0.79
ECW_S67	39	136	7.74	0.72	2.62	0.81
ECW_S68	43	113	8.88	0.88	3.30	0.95
ECW_S69	13	25	3.73	0.93	2.38	0.93
ECW_S70	25	53	6.04	0.83	2.69	0.89
ECW_S71	22	90	4.67	0.86	2.65	0.92
ECW_S73	31	503	4.82	0.62	2.13	0.82
ECE_S76	57	315	9.73	0.84	3.39	0.95
ECE_S77	71	298	12.29	0.82	3.49	0.94
ECE_S78	58	215	10.61	0.89	3.60	0.97
ECE_S79	30	79	6.64	0.87	2.96	0.94
ECE_S80	48	153	9.34	0.86	3.34	0.95
ECE_S82	79	472	12.67	0.71	3.11	0.89
ECE_S83	44	106	9.22	0.87	3.28	0.94
ECE_S84	39	173	7.37	0.77	2.80	0.88
ECE_S85	41	188	7.64	0.77	2.87	0.88
ECE_S86	38	109	7.89	0.85	3.09	0.93
ECE_S89	51	171	9.72	0.80	3.15	0.91
ECE_S90	50	169	9.55	0.80	3.13	0.92
ECE_S91	65	280	11.36	0.86	3.60	0.96
ECE_S93	11	147	2.00	0.71	1.71	0.77
ECE_S94	16	45	3.94	0.80	2.23	0.86
NS_S001	9	43	2.13	0.67	1.46	0.65

Sample ID	Number Of Taxa (S)	Number Of Individuals (N)	Margalef's Richness Index (D)	Pielou's Evenness Index (J')	Shannon-Wiener Index (H')	Simpson's Index of Dominance (1-λ)
NS_S004	12	50	2.81	0.77	1.91	0.80
NS_S008	9	86	1.80	0.62	1.35	0.64
Mean	**44**	**239**	**7.95**	**0.76**	**2.79**	**0.86**
SD	**20**	**164**	**3.10**	**0.14**	**0.69**	**0.14**
Min	**9**	**25**	**1.80**	**0.19**	**0.67**	**0.20**
Max	**99**	**835**	**15.87**	**0.93**	**4.09**	**0.98**
Median	**41**	**188**	**7.89**	**0.79**	**2.87**	**0.91**

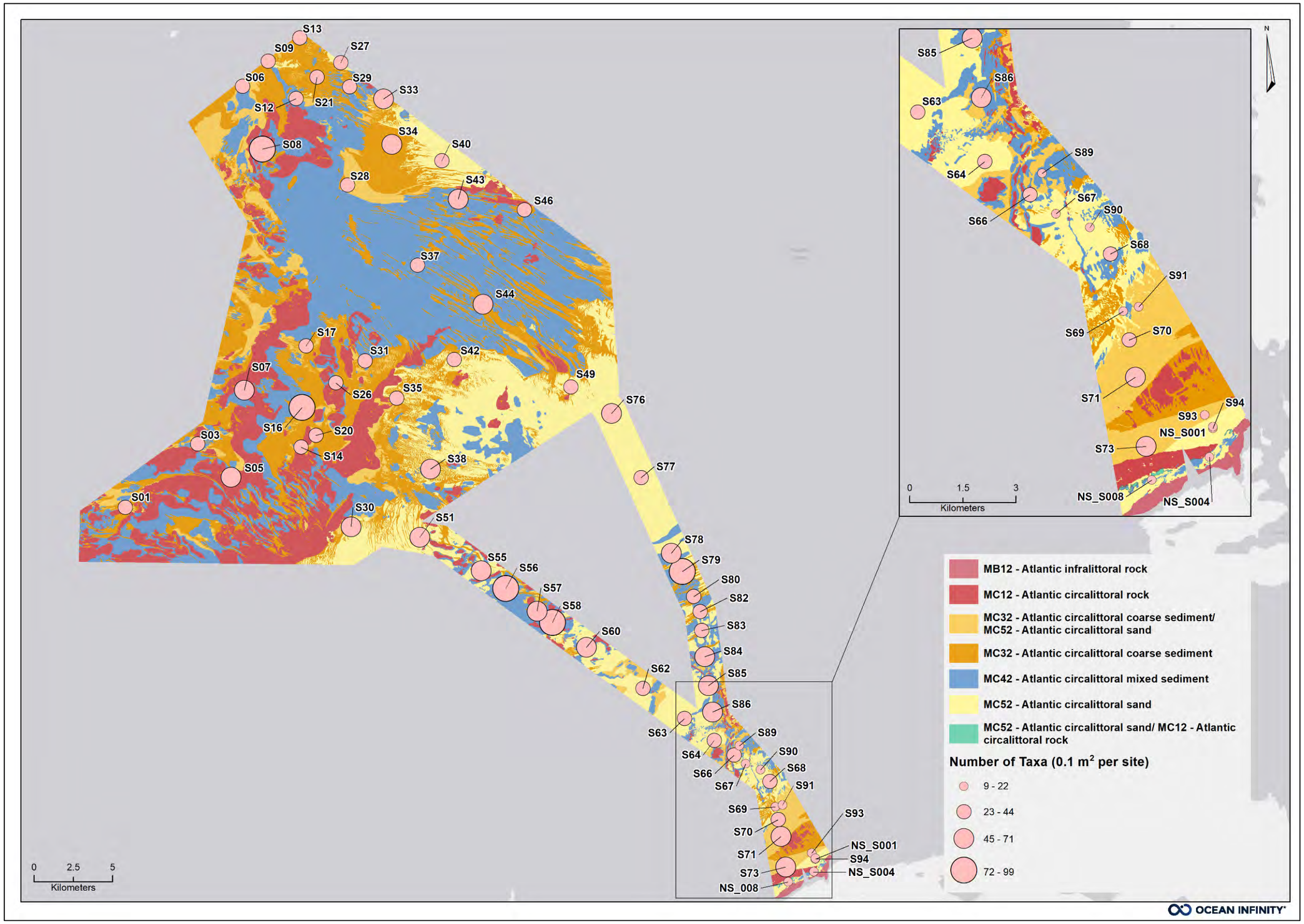


Figure 37 Overview of the Number of Taxa (S) per grab sample site.

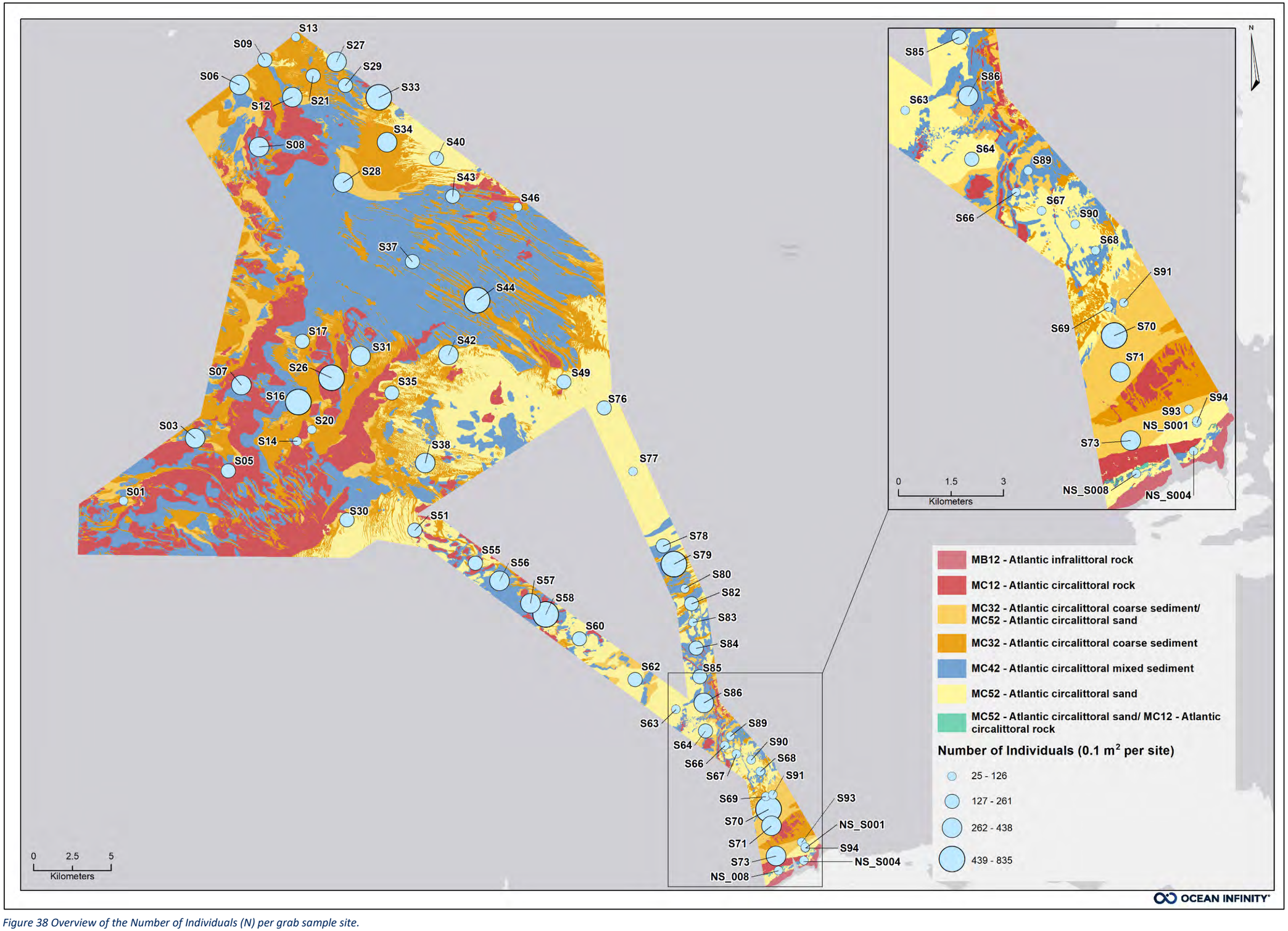


Figure 38 Overview of the Number of Individuals (N) per grab sample site.

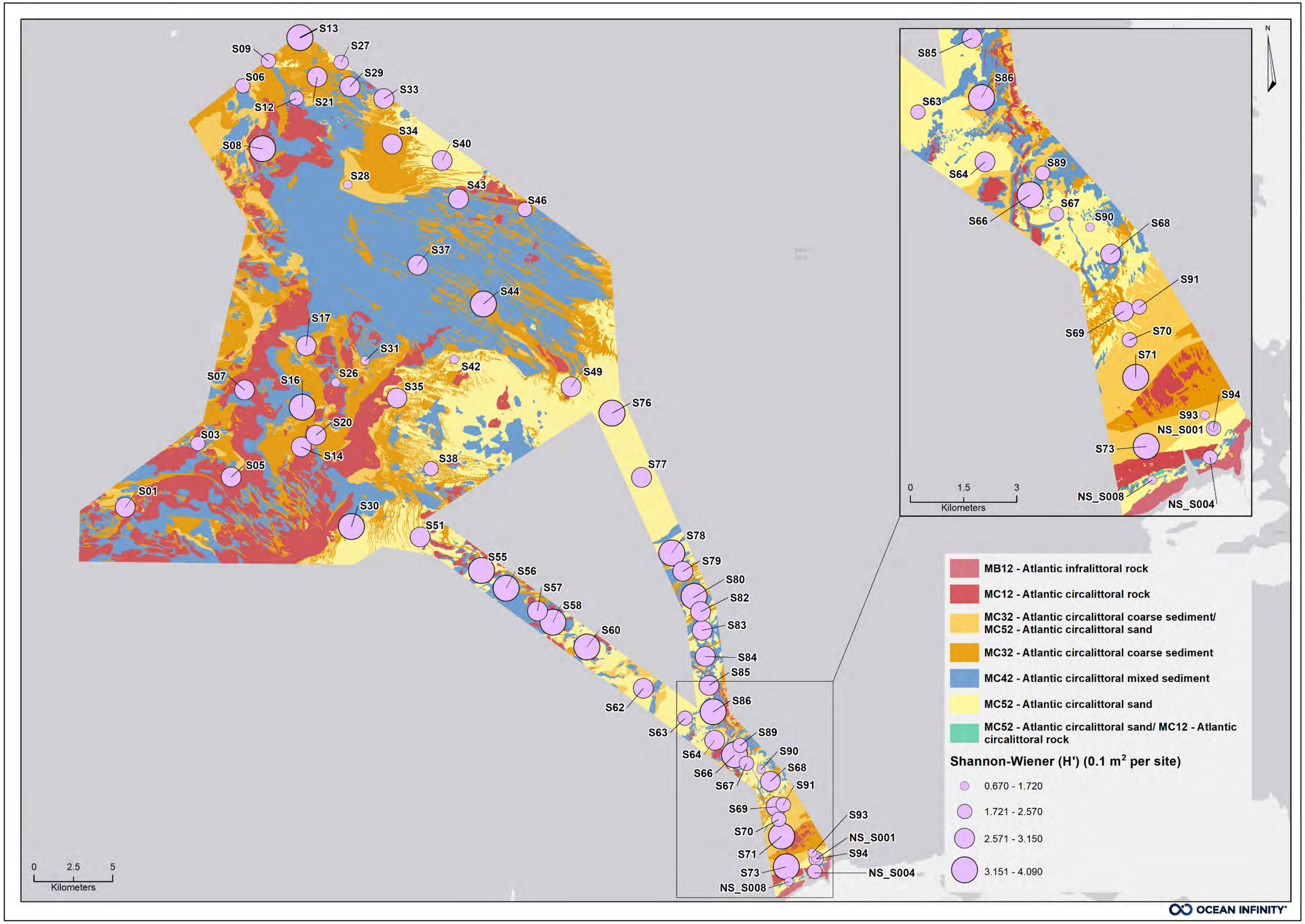


Figure 39 Overview of the Shannon-Wiener Index (H') per grab sample site.

5.6.4 Multivariate Statistical Analyses

Square root transformation was applied to the dataset before calculating the Bray-Curtis similarity measures in the SIMPROF and SIMPER analyses. The transformation was applied to prevent abundant species from influencing the Bray-Curtis similarity index measures excessively and to take the rarer species into account (Clarke & Gorley, PRIMER v7: User Manual/Tutorial. Plymouth: PRIMER-E., 2015). The statistical analyses were based on macrofaunal data derived from the taxonomic analyses of the grab samples.

5.6.5 SIMPROF Cluster Analyses

The SIMPROF analyses of the non-colonial faunal composition produced 21 statistically distinct groups (black lines) and is presented in a hierarchical dendrogram in Figure 40.

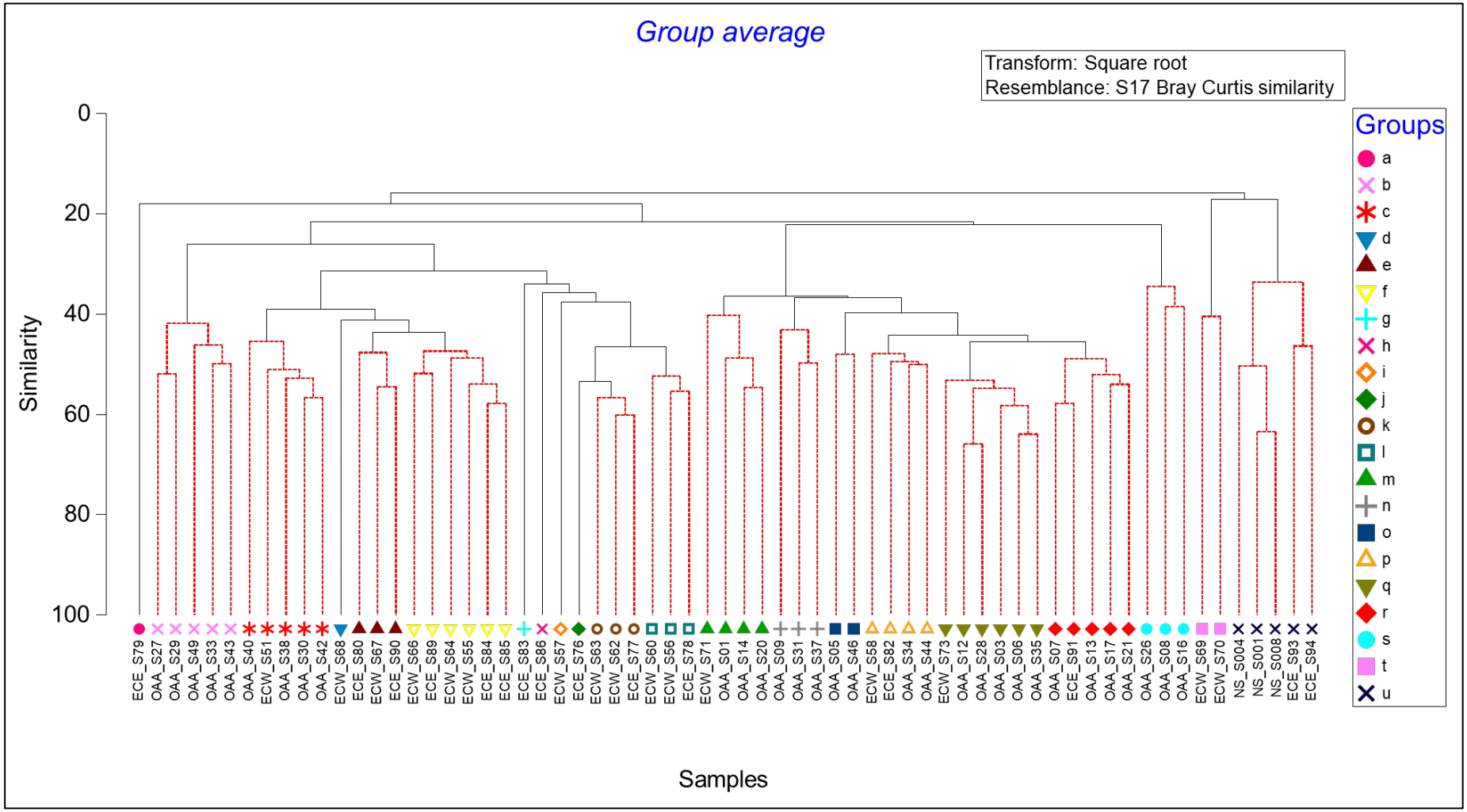


Figure 40 SIMPROF dendrogram of non-colonial faunal composition from grab sample sites.

5.6.6 Non-Metric Multi-Dimensional Scaling (MDS)

The nMDS-plot reflects the dendrogram (Figure 40) and displays the similarity between grab sample sites at 20 % to highlight homogeneous species composition. Sample similarity is further explored in the nMDS-plot in Figure 41.

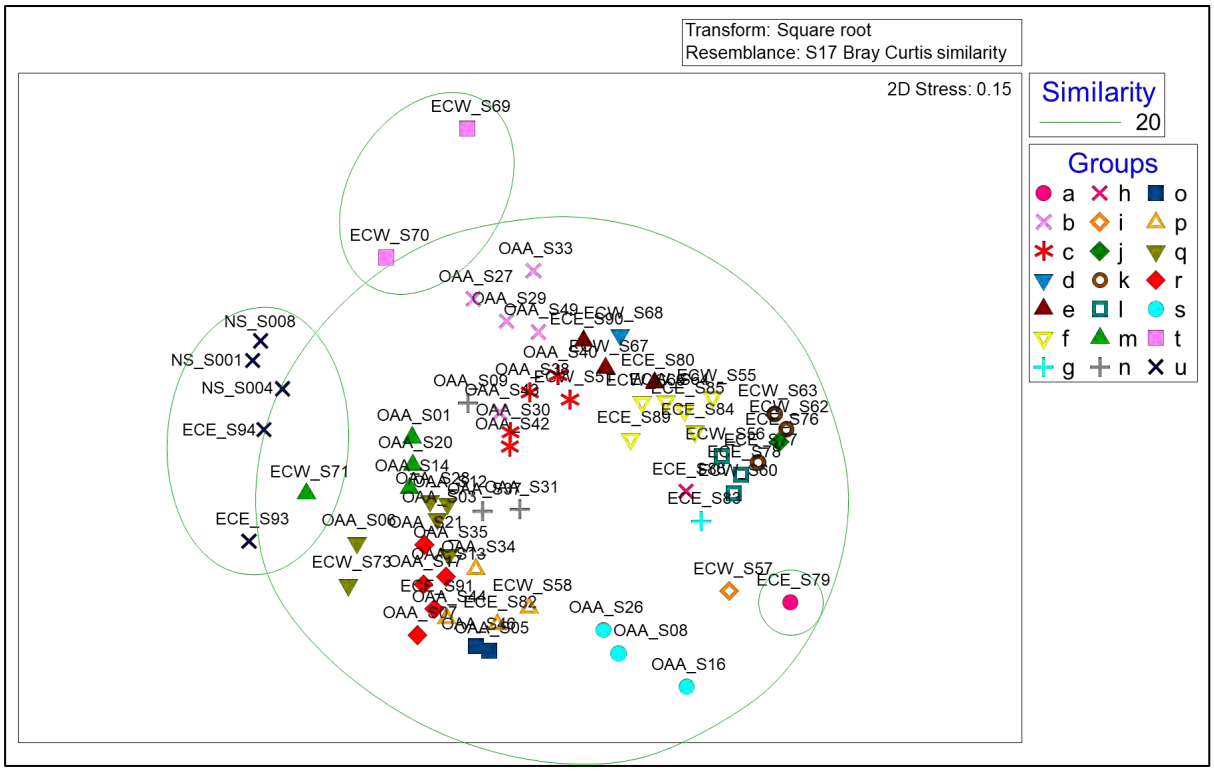


Figure 41 nMDS-plot of non-colonial faunal composition from grab sample sites with group based on the SIMPROF analysis.

5.6.7 SIMPER Results

A SIMPER test, displaying the percentage contribution of the most important species seen in the Bray-Curtis similarity test is presented in Table 38 with species abundance for each SIMPROF group. Average abundance refers to the square root transformed data and is expressed per 0.1 m^2 within the multivariate groups.

Table 38 Summary of characteristics of the non-colonial faunal groups from grab samples derived from the SIMPER test.

Group	Sample ID	Average Depth	Species	Average Abundance	Contribution (%)
a Single sample	ECE_S79	85 m	Less than 2 samples in group	-	-
b Average similarity: 44.58	OAA_S27, OAA_S29, OAA_S33, OAA_S43, OAA_S49	62 m	*Owenia*	16.22	43.52
			Echinocyamus pusillus	3.93	13.35
			Asbjornsenia pygmaea	3.07	9.85
			Timoclea ovata	1.71	5.84
			Goodallia triangularis	1.66	4.09
			Parexogone hebes	1.11	2.74
			Abra prismatica	1.49	1.94
			Unciola planipes	0.85	1.8
			Glycera lapidum (aggregate)	1.20	1.43
			Sthenelais limicola	0.75	1.3

Group	Sample ID	Average Depth	Species	Average Abundance	Contribution (%)
c Average similarity: 49.74	ECW_S51, OAA_S30, OAA_S38, OAA_S40, OAA_S42	70 m	*Echinocyamus pusillus*	6.24	16.03
			Nematoda	4.24	9.02
			Nemertea	2.50	6.15
			Asbjornsenia pygmaea	2.84	6.01
			Abra prismatica	2.45	5.59
			Owenia	4.42	4.91
			Aonides paucibranchiata	1.39	3.42
			Nototropis falcatus	2.19	3.4
			Grania	2.46	3.16
			Aricidea cerrutii	1.25	3.12
d Single sample	ECW_S68	76 m	Less than 2 samples in group	-	-
e Average similarity: 49.97	ECE_S80, ECE_S90, ECW_S67	83 m	*Echinocyamus pusillus*	5.78	12.7
			Exogone verugera	3.93	9.53
			Abra prismatica	3.23	7.47
			Nemertea	2.19	5.18
			Aricidea cerrutii	2.03	4.84
			Polycirrus	1.72	4.35
			Leiochone	1.52	4.04
			Owenia	2.00	4.04
			Ophelia borealis	1.62	3.27
			Unciola planipes	1.80	3.27
f Average similarity: 49.56	ECE_S84, ECE_S85, ECE_S89, ECW_S55, ECW_S64, ECW_S66	85 m	*Echinocyamus pusillus*	7.04	20.19
			Exogone verugera	2.73	7.18
			Owenia	2.59	6.3
			Peresiella clymenoides	2.62	5.22
			Parexogone hebes	2.00	4.72
			Spiophanes kroyeri	1.75	4.67
			Nemertea	2.10	4.62
			Abra prismatica	1.97	4.54
			Aricidea cerrutii	1.87	4.54
			Bathyporeia elegans	1.66	3.99
g Single Sample	ECE_S83	88 m	Less than 2 samples in group	-	-
h Single Sample	ECE_S86	84 m	Less than 2 samples in group	-	-
i Single Sample	ECE_S57	85 m	Less than 2 samples in group	-	-
j Single Sample	ECE_S76	97 m	Less than 2 samples in group	-	-

Group	Sample ID	Average Depth	Species	Average Abundance	Contribution (%)
k Average similarity: 57.85	ECE_S77, ECW_S62, ECW_S63	103 m	*Echinocyamus pusillus*	5.78	7.66
			Spiophanes kroyeri	5.61	7.4
			Scoloplos armiger	4.73	5.59
			Owenia	2.89	4.18
			Peresiella clymenoides	3.08	4.09
			Spiophanes bombyx	2.43	3.18
			Phaxas pellucidus	2.83	3.12
			Pholoe baltica (sensu Petersen)	2.52	3.08
			Harpinia antennaria	2.43	2.81
			Amphiura filiformis	2.16	2.51
l Average similarity: 53.41	ECE_S78, ECW_S56, ECW_S60	93 m	*Echinocyamus pusillus*	5.90	7.18
			Scoloplos armiger	4.19	5.96
			Peresiella clymenoides	4.10	4.67
			Spiophanes kroyeri	3.63	4.2
			Aricidea catherinae	3.68	4.18
			Owenia	3.22	3.73
			Nematoda	3.64	3.56
			Prionospio cf. cirrifera	2.84	3.41
			Galathowenia oculata	2.49	3.39
			Nemertea	2.35	3.2
m Average similarity: 45.50	ECW_S71, OAA_S01, OAA_S14, OAA_S20	62 m	*Polygordius*	3.07	12.73
			Echinocyamus pusillus	2.84	11.88
			Nematoda	3.16	10.91
			Asbjornsenia pygmaea	2.75	10.79
			Glycera lapidum (aggregate)	1.85	8.24
			Goodallia triangularis	2.07	8.06
			Pisione remota	2.19	6.63
			Limatula subauriculata	1.29	5.41
			Crenella decussata	1.48	4.23
			Streptodonta pterochaeta	1.50	3.56
n Average similarity: 45.36	OAA_S09, OAA_S31, OAA_S37	58 m	*Echinocyamus pusillus*	4.47	10.97
			Owenia	4.87	8.64
			Asbjornsenia pygmaea	2.98	7.07
			Nematoda	3.37	6.3
			Glycera lapidum (aggregate)	2.39	6.12
			Aonides paucibranchiata	2.19	5.81
			Nemertea	1.63	4.87
			Goodallia triangularis	2.00	4.07
			Obtusella intersecta	1.28	3.68
			Notomastus	1.47	3.58

Group	Sample ID	Average Depth	Species	Average Abundance	Contribution (%)
o Average similarity: 48.04	OAA_S05, OAA_S46	59 m	Nematoda	8.37	14.15
			Limatula subauriculata	4.74	7.86
			Goodallia triangularis	4.06	6.86
			Echinocyamus pusillus	4.72	6.19
			Timoclea ovata	3.23	5.15
			Obtusella intersecta	2.96	4.2
			Steromphala tumida	3.90	3.84
			Pisione remota	2.32	3.43
			Syllis garciai	2.22	3.43
			Harmothoe impar (aggregate)	1.87	2.97
p Average similarity: 48.81	ECE_S82, ECW_S58, OAA_S34 OAA_S44	76 m	Nematoda	10.66	15.68
			Glycera lapidum (aggregate)	3.59	5.59
			Polygordius	3.79	5.37
			Pisione remota	4.56	5.22
			Grania	3.77	4.36
			Echinocyamus pusillus	3.81	4.03
			Harmothoe impar (aggregate)	3.47	3.6
			Notomastus	2.55	3.37
			Goniadella gracilis	2.87	2.99
			Protodorvillea kefersteini	2.06	2.79
q Average similarity: 56.06	ECW_S73, OAA_S03, OAA_S06, OAA_S12, OAA_S28, OAA_S35	57 m	Nematoda	10.24	19.2
			Pisione remota	6.33	10.63
			Asbjornsenia pygmaea	5.03	7.37
			Polygordius	5.25	6.75
			Echinocyamus pusillus	4.61	6.5
			Glycera lapidum (aggregate)	3.59	5.88
			Goodallia triangularis	3.14	4.76
			Hesionura elongata	3.00	4.63
			Nemertea	2.44	4.43
			Syllis pontxioi	2.70	4.24
r Average similarity: 50.95	ECE_S91, OAA_S07, OAA_S13, OAA_S17, OAA_S21	65 m	Nematoda	5.10	8.43
			Limatula subauriculata	3.79	6.63
			Pisione remota	3.36	6.44
			Polygordius	4.21	6.4
			Echinocyamus pusillus	3.38	5.56
			Glycera lapidum (aggregate)	3.08	4.69
			Syllis pontxioi	2.31	4.62
			Protodorvillea kefersteini	2.64	4.43
			Aonides paucibranchiata	2.30	4.07
			Nototropis vedlomensis	2.26	3.59

Group	Sample ID	Average Depth	Species	Average Abundance	Contribution (%)
s Average similarity: 35.88	OAA_S08, OAA_S16, OAA_S26	62 m	*Echinocyamus pusillus*	3.80	6.36
			Modiolula phaseolina	3.50	4.84
			Harmothoe impar (aggregate)	3.08	4.71
			Glycera lapidum (aggregate)	2.86	4.71
			Nematoda	2.39	4.26
			Spiophanes kroyeri	2.39	4.26
			Stenothoe marina	1.90	3.86
			Notomastus	1.88	3.57
			Verruca stroemia	3.86	3.33
			Owenia	2.43	3.27
t Average similarity: 40.48	ECW_S69, ECW_S70	62 m	*Asbjornsenia pygmaea*	3.18	22.22
			Nototropis falcatus	2.00	19.87
			Sthenelais limicola	1.41	14.05
			Nephtys cirrosa	1.71	14.05
			Ophelia borealis	1.00	9.94
			Pseudocuma simile	1.00	9.94
u Average similarity: 41.29	ECE_S93, ECE_S94, NS_S001, NS_S004, NS_S008	36 m	Nematoda	4.20	34.33
			Hesionura elongata	4.00	27.89
			Travisia forbesii	1.09	7.88
			Asbjornsenia pygmaea	1.69	7.72
			Parexogone hebes	1.08	5.62
			Streptosyllis bidentata	0.77	3.99
			Goodallia triangularis	0.80	3.59

SIMPROF Group **a** comprised a single sample ECE_S79, located within the ECC East. This sample separated from groups **b** – **s** at 18.01 % similarity, primarily due to a lower abundance of *Echinocyamus pusillus* and higher abundance of *Phascolion strombus* in group **a** compared to groups **b** – **s**, but also differences in the abundance of *Galathowenia oculata, Owenia* and presence/absence differences in several less abundant taxa.

Group **b** included five (5) samples (OAA_S27, OAA_S29, OAA_S33, OAA_S43 and OAA_S49) distributed along the eastern edge of the OAA, with an average depth of 62 m. This cluster group separated from groups **c** – **f** at 26.05 % similarity, primarily due to the higher abundance of the tubeworm *Owenia* in group **b**. The average within-group similarity was 44.58 %. The samples were dominated by *Owenia*, which contributed with 43.52 % to within-group similarity. Pea urchin *E. pusillus* and the bivalve *Asbjornsenia pygmaea* followed with 13.35 % and 9.85 % contributions, respectively.

Group **c** consisted of five (5) samples (ECW_S51, OAA_S30, OAA_S38, OAA_S40 and OAA_S42) distributed in the OAA and the ECC West, with an average depth of 70 m. This group separated from groups **d**, **e** and **f** at 39 % similarity, with taxa causing dissimilarity including the higher numbers of *Owenia* and nematodes in group **b** compared to groups **c**, **d** and **e**. The average within-group similarity was 49.74 % and the group was characterised by *E. pusillus*, which was present in higher abundance than in group **a** and contributed 16.03 % to within-group similarity. Followed by nematodes and Nemertea, which contributed 9.02 % and 6.15 % respectively to within-group similarity.

Group d comprised a single sample ECW_S68, located on the ECC West approximately 7 km from the landward end with a depth of 76 m. This sample separated from groups **e** and **f** at 41.16 % similarity, with taxa causing dissimilarity including the lower abundance of *E. pusillus* and higher abundance of *Grania* and *Parexogone hebes* in group **d** compared to groups **e** and **f**. The sample was characterised by the exogonid polychaete *P. hebes*, the bivalve *Abra prismatica* and the polychaete *Travisia forbesii*.

Group **e** consisted of three (3) samples (ECE_S80, ECE_S90 and ECW_S67), distributed along the proposed route corridor with an average depth of 83 m. This group separated from group **f** at 43.67 % similarity, with taxa causing dissimilarity including the capitellid *Peresiella clymenoides*, which was absent in group **e**; along with lower abundances of *P. hebes* and *E. pusillus* in group **e** compared to group **f**. The average within-group similarity for group **e** was 49.97 % and the highest contribution to within-group similarity was *E. pusillus* with 12.7 %, followed by *Exogone verugera* with 9.53 % and *A. prismatica* with 7.47 %.

Cluster group **f** was one of the two largest groups identified, including six (6) samples (ECE_S84, ECE_S85, ECE_S89, ECW_S55, ECW_S64 and ECW_S66) located along the route corridor with an average depth of 83 m. This group had an average within-group similarity of 49.56 % and was characterised by *E. pusillus*, which contributed 20.19 % to within-group similarity; followed by *E. verugera* and *Owenia*, which contributed further with 7.18 % and 6.3 % to within-group similarity, respectively.

SIMPROF cluster groups **g** – **l** separated from groups **c** – **f** at 31.36 % similarity, with taxa causing dissimilarity including higher abundances of *Spiophanes kroyeri, Urothoe elegans, Paradoneis lyra* and *Scoloplos armiger* in groups **g** – **l** compared to groups **c** – **f**.

Group **g** comprised the single sample ECE_S83, located along the ECC East, with a depth of 88 m. This sample separated from groups **h** – **l** at 34.05 % similarity, due to generally lower abundances of taxa including *S. kroyeri, S. armiger, Verruca stroemia* and *Owenia* in group **j** compared to the groups **h** – **l**.

Group **h** consisted of a single sample ECE_S86, located along the ECC East, with a depth of 84 m. Group **h** separated from groups **I** – **l** at 35.73 % similarity, largely due to the absence of taxa such as *Owenia*, *U. elegans* and *Phaxas pellucidus* in group **h**, along with lower abundances of *S. armiger* and *S. kroyeri* in group **h** compared to groups **I** – **l**.

Group **I** consisted of a single sample ECW_S57, located along the ECC West with a depth of 85 m. This group separated from groups **k** and **l** at 37.63 % similarity, with taxa causing dissimilarity including the abundance of *P. lyra, Sabellaria spinulosa* and *Verruca stroemia*, all of which were lower in number or absent entirely from groups **j**, **k** and **l**. Group **f** was diverse, with the third highest number of non-colonial taxa (90) recorded.

SIMPROF group **j** consisted of the single sample ECE_S76, located on the ECC East with a depth of 97 m. This sample separated from group **k** at 53.54 % similarity due to differences in species composition, including the presence of *Papillicardium minimum* and *Gattyana cirrhosa* in Group **j**, both of which were absent from Group **k**; and also the abundances of the polychaetes *Priospio fallax* and *Owenia*, both of which were lower in Group **k** than in Group **j**.

Group **k** included three (3) samples (ECE_S77, ECW_S62 and ECW_S63) distributed along the ECC West, with an average depth of 103 m. This cluster had an average within-group similarity of 57.85 % and the highest contribution to within-group similarity for group **I** was the pea urchin *E. pusillus*, contributing with 7.66 % to within-group similarity; followed by the polychaetes *S. kroyeri, S. armiger* and *Owenia*, which contributed with 7.4 %, 5.59 % and 4.18 %, respectively to with-group similarity.

Group **l** comprised three (3) samples located along the route corridor (ECE_S78, ECW_S56 and ECW_S60), with an average depth of 93 m. This group separated from groups **j** and **k** at 46.5 % similarity, due partly to the lower abundance of *P. pellucidus* in group **l** compared to groups **j** and **k** and partly due to presence/absence differences in several less abundant taxa between the groups. The average within-group similarity was 53.41 % and the highest contribution to within-group similarity for group **l** was the pea urchin *E. pusillus*, contributing with 7.18 %, followed by the polychaetes *S. armiger*, *P. clymenoides and S. kroyeri*, which contributed with 5.96 %, 4.67 % and 4.2 %, respectively.

Groups **m** – **s** separated from groups **b** – **l** at 21.61 % similarity, indicating differences in species assemblages between the two halves of the cluster dendrogram. This division of samples is also visible on the MDS plot in Figure 41, where groups **b** – **l** are distributed towards the upper right of the plot, whereas groups **m** – **s** are distributed towards the lower left of the plot.

Group **m** separated from groups **n** – **r** at 36.52 % similarity, with taxa causing dissimilarity including differences in relative abundances of *Pisione remota*, nematodes, *Polygordius* and *Protodorvillea kefersteini*. Group **m** included four (4) samples, three (3) of which were located in the southwest OAA (OAA_S01, OAA_S14 and OAA_S20) and the fourth ECW_S71 was located on the ECC W, approximately 3.5 km from landfall. These samples had an average depth of 62 m, and the average within-group similarity was 45.5 %. The highest contributions to within-group similarity were from Polygordius, *E. pusillus* and nematodes, contributing 12.73 %, 11.88 % and 10.91 %, respectively.

Group **n** comprised three samples (OAA_S09, OAA_S31 and OAA_S37) distributed through the OAA, with an average depth of 58 m. This group separated from groups **o** – **r** at 36.76 % similarity with taxa causing dissimilarity including *P. remota*, which was absent from group **n**; lower abundance of nematodes in group **n** compared to groups **o** – **r** and differences in the abundance of *Polygordius* and *A. pygmaea* between the groups. Group **n** had an average within-group similarity of 45.36 % and the highest contributing taxa were *E. pusillus, Owenia* and *A. pygmaea* contributed with 10.97 %, 8.64 % and 7.07 %, respectively.

Group o separated from groups **p**, **q** and **r** at a similarity of 39.73 %, with taxa causing dissimilarity including a higher abundance of *Caecum glabrum* and lower abundance of *Polygordius* in group **o** compared to groups **p**, **q** and **r**; along with the presence of *Steromphala tumida* in group **o**, which was absent or only present in low abundance in groups **p**, **q** and **r**. Group **o** included two (2) samples (OAA_S05 and OAA_S46) located towards the northeast and southwest boundaries of the OAA, with an average depth of 59 m. The average within-group similarity was 48.04 % and the group was characterised by nematodes and the bivalve molluscs *Limatula subauriculata* and *Goodallia triangularis*, which contributed 14.15 %, 7.86 % and 6.86 % to within-group similarity, respectively.

Group **p** comprised four (4) samples (ECE_S82, ECW_S58, OAA_S34 and OAA_S44) distributed between the route corridor and the OAA, with an average depth of 76 m. This group separated from groups **q** and **r** at 44.15 % similarity, with taxa causing dissimilarity including the higher abundance of *Owenia* and *Harmothoe impar* aggregate and lower abundance of *G. triangularis* in group **p** compared to groups **q** and **r**. Group **p** had an average within-group similarity of 48.81 %, with the highest contribution to within-group similarity from nematodes with 15.68 %, followed by *G. lapidum* aggregate at 5.59 %, *Polygordius* with 5.37 % and *P. remota* with 5.22 %.

Groups **q** and **r** separated from one another at a similarity of 45.48 %, with taxa causing dissimilarity including higher abundances of nematodes, *A. pygmaea*, *Polygordius* and *P. remota* in group **q** compared to group **r**. Group **q** was one of the two largest cluster groups, including six (6) samples, five (5) of which were spread across the OAA (OAA_S03, OAA_S06, OAA_S12, OAA_S28 and OAA_S35) and the remaining one (ECW_S73) was at the landward end ECC West approximately 1.8 km from the proposed landfall. These samples had an average depth of 57 m and an average within-group similarity of 56.06 %. The dominant taxon in this group was nematodes, which contributed 19.2 % to within-group similarity; followed by *P. remota* and *A. pygmaea*, contributing 10.63 % and 7.37 % to within-group similarity, respectively.

Group **r** comprised five (5) samples, four (4) of which were distributed across the OAA (OAA_S07, OAA_S13, OAA_S17 and OAA_S21) and one (1) (ECE_S91) located on the ECC East approximately 5 km from the landward end. These samples had an average depth of 65 m, and the average within-group similarity was 50.95 %. Nematodes had the highest contribution to within-group similarity at 8.43 %, followed by an abundance of *L. subauriculata* at 6.63 %, *P. remota* at 6.44 % and with *Polygordius* at 6.4 %.

Group **s** separated from groups **m** to **r** at 22.22 % similarity, with taxa causing dissimilarity including *Polygordius* and *P. remota* and *A. pygmaea* all three of which were absent in group **s**; also *V. stroemia* and *Parapleustes monocuspis* were either absent or lower in abundance in groups **m** to **r** compared to group **s**.

Group **s** included three (3) samples (OAA_S08, OAA_S16 and OAA_S26) distributed across the western half of the OAA, with an average depth of 62 m. These samples had relatively high numbers of taxa compared to most of the other groups. The average within-group similarity was 35.88 % with the highest contributions from the taxa *E. pusillus, Modiolula phaseolina*, *H. impar* aggregate and *G. lapidum* aggregate with 6.36 %, 4.84 %, 4.71 % and 4.71 %, respectively.

Groups **t** and **u** show the widest separation from the other samples, separating from groups **a** – **s** at 15.9 % similarity and from one another at 17.17 % similarity. Both groups **t** and **u** included samples located on the proposed cable route close to the landfall end and had the lowest numbers of taxa and individuals compared to the other cluster groups. This split is also visible on the MDS plot in Figure 41, where groups **t** and **u** are distributed away from the other samples and each within their own 20 % similarity contour.

Group **t** comprised two (2) samples (ECW_S69 and ECW_S70) located between 4.6 km and 5.4 km from the landfall and approximately 800 m from one another, with an average depth of 62 m. The average within-group similarity was 40.48 % and the highest contribution to within-group similarity was from the bivalve mollusc *A. pygmaea* followed by the amphipod *Nototropis falcatus* and the scale worm *Sthenelais limicola* with 22.22 %, 19.87 % and 14.05 %, respectively.

Cluster Group **u** included five (5) samples (ECE_S93, ECE_S94, NS_S001, NS_S004 and NS_S008) located closest to the landfall end of the ECC East. These were the shallowest samples, with an average depth of 36 m. The average within-group similarity was 41.29 % and was dominated by the abundance of nematodes, which contributed 34.33 %; followed by *Hesionura elongata* and *T. forbesii*, which contributed further with 27.89 % and 7.88 % to within-group similarity, respectively.

5.6.8 Relationship Between Physical and Biological Data

The relationship between physical and biological data was assessed by applying the BEST analysis from the PRIMER suite. The BEST test identifies which of the variables best explains macrofaunal distribution in the survey area. Square root transformation was applied to the faunal abundance data (expressed per 0.1 m^2) before calculating the Bray-Curtis similarity measures. Normalisation was applied to the physical variables before calculating the Euclidean distance.

A total of 64 sample sites were selected for the BEST analysis, where both physical and biological data was sampled. Selected variables in the BEST test included depth and PSA. Results of the BEST analysis for Single and Multiple variables are presented in Table 39.

Results presented for single variables gave a global correlation (σ) of 0.552 for Depth. The significance level was 1 % which means that the null hypothesis of 'no agreement in multivariate pattern between physical and biological data' can be rejected at p<1%. The variables % Fine sand and % Clay followed with a correlation (σ) of 0.474 and 0.443, respectively.

Results presented for multiple variables gave a global correlation (σ) of 0.675 for the combined variables Depth (m), % Medium Sand, and % Clay. The significance level was 1 % which means that the null hypothesis of 'no agreement in multivariate pattern between physical and biological data' can be rejected at p<1%. The second highest correlation (σ) was 0.672 for the combined variables Depth (m), % Medium Sand, and % Fine Sand. The variables Depth (m) and % Medium Sand were present in all of the top 5 trails.

Chemicals and contaminants variables presented a very weak or negative correlation with the faunal abundance data and are therefore not listed in Table 39.

Table 39 Results of BEST test between physical data and biological data for single and multiple variables.

Max nr of trail variables	Number of variables	Spearman correlation (σ)	Physical Variables
Single variables Global Test (σ): 0.552 Significance: 1%	1	0.552	Depth (m)
	1	0.474	% Fine Sand
	1	0.443	% Clay
	1	0.395	% Very Coarse Silt
	1	0.389	% Very Fine Sand
Multiple variables Global Test (σ): 0.675 Significance: 1%	3	0.675	Depth (m), % Medium Sand, % Clay
	3	0.672	Depth (m), % Medium Sand, % Fine Sand
	4	0.666	Depth (m), % Medium Sand, % Fine Sand, % Clay
	4	0.657	Depth (m), % Very Coarse Sand, % Medium Sand, % Clay
	4	0.657	Depth (m), % Medium Sand, % Fine Sand, % Very Coarse Silt

5.6.9 Multivariate Statistical Analyses EUNIS

Similarities between the macrofaunal data and EUNIS habitats are further explored in a hierarchical dendrogram presented in Figure 42, and in the nMDS-plot, presented in Figure 43. Analyses in Section 5.6.4 are conducted on the faunal composition from grab sampling sites but with groups superimposed with EUNIS habitats.

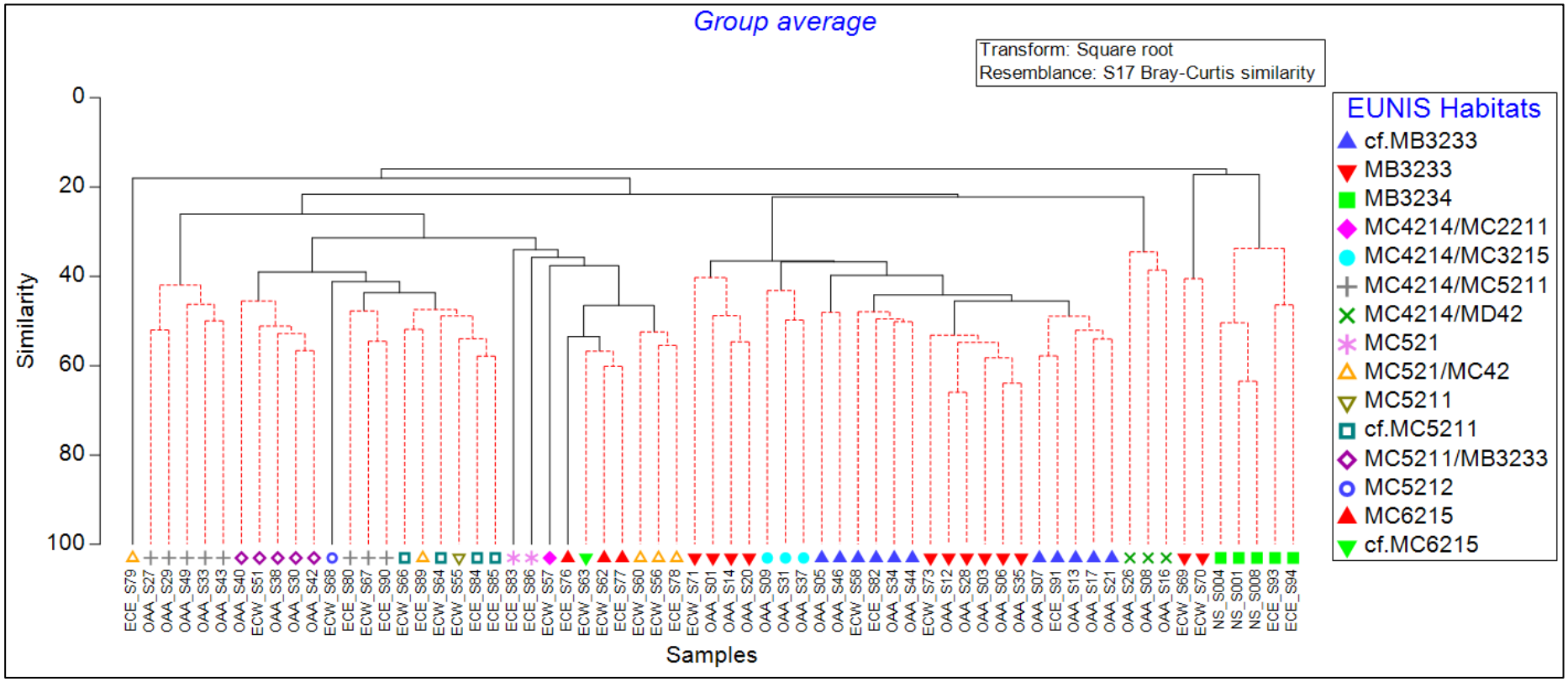


Figure 42 SIMPROF dendrogram of non-colonial faunal composition from grab sampling sites superimposed with EUNIS habitats.

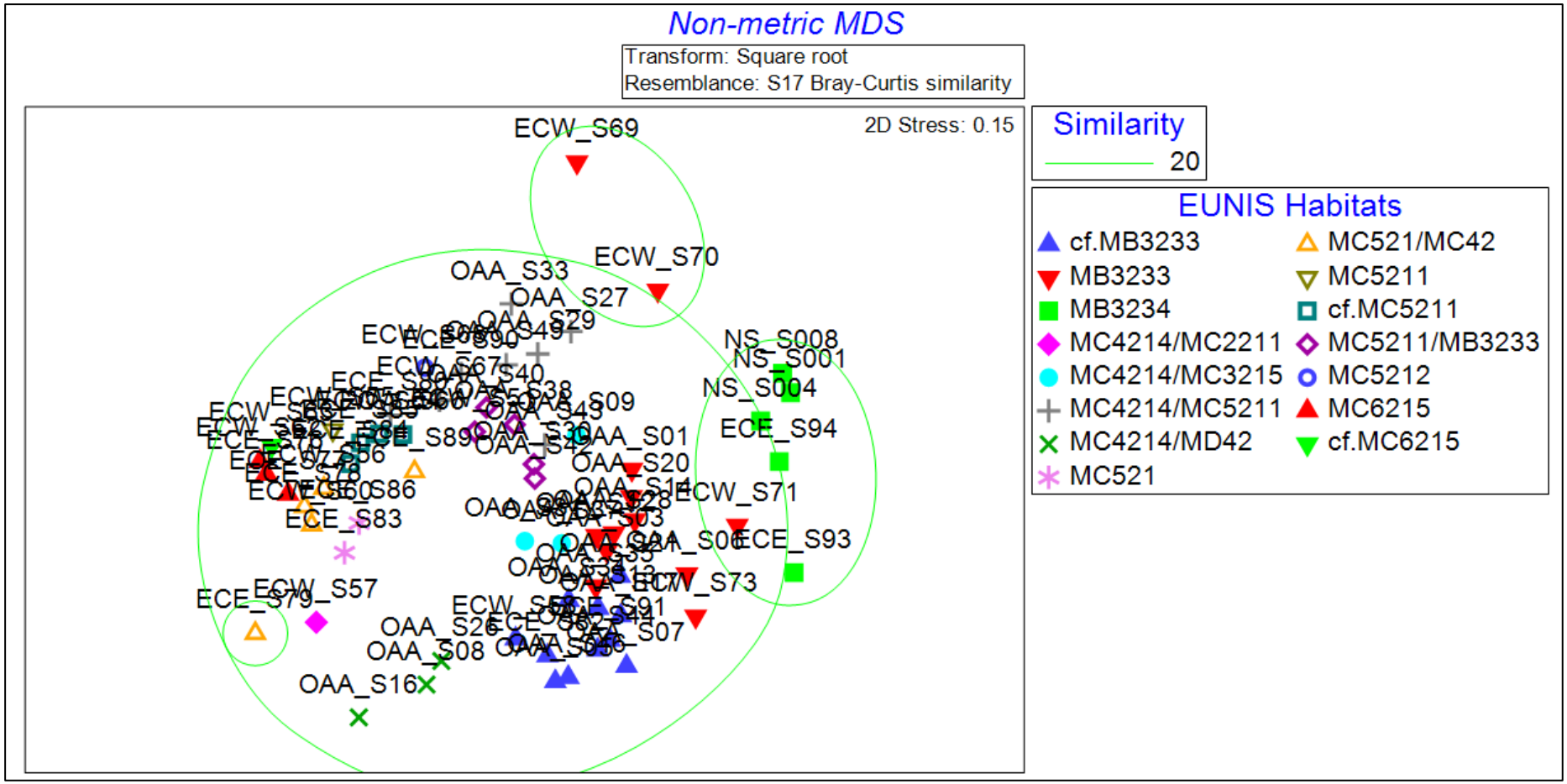


Figure 43 nMDS-plot of non-colonial faunal composition from grab samplings sites superimposed with EUNIS habitats.

5.6.10 Sessile Colonial Epifauna from Grab Samples

The phyletic composition of sessile colonial epifauna identified from grab samples is summarised in Table 40 and illustrated in Figure 44 and Figure 45.

A total of five (5) major phyla were identified. The dominant phylum was Bryozoa which constituted 70 % of the total taxa. Cnidaria followed with 19 % of the total taxa, Porifera with 5 %, Entoprocta with 5 % and Annelida with 0.3 %. In total 84 different taxa were identified.

Abundance was also dominated by Bryozoa with a total of 240 colonies, followed by Cnidaria with 42 colonies and Porifera with 18 colonies. Entoprocta and Annelida contributed with 15 and 1 colony, respectively.

Table 40 Phyletic composition of colonial epifauna from grab samples.

Phylum	Number of Taxa	Abundance of Colonies
Bryozoa	59	240
Cnidaria	16	42
Porifera	4	18
Entoprocta	4	15
Annelida	1	1
Total	**84**	**316**

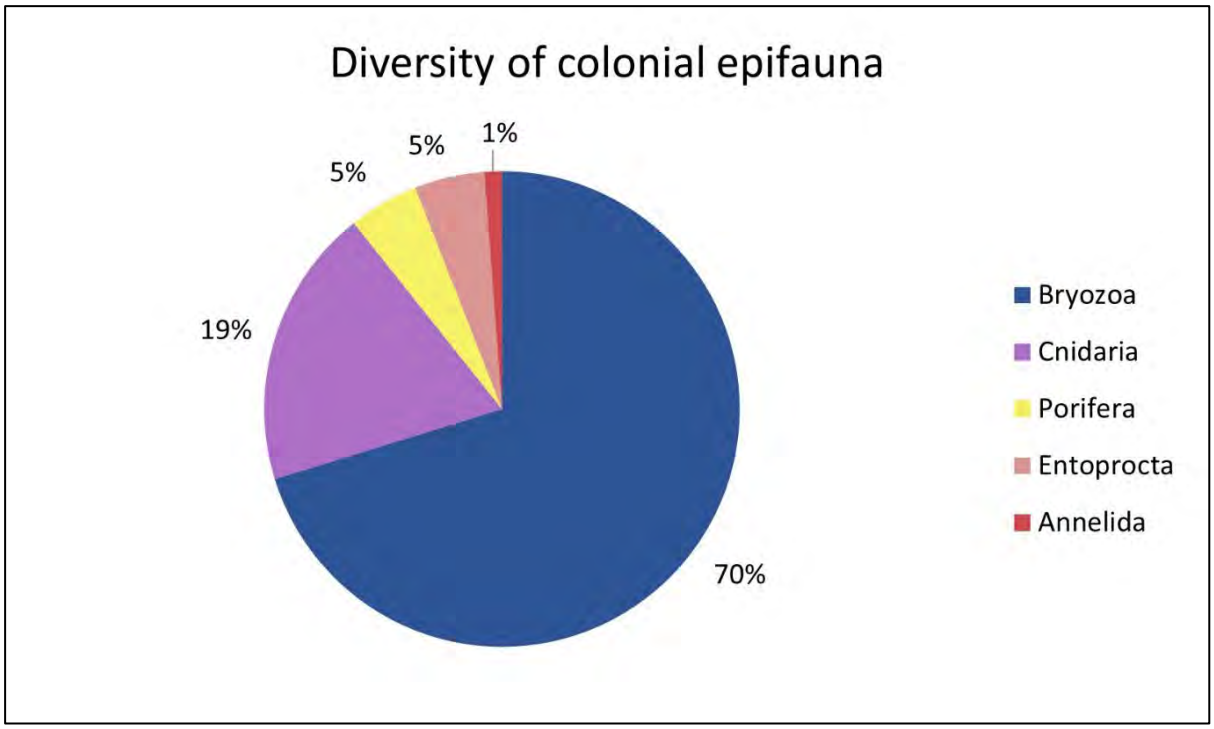


Figure 44 Diversity of colonial epifauna from grab samples.

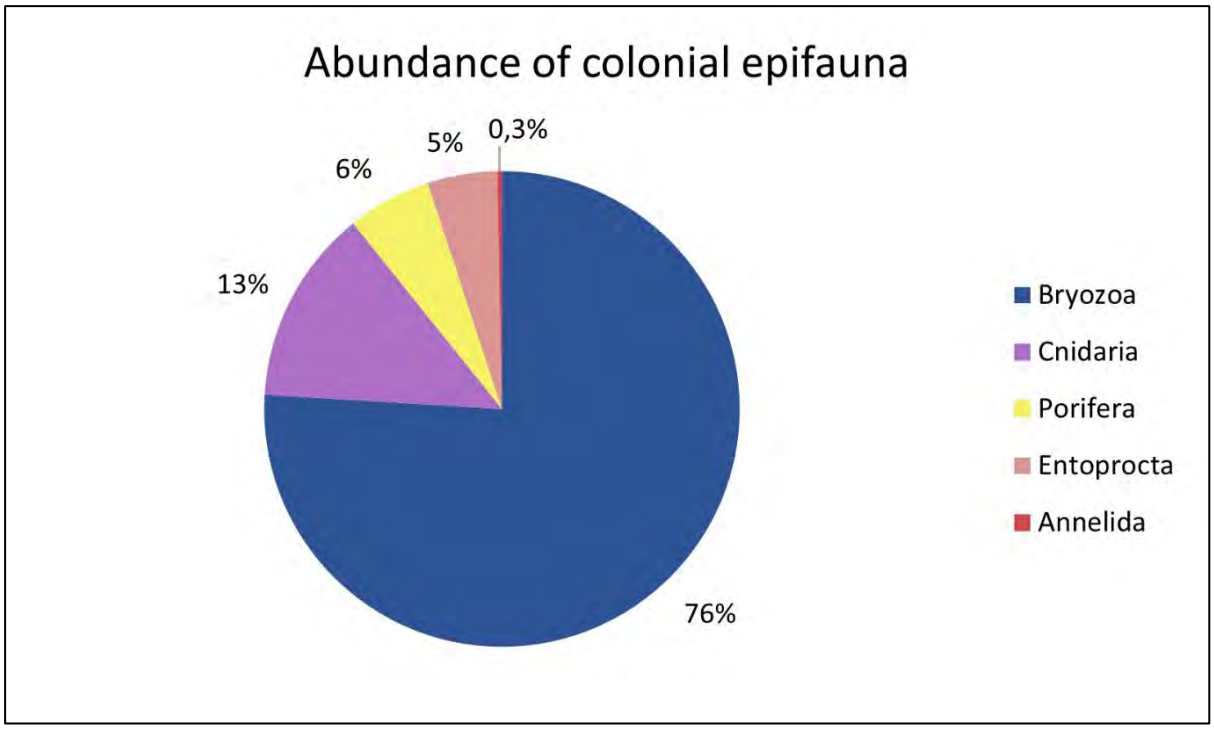


Figure 45 Abundance of colonial epifauna from grab samples.

5.6.11 Biomass

The non-colonial fauna species biomass expressed as blotted wet weight (g per 0.1 m²) is illustrated in Figure 46, and summarised in Table 41. Biomass was grouped into the major groups Annelida, Arthropoda, Chordata, Echinodermata, Mollusca, Nemertea and "Others". The group "Others" included the phyla Cnidaria, Hemichordata, Nematoda, Phoronida and Platyhelminthes.

The biomass was dominated by Mollusca, which accounted for 70 % of the total biomass. This was primarily due to the presence of three (3) large individuals of the dog cockle *Glycymeris glycymeris* in sample ECE_S91, weighing 116 g and constituting 66 % of the total mollusc weight.

The second largest group was Annelida, accounting for 14 % of the total biomass. The high numbers of tubeworm *Owenia* contributed to 54 % of the total Annelida biomass. Echinodermata accounted for 13 % of the total biomass, followed by Chordata, Arthropoda and Nemertea with 1 % respectively. The group "Others" accounted for less than 1 %.

Within the group "Others", Cnidaria constituted 0.12 % of the total biomass. Phoronida contributed 0.10 %, Platyhelminthes 0.10 %, Nematoda 0.02 % and Hemichordata 0.01 % respectively of the total biomass. Non-colonial fauna biomass varied between 0.0343 g/0.1 m² in sample ECE_S94 to 123.8660 g/0.1 m² in sample ECE_S91. The mean biomass across all sites was 3.8445 g/0.1 m² (SD=15.5177). The spatial distribution of biomass across the survey area is illustrated in Figure 47.

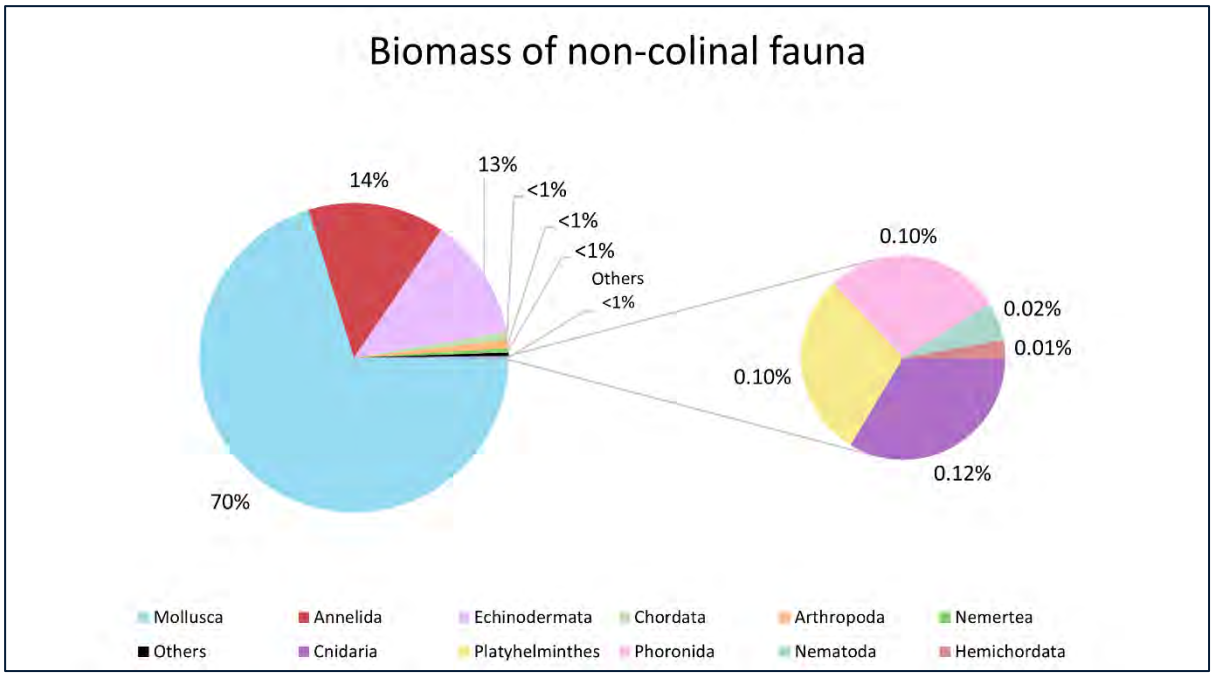


Figure 46 Total biomass (blotted wet weight in g/0.1 m²) composition of major phyla in the left pie chart and group "Others" in the right pie chart.

Table 41 Biomass (blotted wet weight in g/0.1 m²).

Sample ID	Annelida	Arthropoda	Mollusca	Echinodermata	Nemertea	Others	Total
OAA_S01	0.7261	0.0078	0.5142	0.0022	0	0.6264	1.8767
OAA_S03	0.0272	0.0113	0.0609	0.0422	0.0016	0.0056	0.1488
OAA_S05	0.1414	0.0385	0.7619	0.0001	0.0071	0.0204	0.9694
OAA_S06	0.0689	0.0109	0.0542	0.0001	0.0011	0.0058	0.1410
OAA_S07	0.1418	0.0151	0.1034	0.0007	0.0822	0.1063	0.4495
OAA_S08	1.1124	0.3079	0.2264	0.0868	0.0021	0.0001	1.7357
OAA_S09	2.3212	0.0069	0.0094	0.0362	0.0019	0.0147	2.3903
OAA_S12	0.1221	0.0157	0.2322	0.0153	0.0034	0.0055	0.3942
OAA_S13	0.0991	0.3030	0.0787	0.0696	0.0969	0.0795	0.7268
OAA_S14	0.0109	0.0018	0.0936	0.0535	0.0011	0.0001	0.1610
OAA_S16	0.3576	0.2545	3.1396	0.1691	0.0178	0.0476	3.9862
OAA_S17	0.2445	0.0509	0.0222	0.0078	0.0006	0.0317	0.3577
OAA_S20	0.0353	0.0069	0.4718	0.0326	0.0002	0.0001	0.5469
OAA_S21	0.2163	0.0079	0.2032	0.0001	0.0002	0.0920	0.5197
OAA_S26	0.1037	0.0540	1.3078	0.5867	0.0023	0.0013	2.0558
OAA_S27	7.8424	0.0047	0.0953	13.1176	0.0032	0.0473	21.1105
OAA_S28	0.0549	0.0677	0.0816	0.0007	0.0012	0.0003	0.2064
OAA_S29	1.9113	0.0100	0.0728	0.0492	0.0005	0.0001	2.0439
OAA_S30	0.0241	0.0070	0.1153	0.0141	0.0028	0.0015	0.1648
OAA_S31	0.2491	0.0208	0.2381	0.0627	0.0263	0.0008	0.5978

Sample ID	Annelida	Arthropoda	Mollusca	Echinodermata	Nemertea	Others	Total
OAA_S33	1.0576	0.0611	0.1789	0.0055	0	0	1.3031
OAA_S34	3.0865	0.0514	0.4110	0.1040	0.1649	0.0199	3.8377
OAA_S35	0.1934	0.0229	0.0622	0.0155	0.0079	0.0033	0.3052
OAA_S37	0.1344	0.0155	1.2090	0.0708	0.0009	0.0015	1.4321
OAA_S38	0.0675	0.0096	0.0725	0.1742	0.0068	0.0071	0.3377
OAA_S40	0.8035	0.0160	0.3071	8.1899	0.0032	0.0785	9.3982
OAA_S42	0.2253	0.0007	0.1531	0.0397	0.0018	0.0002	0.4208
OAA_S43	1.5203	0.0839	0.1778	0.0312	0.0012	0.0005	1.8149
OAA_S44	0.2365	0.0042	0.3389	0.0212	0.0055	0.0030	0.6093
OAA_S46	0.3274	0.0161	0.0824	0.1834	0.0171	0.1064	0.7328
OAA_S49	0.4663	0.0026	0.2427	0.0567	0.0249	0.0167	0.8099
ECW_S51	0.0863	0.0035	0.1298	0.0323	0.0118	0.0052	0.2689
ECW_S55	0.1391	0.0063	3.0517	0.0610	0.0050	0.0528	3.3159
ECW_S56	0.6014	0.0471	0.0332	0.2988	0.0341	0.0030	1.0176
ECW_S57	0.4916	0.0292	0.3758	0.0122	0.0251	0.0078	0.9417
ECW_S58	0.6694	0.0219	12.6026	0.0036	0.0141	0.0043	13.3159
ECW_S60	1.9215	0.0644	0.4448	0.5918	0.0121	0.0580	3.0926
ECW_S62	0.438	0.0161	3.2028	0.5415	0.0095	0.0200	4.2279
ECW_S63	0.6333	0.0207	1.1638	5.6056	0.0001	0.0177	7.4412
ECW_S64	0.1604	0.0309	0.0390	0.1287	0.0016	0.0001	0.3607
ECW_S66	0.1692	0.0038	0.2842	0.0338	0.0010	0.0092	0.5012
ECW_S67	0.3063	0.0297	0.0218	0.0743	0.0045	0.0275	0.4641
ECW_S68	0.0741	0.0341	0.3066	0.0u	0.0533	0.0034	0.4823
ECW_S69	0.1166	0.0008	0.0246	0.0001	0	0	0.1421
ECW_S70	0.1842	0.0215	0.0654	0.0013	0	0.0001	0.2725
ECW_S71	0.0406	0.0390	0.1079	0.0010	0	0.0001	0.1886
ECW_S73	0.1522	0.0013	1.5532	0.0077	0.0801	0.7340	2.5285
ECE_S76	0.5298	0.0130	1.0934	0.0910	0.0172	0.0062	1.7506
ECE_S77	0.7016	0.0141	1.6740	0.6270	0.0175	0.0362	3.0704
ECE_S78	0.7875	0.0085	0.6351	0.0722	0.0309	0.0010	1.5352
ECE_S79	0.1449	0.0003	0.0784	0.0023	0.0015	0.0001	0.2275
ECE_S80	0.4811	0.0131	0.0506	0.0120	0.0056	0.0235	0.5859
ECE_S82	0.2927	0.0548	10.3008	0.0295	0.0146	0.6232	11.3156
ECE_S83	0.1059	0.0778	0.8473	0.0107	0	0.0056	1.0473
ECE_S84	0.1673	0.0018	0.1647	0.015	0.0139	0.0066	0.3693
ECE_S85	0.2963	0.0109	0.4946	0.0485	0.0299	0.0312	0.9114

Sample ID	Annelida	Arthropoda	Mollusca	Echinodermata	Nemertea	Others	Total
ECE_S86	0.8964	0.0126	0.0292	0.3423	0.0008	0.0001	1.2814
ECE_S89	0.3620	0.0061	0.4971	0.0544	0.0335	0.0046	0.9577
ECE_S90	0.2013	0.0072	0.1444	0.0306	0.1858	0.0351	0.6044
ECE_S91	0.4087	0.0430	123.3823	0.0033	0.0286	0.0001	123.8660
ECE_S93	0.0141	0.0001	0.0334	0	0.0010	0.0003	0.0489
ECE_S94	0.0155	0.0012	0.0031	0.0001	0.0143	0.0001	0.0343
NS_S001	0.0336	0.0323	0.0293	0	0	0.0001	0.0953
NS_S004	0.0611	0	1.6190	0.0001	0	0.0001	1.6803
NS_S008	0.2265	0	0.1384	0	0	0.0002	0.3651
Total	**35.8395**	**2.1544**	**175.7405**	**31.9829**	**1.1341**	**3.0417**	**249.8931**
Mean	**0.5514**	**0.0331**	**2.7037**	**0.4920**	**0.0174**	**0.0468**	**3.8445**
SD	**1.0874**	**0.0605**	**15.3399**	**2.0024**	**0.0347**	**0.1392**	**15.5177**
Min	**0.0109**	**0.0000**	**0.0031**	**0.0000**	**0.0000**	**0.0000**	**0.0343**
Max	**7.8424**	**0.3079**	**123.3823**	**13.1176**	**0.1858**	**0.7340**	**123.8660**
Median	**0.2253**	**0.0141**	**0.1789**	**0.0323**	**0.0034**	**0.0055**	**0.7328**

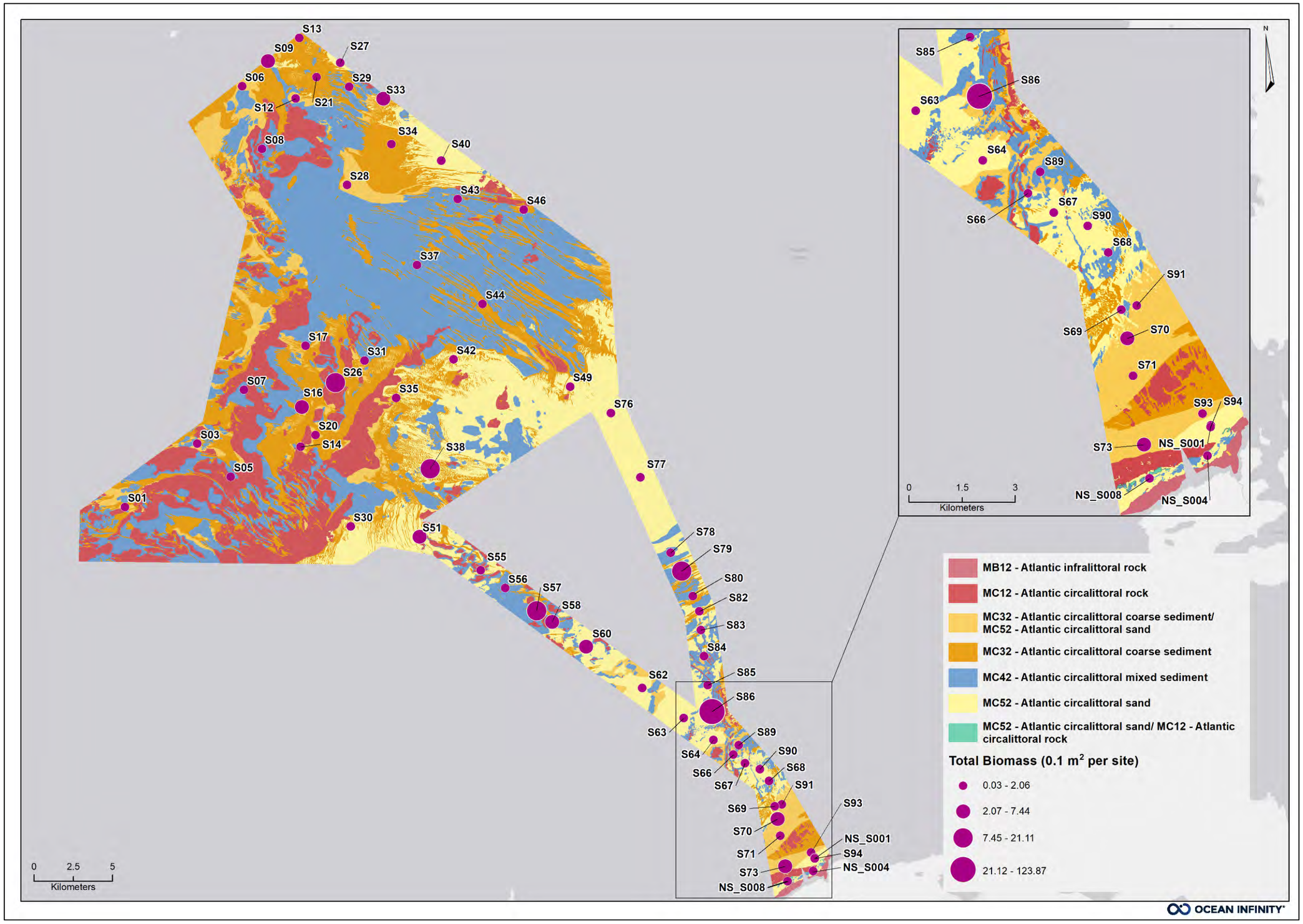


Figure 47 Overview of the total biomass (g/0.1 m2) per grab sample site.

5.7 Epibenthic Fauna from Visual Survey

The results from the analyses of the stills from grab sample sites and transects presented habitats generally dominated by Atlantic circalittoral mixed sediment. Conspicuous fauna were arthropods, bryozoans and echinoderms mostly associated with the mixed substrate. Eight (8) out of the total 108 sites had no fauna recorded in the stills acquired (ECE_S94, ECW_S69, OAA_S27, OAA_S30, NS_S001, NS_S002, NS_S004 and NS_S008). These sites showed habitats comprising **MC52** – Atlantic circalittoral sand, **MC32 –** Atlantic circalittoral coarse sediment/**MC52** – Atlantic circalittoral sand, and **MC42** – Atlantic circalittoral mixed sediment.

The top ten sites with the highest number of taxa are presented in Table 42. The average number of taxa was 18 per site. Figure 48 presents a still photo from site OAA_S10, which had the greatest diversity of all sites.

Table 42 Top sites with the greatest species diversity.

Phylum/Site ID	OAA_S10	ECW_S53	ECE_S92	OAA_S32	ECW_S54	OAA_S08	OAA_T47	OAA_T97	ECE_S84	ECW_S65
Annelida	2	3	2	2	3	2	2	2	2	2
Arthropoda	12	10	10	7	9	4	5	12	7	9
Bryozoa	11	10	8	11	9	11	9	6	11	12
Chordata	4	3	1	2	2	3	4		3	2
Cnidaria	8	9	8	14	9	8	12	14	8	7
Echinodermata	2	2	4	1	2	1	1		2	
Mollusca	10	4	10	6	5	8	6	5	6	5
Porifera	2	5	1	1	3	3	1			2
Rhodophyta								1		
Total No. of Taxa	**51**	**46**	**44**	**44**	**42**	**40**	**40**	**40**	**39**	**39**

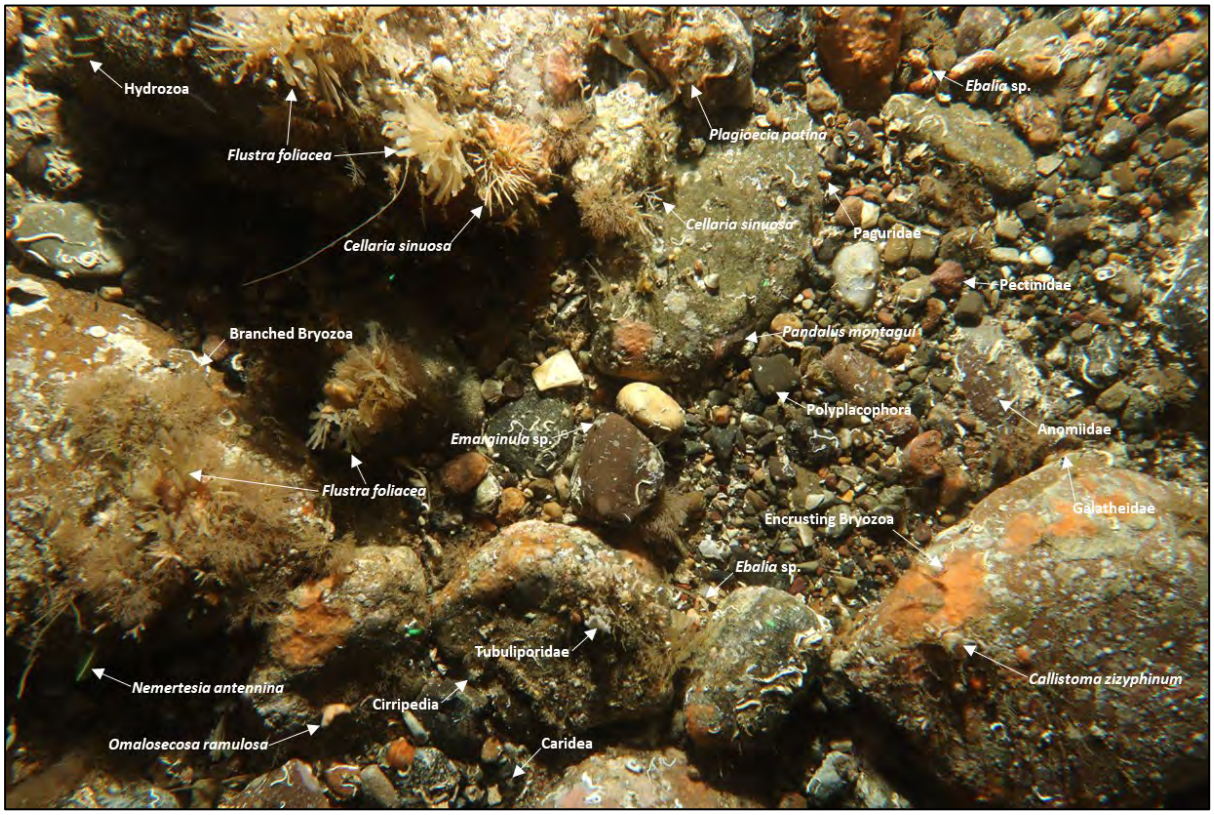


Figure 48 Site photo OI_164_GR_ENV_OAA_S10_4, presents the greatest species diversity from the visual survey.

5.7.1 Non-Colonial Epibenthic Fauna in Site Stills

The total abundance of the number of individuals recorded from the different phyla from the stills acquired is presented in Figure 49. The group "Others" included phyla Nemertea and Porifera. The most abundant phylum was Mollusca, followed by Arthropoda and Cnidaria, which contributed 37 %, 27 % and 18 % respectively. These three phyla contributed to 82 % of the recorded non-colonial individuals.

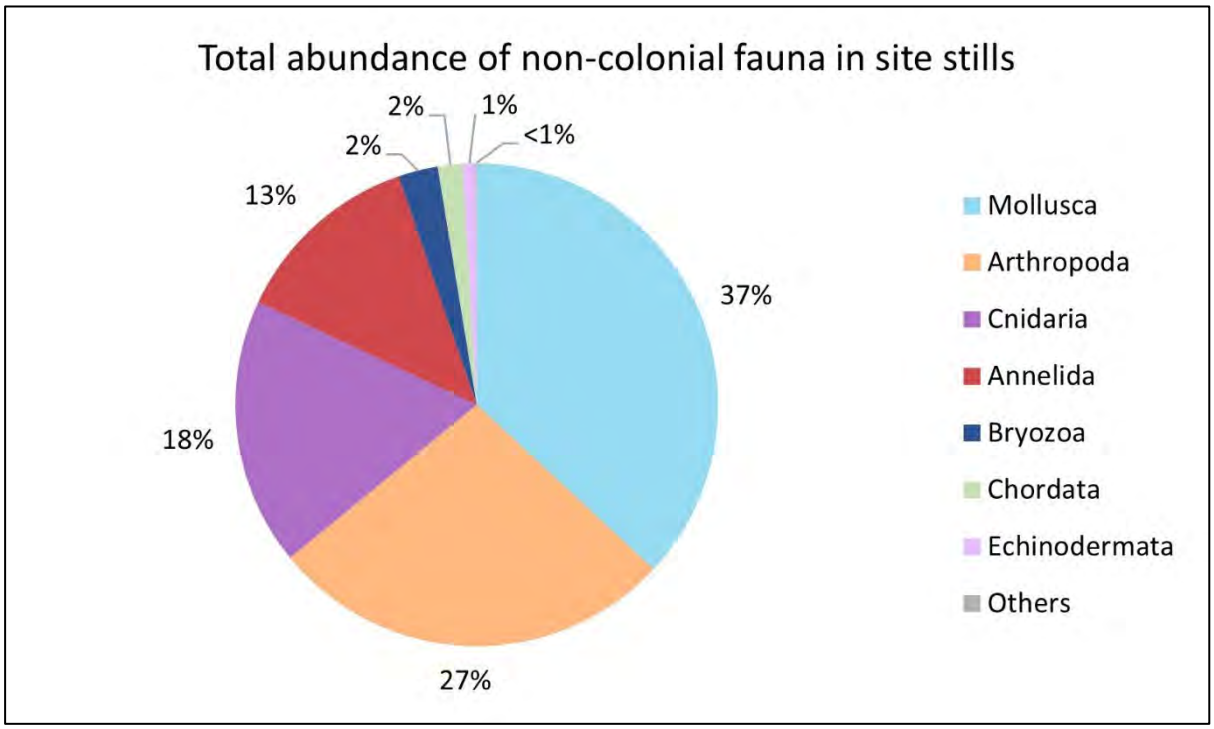


Figure 49 Total abundance of non-colonial fauna in site stills.

The average abundance, expressed as individuals per square meter (ind./m^2), varied from zero (0) (ind./m^2) to 371 (ind./m^2) in OAA_T47. The average non-colonial fauna abundance per site was 65 (SD=89) (ind./m^2). The average abundance expressed as (ind./m^2) for each site is presented per phylum in Figure 50.

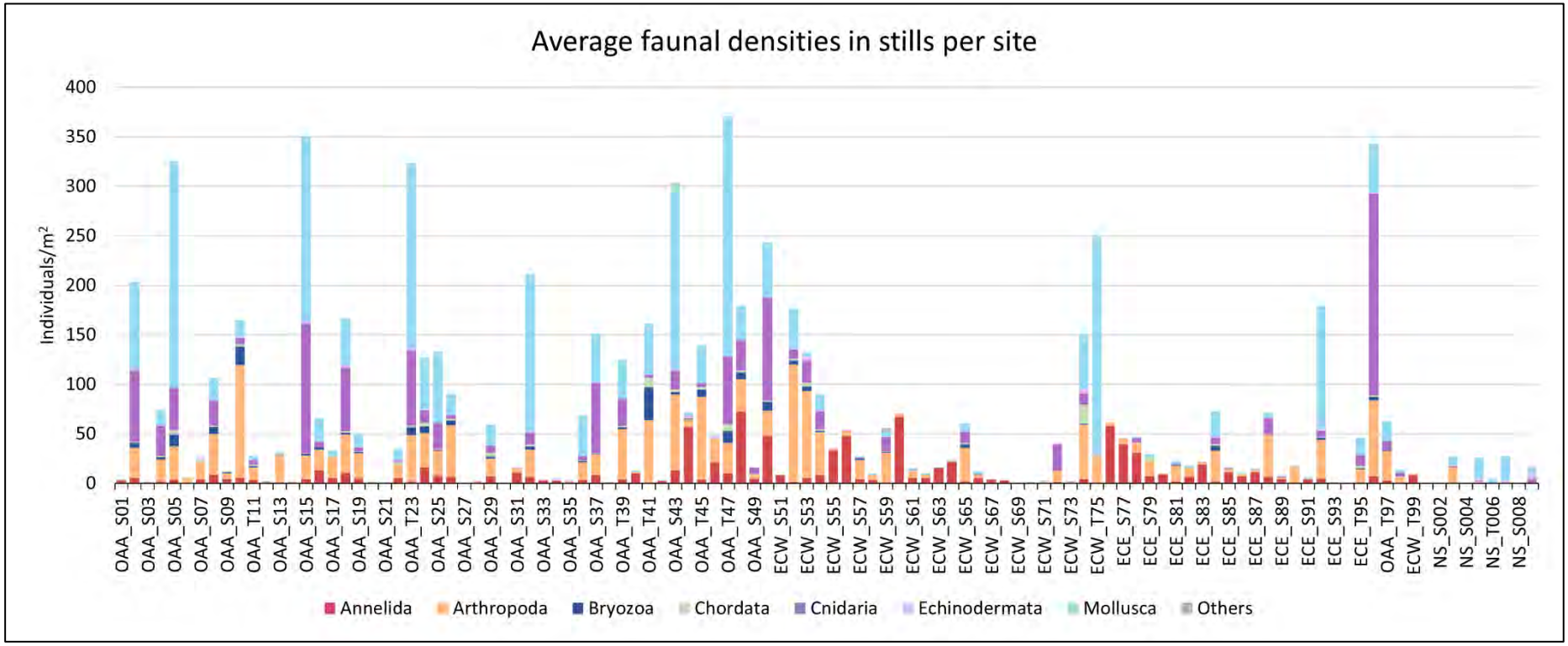


Figure 50 Average faunal densities (ind./m²) in stills per site.

5.7.2 Colonial Epifauna in Site Stills

The total coverage of colonial species, recorded in site stills is presented in Figure 51. The group "Others" included the phylum Annelida, Chordata and Mollusca. Phylum Bryozoa represented the phylum with taxa covering the largest surface area, with a total contribution of 61 %. Cnidaria, Ochrophyta and Arthropoda contributed 25 %, 5 % and 4 % of the recorded taxa, respectively. Rhodophyta, Porifera, and Others followed by 3 %, 1 % and 1 %, respectively. The coverage of colonial fauna varied from 0 % to 49 % (ECW_T74). The average colonial faunal coverage expressed per site was 8 % (SD= 11 %).

The average coverage per site, for each phylum, is presented in Figure 52. Bryozoa dominated the coverage of colonial fauna in site stills and a separate comparison was conducted excluding Bryozoa from the dataset. The average coverage per site, excluding Bryozoa is presented in Figure 53.

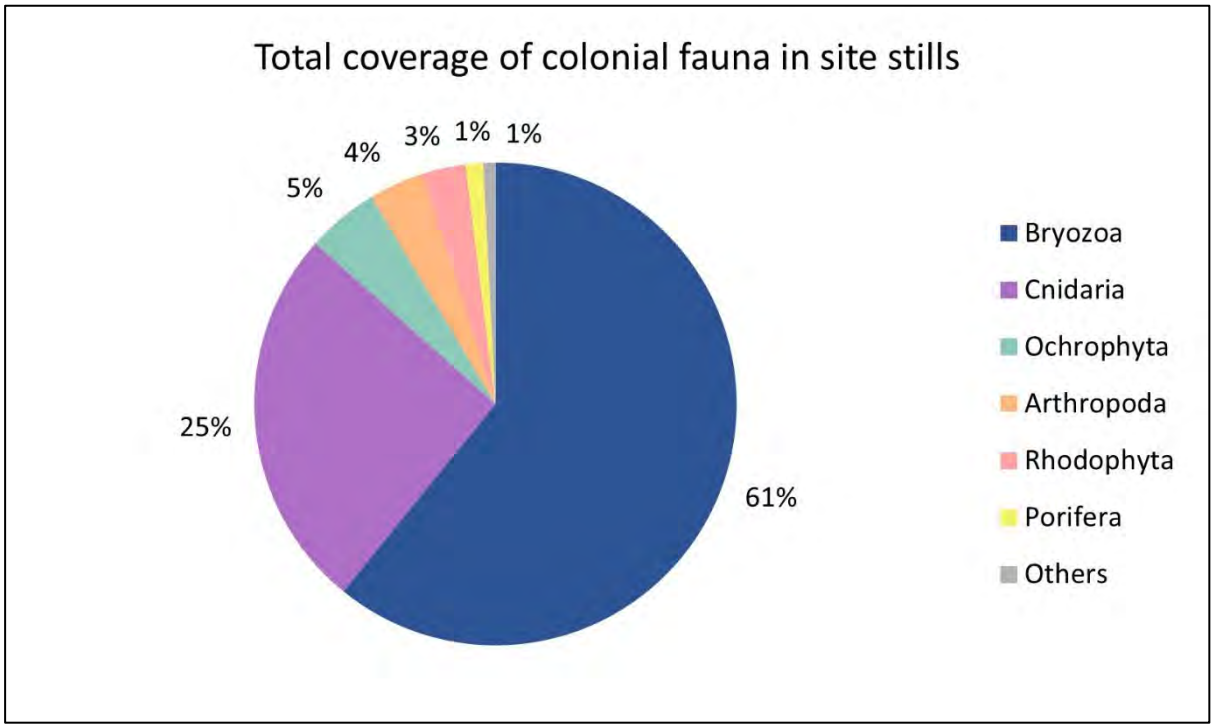


Figure 51 Total coverage of colonial fauna in site stills.

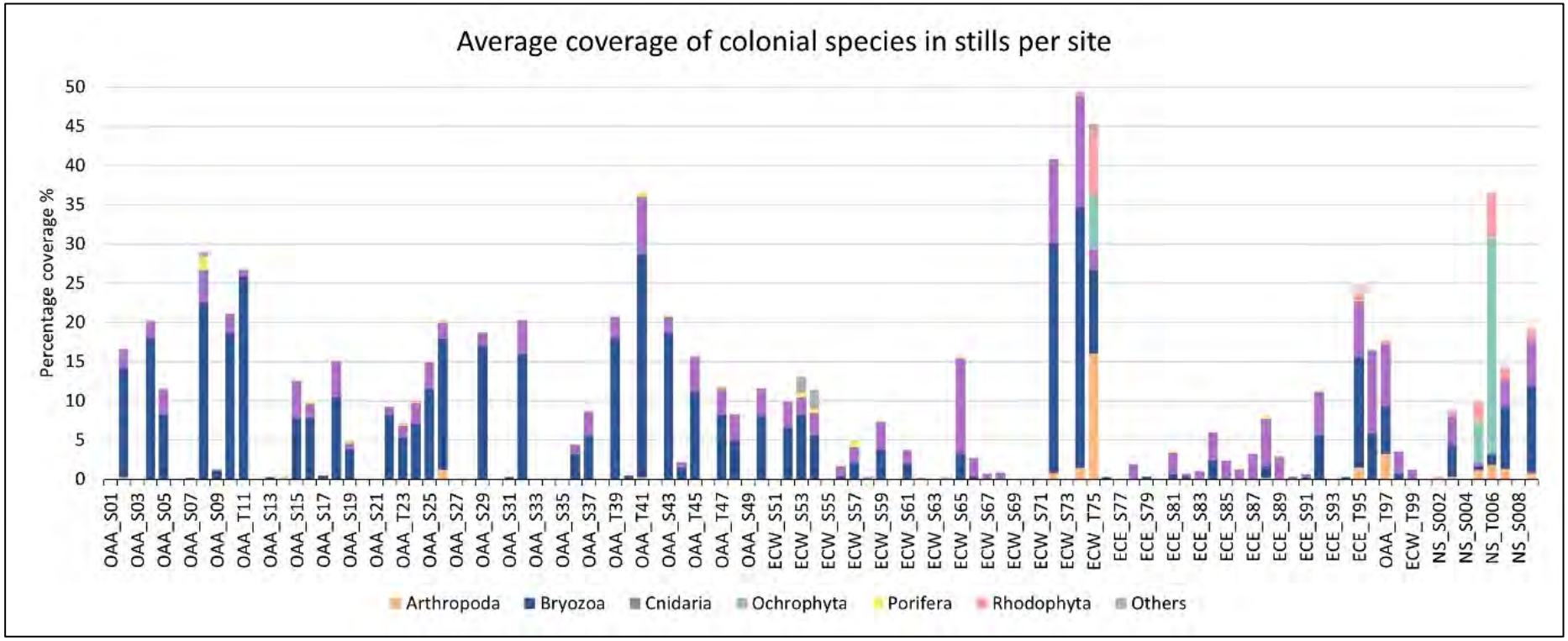


Figure 52 Average percentage coverage per m² for colonial fauna in stills per site.

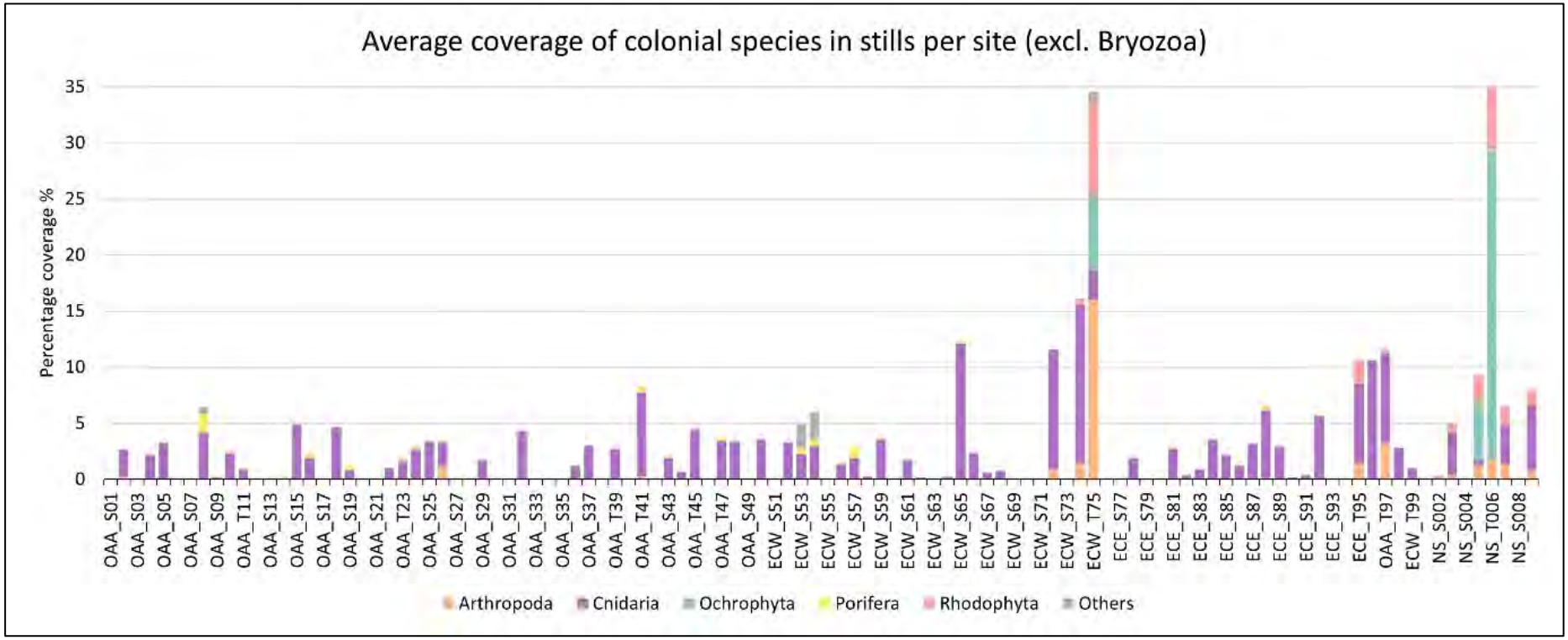


Figure 53 Average percentage coverage per m² for colonial fauna in stills per site, excluding the phylum Bryozoa.

5.8 CTD and Turbidity

Water sampling was planned offshore at 20 sites, with four (4) of these sites sampled during neap and spring tide, in addition to five (5) nearshore sites. CTD profiles and water samples were successfully sampled from all planned sites. An overview map of the water sample sites is presented in Figure 54 with a summary of the CTD profile values and turbidity results presented in Table 43.

The plotted CTD and TSS profiles are presented in detail in Appendix H with positions detailed in Appendix B.

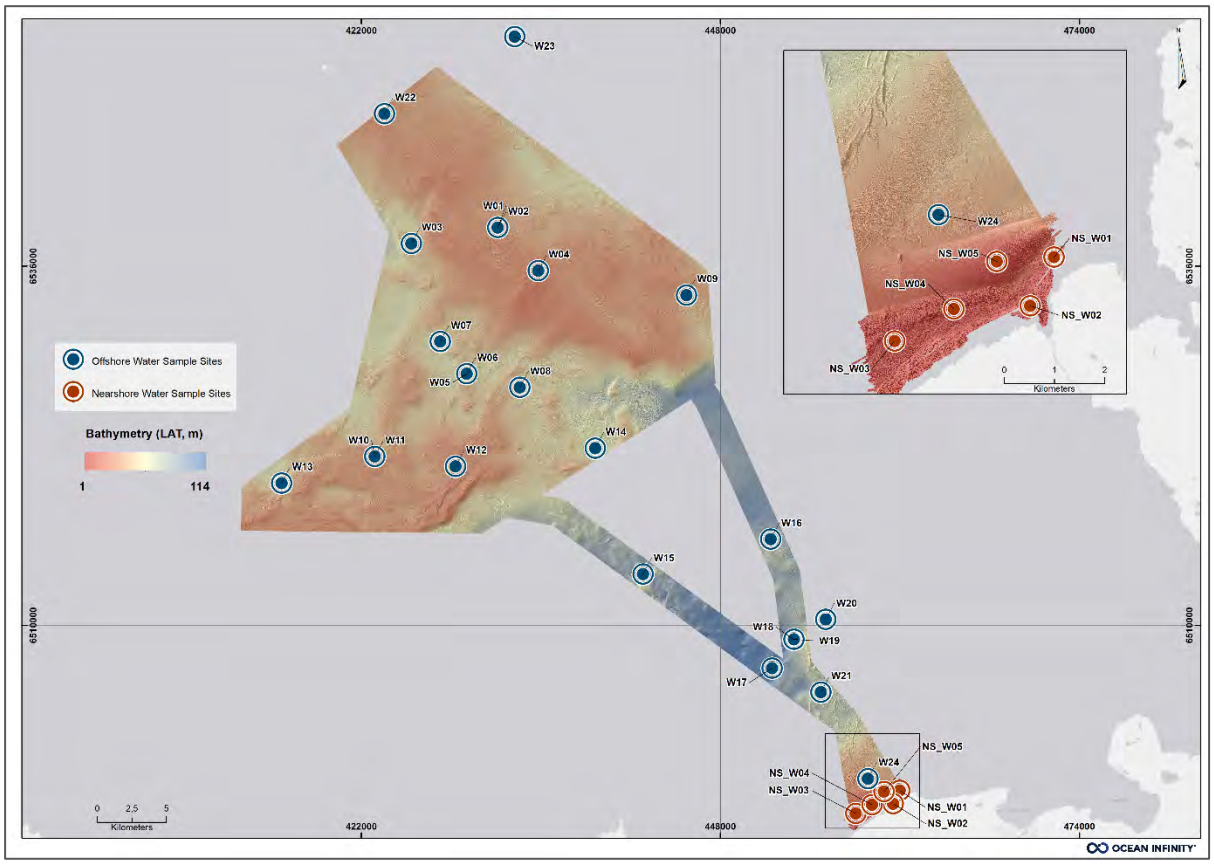


Figure 54 Overview of the distribution of water sample sites.

The results presented various levels of salinity ranging from 30.7 – 35.2 PSU, with an average salinity of 34.8 PSU. This aligns with historical measurements in Scottish waters (33.1 – 35.2 PSU) (Marine Scotland, 2020). Density ranged between 1021.5 – 1026.8 kg/m^3, temperature between 12.3 – 14.2 °C and turbidity between -1.4 – 15.1 mg/l. The trends exhibited in the CTD profiles for salinity, temperature, density, and turbidity were consistent across sampling sites.

The analysed water samples show clear conditions and low turbidity in the OAA and the ECC. Values were below 1 NTU at most of the sites and were occasionally above 3 NTU. The values were occasionally negative.

The turbidity sensor 'Eureka Trimeter c/w Turbidity 200M' has a stated minimum accuracy of 0.3 NTU. When there are no particles present in the water column to reflect light from the sensor, the instrument can indicate close to zero NTUs and with an accuracy of 0.3 NTUs the results yield can be displayed as negative.

Turbidity data has been converted from NTU into mg/l by multiplying by a conversion factor (3.4216).

Table 43 Summary of the CTD profile and Turbidity results of water sample sites.

Salinity (PSU)		Density (kg/m^3)		Temperature (°C)		Turbidity (mg/l)	
MIN	MAX	MIN	MAX	MIN	MAX	MIN	MAX
30.7	35.2	1021.5	1026.8	12.3	14.2	-1.4	15.1

5.8.1 Total Suspended Solids

In addition to the sensory turbidity measures, water samples were collected to analyse levels of Total Suspended Solids (TSS) in the Top, Middle and Bottom of the water column (Figure 55). The majority of the water samples showed values below 5 mg/l, which is below the instrument threshold and are therefore not presented in Figure 55. Sites with higher values were all located along the ECC and in particular in the area where the two routes intersect and join (W15 – W21). The TSS values ranged between <5 mg/l – 35 mg/l at site ECW_W15 Top.

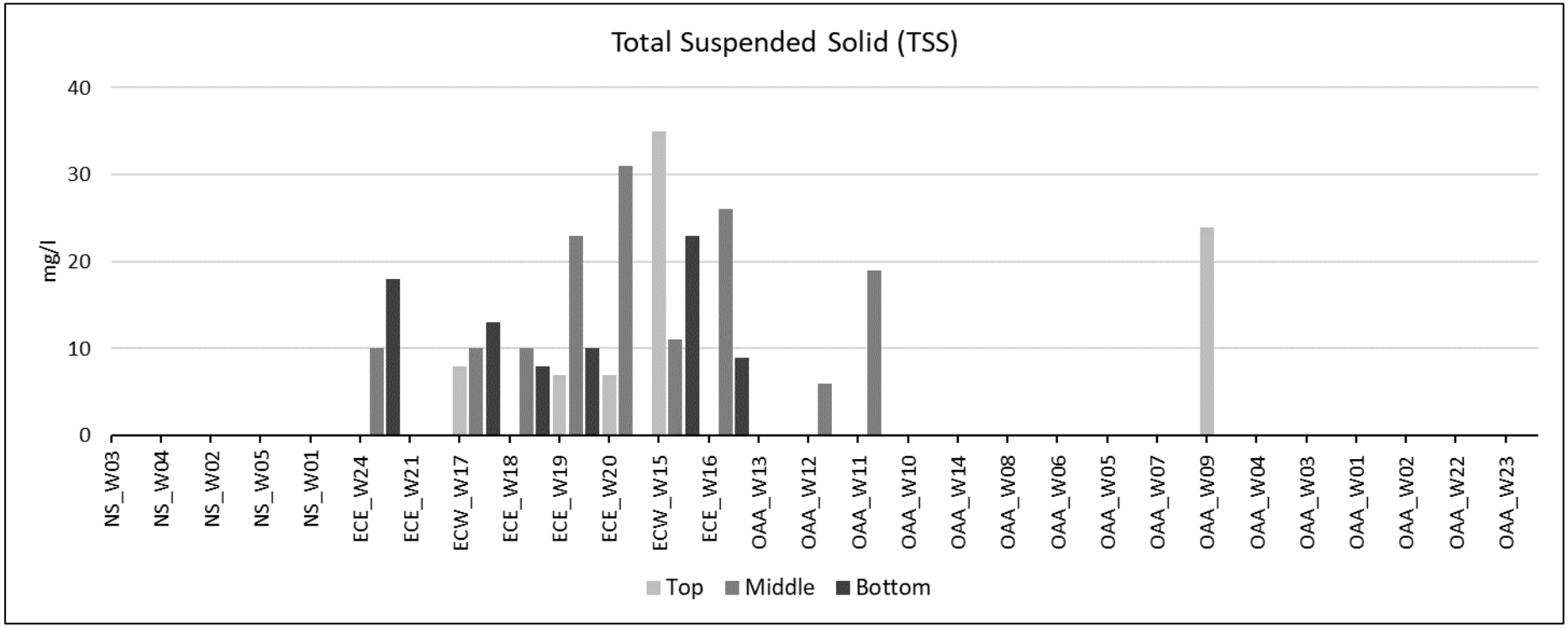


Figure 55 TSS levels (mg/l) for water samples sites, sorted by increasing northing.

5.9 eDNA

Water sampling for eDNA analyses was acquired at 20 sites (Figure 54) and samples were collected from the Top and Bottom of the water column at each of these sites. The results are presented in four standalone reports (Appendix I). A total of four (4) assays were targeted; Vertebrates (12S gene), Fish (12S gene), Marine Mammals (16S gene) and Eukaryotes which include Invertebrates (18S gene).

No comparisons between these analyses and the taxonomic analyses of the faunal grab samples have been conducted primarily due to that eDNA was not collected from sediment but from the water column.

The water column may hold genetic material from a specific species but that does not mean that the species is present. The water column may contain small amounts of DNA resulting in, too few detections, providing a below-threshold reading. Furthermore, the DNA may have been transported by another organism and dispersed through physical processes.

Any comparison could likely result in notable dissimilarities in quantities, false negatives and false positives. No eDNA sampling or analysis was carried out on the nearshore water samples.

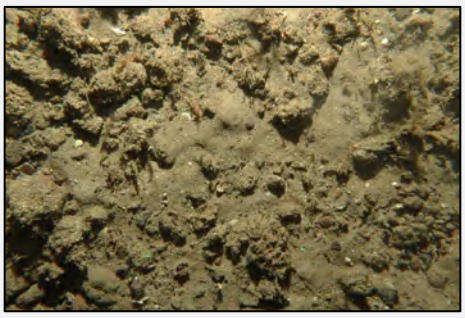
6. Potential Habitats and Taxa of Conservation Importance

The habitats and species corresponding to those defined in the EC's Habitats Directive, the OSPAR List of Threatened and/or Declining Species and Habitats, Scottish PMF, and SBL are listed in Table 44 and Table 57.

In Table 44, the Resemblance to Stony Reef (Low/Low to Medium/Medium/Potential) refers to the polygon that a site/transect is intersecting, not the resemblance a single individual site/transect has been assessed to hold (Section 6.1).

Table 44 Potential habitats of conservation identified within the survey area.

Image	Annex I	OSPAR/PMF/ SBL	Site ID
	Annex I (1170) Bedrock Reefs Habitats Directive		T74, T75 and T95 NS_T003, NS_T005, NS_T006, NS_T007 and NS_T009
	Annex I (1170) Stony Reefs Low Habitats Directive		S16
	Annex I (1170) Stony Reefs Low to Medium Habitats Directive		S04, S08, S18, S19, S24, S25, S26, S29, S36, S37, S43, S50, S54, S59 and S61 T11, T22, T41, T97 and T98
	Annex I (1170) Stony Reefs Medium Habitats Directive		S02, S05, S10, S15, S32, S48, S53, S65 and S84 T23, T39, T45, T47, T52, T72, T88, T96 and T97

Image	Annex I	OSPAR/PMF/ SBL	Site ID
	Annex I (1170) Potential Stony Reefs Habitats Directive		S57 and S84 T99
		Offshore Subtidal Sands and Gravels PMF Habitat & Subtidal Sand and Gravels SBL Habitat	S01, S03, S06, S07, S09, S12, S13, S14, S17, S19, S20, S21, S27, S28, S30, S31, S33, S34, S35, S38, S40, S42, S44, S46, S49, S51, S55, S56, S58, S60, S62, S63, S64, S66, S67, S68, S69, S70, S71, S73, S76, S77, S79, S80, S82, S86, S90. S91, S92, S93 and S94 T22, T72 and T99 NS_S001, NS_S002, NS_S004, NS_S008
		Kelp Beds PMF Habitat	T75 NS_T005, NS_T006

6.1 EC Habitats Directive

6.1.1 Stony Reefs

The stony reef areas were assessed in accordance with the criteria as outlined in JNCC Report No.432 (2009) and JNCC Report No.656 by Golding *et al* (2020) and Brazier (2020).

Guidance for standardising an approach to the assessment criteria is introduced by the JNCC in Report No 656 (Golding, Albrecht, & McBreen, 2020) to align the interpretation of Composition, Elevation, Extent and Biota, with regards to the application of Annex I Stony Reefs. Stony reefs are generally divided into Clast supported (cobbles neighbouring cobbles) and Matrix supported (intermediate fine sediments are present) reefs.

With regards to Composition, the guidance states that there should be at least 10 % rocky substrates with cobbles greater than 64 mm in diameter or boulders greater than 256 mm in diameter. These 10 % should be considered across the entirety of the area of interest or at the minimum extent of 25m^2. As most reefs are patchy by nature, and although the composition does not act as a direct measure of patchiness, it is advised that areas absent of cobbles and boulders be considered to account for the present patchiness.

Elevation is often ascribed to a distinct elevation compared to the surrounding environs but considered “distinct” when greater than 64 mm. It is further thought that a matrix-supported composition of reef, where the cobbles and boulders could be partially buried, may function as a reef and comprise associated reef communities whilst exhibiting an elevation smaller than 64 mm as long as the diameter is at least 64 mm.

When considering Extent, the area of interest should be a minimum of 25m^2 and include intermediate areas that may measure less than 25m^2.

Biota should be dominated by epifaunal species although it should be recognised that some areas that are subject to disturbance or sand scour, thus resulting in a limited epifaunal community, may still function as a reef. This criterion is under development to ascertain what type of communities most often are associated with reefs and whether any specific species contribute to the stability of the reef.

Should an area meet these four criteria it is considered to meet the minimum of Low Resemblance to Annex I Stony Reef, while if the area exceeds the Low Resemblance for a majority of these it would be considered to be a Medium Resemblance to Annex I Stony Reef.

For the purpose of mapping the Stony Reefs within this report, the assessment was divided into four categories of Stony Reefs resemblance: Potential Reef, Low Resemblance, Low to Medium Resemblance and Medium. Low to Medium Resemblance was assigned to larger areas exhibiting small-scale variation where the delineation between Low and Medium reef was not distinct. These large areas of Low to Medium Resemblance comprise sites and transects individually assess as Low or Medium Resemblance.

Areas of Low Resemblance were all deemed to qualify as Annex I, based on the guidance by Brazier and Golding *et al* (2020; 2020) with regard to reef habitats.

The assessments and delineations are based on ground truthing imagery and grab sample composition. A collective assessment was then extrapolated based on the textural similarity, as interpreted from SSS data, and elevation, as interpreted from MBES data to the surrounding areas.

Each site and transect was assessed on an individual level, and each acquired image for processing was assessed (Table 45) against Composition, Elevation, Extent, and Biota as outlined in the methodology in Section 4.7. Biota was divided into three categories; dominated by infauna, epifauna <80 % and epifauna >80 % as outlined in Irving (2009).

Table 45 Example of an individual site assessment at S48, per image acquired.

Sample ID	Elevation (m)	Composition (%)	Elevation vs Composition	Area m² (GIS)	Biota	Final Resemblance
OAA_S48_1	0.064-5	30	Low	>25	Epifauna <80 %	Low
OAA_S48_2	0.064-5	40	Medium	>25	Epifauna <80 %	Medium
OAA_S48_3	0.064-5	35	Low	>25	Epifauna <80 %	Low
OAA_S48_4	0.064-5	35	Low	>25	Epifauna <80 %	Low
OAA_S48_5	0.064-5	45	Medium	>25	Epifauna <80 %	Medium
OAA_S48_6	0.064-5	70	Medium	>25	Epifauna <80 %	Medium
OAA_S48_7	0.064-5	70	Medium	>25	Epifauna <80 %	Medium
OAA_S48_8	0.064-5	80	Medium	>25	Epifauna <80 %	Medium
OAA_S48_9	0.064-5	70	Medium	>25	Epifauna <80 %	Medium
OAA_S48_10	0.064-5	65	Medium	>25	Epifauna <80 %	Medium
OAA_S48_11	0.064-5	65	Medium	>25	Epifauna <80 %	Medium

The OAA area mainly comprises densely packed coarse sediments associated with ripple scour depressions as well as rocky substrates, supported by the backscatter data (Figure 56), which returned the highest intensity values (-7 to – 14 dB) in these areas. The signature decibel values do not deviate significantly from one another between these two categories thus no delineation based on decibel value was conclusive.

The overall high intensity of the backscatter impacted the delineations, with regards to areas of “Stony Reefs” as many of the areas comprising accumulation of densely packed coarse sand, gravel and pebbles have an equally, or occasionally, stronger return than the rocky habitats.

The southernmost areas of the OAA exhibit lower intensity (-17 to -27 dB), in line with ground truthing data, associated with finer sediments. Similar interval ranges are denoted in the ECC West and ECC East with a generally increasing intensity southward toward the coastline (Figure 57). Nearshore backscatter data was subject to operational and technical difficulties resulting in noise disturbances in the dataset and was excluded from this analysis.

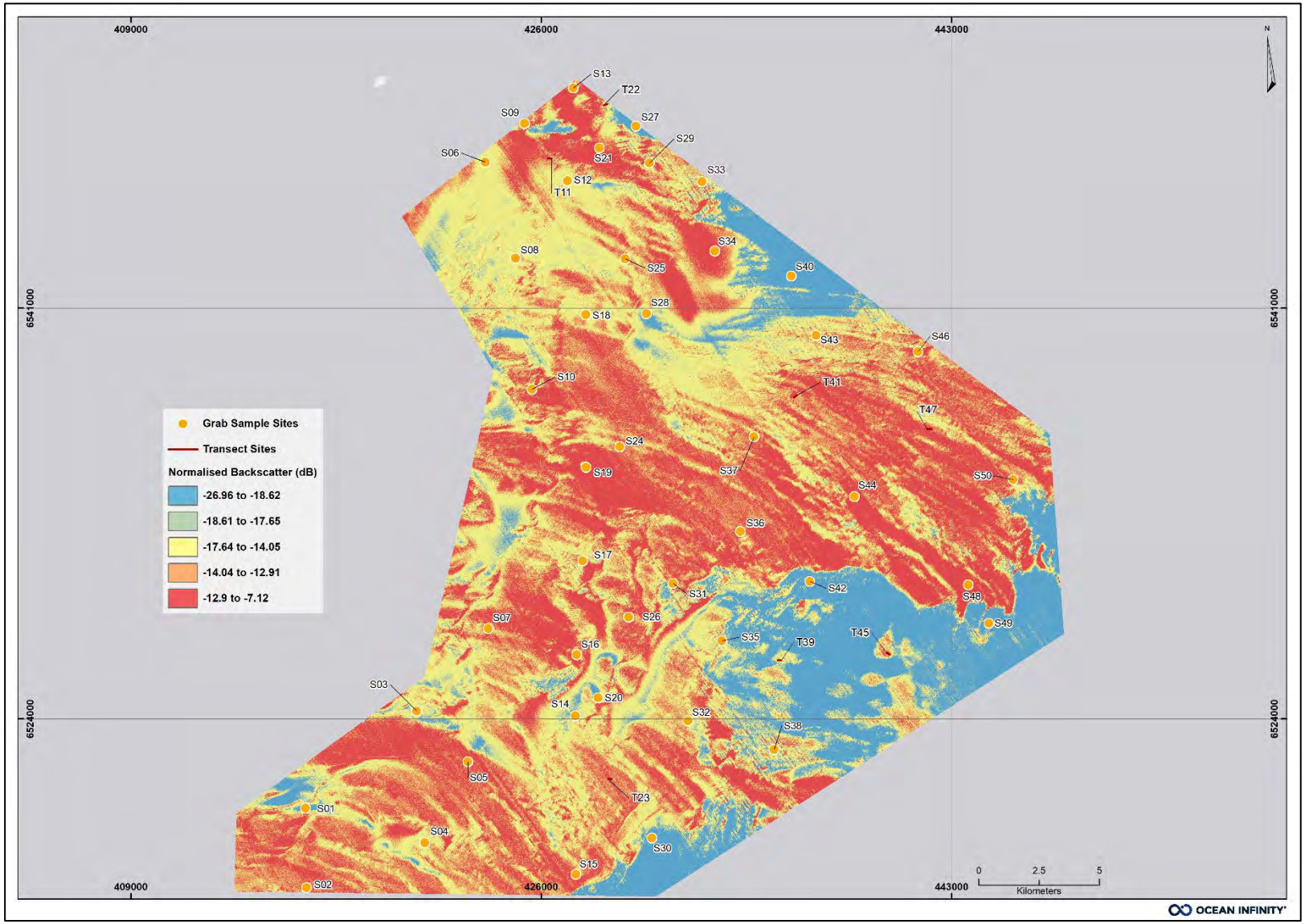


Figure 56 Overview of offshore backscatter data in the OAA.

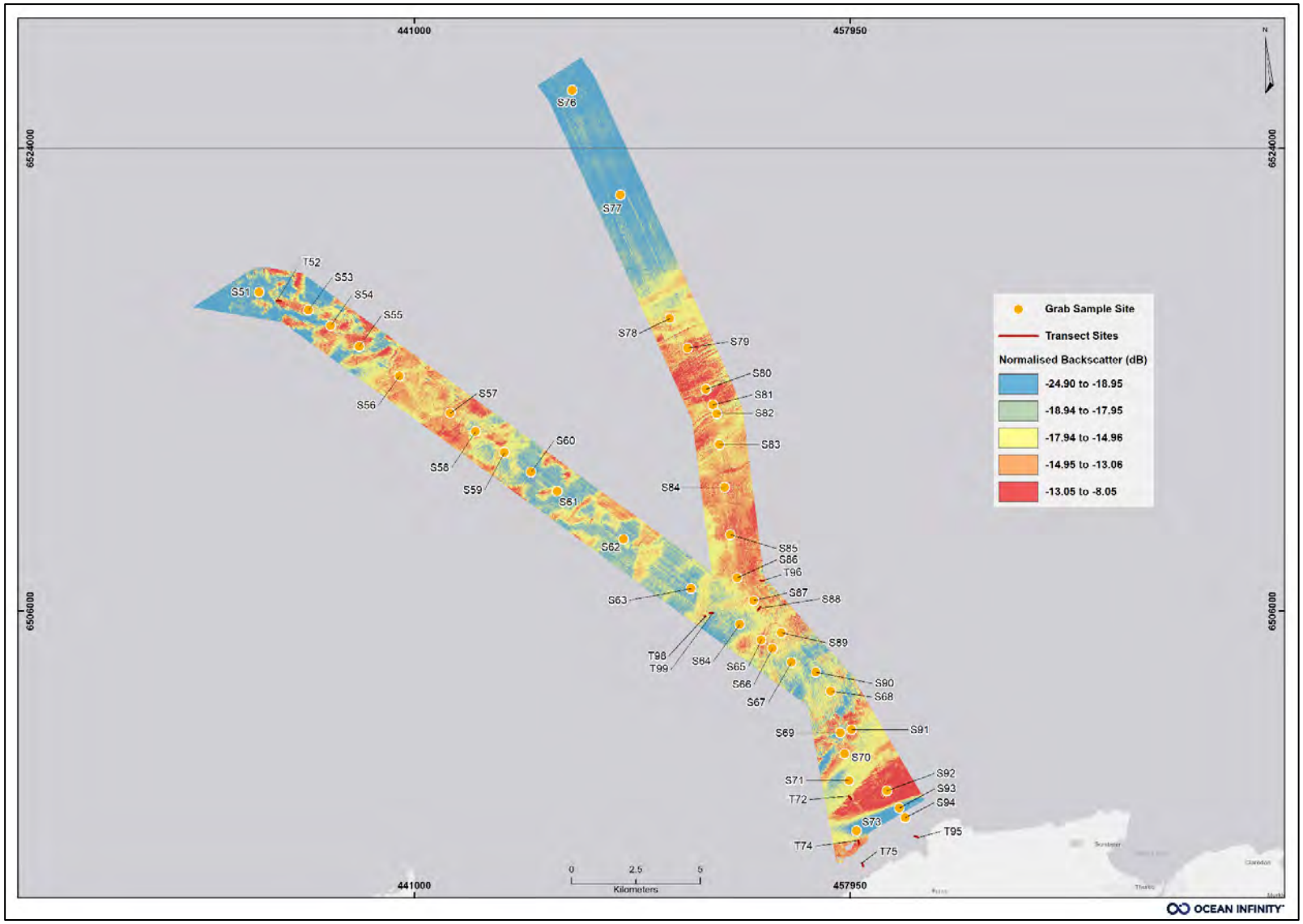


Figure 57 Overview of offshore backscatter data in the ECC East and ECC West. Due to technical and operational difficulties backscatter data is not available for the nearshore area. However, the nearshore area has been interpreted as described in Section 4.5.3.

Annex I (1170) Reefs – Potential Stony Reefs

Annex I (1170) Reefs – Potential Stony Reefs have been assigned in areas comprising a mosaic of mobile and non-mobile substrates as well as rocky substrates. These are primarily delineated based on interpretations of geophysical data. It is possible that areas within these boundaries would meet the extent and elevation criteria of Stony Reefs (Irving, 2009) based on the rocky nature of the survey area but a Resemblance assessment cannot be based on geophysical data solely.

The areas delineated as Potential Stony Reefs were primarily assessed based on the coarseness and density of rocky substrates present, as interpreted in the SSS data, as well as the textural similarity to areas assessed and ground-truthed as Stony Reef.

Areas of Potential Reefs have been identified throughout the OAA, mainly the western section, as well as the ECC West and ECC East route corridors (Figure 58).

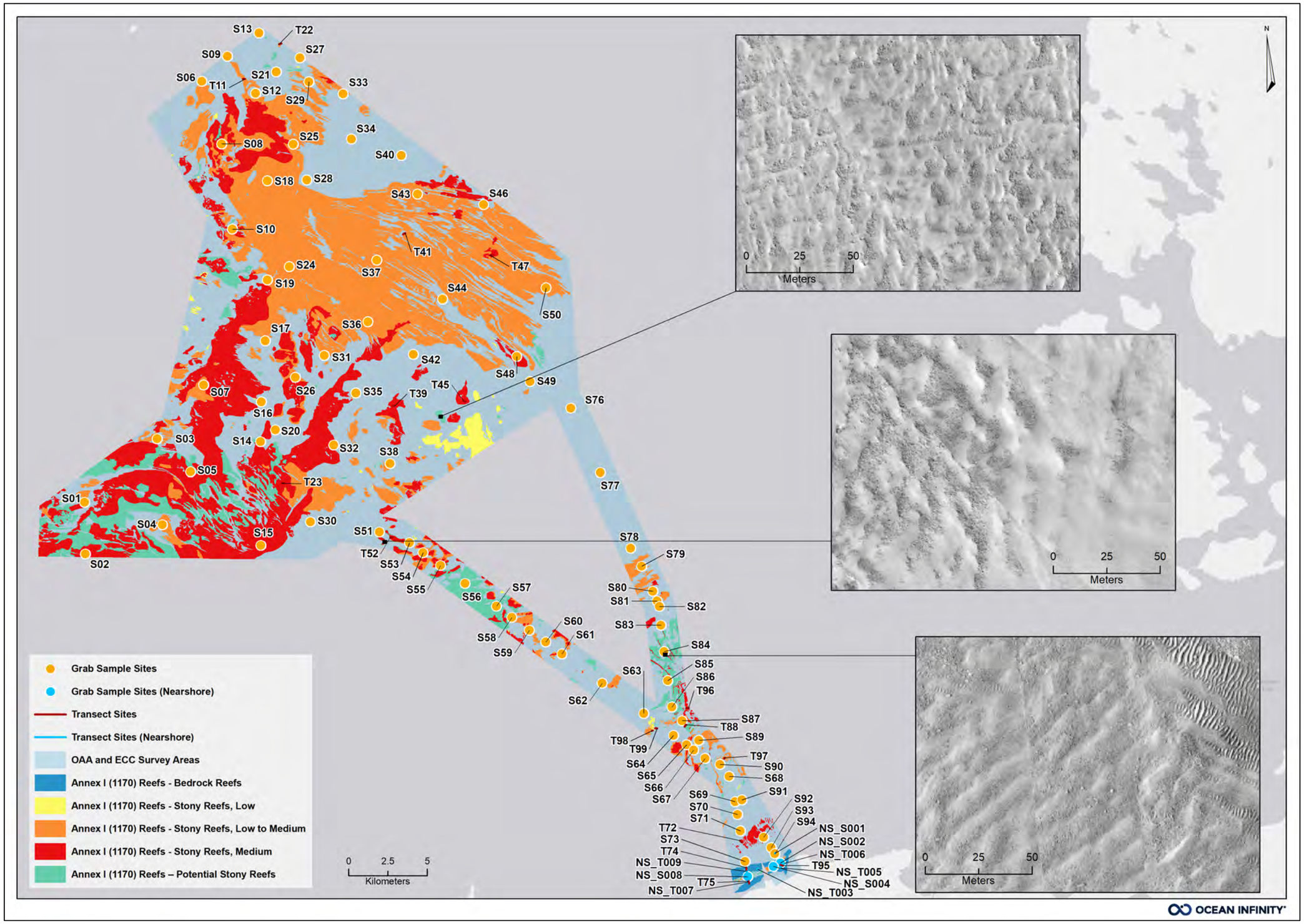


Figure 58 Example of SSS data at potential Stony Reefs areas.

Annex I (1170) Reefs – Stony Reefs, Low Resemblance

Annex I (1170) Reefs – Stony Reefs of Low Resemblance, have predominantly been classified in areas comprising mixed sediments that meet the Annex I criteria, as detailed by Goulding (2020) and Brazier (2020). Each image from each grab sample site and each transect was assessed individually against the four main criteria of elevation, composition, extent, and biota.

The majority of these Low Resemblance reefs are considered to be matrix-supported with an overall low composition, high patchiness, and very variable elevation at each of the sites (Table 46) with partly buried cobbles and boulders. Scattered smaller areas of Low Reefs have been identified throughout the OAA, ECC West and the area where the route survey corridors overlap. A larger area of Low Resemblance is located at the OAA's south-eastern border (Figure *59*). The interpreted Stony Reefs of Low Resemblance is further exemplified through the acquired ground truthing data from Table 47 to Table 50.

The fauna present was abundant and dominated to a high degree by scour-tolerant *Flustra foliacea*, *Securiflustra securifrons*, *Alcyonium digitatum*, and *Caryophyllia (Caryophyllia) smithii* including, and to a lesser degree, poriferans and hydrozoans.

Table 46 Summary of sites and transects of Low Stony Reefs resemblance.

Site ID	Elevation				Composition %				Final Resemblance
	Not a Reef	Low	Medium	High	0-9 Not a Reef	10-39 Low	40-94 Medium	95-100 High	
T11		X	X		X	X	X		Low
T22	X	X	X		X	X			Low
T88	X	X	X		X	X	X		Low
T98	X	X	X		X	X	X		Low
S04		X	X			X	X		Low
S08		X	X			X	X		Low
S16	X	X				X	X		Low
S18		X	X			X	X		Low
S19		X				X	X		Low
S24			X			X	X		Low
S25	X		X		X	X	X		Low
S26	X	X	X		X	X	X		Low
S29	X	X	X		X	X	X		Low
S36		X	X		X	X			Low
S37		X	X		X	X	X		Low
S44		X			X	X			Low
S53			X			X	X		Low
S54			X			X	X		Low
S57		X	X		X	X			Low
S59		X	X		X	X			Low
S61	X	X	X		X	X			Low
S92			X			X	X		Low

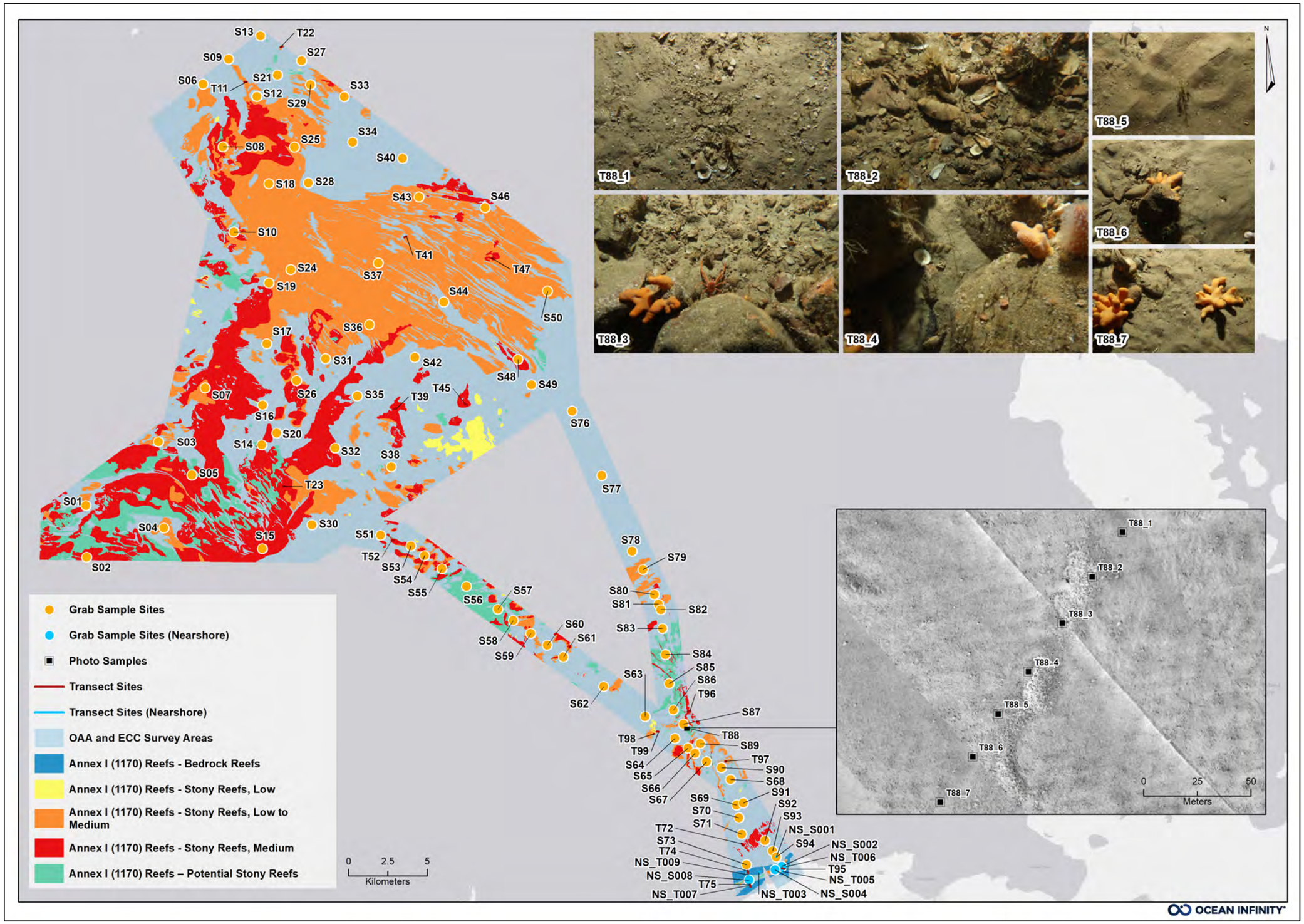


Figure 59 Overview of Annex I (1170) classed as Low Resemblance Reefs , together with example stills and SSS data at transects T88.

Table 47 Example imagery acquired every 5 m at S04.

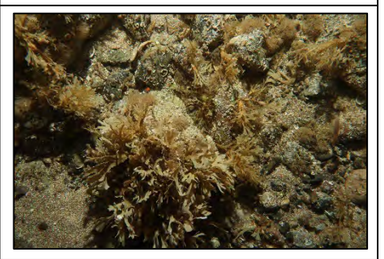
Site: S04 Area: OAA Image order: Left to right acquisition order					
					

Table 48 Example imagery acquired every 5 m at S08.

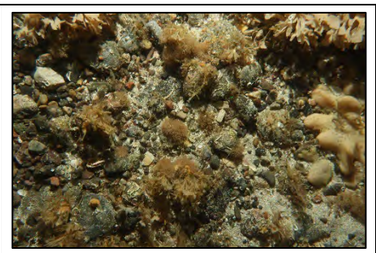
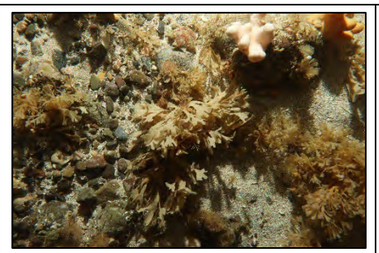
Site: S08 Area: OAA Image order: Left to right acquisition order					
					

Table 49 Example imagery acquired every 5 m at S16.

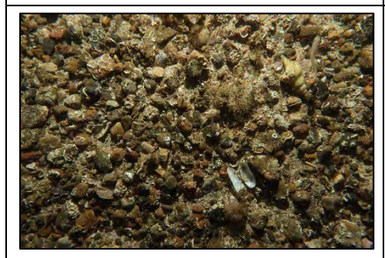
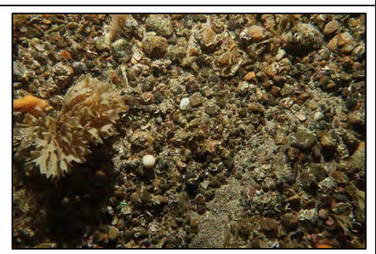
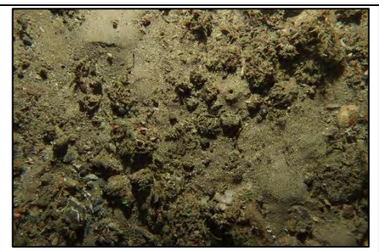
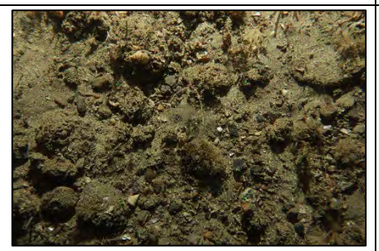
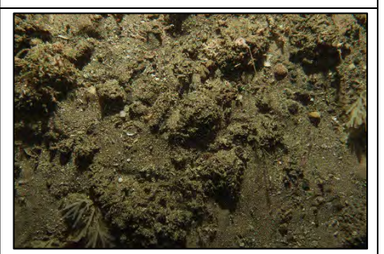
Site: S16 Area: OAA Image order: Left to right acquisition order					
					

Table 50 Example imagery acquired every 5 m at S53.

Site: S53 Area: ECW Image order: Left to right acquisition order					
					

Annex I (1170) Reefs – Stony Reefs, Medium Resemblance

Annex I (1170) Reefs – Stony Reefs, Medium Resemblance, have been classified in areas comprising homogenous rocky substrates. Each image from each grab sample site and each transect was assessed individually against the main criteria of elevation, composition, extent and biota.

The majority of these Medium Resemblance Reefs are clast supported with an overall medium composition and medium elevation, although variable, at each of the sites and transects (Table 51).

Areas of Medium Resemblance have been identified throughout the OAA and both route corridors, however, it has primarily been identified in the southwestern OAA (Figure 60). The interpreted Stony Reefs of Medium Resemblance are further exemplified through the acquired ground truthing data in Table 52 to Table 55.

The fauna present was dominated by a variety of bryozoan species, mainly *Flustra foliacea*, *Securiflustra securifrons*, and the mollusc Anomiidae. Cnidarian species were also prevalent and primarily constituted *Caryophyllia (Caryophyllia) smithii* and *Alcyonium digitatum*. Another common phylum was Arthropoda which predominantly comprised *Pandalus montagui, Munida sp*., Galatheoidea and Paguridae.

Table 51 Summary of sites and transects of Medium Stony Reefs resemblance.

Site ID	Elevation				Composition %				Final Resemblance
	Not a Reef	Low	Medium	High	0-9 Not a Reef	10-39 Low	40-94 Medium	95-100 High	
T23			X			X	X		Medium
T39	X		X		X	X	X		Medium
T41			X			X	X		Medium
T45			X			X	X		Medium
T47		X	X			X	X	X	Medium
T52			X			X	X		Medium
T72	X		X		X		X	X	Medium
T74			X				X	X	Medium
T96	X		X		X		X		Medium
T97		X	X			X	X		Medium
S02			X			X	X		Medium
S05		X	X			X	X		Medium
S10			X				X		Medium
S15		X	X			X	X		Medium
S32	X	X	X		X	X	X		Medium
S43	X	X	X		X	X	X		Medium
S48			X			X	X		Medium
S50			X			X	X		Medium
S65			X			X	X		Medium
S84	X		X		X	X	X		Medium

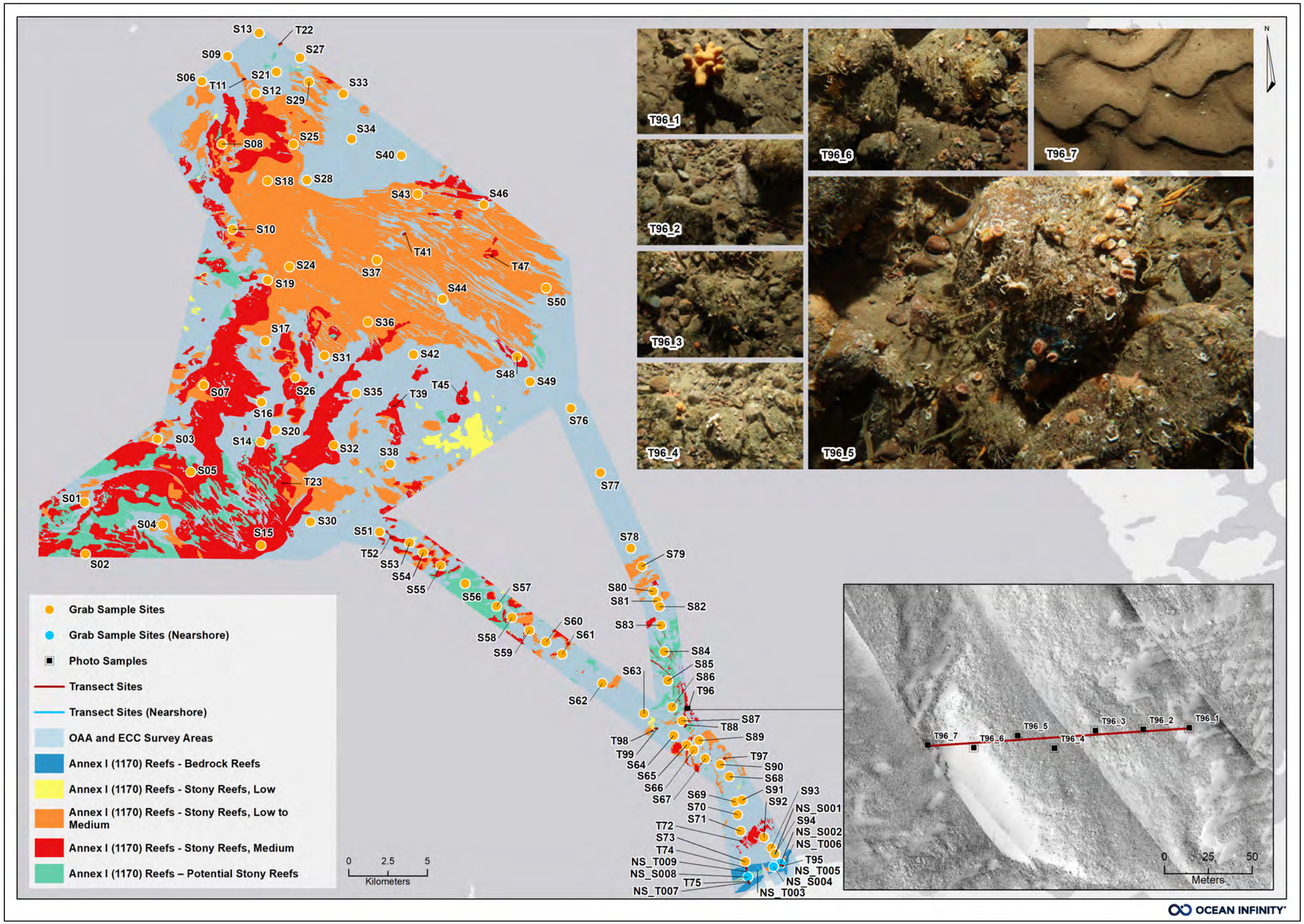


Figure 60 Overview of Annex I (1170) classed as Medium Resemblance Reefs, together with example stills and SSS data at transects T96.

Table 52 Example imagery acquired every 5 m at S02.

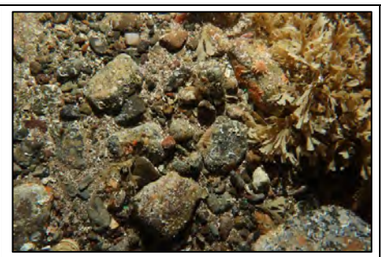
Site: S02 Area: OAA Image order: Left to right acquisition order					
					

Table 53 Example imagery acquired every 5 m at S15.

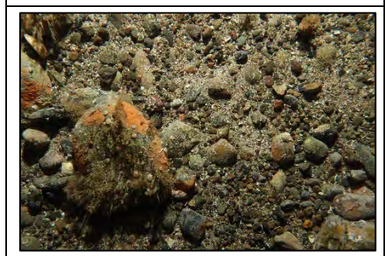
Site: S15 Area: OAA Image order: Left to right acquisition order					
					

Table 54 Example imagery acquired every 5 m at S65.

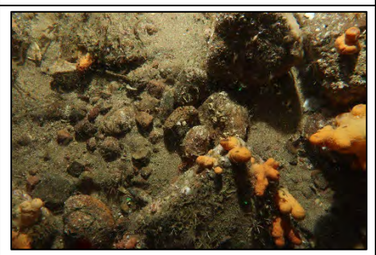
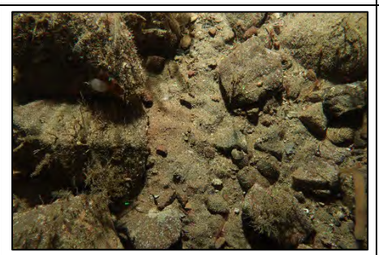
Site: S65 Area: ECW Image order: Left to right acquisition order					
						

Table 55 Example imagery acquired every 5 m at S84.

Site: S84 Area: ECE Image order: Left to right acquisition order					
						

Annex I (1170) Reefs – Stony Reefs, Low to Medium Resemblance

Annex I (1170) Reefs – Stony Reefs, Low to Medium Resemblance, has been delineated in areas comprising heterogeneous rocky substrates with varying density and/or elevation. Low to Medium Resemblance areas comprise individual transects and/or sites which have been assessed as either Low or Medium Resemblance but where the geophysical data is too similar and a clear boundary between these two categories cannot be established.

These areas have been identified across the OAA and both route corridors, including a large area of variably rocky seabed located in the central and northeastern OAA.

Rugosity

In an attempt to provide further information on the characteristics and patchiness of the "Low to Medium" Resemblance Stony Reefs within the eastern OAA, a Vector Ruggedness Measure (VRM) was utilised to create a seabed roughness model, Rugosity.

The model, created in ArcGIS, provides indicative trends with regard to seabed variability and was used to model the variability within the delineated area of the "Low to Medium" polygon. The approach aimed at quantifying and further differentiating between patches of Low and Medium Resemblance.

The interval values presented within this report are aligned and based on the ground-truthing imagery data and grab samples acquired during the survey. The value intervals have been divided into three categories of interpretation, Category I represents the Reef Features Likely Absent whereas Categories II and III represent Reef Features Likely Present (Table 56).

The VRM model provided indicative trends with regard to the likely presence of Stony Reefs (Figure 61), however, it should be noted that there are limiting factors due to the morphologically different ripple features present in the survey area which impacts the computed variability i.e., increasing it. This has, in part, been mitigated by only using the relevant data where all features not identified as Stony Reefs have been removed prior to the VRM being computed.

The VRM intervals were adjusted manually (by reviewing the statics and spread of values within different areas) based on the heterogeneity, or lack thereof, as interpreted from the ground-truthing-, visual- and acoustic datasets. Category I "Reef Features Likely Absent" was assigned to areas that appear rather featureless and are interpreted to comprise gravel and/or coarse sand.

Table 56 VRM model intervals.

VRM Values (Unitless)	ID	Description	Category
0.000001073 – 0.000045		Reef Features Likely Absent	I
0.000045 – 0.00023		Reef Features Likely Present	II
0.00023 – 0.0027		Reef Features Likely Present	III

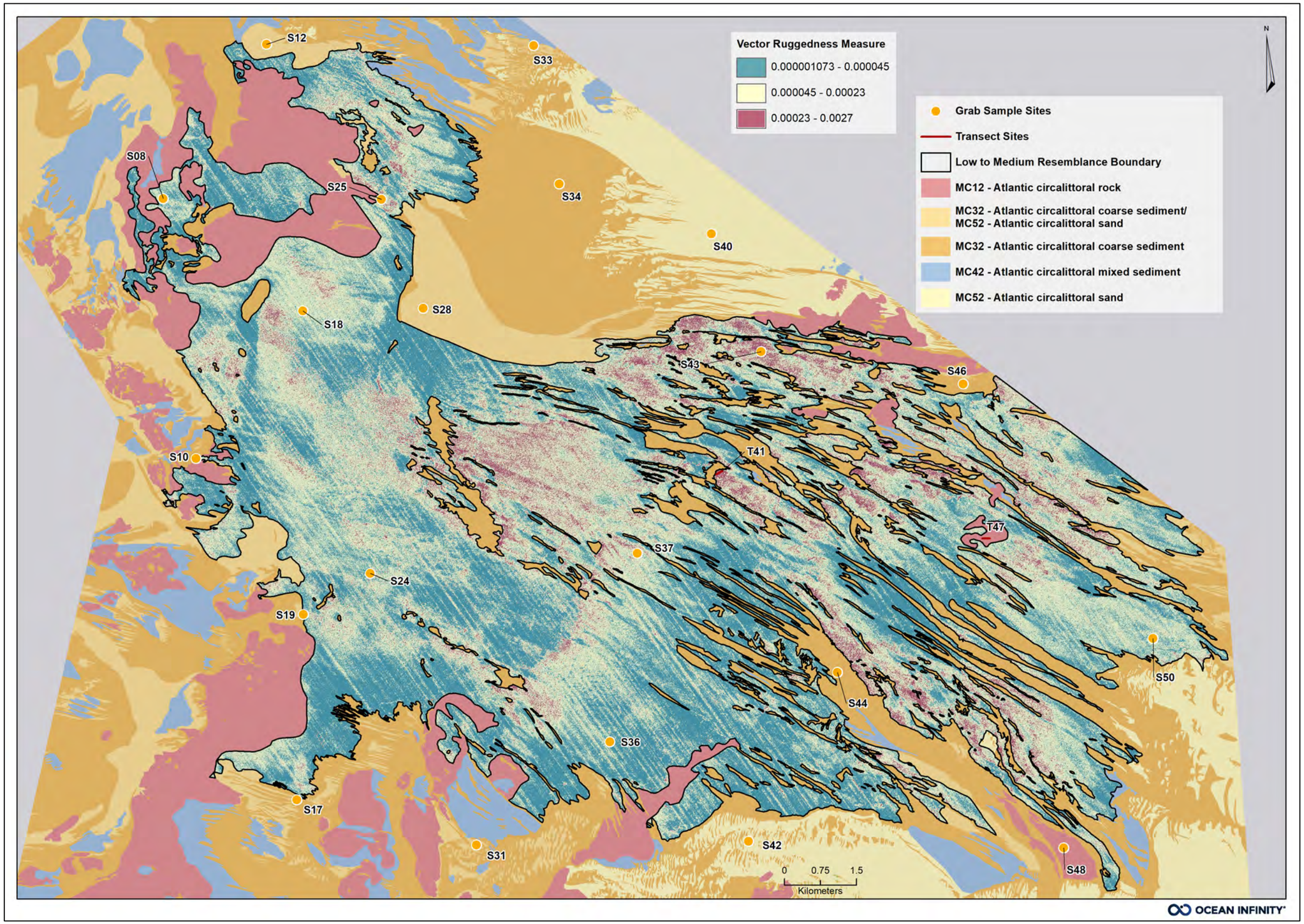


Figure 61 VRM model superimposed on the classified habitats within the eastern OAA.

The presence of Stony Reefs, based on acquired data, is interpreted to be represented by higher relative variability within the modelled area. The produced model indicates that variability can be tied to Stony Reefs, to an extent (Figure 62). While it is not possible to delineate between the Low and Medium Resemblance, mainly due to patchiness being a natural progression, the VRM indicates that the flattest areas i.e., lowest variability, Category I (Figure 63), are unlikely Reefs. Category II are areas interpreted as likely to be associated with Reefs. Category III, also likely to be associated with Reefs, is interpreted to represent mottled seabed features, as noted in the SSS, and where rapid variations on a small scale were further noted in the MBES data (Figure 64).

The extrapolated and delineated seabed surface assessed as Low and Medium Stony Reefs Resemblance covered an area of approximately 156 km^2. Although established that the reef features within this area are patchy a delineation on that level was not feasible. The Rugosity, and the inherent variability interpreted to be associated with the ground-truthed Stony Reefs, indicates that the likely coverage of Stony Reefs within this area covers an area of approximately 93 km^2, 60 % of the area.

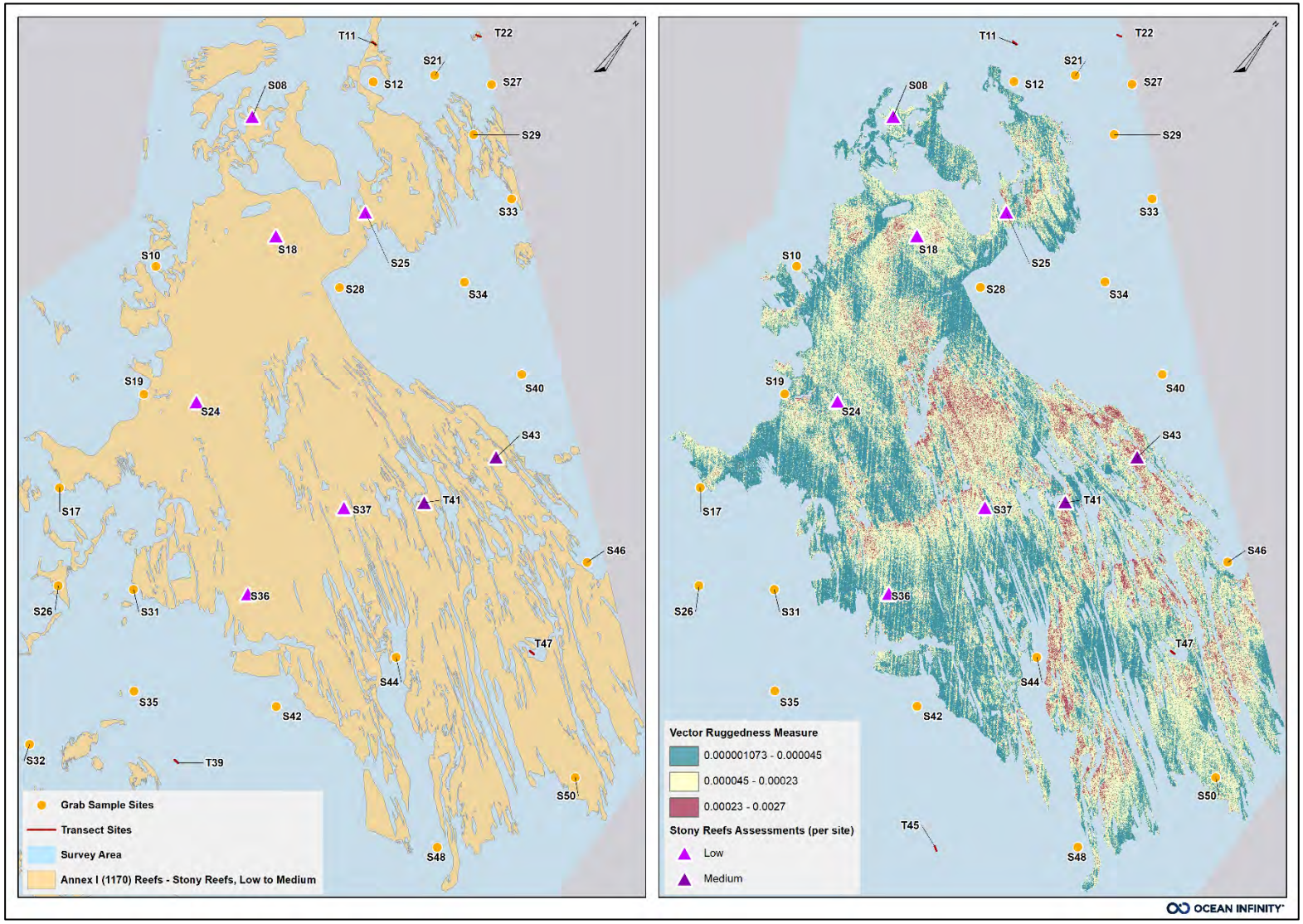


Figure 62 Comparison of delineation of the Low to Medium Resemblance Stony Reefs.

Note: The data frame has been rotated 320 degrees to allow for better comparability of the charts.

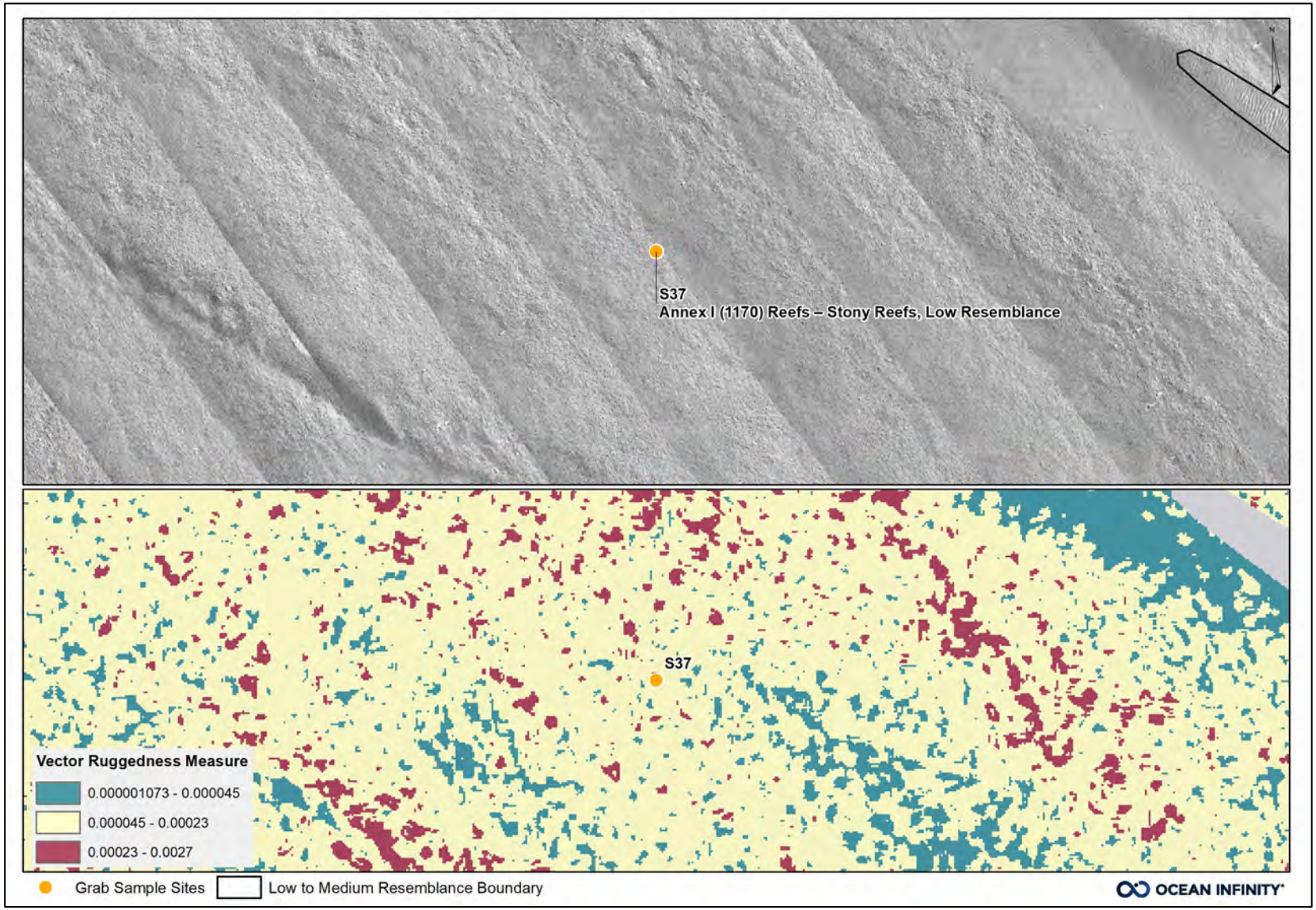


Figure 63 Site S37 illustrated with SSS and VRM.

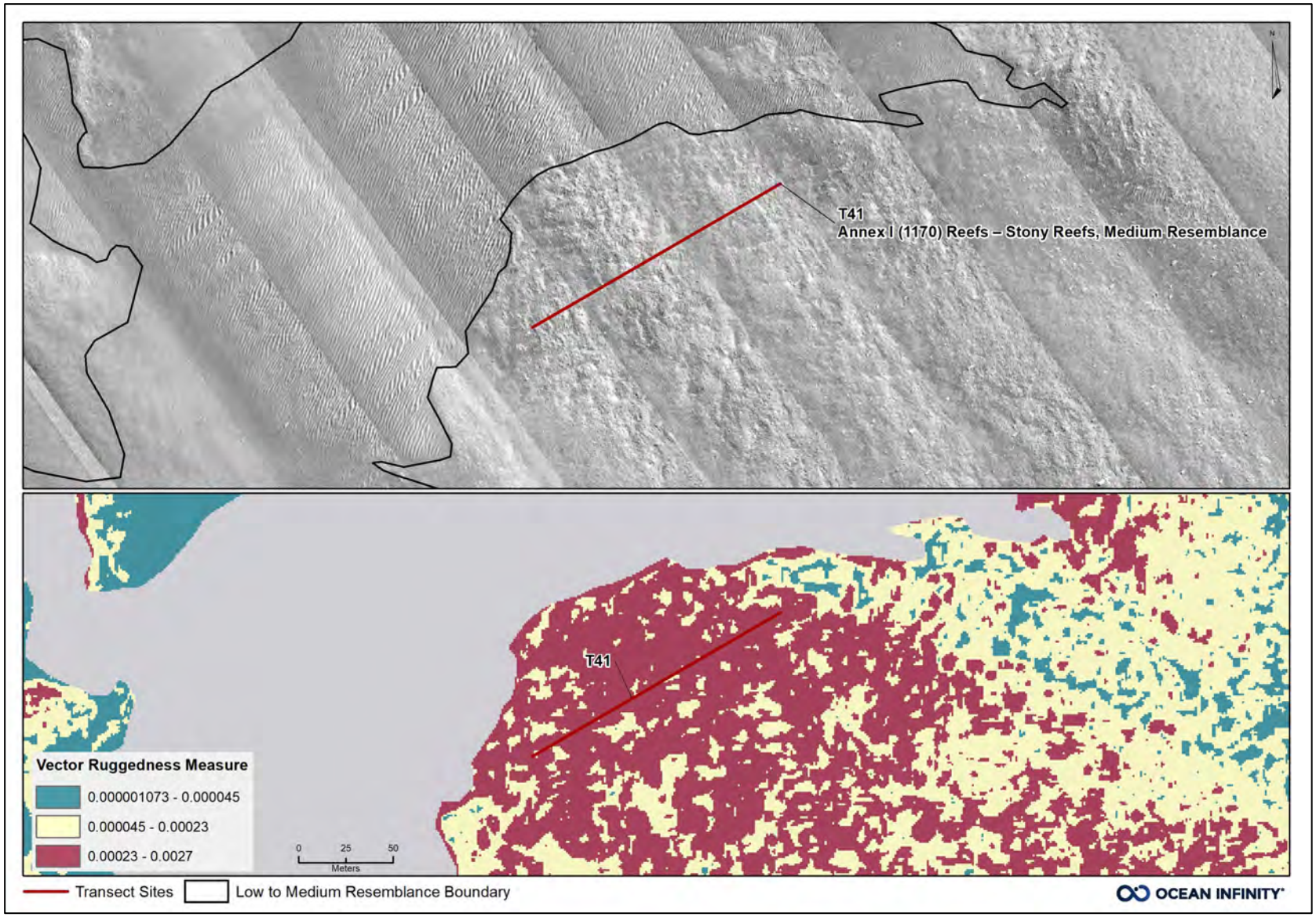


Figure 64 Transect T41 illustrated with SSS and VRM.

The 10 most abundant taxa at sites/transects present at Stony Reef features are presented in Figure 65 and Figure 66. The most abundant taxa in the non-colonial epifauna densities identified in stills were Anomiidae. Site OAA_S43 presented the highest average abundance at the sites/transects present at stony reef features. *Flustra foliacea* was the dominating colonial taxa identified in stills. Site OAA_T41 presented the highest average coverage at the sites/transects present at Stony Reef features.

A full list of the species identified in sites/transects is presented in Appendix C and Appendix D.

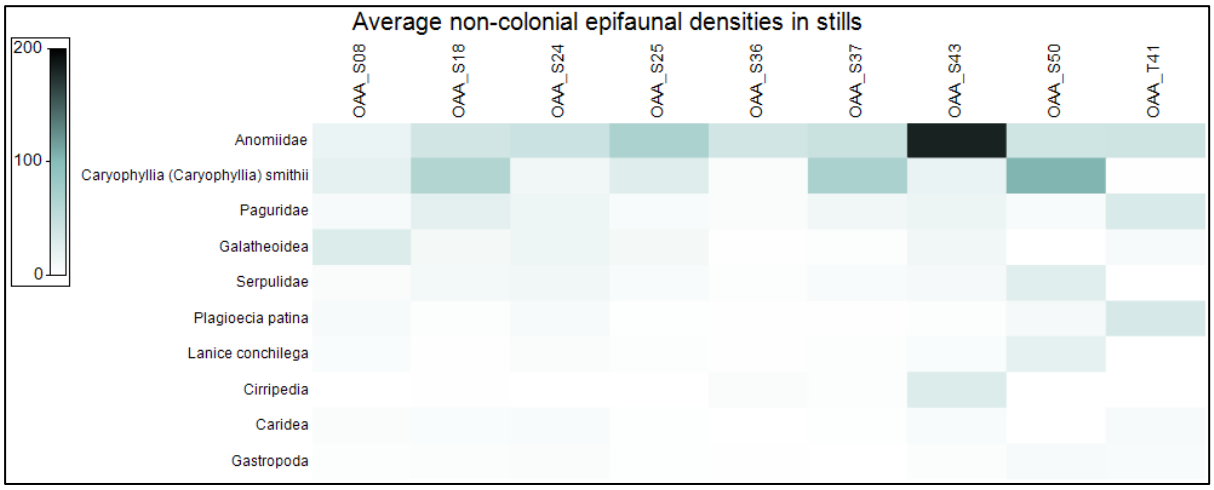


Figure 65 Heatmap illustrating the 10 most abundant non-colonial epifaunal taxa in stills.

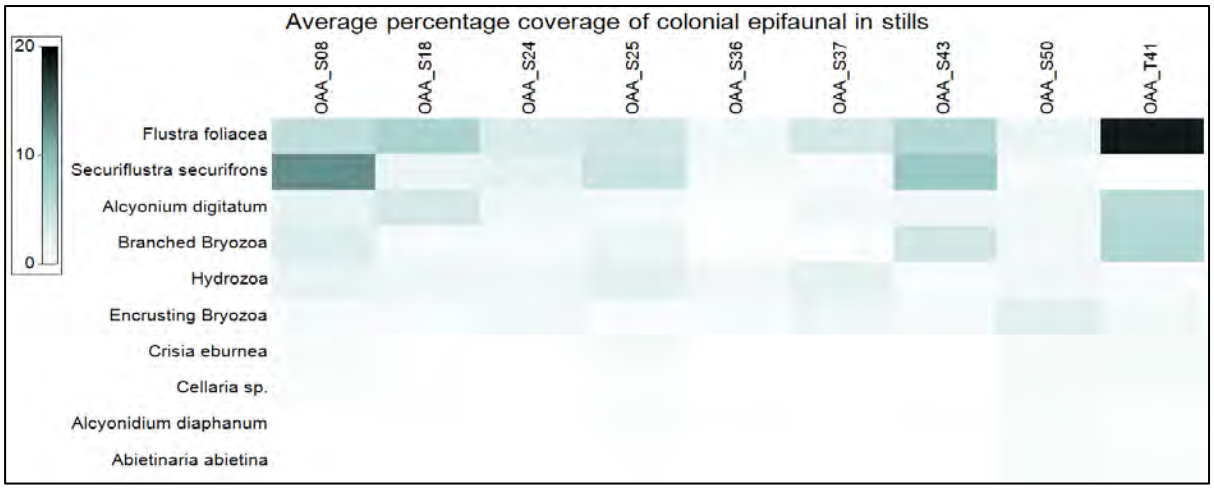


Figure 66 Heatmap illustrating the 10 most abundant colonial epifaunal taxa in stills.

6.1.2 Bedrock Reefs

Annex I (1170) Reefs – Bedrock Reefs have been classified in the areas bordering both route corridor landfalls (Figure 67). Two extensive areas covering the width of both route corridors were classified as **MB12** – Atlantic infralittoral rock and **MC12** – Atlantic circalittoral rock.

The bedrock is predominantly exposed and where covered the surface composition is variable, from veneers of sand and gravel, cobble and boulders to Kelp and Hydrozoan turfs.

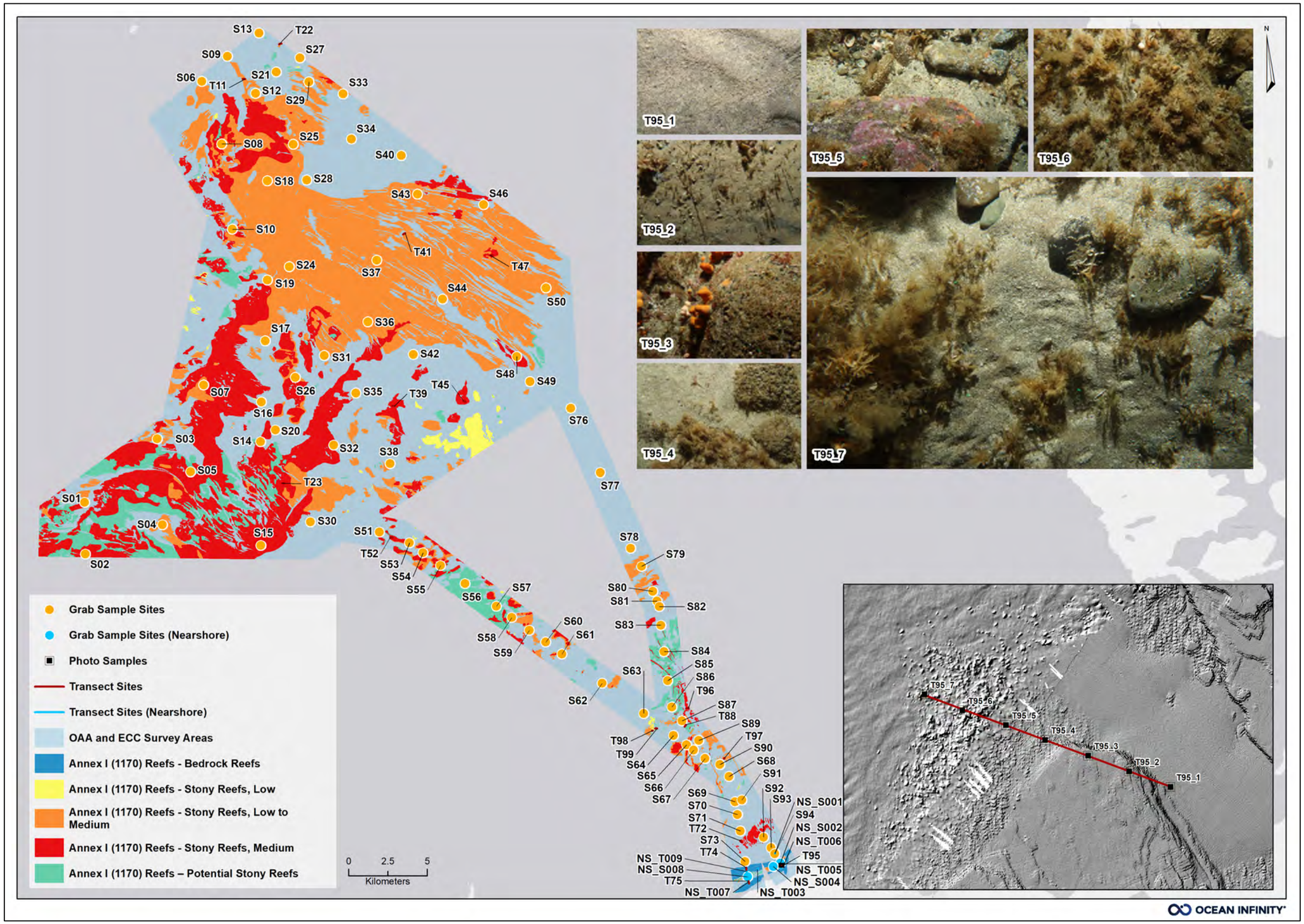


Figure 67 Overview of Annex I (1170) Bedrock Reefs, together with example stills and MBES Hillshade Relief data at transects T95.

6.1.3 Biogenic Reefs

Ross worm Sabellaria spinulosa

Ross worm *S. spinulosa* was identified at five (5) locations (S26, S53, S54, T45 and T52) primarily encrusting on cobbles and boulders. Three of these sites (S53, S54 and T52) are classified as **MC128** – *Sabellaria* on Atlantic circalittoral rock. Site S26 was classified as **MC42** – Atlantic circalittoral mixed sediment and transect T45 was classified as **MC12** – Atlantic circalittoral rock.

Due to the habitat exhibiting the same geophysical signature as the surrounding rocky habitat, **MC12**, no delineations nor extrapolation was applied. Furthermore, the identified *S. spinulosa* did not qualify as Annex I (1170) – Reefs, Biogenic Reefs due to the fact that the supporting substrate is geogenic and not biogenic.

Additionally, a total of 90 individual specimens of *S. spinulosa* were identified at eight (8) grab sample sites; OAA_S08, OAA_S16, OAA_S19_US, OAA_S26, ECW_S54_US, ECW_S57, ECW_S59 and ECE_S81.

6.2 Other Habitats of Conservation Importance

Kelp Beds (PMF)

The PMF habitat Kelp beds are classified at transect T75 as well as nearshore transects NS_T005 and NS_T006, corresponding to EUNIS habitat **MB121** – Kelp and seaweed communities on Atlantic infralittoral rock. Transect T75 is located on the exposed bedrock close to the landfall of ECC West and NS_T005 and NS_T006 are located on the exposed bedrock close to the landfall of ECC East.

Offshore Subtidal Sands and Gravels (PMF) and Subtidal Sand and Gravels (SBL)

The PMF habitat Offshore subtidal sands and gravels and SBL habitat Subtidal sands and gravels were both identified across large parts of the OAA and the majority of both ECC East and ECC West (Tyler-Walters, et al., 2016; Scottish Biodiversity Forum, 2012). It is the most common subtidal habitat around the British Isles and includes a wide variety of sediments across a wide depth range. All subtype habitats of **MC32** – Atlantic circalittoral coarse sediment and **MC52** – Atlantic circalittoral sand including habitat complexes where such habitats are included have been interpreted to qualify as Subtidal Sands and Gravels.

6.3 Taxa of Conservation Importance

A summary of the OSPAR List of Threatened and/or Declining Species and Habitats, Scottish PMF, and Scottish Biodiversity List, are listed in Table 57 and illustrated in Figure 68.

Table 57 Potential taxa of conservation identified within the survey area.

Image	OSPAR/PMF/SBL	Site ID
	Octocorallia SBL Taxa	S02, S04, S05, S08, S15, S16, S18, S24, S25, S29, S32, S36, S37, S43, S44, S48, S50, S59, S65, S78, S85, S86, S87, S89 and S92 T11, T22, T39, T41, T45, T47, T52, T72, T74, T75, T88, T95, T96, T97, T98 and T99 NS_T003, NS_T007 and NS_T009

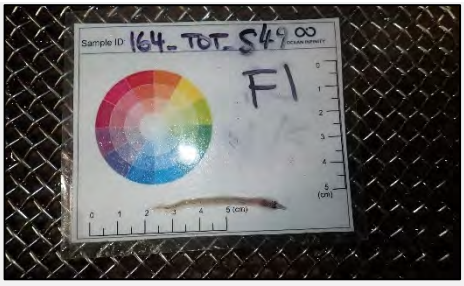
Image	OSPAR/PMF/SBL	Site ID
	*Ammodytes* sp. (including juveniles) PMF and SBL Taxa	S01, S05, S07, S12, S14, S18, S20, S29, S33, S49, S67, S73 and S82
	*Dipturus batis* complex OSPAR, PMF and SBL Taxa	S84
	*Raja clavata* OSPAR and SBL Taxa	S35
	*Lophius piscatorius* PMF and SBL Taxa	T96
	*Molva molva* PMF and SBL Taxa	S05, S65 and T96
	*Merlangius merlangus* PMF and SBL Taxa	S65 and S84

Image	OSPAR/PMF/SBL	Site ID
	*Gadus morhua* OSPAR, PMF and SBL Taxa	S46, S64 and S65
	*Arctica islandica* (including juveniles) OSPAR and PMF Taxa	S01, S20, S27, S30, S31, S33, S38, S40, S42, S49, S51, S54, S60, S62, S63, S76, S77, S81 and S84
	*Trisopterus esmarkii* PMF and SBL Taxa	S05, S18, S43 and S76
No image available	*Pecten maximus* Commercially important Taxa	S36
No image available	*Aequipecten opercularis* Commercially important Taxa	S16 and S59
No image available	*Tamarisca tamarisca* SBL Taxa	S08, S16, S57 and S81
No image available	*Ceratia proxima* SBL Taxa	S76

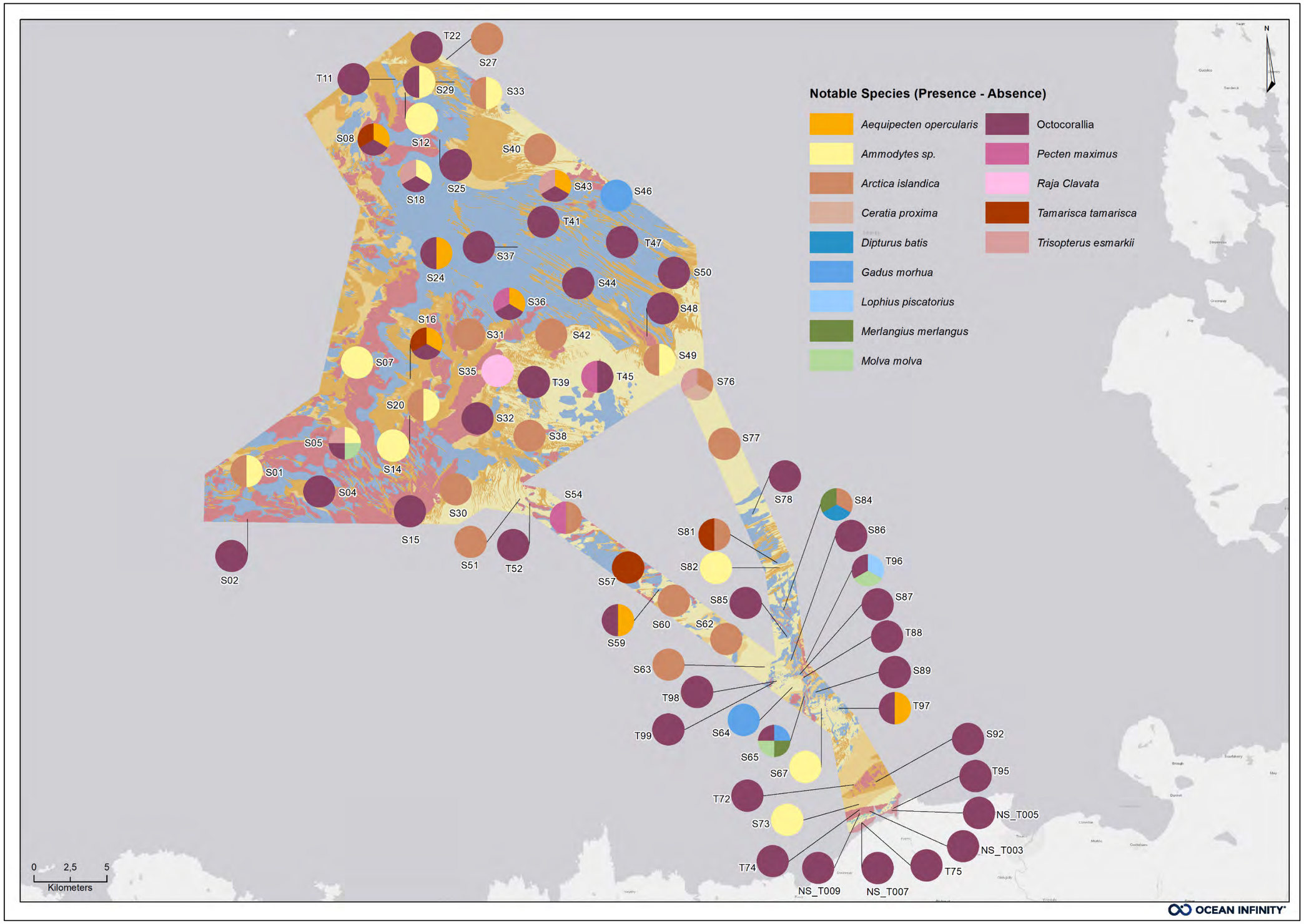


Figure 68 Overview of potential taxa of conservation identified within the survey.

6.3.1 Notable Taxa

Horse mussel *Modiolus modiolus* (PMF, OSPAR & SBL)

The horse mussel *Modiolus modiolus* represents a priority habitat if reef-forming. One (1) juvenile individual was identified at grab sample site ECW_S64.

No *M. modiolus* beds (reefs) were identified within the survey area.

Ocean quahog (PMF & OSPAR)

Adult ocean quahog *Arctica islandica* was identified in the grab samples at OAA_S01, OAA_S20, OAA_S27, OAA_S30, OAA_S31, OAA_S33, OAA_S38, OAA_S40, OAA_S42, OAA_S49, ECW_S51, ECW_S54, ECW_S60, ECW_S62, ECW_S63, ECE_S76, ECE_S77, ECE_S81 and ECE_S84 (Figure 69). Juvenile *A. islandica* were identified at all grab sample sites but for ECE_S77 which was identified in the site imagery acquired.

A total of 2 adults and 51 juveniles were identified in the grab samples across the survey area. The adult specimen sampled at site ECW_S63 was released back. *A. islandica* is typically found in sand/sandy mud or coarse sand habitats from the low intertidal zone to 400 m throughout the North Sea. The broad-scale habitats where *A. islandica* was identified were predominantly either circalittoral sand, **MC52**, or a gravelly sand matrix, **MC32/MC52**. Sites ECW_S54 and ECE_S81 are located in **MC42** with ECE_S84 at the edge of an area classified as **MC12** (Figure 70).

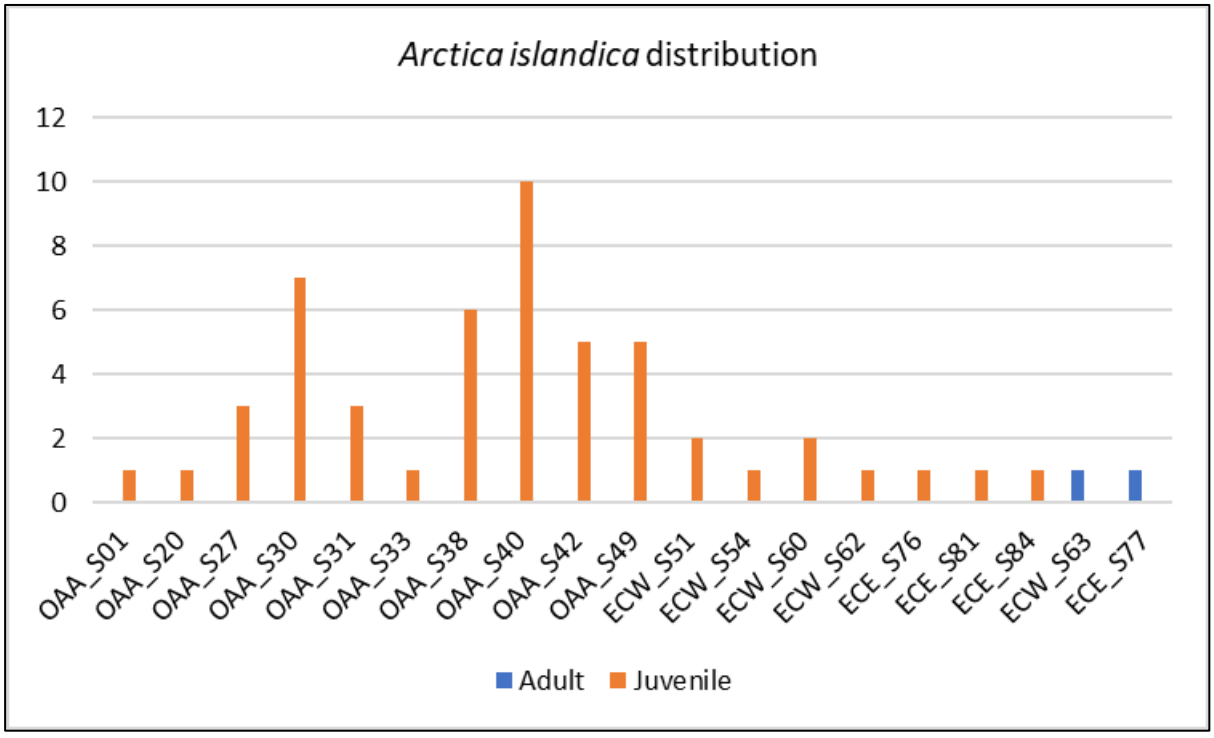


Figure 69 Total abundance of A. islandica per site.

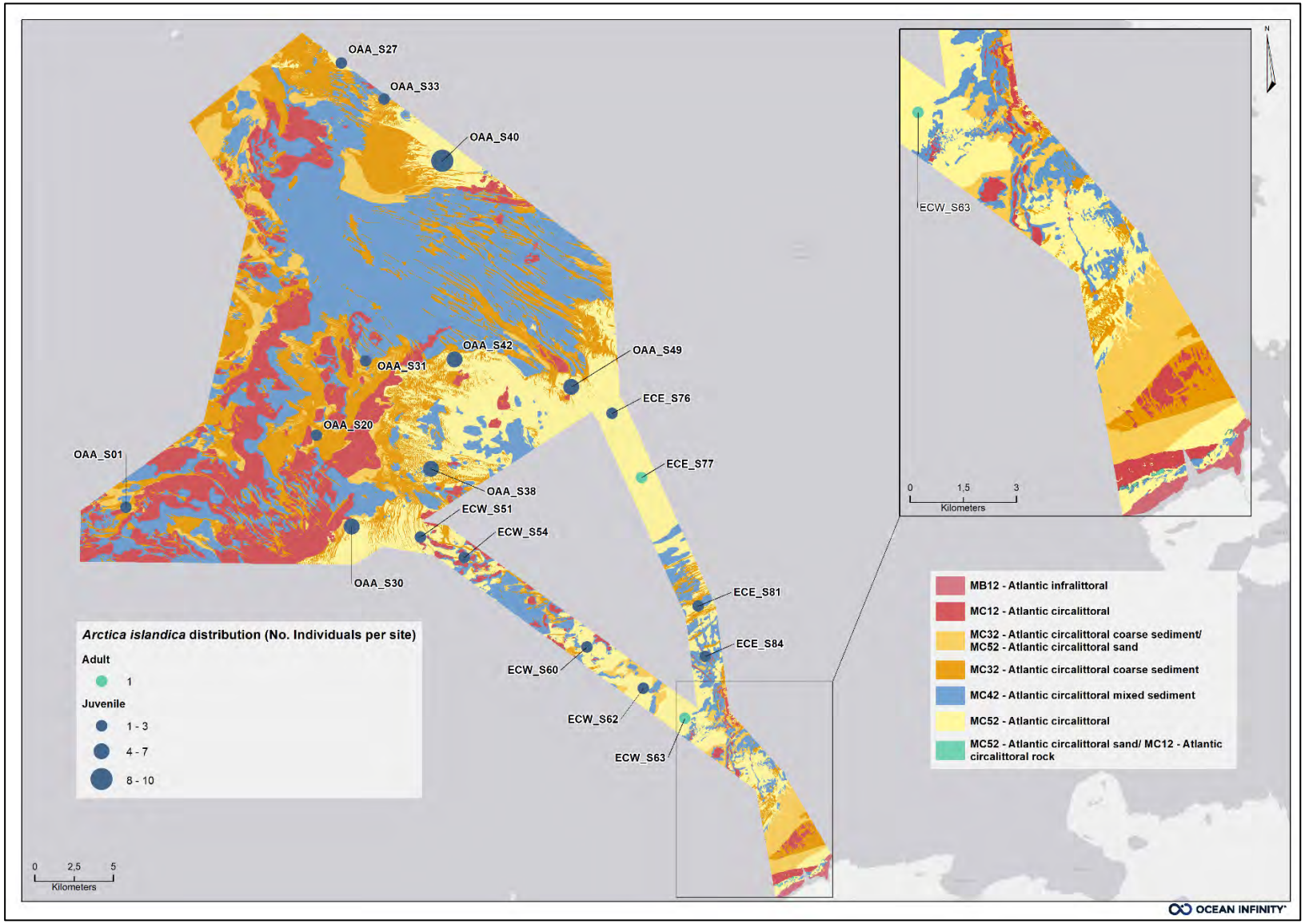


Figure 70 Arctica islandica *distribution.*

6.3.2 Invasive and Non-Native Taxa

No invasive non-native taxa but one (1) non-native taxa were identified during the survey. Non-native taxa do not naturally occur in an area but have been introduced intentionally or deliberately by humans. Non-native taxa do not harm the ecosystem where it's been introduced in contrast to invasive non-native taxa. Invasive non-native taxa are a threat to the ecosystem, the economy and/or the health of humans and/or animals (Directorate-General for Environment, 2023).

The non-native polychaete *Goniadella gracilis* was identified at twenty-three (23) different grab sample sites with a total of 80 individuals (Table 58). The species was described from eastern North America, and the first British records are from 1970 in Liverpool Bay (Eno, Clark, & Sanderson, 1997).

Table 58 Non-native taxa identified during the survey.

Non-native taxa	Grab Sample Site	Abundance/0.1 m^2
Goniadella gracilis	OAA_S03	1
	OAA_S06	2
	OAA_S09	Fragment
	OAA_S12	4
	OAA_S13	1
	OAA_S14	5
	OAA_S16	1
	OAA_S17	4
	OAA_S24	1
	OAA_S31	2
	OAA_S34	17
	OAA_S35	3
	OAA_S37	1
	OAA_S42	1
	OAA_S43	2
	OAA_S44	1
	OAA_S46	5
	ECW_S58	19
	ECW_S70	1
	ECE_S79	2
	ECE_S82	4
	ECE_S87	1
	ECE_S91	2

6.4 MESH Confidence Assessment

The MESH confidence assessment yielded seven (7) different scores: Polygons with no ground-truthing scored 53, polygons with only Vibrocores (VC) scored 76, polygons with only imagery data (grab sites/transects) scored 83 on a soft substrate and 87 on hard substrate. On three occasions, images and VC or PSA were part of the ground-truthing and scored 85.

Habitat **MB12** – Atlantic infralittoral rock only comprised imagery on hard substrate and less than three (3) samples per habitat thus scoring 86. Sites ground-truthed by grab sampling combined with PSA and imagery had the highest score of 97.

Polygons lacking ground-truthed data could obtain a maximum score of 61, whereas polygons with ground-truthed data could obtain a maximum score of 100.

A summary of the MESH confidence scores is presented in Table 59.

Table 59 Summary of MESH confidence assessment scores.

Sampling Method	Mesh Score	Maximum Score	No. of Polygons	Sum of Area (km^2)	Percentage of Total Area
No ground-truth data	53	58	5593	266.1	34.1 %
Vibrocore	76	100	5	2.1	0.3 %
Seabed imagery (Soft)	83	100	19	7.2	0.9 %
Seabed imagery and PSA/VC (Soft)	85	100	3	7.4	0.9 %
Seabed imagery (Hard*+ <3**)	86	100	2	1.3	0.2 %
Seabed imagery (Hard*)	87	100	15	62.9	8.1 %
Grab sampling, PSA and seabed imagery	97	100	51	433.5	55.5 %

** Mesh scoring and weight system give a higher score to imagery on hard substrate than on soft substrate.*

*** Less than three (3) samples per habitat.*

The sampling method consisting of grab, imagery, and PSA, with a MESH confidence score of 97 represented the largest coverage, more than half (55 %) of the survey area, consisting of 51 polygons. Most prominent in the east part of the OAA and northern part of ECC East lesser and more scattered in the west part of OAA and ECC West (Figure 71).

Close to a third (34.1 %) consisted of habitats with a MESH confidence score of 53, consisting of 5597 polygons with no ground truth data. These areas are scattered throughout OAA and ECCs, most prominent in the western part of OAA stretching from south to north as well as along the ECC West and ECC East (Figure 71).

At 8.1 % coverage with a MESH confidence of 87 are 15 polygons containing seabed images on hard substrate. The final four (4) sampling methods with a mesh confidence score of 76, 83, 85 and 86, covered an area of 2.30 % spread over 29 polygons.

Figure 71 Overview of MESH score areas based on habitat polygons.

7. Discussion

The majority of the sampled sites share components, to a varying degree, of subtypes to **MC52** – Atlantic circalittoral sand, **MC32** – Atlantic circalittoral coarse sediment and **MC42** – Atlantic circalittoral mixed sediment as well as **MC53** – Atlantic circalittoral rock. Two habitat complexes were assigned to provide higher resolution in the delineation of the broadscale habitats: **MC32** – Atlantic circalittoral coarse sediment/**MC52** – Atlantic circalittoral sand, and **MC52** – Atlantic circalittoral sand/**MC12** – Atlantic circalittoral rock.

These complexes were assigned to differentiate between areas comprising coarse sand and those comprising coarse sediments such as gravel and pebbles as these would likely exhibit different faunal compositions but are grouped within the same EUNIS classification of **MC32** – Atlantic circalittoral coarse sediment.

Each of the grab sample sites was further classified individually and to a higher level where possible. It was deemed most appropriate to present these sample-specific habitats separate from the broad-scale habitats due to the heterogeneity of the area. The taxonomic assemblage from the grab samples indicates that the most commonly occurring habitat was **MB3233** – *Moerella* spp. With venerid bivalves in Atlantic infralittoral gravelly sand and a variant of this habitat with a low presence of *Asbjornsenia pygmaea* followed by habitat complexes **MC4214** – *Flustra foliacea* and *Hydrallmania falcata* on tide-swept circalittoral mixed sediment/ **MC5211** – *Echinocyamus pusillus*, *Ophelia borealis* and *Abra prismatica* in circalittoral fine sand and **MC521** – Faunal communities of Atlantic circalittoral sand/**MC42** – Atlantic offshore circalittoral mixed sediment.

The Total Organic Matter (TOM) and Total Organic Carbon (TOC) analyses results indicate a low fraction of TOM and TOC in the sediment at all grab sample sites. TOC levels were within the expected range of surface sediments of 0 – 2 % (Smeaton, Hunt, Turrell, & Austin, 2021). The highest levels of TOM, >= 2 %, were identified at the sites S42, S73, S76, S77 and site S91.

There was no apparent correlation between TOM and Mud contents, with the two sites with the highest TOM concentration (ECW_S73 and ECE_S76) having both low and high (relatively) Mud contents of 0.3 % and 8.2 %, respectively. Whilst depth and Mud content showed some correlative trends, the depth did not correlate with TOM content, with sites ECW_S73 and ECE_S76 being located at a depth of 42 m and 97 m, respectively (Figure 72). The highest TOC content was highest at sites ECW_S54 and ECW_S56, however, no correlations were identified for TOC and any other variable.

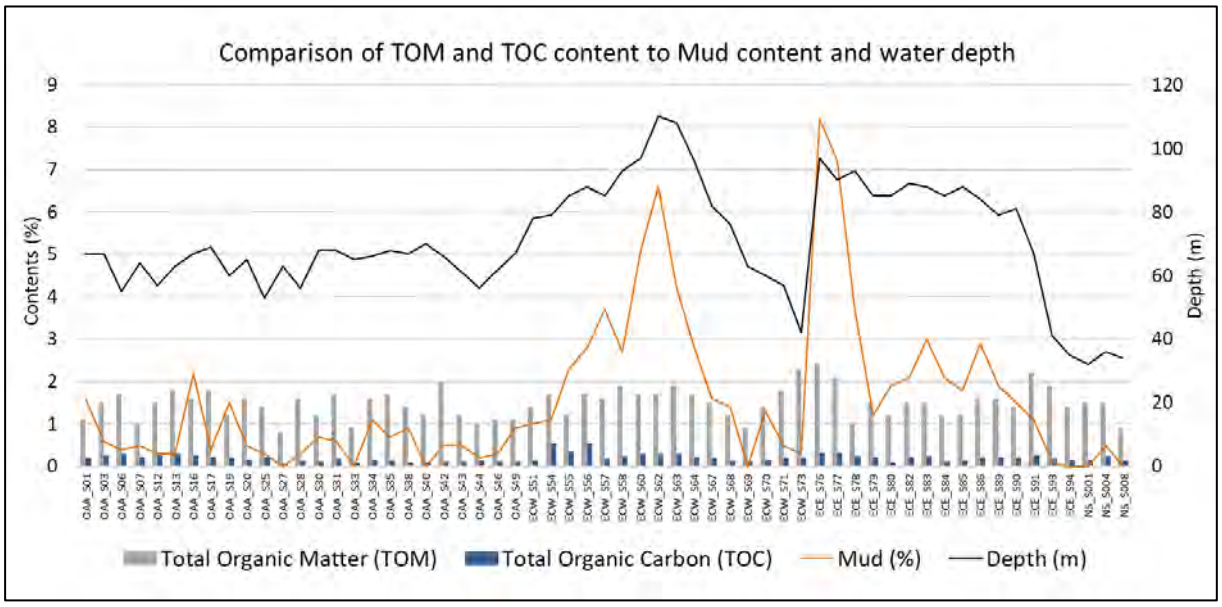


Figure 72 Comparison of Total Organic Matter and Total Organic Carbon.

Brominated Flame Retardants levels, although low within the current survey, showed that congener BDE209 was generally higher in concentration compared to the other congeners and had a notably higher concentration at 8.75 µg/kg at site S76. The lower threshold of NEA's class 2 – Good, as well as the OSPAR BAC threshold values, were exceeded for one or multiple congeners at 18 of the 29 sampled sites. Research published by the Scottish Marine Assessment (Marine Scotland Assessment, 2020) indicates that the levels of PBDE congeners in Scottish waters are above natural levels but mostly below the thresholds at which adverse effects are noted to occur. The mean concentrations were also noted to be above BAC. The assessments in Scottish waters, conducted between 1999 and 2018, show that the mean concentrations of PBDEs, in general, appeared stable in some regions while decreasing in others.

The most abundant taxon was the annelid *Owenia*, with a total of 2332 individuals recorded, and the taxon occurred in 77 % of the grab samples. *Owenia* occurred both in the OAA area and along the ECCs, which is reflected in Figure 36. The greatest abundances occurred in the northeast OAA area in the habitats **MC32** – Atlantic circalittoral coarse sediment, **MC32** – Atlantic circalittoral coarse sediment/**MC52** – Atlantic circalittoral sand and **MC52** – Atlantic circalittoral sand. Nematoda had a total abundance of 1856 individuals and occurred extensively throughout the survey area (92 % of the grab samples). Nematoda is a broad taxonomic classification, and its species inhabit various kinds of habitats, as can be seen in Figure 36.

Pielou's Evenness Index and Simpson's Index of Dominance presented overall high values with limited variation, with mean values of 0.76 and 0.86, respectively. This indicates a relatively even distribution of individuals across the different taxa in the samples. Shannon-Wiener Index and Margalef's Diversity Index varied moderately, with Margalef's Diversity Index strong correlation with the species richness (R^2=0.95). The variation seen in the Shannon-Weiner Index is mainly due to the tenfold variation in species richness, as the Pielou's Evenness Index (being an expression of a given Shannon-Weiner Index value which is divided by the theoretical maximum value with that number of taxa) is less variable. However, the samples with the lowest Shannon-Weiner Index values all have Pielou's Evenness Index values which are below the mean value, indicating that these samples are more influenced by the low evenness compared to the rest of the samples (Table 37). The variation in Shannon-Weiner Index does not show any clear trends with geographical location, sediment composition, depth or contaminants.

The SIMPROF analysis produced 21 statistically significant groups. The large number of groups seen in the SIMPROF analysis shows that the survey area stretches over a varied and complex seabed with many different habitats. A majority of these groups were based on grab samples that were spatially closer to each other within the survey area, and classified in similar habitats, indicating the possibility of some between-site homogeneity. This is further reflected in the SIMPROF analysis superimposed with EUNIS classifications in Section 5.6.9.

The main driving variables for the faunal assemblages are likely depth and substrate, as these variables together constituted the best correlation with the macrofaunal distribution in the survey area, which is presented in the BEST analysis in Section 5.6.8.

In theory, strongly correlated variables tend to increase or decrease together (Taylor, 1990). The results presented a strong correlation (0.675) between the combined variables Depth, % Medium Sand and % Clay and these physical variables are likely to be the main factors influencing the distribution of the faunal communities. The relationship between sediment composition and the faunal communities has long been known (Sanders, 1958). The depth presented the best correlation both in the single, where each variable was tested separately and in the multiple variable tests, emphasising depth playing a major role in forming the faunal communities.

To test the similarities further, statistically, between the OAA and ECC sample sites, additional multivariate analyses were conducted separately for the different areas. The statistical analyses were based on the macrofaunal data derived from the taxonomic analyses of the grab samples. Square root transformation was applied to the dataset before calculating the Bray-Curtis similarity measures in the SIMPROF analyses.

The SIMPROF analyses conducted for the OAA and ECC grab sample sites produced five (5) and 11 statistically distinct groups (black lines) respectively and are presented in the hierarchical dendrograms in Figure 73 and Figure 75. Sample similarity is further explored in the nMDS-plots in Figure 74 and Figure 76.

Differences seen in the additional multivariate analyses conducted, compared to the incorporated analysis with all sites, were a decrease in the number of SIMPROF groups. Differences seen in the nMDS-plots for the OAA and ECC grab sample sites were lower stress values, indicating an even better ordination with less prospect of a misleading interpretation.

The outlier group in the OAA sites dataset, group **a**, was characterised by not having any dominant taxa contributing to high similarities, with *E. pusillus* being the top contributor with just over 6 %. This notable lack of a dominant taxa can be seen in Figure 36 where most sites have a dominant taxon, which is lacking for the sites belonging to group **a**. The sites are further classified as the same habitat complex **MC4214** - *Flustra foliacea* and *Hydrallmania falcata* on tide-swept circalittoral mixed sediment/**MD42** -Atlantic offshore circalittoral mixed sediment and are the only sites classified to this complex, further supporting these sites comprising a separate group (Table 26).

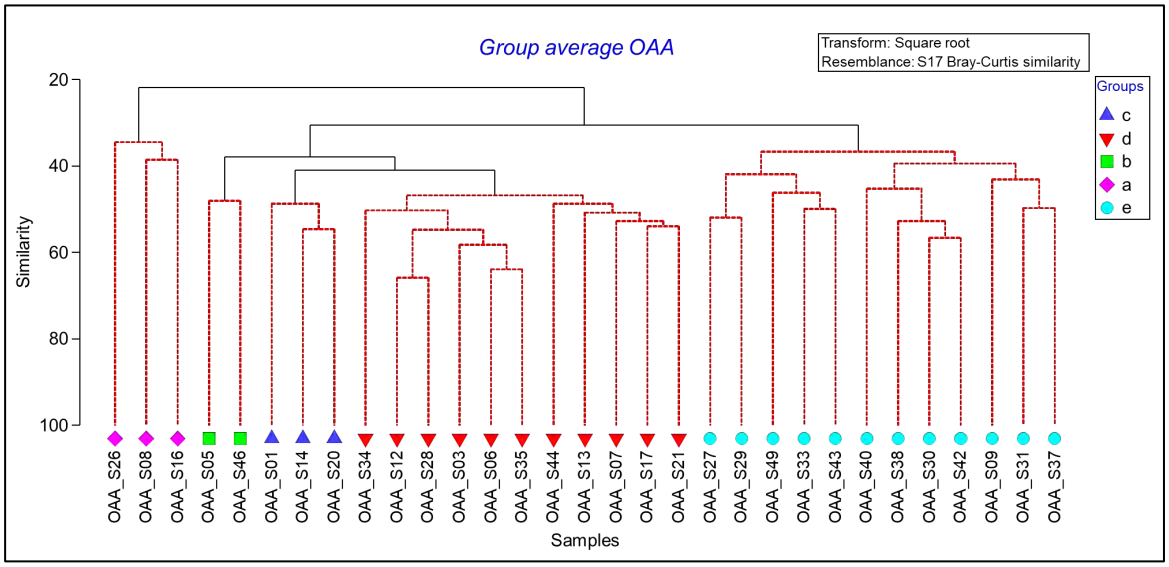


Figure 73 SIMPROF dendrogram of non-colonial faunal composition from OAA grab sample sites.

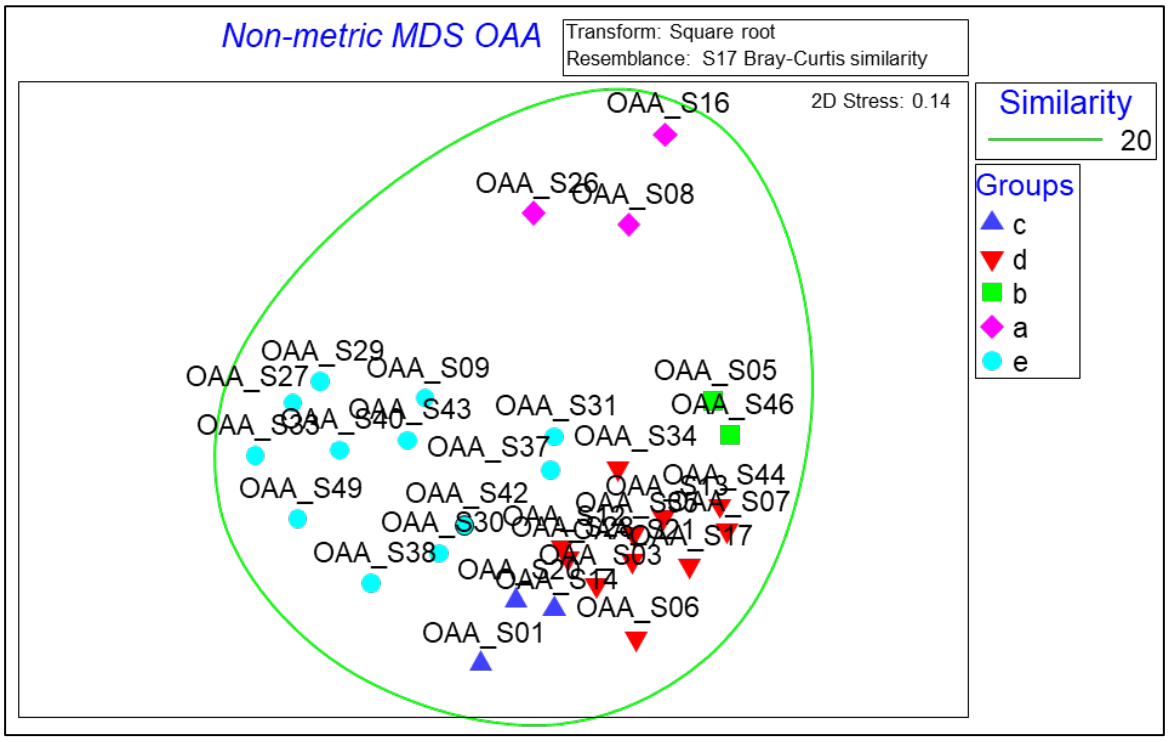


Figure 74 nMDS-plot of non-colonial faunal composition from OAA grab sample sites with group based on the SIMPROF analysis.

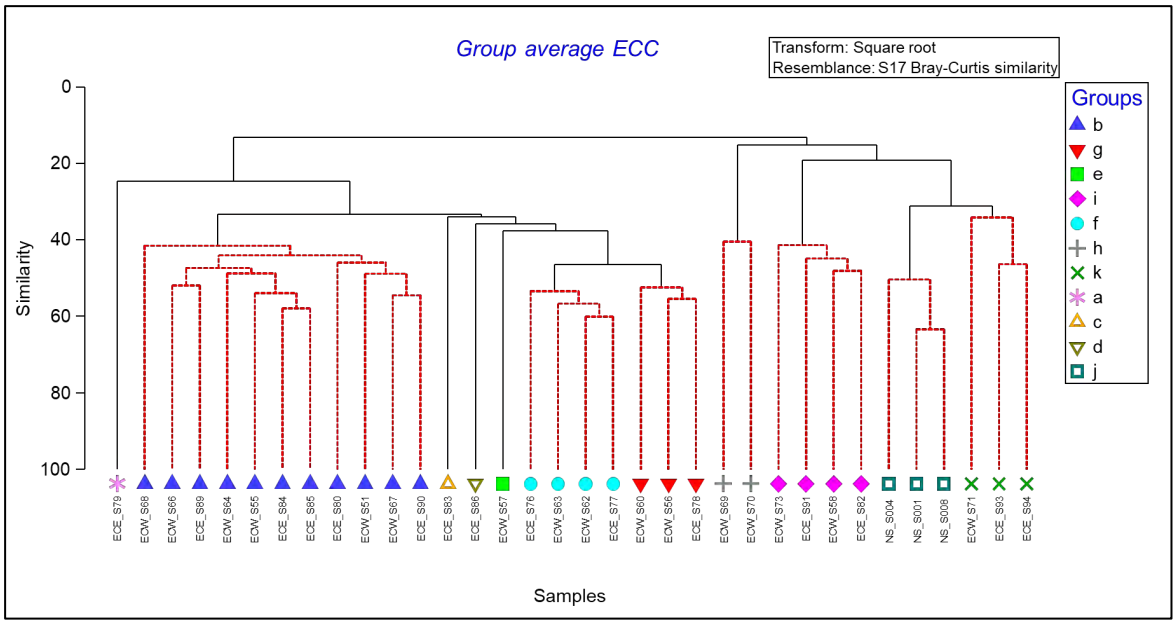


Figure 75 SIMPROF dendrogram of non-colonial faunal composition from ECC grab sample sites.

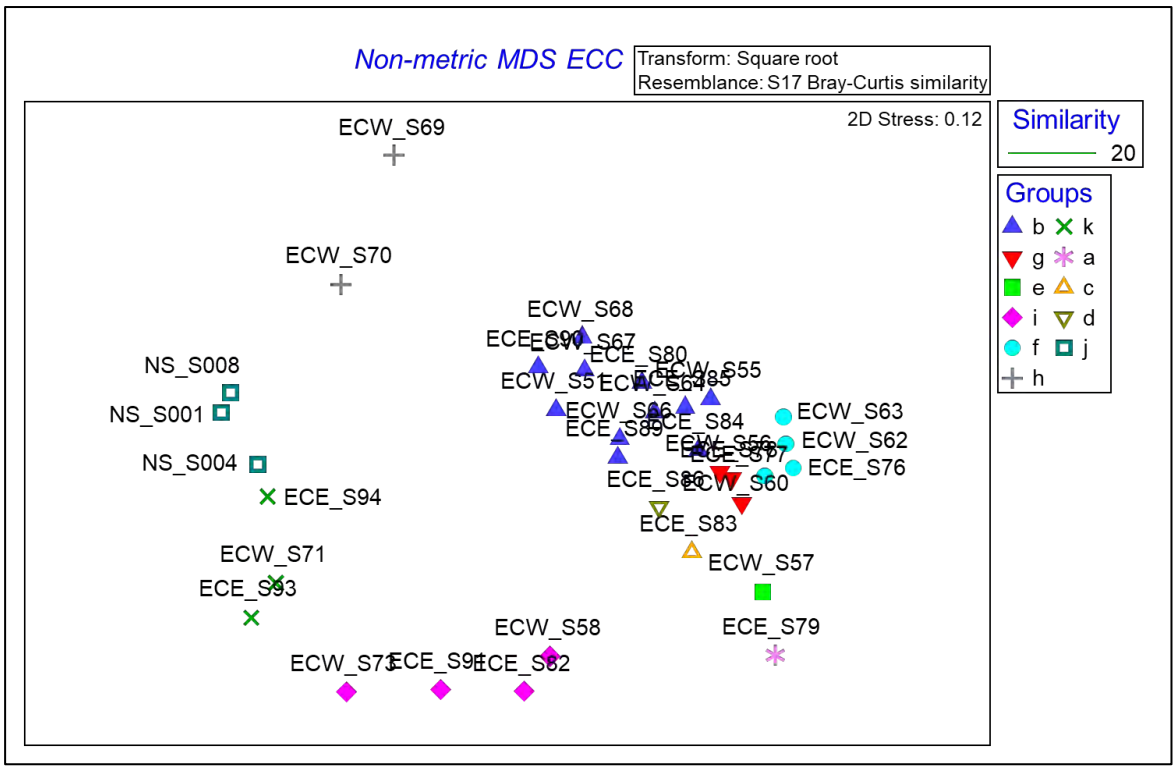


Figure 76 nMDS-plot of non-colonial faunal composition from ECC grab sample sites with groups based on the SIMPROF analysis.

Density measurements from the CTD ranged from approximately 1021 to 1026 kg/m^3, and no strong pycnoclines were evident. Temperature exhibited limited variation, particularly at the nearshore sites where the temperature remained around 12.3 ^{o}C. Offshore temperatures showed a greater variation but typically declined by no more than 0.5 ^{o}C with increased depth. The lower salinity measurements (31 – 32 PSU) were all obtained within the first one (1) meter of the water column and are potentially a result of the CTD instrument stabilising as it entered the water. The turbidity measurements from the CTD were variable and low throughout the survey area, ranging from -1.4 to 15.1 mg/l with higher concentrations in the ECCs. Likewise, TSS was also low, typically <5 mg/l at most sites but higher at sites W15 – W20 and W24 in the ECCs. These higher concentrations may be attributed to the closer proximity to the coastline and the influence of sedimentation.

8. Conclusions

Benthic offshore sampling was performed at 99 sites, out of which 73 sites were sampled using a combination of DDV together with grab sampling, and 26 (17 standalone DDV transects and 9 grab sites) sites were sampled using only DDV. In addition to the benthic sampling, water sampling, together with CTD profiling, was performed at 20 sites.

Benthic nearshore sampling was performed at nine (9) sites, out of which four (4) were sampled using a combination of DDV together with grab sampling, and five (5) sites were sampled only using DDV. In addition to the benthic sampling, water sampling, together with CTD profiling, was performed at five (5) sites.

All sampling was conducted as part of the Benthic Environmental survey for the West of Orkney offshore windfarm, located 28 km west of Hoy, Orkney, Scotland.

The bathymetry of the survey area was typically gently sloping, with gradients around 0.8° throughout most of the survey area. Boulders, often associated with scour, were observed throughout the OAA and the ECE. Megaripples, also associated with scour were also present throughout the OAA, ECE and ECW. Bedforms in the ECE and ECW exhibited north-south and east-west orientations, suggesting the occurrence of complex flow conditions in the area.

The seabed in the survey area comprises a complex and heterogeneous mosaic of mainly coarse and rocky substrates. A total of seven (7) EUNIS habitats and two (2) habitat complexes were identified within the survey area. The taxonomic assemblages from the acquired grab sample data indicate the presence of 15 sample-specific habitats across the survey area, including 6 transitional habitat complexes.

Two sub-types of Annex I (1170) - Reefs habitat, were interpreted to be present, Stony Reefs and Bedrock Reefs. Bedrock reefs were identified in the Nearshore areas of both the EEC West and ECC East. Stony Reefs were identified within the OAA as well as both ECCs. The majority of the Low to Medium Resemblance Reefs are located in the OAA whereas the Potential Reefs are mainly located in the ECCs and the western section of the OAA.

A total of four (4) transects (T11, T22, T88 and T98) and 18 grab samples sites (S04, S08, S16, S18, S19, S24, S25, S26, S29, S36, S37, S44, S53, S54, S57, S59, S61 and S92) are assessed to qualify as Annex I (1170) Reefs - Stony Reefs, Low Resemblance.

A total of ten transects (T23, T39, T41, T45, T47, T52, T72, T74, T96 and T97) and ten grab samples sites (S02, S05, S10, S15, S32, S43, S48, S50, S65 and S84) are assessed to qualify as Annex I (1170) Reefs - Stony Reefs, Medium Resemblance.

The PMF habitat Offshore subtidal sands and gravels and the SBL habitat Subtidal sands and gravels were identified across large areas of the OAA and within both the ECC route corridors. The PMF habitat Kelp Beds was identified at the bedrock exposures in close proximity to the landfall of the ECC West route corridor.

Fourteen taxa of conservation importance and one (1) non-native species were identified within the survey area.

The sediment composition had limited variation throughout the survey area. Sand and Gravel were the dominant sediment fractions in the samples from the OAA and Sand dominated the ECC. The PCA mainly grouped the sites based on the gravel-to-sand ratio and to a lesser extent on mud content.

Metal concentrations were generally low, with grab samples exceeding some threshold values for arsenic and/or nickel at 13 grab sample sites. Total Organic Matter and Total Organic Carbon both varied throughout the survey area; however, no trends nor correlations were identified. Hydrocarbon content was generally low but variable, with higher concentrations noted in the Nearshore samples. Polycyclic Aromatic Hydrocarbon concentrations exceeded threshold values for several congeners and ΣEPA 16 PAH in samples from five (5) sites. Concentrations of PCBs were low and exceeded the LoD in samples from seven (7) sites, all of which exceeded threshold values. Organotin (MBT, DBT, TBT) concentrations were below the detection limit at all sampled sites. Organochloride pesticide concentrations were low and exceeded the LoD in three (3) of the samples. Concentrations of PBDEs were generally low, with grab samples exceeding threshold values at 18 sites.

The phyletic composition from grab samples, regarding both the total number of taxa and abundance, was dominated by annelids. The two most abundant taxa were the annelid Owenia and the roundworm Nematoda. *Owenia* had a total abundance of 2332 individuals and occurred in 77 % of the grab samples. Nematoda had a total abundance of 1856 individuals and occurred in 92 % of the grab samples.

Pielou's Evenness index and Simpson's Index of Dominance had a limited variation, whereas Margalef's Richness Index and Shannon-Wiener index presented slightly higher variation across the grab samples. The number of taxa and the number of individuals varied between 9 - 99 taxa and 25 - 835 (ind./m^2), respectively per grab sampling site. The SIMPROF analysis of the non-colonial faunal composition produced 21 statistically distinct groups. The sample similarity explored in the nMDS-plot presented a stress value of 0.15 which is a relatively good ordination with a low prospect of a misleading interpretation, the nMDS might have some contradicts with the resemblance matrix.

In the results of the BEST analysis limited to a single variable, Depth was the most distinguished variable with a global correlation (σ) of 0.552 and was the statistically significant variable for the distribution of the biological data. The strength of this correlation is considered moderate (Taylor, 1990).

In the results of the BEST analysis using multiple variables, Depth, % Medium Sand and % Clay together were the most distinguished variables with a global correlation (σ) of 0.675 and were statistically significant variables for the distribution of the biological data. The strength in this correlation is considered strong (Taylor, 1990).

The non-colonial fauna species biomass was dominated by Mollusca with 70 % of the total biomass, followed by Annelida with 14 %. Non-colonial fauna biomass varied between 0.0343 g/0.1 m^2 in sample ECE_S94, to 123.8660 g/0.1 m^2 in sample ECE_S91. The non-colonial fauna biomass expressed as mean value across all grab samples sites was 3.8445 g/0.1 m^2 (SD=15.5177). Bryozoa dominated the phyletic composition of the sessile colonial epifauna in grab samples, both regarding the number of taxa and abundance of colonies.

Grab sampling site OAA_S10 presented the greatest species diversity from the analysis of the stills, with a total of 51 taxa. The most abundant phyla of non-colonial fauna in stills were Mollusca with 37 %, followed by Arthropoda and Cnidaria with 27 % and 18 %, respectively. The average abundances of non-colonial fauna varied from zero (0) ind./m^2 to 371 ind./m^2 at site OAA_T47. The average non-colonial fauna abundance per grab sample site still was 65 (SD=89) ind./m^2. Bryozoa represented the phylum with taxa covering the largest surface area, with 61 %. Cnidaria and Ochrophyta contributed 25 % and 5 % of the recorded taxa in stills, respectively. Arthropoda, Rhodophyta, Porifera and Others followed with 4 %, 3 %, 1 % and 1 %, respectively. The coverage of colonial fauna from stills varied from 0 % to 49.42 % (ECW_T74). The average cover of the fauna was 7.5 % (SD=10.65 %).

The CTD results showed that salinity, temperature, and density were consistent across sampled sites and showed limited variation. Turbidity and TSS values were low throughout the survey area, exhibiting greater concentrations in the ECCs. Site ECW_W15 exhibited the highest TSS concentration of 35 mg/l in the samples acquired closest to the surface.

The majority of the delineated habitat polygons had ground-truthed data collected, resulting in that 66 % of the survey area achieving a MESH confidence score of ≥76, with 38 % scoring ≥90. The remaining 34 % of the survey area had a confidence score of 53, as these polygons lacked ground-truthed data. The overall high scores indicate a robust assessment and confidence in the habitats identified within the OAA and ECCs. Considering the large number of polygons within the non-ground-truthed data compared to the area (km^2), the assessment would further confirm the heterogeneity of the area and it would be difficult to justify more ground-truthing in order to gain a higher overall score.

9. Reservations and Recommendations

The results detailed within this report are based on the data derived from the faunal grab sampling and still imagery data together with sediment, water and contaminant analyses from each sample site investigated within this survey. The data has been reviewed in conjunction with the geophysical data (SSS and MBES) and interpretations. It should be taken into account that there is a natural limitation in the accuracy of interpretations. Where considered applicable, the sampling results have been extrapolated to surrounding areas exhibiting similarity as interpreted from the geophysical data.

The definition of a "Reef" is not defined within the EC Habitats Directive. Areas interpreted as potential stony reefs in this report are based on methods defined in the JNCC report No. 432 "The identification of the main characteristics of stony reef habitats under the Habitats Directive" (Irving, 2009) and JNCC report No. 656 "Refining the criteria for defining areas with a 'low resemblance' to Annex I stony reef" (Golding, Albrecht, & McBreen, 2020). All areas interpreted as bedrock were considered to be reefs, as there is no grading system of "reefiness" of bedrock reefs.

The EUNIS habitat "**MB3233** – *Moerella* spp. with venerid bivalves in Atlantic infralittoral gravelly sand" was identified in the survey area. One of the key characteristics to this habitat is the species *Moerella pygmaea*. It should be noted that the accepted name for the taxa has changed from *M. pygmaea* to *Asbjornsenia pygmaea* and that this name change is, as of writing this report, not reflected in the EUNIS habitat **MB3233** description.

The output from the PRIMER analysis was used as a guidance when assigning habitat codes for each sample. Please note that the analysis in PRIMER when using no replicates often results in less certain results (lower statistical power), thus the multivariate analysis is only regarded as a guidance.

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VERTEBRATE METABARCODING **RESULTS**

Order number:	SO01060
Report number:	NM-ABO743
Company:	APEM Ltd
Contact:	Chris Ashelby
Project:	Total Energies - West of Orkney Wind farm
Sample type:	NatureMetrics eDNA disk filter
Date of report:	27-Jan-2023
Number of samples:	40

Thank you for sending your samples for analysis by NatureMetrics. Your samples have been metabarcoded following our eDNA survey - Vertebrate workflow. **A taxon-by-sample table of your samples is attached to this report (NM-ABO743.SO01060.Vertebrate.xlsx)**. Each row in the table represents one taxon (OTU), shown with the lowest possible taxonomic assignment based on currently available reference data. Each column represents a sample, showing the percentage of sequence reads per detected OTU (Table 1) and the number of sequence reads per detected OTU (Table 2) in that sample. Care should be taken in interpreting the numbers in terms of relative species abundance, but a high sequence proportion can be interpreted as lending greater confidence to a detection. This report contains biodiversity information that may be sensitive, particularly with respect to endangered or protected species. It is the responsibility of the client to ensure that due consideration is given to the data and that the information is shared in a responsible way.

Here we present an overview of the key results, followed by a more detailed report that starts with the taxonomic composition of the samples followed by a more detailed look at the steps taken to extract, amplify, sequence, and analyse your DNA. A glossary for terms in bold is provided at the end of the report to define key terms used within the report.

OVERVIEW OF YOUR RESULTS

- A total of 42 taxa were detected.
- Average taxon richness was 5.67 and ranged from 2 to 15.
- Most abundant sequences: Gadidae sp.
- Most commonly detected taxa: Gadidae sp., poor cod (*Trisopterus minutus*) and Atlantic mackerel (*Scomber scombrus*).
- Species of note: Atlantic horse mackerel (*Trachurus trachurus* - Vulnerable).
- Vertebrate sequence data were obtained from 33 of 40 eDNA sequences.

FULL REPORT

Sample composition

A total of 42 taxa were detected (**Table 1** and **Table 2**). 64.3% (27 taxa) were at least 99% similar to a species in the global reference databases, and species names are suggested. The remaining taxa were identified to the lowest possible taxonomic level: 14.3% to genus (6 taxa), and the remainder to family (9 taxa). A total of 32 unique fish, 3 birds and 7 mammals were detected. The taxa belong to 14 orders, 25 families, and 31 genera.

Species of note include the: Atlantic horse mackerel (*Trachurus trachurus* - Vulnerable).

The average taxon richness was 5.67 and ranged from 2 ('164_TOT_eDNA_W08_TOP', '164_TOT_eDNA_W13_TOP', '164_TOT_eDNA_W20_TOP') to 15 ('164_TOT_eDNA_W03_BOT'). The relative proportion of the sequences found in each of the samples is shown in **Figure 2**, **Figure 3**, **Table 1** and **Table 2** and the diversity is summarised in **Table 3** and **Table 4**.

A species in the family Gadidae, which accounted for 20.2% of the total sequence reads, was among the most abundant in terms of sequences. Among the most commonly detected species were Gadidae sp., poor cod (*Trisopterus minutus*) and Atlantic mackerel (*Scomber scombrus*), which were detected in 25, 21 and 16 samples, respectively.

Note that *Apodemus sylvaticus* (European woodmouse) is an unexpected detection in marine samples, however, we do not suspect any internal contamination (as the lab controls are clean, and this is not a species we see in high frequency or abundance in other samples). The species has been detected in the same sample (164_TOT_eDNA_W09_TOP) with the mammal and vertebrate assays, we believe that this is a real detection in the sample, likely to have originated during sample handling (e.g. mice on the marine vessels).

High-quality vertebrate sequence data were obtained for 10 of the 40 eDNA samples. eDNA metabarcoding of vertebrates was not successful for '164_TOT_eDNA_W15_TOP', which failed to amplify despite troubleshooting. 23 Samples produced fewer than expected target reads; results for these samples are therefore considered tentative as they may not reflect the full range of vertebrate species diversity in these sample(s). The total number of target sequences in the remaining 6 samples were below our threshold for reporting and the corresponding detections are not reported. The report status of all samples is summarised in ***Table 5.***

All laboratory controls behaved as expected.

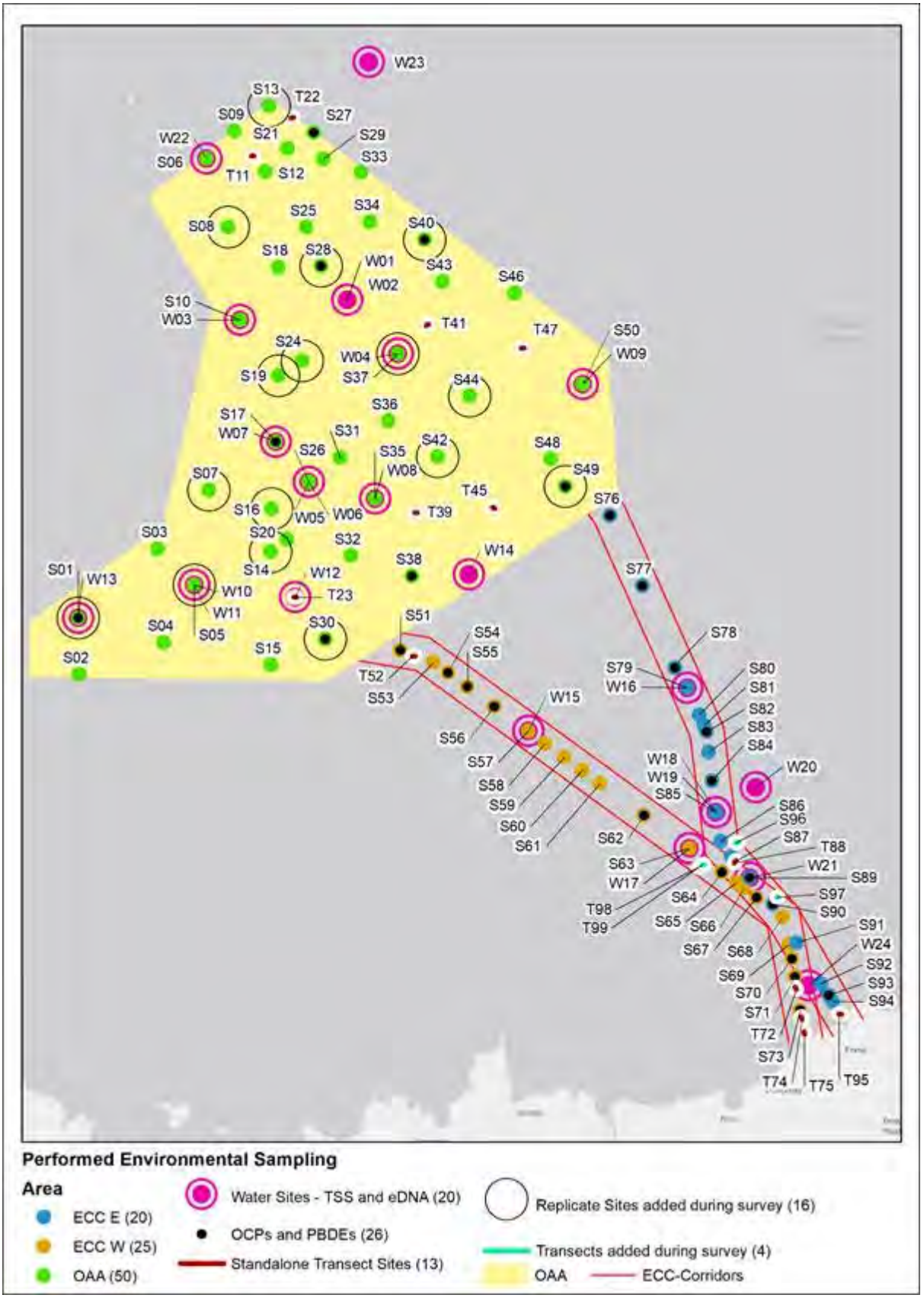

Figure 1. Sampling locations for West of Orkney offshore wind farm.

Table 1 (attached separately). Taxon-by-sample table by read proportion.

Table 2 (attached separately). Taxon-by-sample table by read count.

Figure 2 (attached separately). The proportion of the sequencing output allocated to the different taxa (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each taxon for that sample. The size of the bubble is relative to the number of sequences from all taxa detected in that sample.

Figure 3 (next page). The proportion of the sequencing output allocated to the different families (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each family for that sample. The size of the bubble is relative to the number of sequences from all families detected in that sample.

NATURE METRICS
DNA-BASED MONITORING

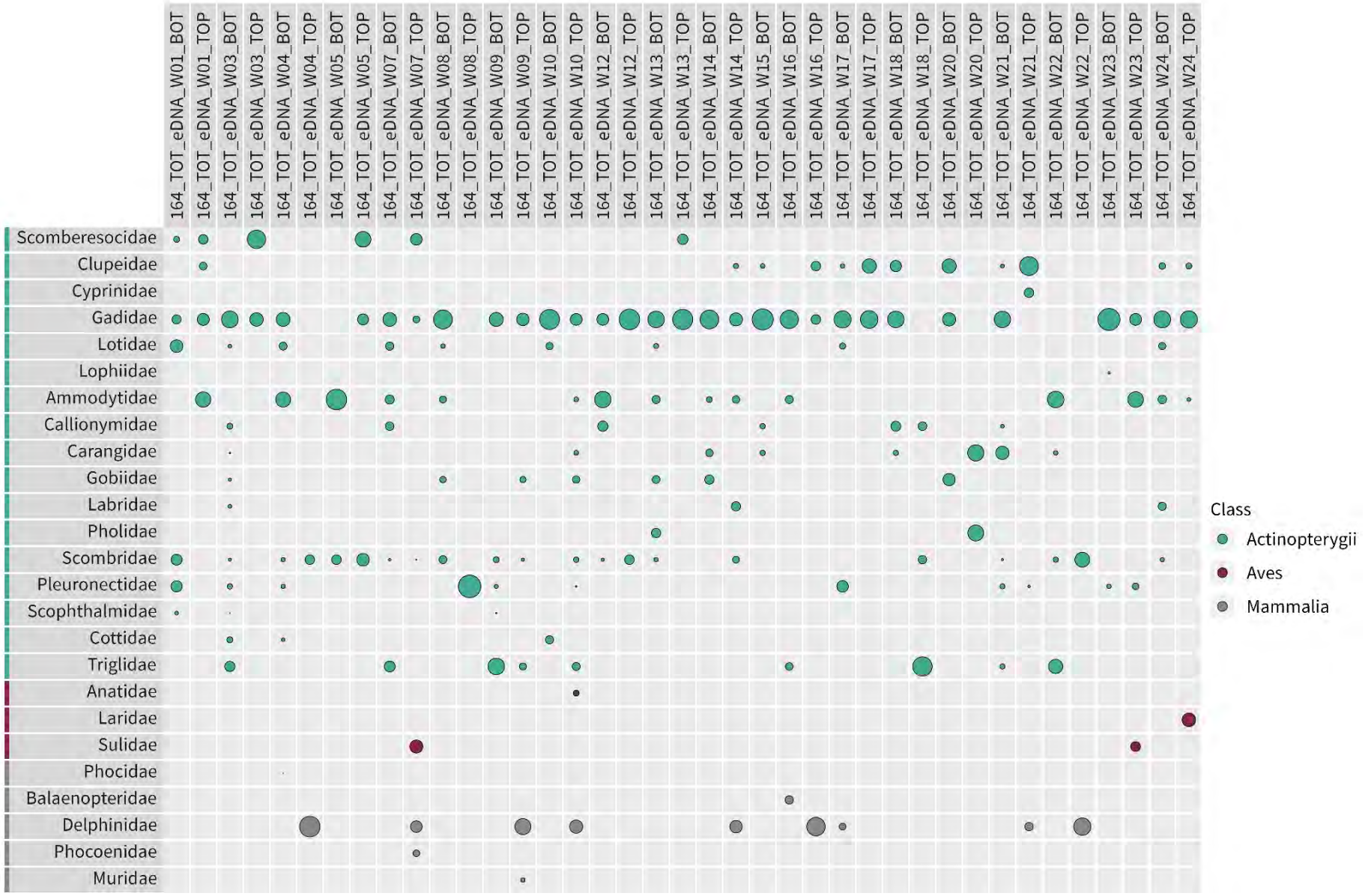

Table 3. Taxon richness among the samples.

Sample ID	Class	Order	Family	Genus	Taxa (Species)
164_TOT_eDNA_W01_BOT	1	4	6	3	7 (3)
164_TOT_eDNA_W01_TOP	1	4	4	1	4 (1)
164_TOT_eDNA_W03_BOT	1	4	11	11	15 (11)
164_TOT_eDNA_W03_TOP	1	2	2	1	3 (1)
164_TOT_eDNA_W04_BOT	2	5	7	5	9 (6)
164_TOT_eDNA_W05_TOP	1	3	3	2	3 (2)
164_TOT_eDNA_W07_BOT	1	3	6	5	7 (5)
164_TOT_eDNA_W07_TOP	3	5	6	4	6 (4)
164_TOT_eDNA_W08_BOT	1	2	5	4	5 (4)
164_TOT_eDNA_W08_TOP	1	1	1	1	2 (1)
164_TOT_eDNA_W09_BOT	1	4	5	3	6 (3)
164_TOT_eDNA_W09_TOP	2	5	6	7	9 (5)
164_TOT_eDNA_W10_BOT	1	2	3	3	4 (2)
164_TOT_eDNA_W10_TOP	3	6	9	6	10 (6)
164_TOT_eDNA_W12_BOT	1	2	4	4	5 (4)
164_TOT_eDNA_W13_BOT	1	2	6	6	7 (5)
164_TOT_eDNA_W13_TOP	1	2	2	1	2 (1)
164_TOT_eDNA_W14_BOT	1	2	4	4	5 (3)
164_TOT_eDNA_W14_TOP	2	4	6	5	7 (5)
164_TOT_eDNA_W15_BOT	1	3	4	3	6 (3)
164_TOT_eDNA_W16_BOT	2	4	4	3	6 (4)
164_TOT_eDNA_W16_TOP	2	3	3	1	3 (1)
164_TOT_eDNA_W17_BOT	2	4	5	4	6 (4)
164_TOT_eDNA_W18_BOT	1	3	4	3	5 (3)
164_TOT_eDNA_W18_TOP	1	2	3	2	3 (1)
164_TOT_eDNA_W20_TOP	1	1	2	2	2 (2)
164_TOT_eDNA_W21_BOT	1	5	7	4	8 (4)
164_TOT_eDNA_W21_TOP	2	4	4	3	4 (3)
164_TOT_eDNA_W22_BOT	1	2	4	4	5 (3)
164_TOT_eDNA_W23_BOT	1	3	3	3	5 (4)
164_TOT_eDNA_W23_TOP	2	4	4	3	4 (3)
164_TOT_eDNA_W24_BOT	1	3	6	7	9 (6)
164_TOT_eDNA_W24_TOP	2	4	4	2	5 (1)

Table 4 (attached separately). The frequency of occurrence of all detected families. Numbers correspond to the number of taxa belonging to those families in those samples.

METHODS

DNA from each filter was extracted using a commercial DNA extraction kit with a protocol modified to increase DNA yields. An extraction blank was also processed for the extraction batch. DNA was purified to remove PCR inhibitors using a commercial purification kit.

Comment: DNA yields were as expected.

Purified DNAs were amplified with PCR for a hypervariable region of the 12S rRNA gene to target vertebrates as part of the eDNA survey - Vertebrate workflow. Our standard analysis includes 12 replicate PCRs per sample.

All PCRs were performed in the presence of both a negative control and a positive control sample (a mock community with a known composition). Amplification success was determined by gel electrophoresis.

Comment: PCR reactions were successful for 39 of 40 samples. Electrophoresis bands were strong and of the expected size. Sample '164_TOT_eDNA_W15_TOP' failed to amplify despite troubleshooting steps. Overall, 4-12 successful PCRs replicates were obtained for each of the 39 samples submitted for sequencing. No bands were observed on electrophoresis gels for the extraction blank or negative controls.

PCR replicates were pooled and purified, and sequencing adapters were added. Success was determined by gel electrophoresis.

Comment: All samples were successfully indexed, electrophoresis bands were strong and of the expected size. No repeat reactions were necessary.

Amplicons were purified and checked by gel electrophoresis, these were then quantified using a Qubit high sensitivity kit according to the manufacturer's protocol.

Comment: All amplicons were successfully purified.

All purified index PCRs were pooled into a final library with equal concentrations. The final library was sequenced using an Illumina MiSeq V3 kit at 10.5 pM with a 20% PhiX spike in.

Sequence data were processed using a custom bioinformatics workflow for quality filtering, OTU clustering, and taxonomic assignment.

Comment: Both negative and positive controls were as expected. Very few sequences were discarded prior to dereplication, which is indicative of high-quality data with minimal PCR and sequencing errors. A total of 285,230 high-quality sequences were included in the final dataset.

Consensus taxonomic assignments were made for each OTU using sequence similarity searches against the NCBI nt (GenBank) reference database. Assignments were made to the lowest possible taxonomic level where there was consistency in the matches. Conflicts were flagged and resolved manually. Minimum similarity thresholds of 99%, 97%, and 95% were used for species-, genus- and higher-level assignments respectively. In cases where there were equally good matches to multiple species, public records from GBIF were used to assess which were most likely to be present in the

United Kingdom. Higher-level taxonomic identifications or multiple potential identifications were reported in cases that could not be resolved in this way.

The OTU table was then filtered to remove low abundance OTUs from each sample (<0.05% or <10 reads, whichever is the greater threshold for the sample). Unidentified, non-target, and common contaminant sequences were then removed.

Note that unidentified or misidentified taxa can result from incomplete or incorrect reference databases, and taxa may be missed due to low quality DNA, environmental contaminants, or the dominance of other species in the sample.

Please note that the abundance of taxa cannot be directly inferred from the proportion of total sequence reads. While the proportion of sequence reads is a consequence of abundance, it is also impacted by biomass, activity, surface area, condition, distance from the physical sample, primer bias, and species-specific variation in the genome.

Table 5 (attached separately). Sample information table.

END OF REPORT

Report issued by: Ben Jones

Contact: team@naturemetrics.co.uk

GLOSSARY

adapter	short, artificially synthesised nucleotide sequence which attaches to the ends of the target DNA or RNA sequences prior to sequencing. They are typically used to aid in attachment of the target sequence to other functional molecules/sequences.
amplicon	A DNA sequence which is the product of PCR amplification.
bioinformatics	An interface between genetics, computational biology, statistics, and programming in which DNA or other biological data is processed, analysed and integrated into research or communications.
bioinformatics workflow	Refers to a data processing workflow that takes the raw sequence data from high-throughput sequencing (often 20 million sequences or more) and transforms it into usable ecological data. Key steps for metabarcoding workflows include quality filtering, trimming, merging paired ends, removal of sequencing errors such as chimeras, clustering of similar sequences into molecular Operational Taxonomic Units, and matching one sequence from each cluster against a reference database. The output is a OTU-by-sample table showing how many sequences from each sample were assigned to each OTU.
BMWP	Short for biological monitoring working party, an index that can be used to measure water quality by scoring the presence of aquatic invertebrate indicator taxa. The index is reliant on taxa that are less tolerant of polluted water bodies (e.g. Ephemeroptera, Plecoptera, Trichoptera).
BOLD	Barcode Of Life Database; a specialised database of eukaryote COI reference sequences.
contaminant sequences	The sensitivity of high-throughput sequencing of eDNA means that contamination is always a concern that needs to be minimised. The sources of contamination are threefold: Natural - Examples of natural contaminants include: frequent visitors to site, faecal discharge from predators, livestock, wastewater, and fishing bait. This type of contamination is typically unavoidable and very difficult to quantify. Sequences of this type are typically flagged and conservatively removed from the sequencing output. Typical contaminant species include cow, pig, dog, cat, sheep, etc. Sampling - Human contamination of sampling equipment can reduce the efficiency of the sequencing. This type of

	contamination can be minimised by stringent contamination protocols, such as PPE. Laboratory - Residual DNA can contaminate other samples processed at the same time in other labs. At NatureMetrics this is mitigated by a designated eDNA laboratory, strict decontamination procedures, negative controls, and good laboratory practices.
dereplication	The identification of unique sequences so that only one copy of each sequence is reported.
eBioAtlas	A global partnership between IUCN and NatureMetrics to map the world's biodiversity using DNA from water samples as a foundation for the Global Biodiversity Framework and to enable IUCN Red List Assessments.
eDNA	Short for 'environmental DNA'. Refers to DNA deposited in the environment through excretion, shedding, mucous secretions, saliva etc. This can be collected in environmental samples (e.g. water, sediment) and used to identify the organisms that it originated from. eDNA in water is broken down by environmental processes over a period of days to weeks. It can travel some distance from the point at which it was released from the organism, particularly in running water. eDNA is sampled in low concentrations and can be degraded (i.e. broken into short fragments), which limits the analysis options.
extraction blank	A DNA extraction with no sample added to assess potential contamination during the DNA extraction process.
gel electrophoresis	The process in which DNA is separated according to size and electrical charge via an electric current, while in a gel. The process is used to confirm the successful amplification of a specifically sized fragment of DNA.
high-throughput sequencing	Technology developed in the 2000s that produces millions of sequences in parallel. Enables thousands of different organisms from a mixture of species to be sequenced at once, so community DNA can be sequenced. Various different technologies exist to do this, but the most commonly used platform is Illumina's MiSeq. Also known as Next-Generation Sequencing (NGS) or parallel sequencing.
inhibitors/inhibition	Naturally-occurring chemicals/compounds that cause DNA amplification to fail, potentially resulting in false negative results. Common inhibitors include tannins, humic acids and other organic compounds. Inhibitors can be overcome by either diluting the DNA (and the inhibitors) or by additional cleaning of the DNA, but

dilution carries the risk of reducing the DNA concentration below the limits of detection. At NatureMetrics, inhibition is removed using a commercial purification kit.

invasive

Invasive species are defined using GRIIS (Global Register of Introduced and Invasive Species) which is a checklist of Introduced and Invasive species for each country. The IUCN describes an Introduced species as a species outside of its natural range and dispersal potential, and an Invasive species as an introduced species which becomes established in a habitat, is an agent of change or threatens native biological diversity.

IUCN Red List

The IUCN (International Union for the Conservation of Nature) is a global union of government and civil organisations that disseminates information to assist conservation. The IUCN Red List of Threatened Species is an inventory of the conservation status of over 100,000 species worldwide. The Red List evaluates data such as population trends, geographic range and the number of mature individuals in order to categorise species based on their extinction risk:

Extinct (EX) - No individual of this species remains alive.

Extinct in the Wild (EW) - Surviving individuals are only found in captivity.

Critically Endangered (CE) - species faces an extremely high risk of extinction in the wild. e.g. Population size estimated at fewer than 50 mature individuals.

Endangered (EN) - species faces a very high risk of extinction in the wild. e.g. Population size estimated at fewer than 250 mature individuals.

Vulnerable (VU) - species faces a high risk of extinction in the wild. e.g. Population size estimated at fewer than 10,000 mature individuals and declining.

Near Threatened (NT) - species is below the threshold for any of the threatened categories (CE, E, V) but is close to this threshold or is expected to pass it in the near future.

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Data Deficient (DD) - There is currently insufficient data available to make an assessment of extinction risk. This is not a threat category - when more data becomes available the species may be recategorised as threatened.

Jaccard similarity index

This index is a calculation that compares two samples to see which taxa are shared and which are distinct. The higher the percentage,

	the more similar two samples are in their community composition.
metabarcoding	Refers to identification of species assemblages from community DNA using barcode genes. PCR is carried out with non-specific primers, followed by high-throughput sequencing and bioinformatics processing. Can identify hundreds of species in each sample, and 100+ different samples can be processed in parallel to reduce sequencing cost.
NCBI nt	National Centre for Biotechnology Information nucleotide database; a general reference database.
negative control	Used to determine whether PCR reactions are contaminated.
NMDS	Non-metric multidimensional scaling (NMDS) is a method that allows visualisation of the similarity of each sample to one another. The dissimilarity between each sample is calculated, taking into account shared taxa (Jaccard similarity index), and then configured into a 2D ordinal space that allows the similarity-based relationship between each sample to be plotted. Samples which are closer together are more similar to one another in terms of community composition, while samples which are further apart are less similar. This type of clustering analysis allows you to see if certain types of samples, for example, those from a particular habitat type, are more clustered together and therefore more similar to one another compared to other groups.
nucleotide	An individual unit of genetic material which, when strung together constitutes a DNA (or RNA) strand/sequence.
OTU	Operational Taxonomic Unit; similar sequences are clustered into OTUs at a defined similarity threshold. OTUs are approximately equivalent to species and are treated as such in our analyses. Species-level taxonomic assignments may or may not be possible, depending on the availability of reference sequences and the similarity between closely related species in the amplified marker. It may be possible to refine the taxonomic assignment for an OTU later as more sequences are added to reference databases.
PCR	Polymerase Chain Reaction; a process by which millions of copies of a particular DNA segment are produced through a series of heating and cooling steps. Known as an 'amplification' process. One of the most common processes in molecular biology and a precursor to most sequencing-based analyses.

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primers	Short sections of synthesised DNA that bind to either end of the DNA segment to be amplified by PCR. Can be designed to be totally specific to a particular species (so that only that species' DNA will be amplified from a community DNA sample), or to be very general so that a wide range of species' DNA will be amplified. Good design of primers is one of the critical factors in DNA-based monitoring.
rarefaction curve	A plot showing the number of taxa as a function of the sequencing depth (number of reads). Rarefaction curves grow rapidly at first as common species are found then reach a plateau as only the rarest species remain to be detected. Rarefaction curves can provide an indication as to whether the species being studied have been comprehensively sampled.
rarefy	A normalisation technique which transforms the data to remove biases associated with uneven sampling depth (number of reads) across samples. The sampling depth of each sample is standardised to a specified number of reads (usually that of the sample with the lowest depth) by random resampling.
reference databases	Over time, the DNA sequences of many species have been compiled into publicly accessible databases by scientists from around the world. These databases serve as a reference against which unknown sequences can be queried to obtain a species identification. The most commonly accessed database is NCBI, which is maintained by the US National Institute of Health. Anyone can search for DNA sequences at https://www.ncbi.nlm.nih.gov.
richness	The total number of taxa within a sample.
rRNA	Ribosomal RNA.
SAC species	Typically the presence of these species potentially elevates the conservation status of a site to a Special Area of Conservation (SAC). Special Areas of Conservation (SACs) are strictly protected sites designated under the EC Habitats Directive.
sequence(s)	A DNA sequence is made up of four nucleotide bases represented by the letters A, T, C & G. The precise order of these letters is used to compare genetic similarity among individuals or species and to identify species using reference databases. In high-throughput sequencing analyses (e.g. metabarcoding), many identical copies of the same sequence are obtained for each species in the sample. The number of copies obtained per species is known as the number of sequence reads, and this is often -

	although not always - related to the relative abundance of the species.
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taxonomy	The branch of science concerned with classification of organisms. species (s./pl.) - A group of genetically similar organisms that show a high degree of overall similarity in many independent characteristics. Related species are grouped together into progressively larger taxonomic units, from genus to kingdom. Homo sapiens (human) is an example of a species. genus (s.) / genera (pl.) - A group of closely related species. Each genus can include one or more species. Homo is an example of a genus. family (s.) / families (pl.) - A group of closely related genera. Homo sapiens is in the Family Hominidae (great apes). order (s.) / orders (pl.) - A group of closely related families. Homo sapiens is in the Order Primates. class (s.) / classes (pl.) - A group of closely related orders. Homo sapiens is in the Class Mammalia. phylum (s.) / phyla (pl.) - A group of closely related classes. Homo sapiens is in the Phylum Chordata.
UKBAP species	UK Biodiversity Action Plan species have been identified as being the most threatened and requiring conservation action under the UK Biodiversity Action Plan.
UNITE	A ribosomal RNA database for identification of fungi.

MARINE WATER EUKARYOTES METABARCODING **RESULTS**

Order number:	SO01060
Report number:	NM-AFU619
Company:	APEM Ltd
Contact:	Chris Ashelby
Project:	Total Energies - West of Orkney Wind farm
Sample type:	NatureMetrics eDNA disk filter
Date of report:	27-Jan-2023
Number of samples:	20

Thank you for sending your samples for analysis by NatureMetrics. Your samples have been metabarcoded following our eDNA survey - Marine eukaryotes from water workflow. **A taxon-by-sample table of your samples is attached to this report (NM-AFU619.SO01060.Marine water eukaryotes.xlsx)**. Each row in the table represents one taxon (OTU), shown with the lowest possible taxonomic assignment based on currently available reference data. Each column represents a sample, showing the percentage of sequence reads per detected OTU (Table 1) and the number of sequence reads per detected OTU (Table 2) in that sample. Care should be taken in interpreting the numbers in terms of relative species abundance, but a high sequence proportion can be interpreted as lending greater confidence to a detection. This report contains biodiversity information that may be sensitive, particularly with respect to endangered or protected species. It is the responsibility of the client to ensure that due consideration is given to the data and that the information is shared in a responsible way.

Here we present an overview of the key results, followed by a more detailed report that starts with the taxonomic composition of the samples followed by a more detailed look at the steps taken to extract, amplify, sequence, and analyse your DNA. A glossary for terms in bold is provided at the end of the report to define key terms used within the report.

OVERVIEW OF YOUR RESULTS

- A total of 407 taxa were detected
- Average taxon richness was 143 and ranged from 67 to 179
- Most abundant sequence: Siphonophorae sp.
- Most commonly detected taxa: 7 species were detected in all 20 samples

FULL REPORT

Sample composition

A total of 407 taxa were detected (**Table 1** and **Table 2**). 16% (65 taxa) were at least 99% similar to a species in the global reference databases, and species names are suggested. The remaining taxa were identified to the lowest possible taxonomic level: 23.8% to genus (97 taxa), 24.1% to family (98 taxa), 13% to order (53 taxa), 17.7% to class (72 taxa), 4.9% to phylum (20 taxa) and the remainder to kingdom (2 taxa). The taxa belong to 28 phyla, 44 classes, 95 orders, 129 families, and 96 genera.

The average taxon richness was 143 and ranged from 67 ('164_TOT_eDNA_W22_BOT') to 179 ('164_TOT_eDNA_W20_BOT'). The relative proportion of the sequences found in each of the samples is shown in **Figure 2**, **Figure 3**, **Table 1** and **Table 2** and the diversity is summarised in **Table 3** and **Table 4**.

A species in the order Siphonophorae, which accounted for 17.5% of the total sequence reads, was among the most abundant in terms of sequences. Among the most commonly detected taxa were *Acartia clausii*, *Bathycoccus prasinos, Oithona similis*, *Picomonas judraskeda, Pseudocalanus* elongatus, *Strombidium caudispina*, and *Teleaulax amphioxeia*, which were detected in all 20 samples.

No IUCN Red List taxa were detected using this assay.

High-quality marine eukaryote sequence data were obtained for all 20 of the eDNA samples.

All laboratory controls behaved as expected.

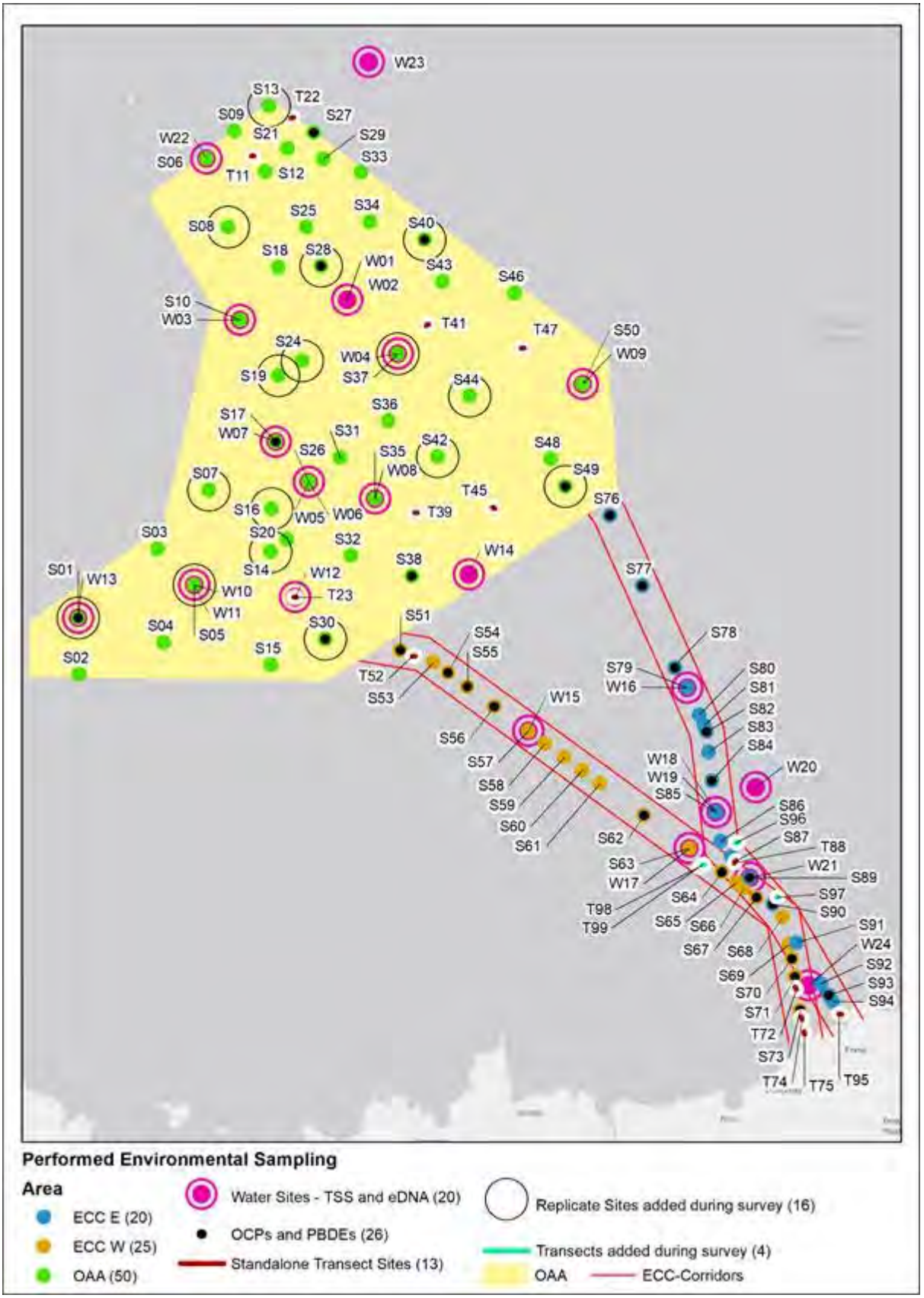

Figure 1. Sampling locations for West of Orkney offshore wind farm.

Table 1 (attached separately). Taxon-by-sample table by read proportion.

Table 2 (attached separately). Taxon-by-sample table by read count.

Figure 2 (attached separately). The proportion of the sequencing output allocated to the different taxa (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each taxon for that sample. The size of the bubble is relative to the number of sequences from all taxa detected in that sample.

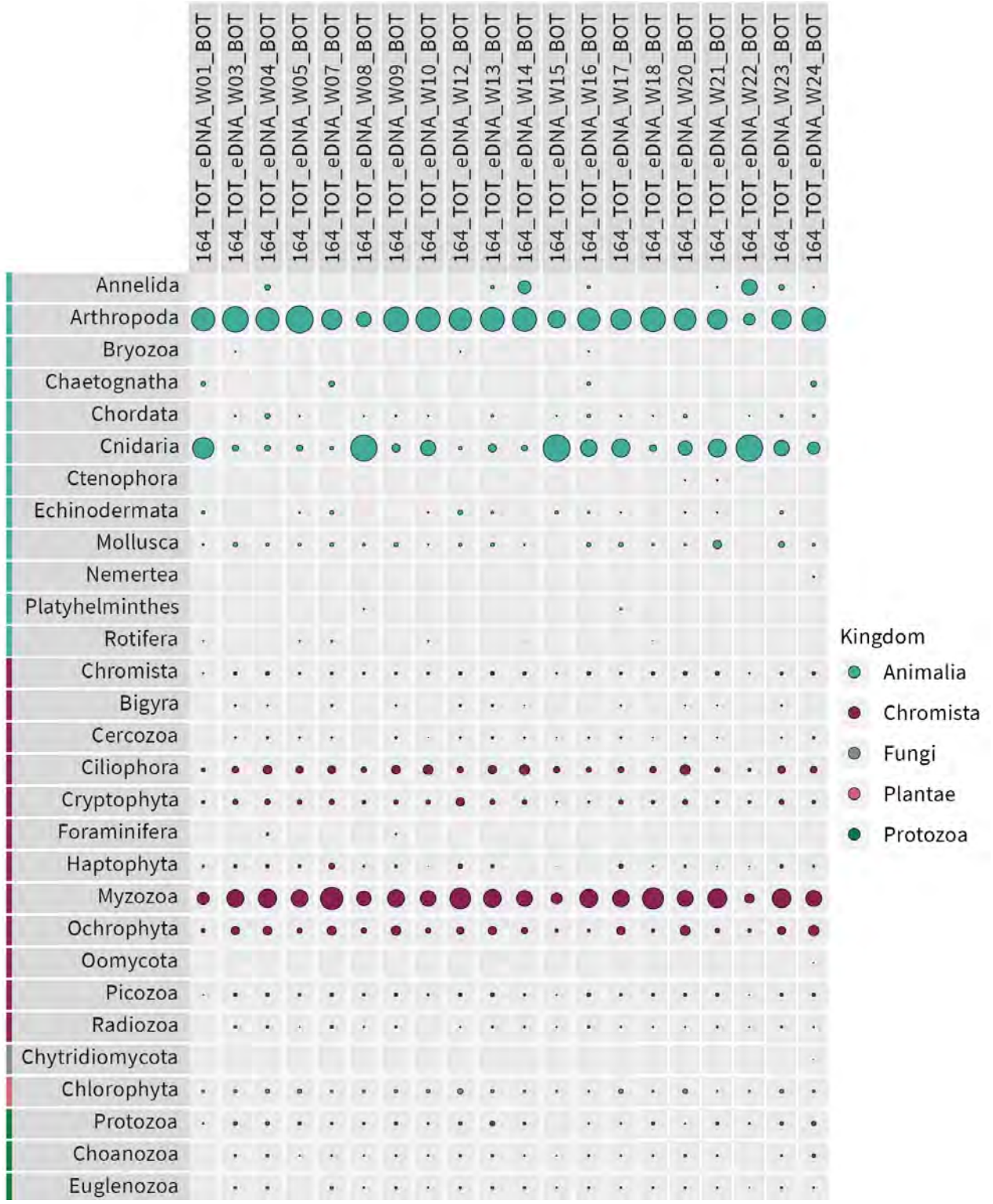

Figure 3. The proportion of the sequencing output allocated to the different phyla (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each phylum for that sample. The size of the bubble is relative to the number of sequences from all phyla detected in that sample.

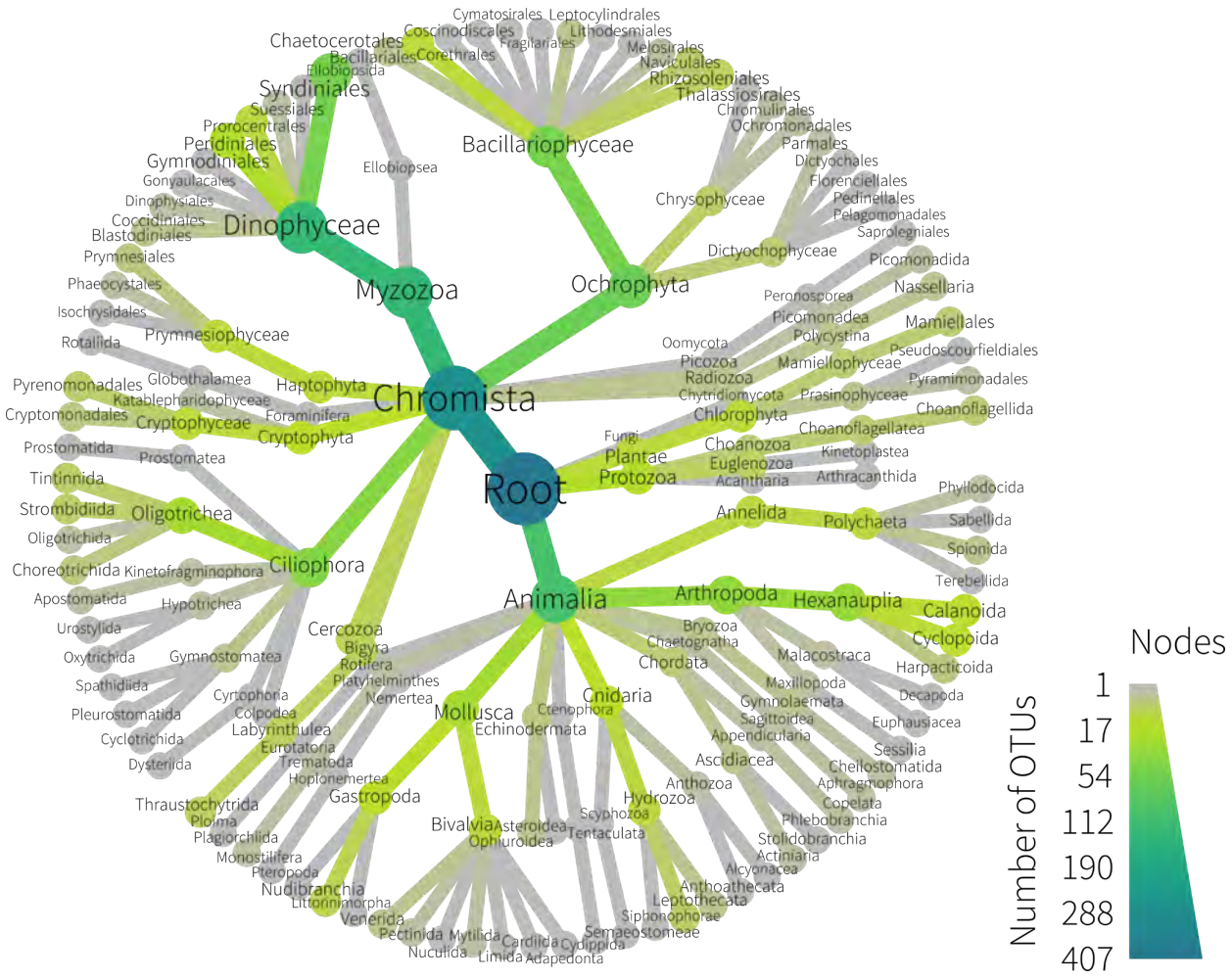

Figure 4. A taxonomic heat tree showing the number of OTUs across all samples down to the order rank. Each node (the circles) is a taxon and the edges (lines) show hierarchical relationships between taxa. The colour scale and the relative width of the node represent the number of taxa at each level.

Table 3. Taxon richness among the samples.

Sample ID	Class	Order	Family	Genus	Taxa (Species)
164_TOT_eDNA_W01_BOT	17	32	39	31	92 (14)
164_TOT_eDNA_W03_BOT	28	49	62	51	169 (36)
164_TOT_eDNA_W04_BOT	27	51	66	54	177 (37)
164_TOT_eDNA_W05_BOT	22	41	55	43	147 (27)
164_TOT_eDNA_W07_BOT	25	47	61	49	166 (27)
164_TOT_eDNA_W08_BOT	19	37	48	39	118 (22)
164_TOT_eDNA_W09_BOT	25	48	60	50	167 (33)
164_TOT_eDNA_W10_BOT	20	37	52	43	135 (25)
164_TOT_eDNA_W12_BOT	24	43	60	49	177 (33)
164_TOT_eDNA_W13_BOT	27	51	66	51	178 (33)
164_TOT_eDNA_W14_BOT	24	39	52	40	125 (23)
164_TOT_eDNA_W15_BOT	15	25	33	26	73 (16)
164_TOT_eDNA_W16_BOT	23	33	43	37	128 (21)
164_TOT_eDNA_W17_BOT	23	45	56	42	160 (32)
164_TOT_eDNA_W18_BOT	22	35	52	40	138 (23)
164_TOT_eDNA_W20_BOT	25	44	58	50	179 (37)
164_TOT_eDNA_W21_BOT	24	37	46	34	127 (22)
164_TOT_eDNA_W22_BOT	15	24	32	28	67 (15)
164_TOT_eDNA_W23_BOT	27	52	62	50	165 (33)
164_TOT_eDNA_W24_BOT	23	43	62	50	172 (33)

Table 4 (attached separately). The frequency of occurrence of all detected families. Numbers correspond to the number of taxa belonging to those families in those samples.

METHODS

DNA from each filter was extracted using a commercial DNA extraction kit with a protocol modified to increase DNA yields. An extraction blank was also processed for the extraction batch. DNA was purified to remove PCR inhibitors using a commercial purification kit.

Comment: DNA yields were as expected.

Purified DNAs were amplified with PCR for a hypervariable region of the 18S rRNA gene to target marine eukaryotes as part of the eDNA survey - Marine eukaryotes from water workflow. Our standard analysis includes 3 replicate PCRs per sample.

All PCRs were performed in the presence of both a negative control and a positive control sample. Amplification success was determined by gel electrophoresis.

Comment: PCR reactions were successful for all 20 samples. Electrophoresis bands were strong and of the expected size. Overall, 3 successful PCRs replicates were obtained for each of the 20 samples submitted for sequencing. No bands were observed on electrophoresis gels for the extraction blank or negative controls.

PCR replicates were pooled and purified, and sequencing adapters were added. Success was determined by gel electrophoresis.

Comment: All samples were successfully indexed, electrophoresis bands were strong and of the expected size. No repeat reactions were necessary.

Amplicons were purified and checked by gel electrophoresis, these were then quantified using a Qubit high sensitivity kit according to the manufacturer's protocol.

Comment: All amplicons were successfully purified.

All purified index PCRs were pooled into a final library with equal concentrations. The final library was sequenced using an Illumina MiSeq V3 kit at 10.5 pM with a 20% PhiX spike in.

Sequence data were processed using a custom bioinformatics workflow for quality filtering, OTU clustering, and taxonomic assignment.

Comment: Both negative and positive controls were as expected. Very few sequences were discarded prior to dereplication, which is indicative of high-quality data with minimal PCR and sequencing errors. A total of 937,652 high-quality sequences were included in the final dataset.

Consensus taxonomic assignments were made for each OTU using sequence similarity searches against two reference databases appropriate for the dataset, one generic (NCBI *nt*) and one specialised (SILVA 18S v138.1). The GBIF taxonomic backbone was used for consistency between databases. Results from both searches were combined and assignments made to the lowest possible taxonomic level where there was consistency in the matches. Conflicts were flagged and resolved manually. Minimum similarity thresholds of 98%, 95%, and 92% were required for species-, genus-, and higher-level assignments respectively. Identifications that were based on fewer than three reference matches have been flagged.

The OTU table was then filtered to remove low abundance OTUs from each sample (<0.035% or <10 reads, whichever is the greater threshold for the sample). Unidentified, non-target, and common contaminant sequences were then removed.

Note that unidentified or misidentified taxa can result from incomplete or incorrect reference databases, and taxa may be missed due to low quality DNA, environmental contaminants, or the dominance of other species in the sample.

Please note that the abundance of taxa cannot be directly inferred from the proportion of total sequence reads. While the proportion of sequence reads is a consequence of abundance, it is also impacted by biomass, activity, surface area, condition, distance from the physical sample, primer bias, and species-specific variation in the genome.

Table 5 (attached separately). Sample information table.

END OF REPORT

Report issued by: Ben Jones

Contact: team@naturemetrics.co.uk

GLOSSARY

adapter	short, artificially synthesised nucleotide sequence which attaches to the ends of the target DNA or RNA sequences prior to sequencing. They are typically used to aid in attachment of the target sequence to other functional molecules/sequences.
amplicon	A DNA sequence which is the product of PCR amplification.
bioinformatics	An interface between genetics, computational biology, statistics, and programming in which DNA or other biological data is processed, analysed and integrated into research or communications.
bioinformatics workflow	Refers to a data processing workflow that takes the raw sequence data from high-throughput sequencing (often 20 million sequences or more) and transforms it into usable ecological data. Key steps for metabarcoding workflows include quality filtering, trimming, merging paired ends, removal of sequencing errors such as chimeras, clustering of similar sequences into molecular Operational Taxonomic Units, and matching one sequence from each cluster against a reference database. The output is a OTU-by-sample table showing how many sequences from each sample were assigned to each OTU.
BMWP	Short for biological monitoring working party, an index that can be used to measure water quality by scoring the presence of aquatic invertebrate indicator taxa. The index is reliant on taxa that are less tolerant of polluted water bodies (e.g. Ephemeroptera, Plecoptera, Trichoptera).
BOLD	Barcode Of Life Database; a specialised database of eukaryote COI reference sequences.
contaminant sequences	The sensitivity of high-throughput sequencing of eDNA means that contamination is always a concern that needs to be minimised. The sources of contamination are threefold: Natural - Examples of natural contaminants include: frequent visitors to site, faecal discharge from predators, livestock, wastewater, and fishing bait. This type of contamination is typically unavoidable and very difficult to quantify. Sequences of this type are typically flagged and conservatively removed from the sequencing output. Typical contaminant species include cow, pig, dog, cat, sheep, etc. Sampling - Human contamination of sampling equipment can reduce the efficiency of the sequencing. This type of

	contamination can be minimised by stringent contamination protocols, such as PPE. Laboratory - Residual DNA can contaminate other samples processed at the same time in other labs. At NatureMetrics this is mitigated by a designated eDNA laboratory, strict decontamination procedures, negative controls, and good laboratory practices.
dereplication	The identification of unique sequences so that only one copy of each sequence is reported.
eBioAtlas	A global partnership between IUCN and NatureMetrics to map the world's biodiversity using DNA from water samples as a foundation for the Global Biodiversity Framework and to enable IUCN Red List Assessments.
eDNA	Short for 'environmental DNA'. Refers to DNA deposited in the environment through excretion, shedding, mucous secretions, saliva etc. This can be collected in environmental samples (e.g. water, sediment) and used to identify the organisms that it originated from. eDNA in water is broken down by environmental processes over a period of days to weeks. It can travel some distance from the point at which it was released from the organism, particularly in running water. eDNA is sampled in low concentrations and can be degraded (i.e. broken into short fragments), which limits the analysis options.
extraction blank	A DNA extraction with no sample added to assess potential contamination during the DNA extraction process.
gel electrophoresis	The process in which DNA is separated according to size and electrical charge via an electric current, while in a gel. The process is used to confirm the successful amplification of a specifically sized fragment of DNA.
high-throughput sequencing	Technology developed in the 2000s that produces millions of sequences in parallel. Enables thousands of different organisms from a mixture of species to be sequenced at once, so community DNA can be sequenced. Various different technologies exist to do this, but the most commonly used platform is Illumina's MiSeq. Also known as Next-Generation Sequencing (NGS) or parallel sequencing.
inhibitors/inhibition	Naturally-occurring chemicals/compounds that cause DNA amplification to fail, potentially resulting in false negative results. Common inhibitors include tannins, humic acids and other organic compounds. Inhibitors can be overcome by either diluting the DNA (and the inhibitors) or by additional cleaning of the DNA, but

dilution carries the risk of reducing the DNA concentration below the limits of detection. At NatureMetrics, inhibition is removed using a commercial purification kit.

invasive

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rarefy	A normalisation technique which transforms the data to remove biases associated with uneven sampling depth (number of reads) across samples. The sampling depth of each sample is standardised to a specified number of reads (usually that of the sample with the lowest depth) by random resampling.
reference databases	Over time, the DNA sequences of many species have been compiled into publicly accessible databases by scientists from around the world. These databases serve as a reference against which unknown sequences can be queried to obtain a species identification. The most commonly accessed database is NCBI, which is maintained by the US National Institute of Health. Anyone can search for DNA sequences at https://www.ncbi.nlm.nih.gov.
richness	The total number of taxa within a sample.
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UKBAP species	UK Biodiversity Action Plan species have been identified as being the most threatened and requiring conservation action under the UK Biodiversity Action Plan.
UNITE	A ribosomal RNA database for identification of fungi.

MAMMAL METABARCODING **RESULTS**

Order number:	SO01060
Report number:	NM-LFT120
Company:	APEM Ltd
Contact:	Chris Ashelby
Project:	Total Energies - West of Orkney Wind farm
Sample type:	NatureMetrics eDNA disk filter
Date of report:	27-Jan-2023
Number of samples:	40

Thank you for sending your samples for analysis by NatureMetrics. Your samples have been metabarcoded following our eDNA survey - Mammal workflow. **A taxon-by-sample table of your samples is attached to this report (NM-LFT120.SO01060.Mammal.xlsx)**. Each row in the table represents one taxon (OTU), shown with the lowest possible taxonomic assignment based on currently available reference data. Each column represents a sample, showing the percentage of sequence reads per detected OTU (Table 1) and the number of sequence reads per detected OTU (Table 2) in that sample. Care should be taken in interpreting the numbers in terms of relative species abundance, but a high sequence proportion can be interpreted as lending greater confidence to a detection. This report contains biodiversity information that may be sensitive, particularly with respect to endangered or protected species. It is the responsibility of the client to ensure that due consideration is given to the data and that the information is shared in a responsible way.

Here we present an overview of the key results, followed by a more detailed report that starts with the taxonomic composition of the samples followed by a more detailed look at the steps taken to extract, amplify, sequence, and analyse your DNA. A glossary for terms in bold is provided at the end of the report to define key terms used within the report.

OVERVIEW OF YOUR RESULTS

- A total of 7 taxa were detected.
- Average taxon richness was 1.78 and ranged from 1 to 4.
- Most abundant sequences: common bottlenose dolphin (*Tursiops truncatus*).
- Most commonly detected taxa: common dolphin/striped dolphin (*Delphinus delphis*/*Stenella coeruleoalba*), common bottlenose dolphin (*Tursiops truncatus*) and grey seal (*Halichoerus grypus*).
- Mammal sequence data were obtained from 9 of 40 eDNA sequences.

FULL REPORT

Sample composition

A total of 7 taxa were detected (**Table 1** and **Table 2**). 85.7% (6 taxa) were at least 99% similar to a species in the global reference databases, and species names are suggested. The remaining taxon was identified to family. The taxa belong to 3 orders, 5 families, and 6 genera.

The average taxon richness was 1.78 and ranged from 1 ('164_TOT_eDNA_W12_BOT, '164_TOT_eDNA_W13_BOT', '164_TOT_eDNA_W15_BOT', & '164_TOT_eDNA_W18_TOP') to 4 ('164_TOT_eDNA_W09_TOP'). The relative proportion of the sequences found in each of the samples is shown in **Figure 2**, **Table 1** and **Table 2** and the diversity is summarised in **Table 3** and **Table 4**.

Common bottlenose dolphin (*Tursiops truncatus*), which accounted for 39.9% of the total sequence reads, was among the most abundant in terms of sequences. Among the most commonly detected species were common dolphin/striped dolphin (*Delphinus delphis/Stenella coeruleoalba*), common bottlenose dolphin (*Tursiops truncatus*) and grey seal (*Halichoerus grypus*), which were detected in 5, 4 and 3 samples, respectively.

Note that *Apodemus sylvaticus* (European woodmouse) is an unexpected detection in marine samples, however, we do not suspect any internal contamination (as the lab controls are clean, and this is not a species we see in high frequency or abundance in other samples). The species has been detected in the same sample (164_TOT_eDNA_W09_TOP) with the mammal and vertebrate assays, we believe that this is a real detection in the sample, likely to have originated during sample handling (e.g. mice on the marine vessels).

High-quality mammal sequence data were obtained for 9 of the 40 eDNA samples. eDNA metabarcoding of mammals was not successful for '164_TOT_eDNA_W15_TOP', which failed to amplify despite troubleshooting. The remaining 30 samples did not produce any target reads and have not been reported. The report status of all samples is summarised in ***Table 5***

All laboratory controls behaved as expected.

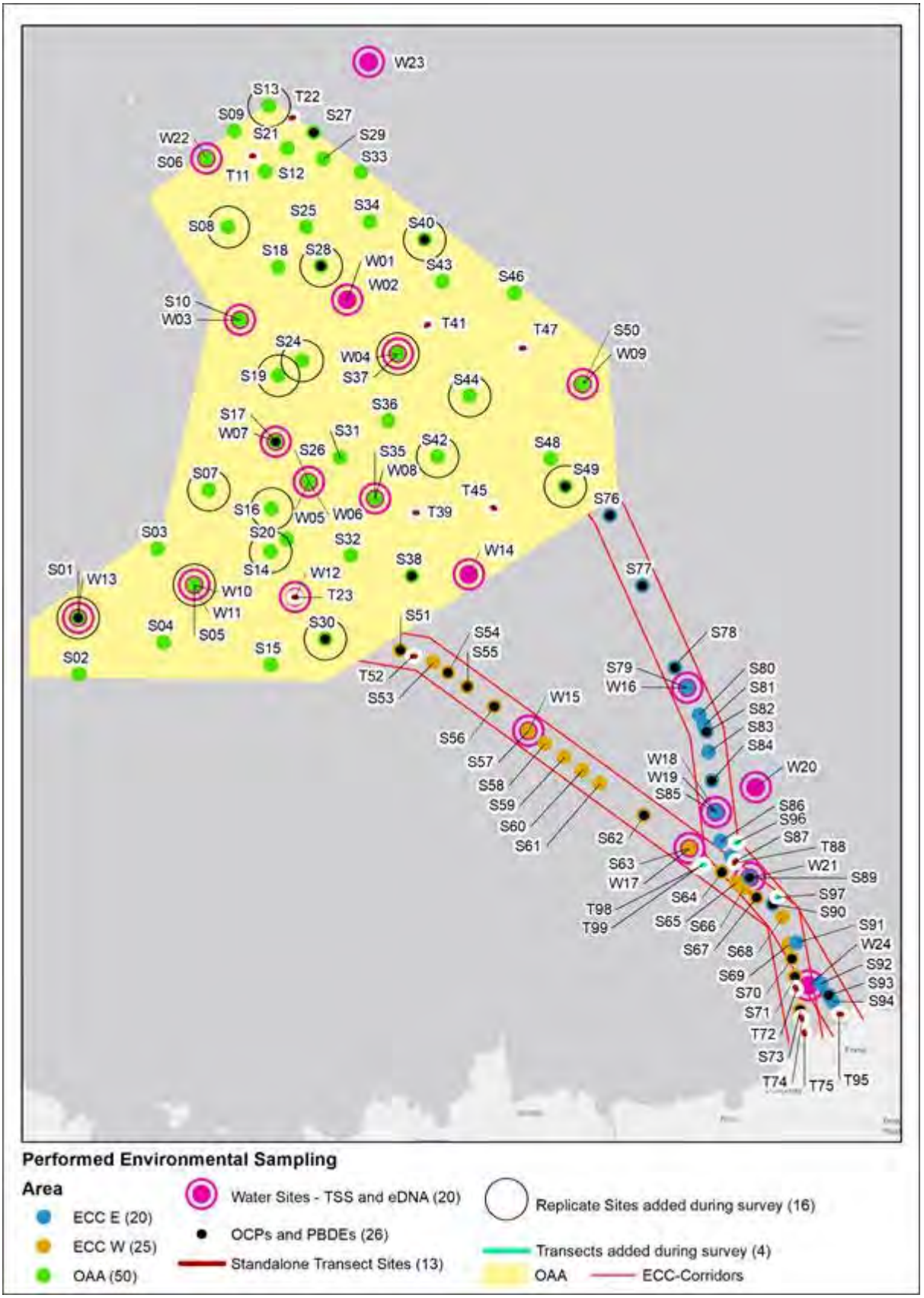

Figure 1. Sampling locations for West of Orkney offshore wind farm.

Table 1 (attached separately). Taxon-by-sample table by read proportion.

Table 2 (attached separately). Taxon-by-sample table by read count.

Figure 2 (attached separately). The proportion of the sequencing output allocated to the different taxa (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each taxon for that sample. The size of the bubble is relative to the number of sequences from all taxa detected in that sample.

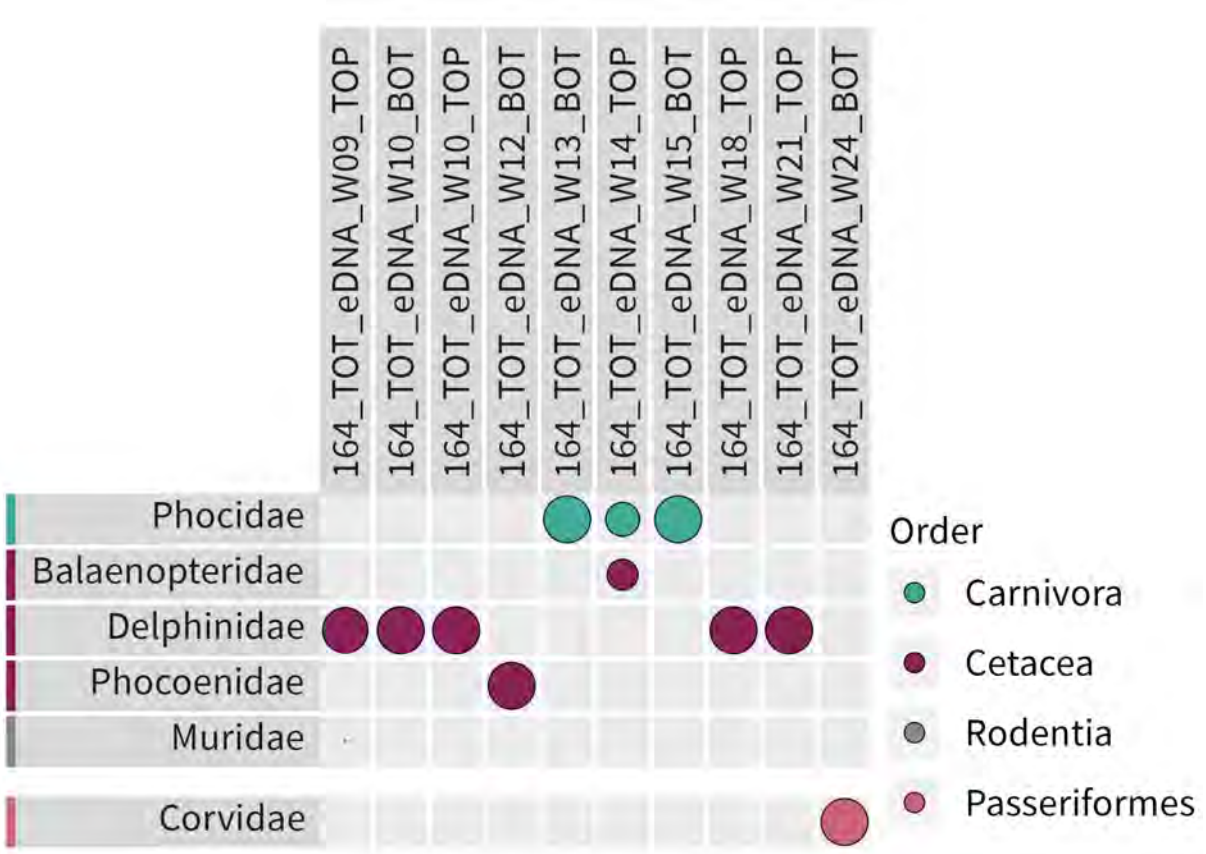

Figure 3. The proportion of the sequencing output allocated to the different families (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each family for that sample. The size of the bubble is relative to the number of sequences from all families detected in that sample.

Table 3. Taxon richness among the samples.

Sample ID	Class	Order	Family	Genus	Taxa (Species)
164_TOT_eDNA_W09_TOP	1	2	2	3	4 (3)
164_TOT_eDNA_W10_BOT	1	1	1	1	2 (1)
164_TOT_eDNA_W10_TOP	1	1	1	1	2 (1)
164_TOT_eDNA_W12_BOT	1	1	1	1	1 (1)
164_TOT_eDNA_W13_BOT	1	1	1	1	1 (1)
164_TOT_eDNA_W14_TOP	1	2	2	2	2 (2)
164_TOT_eDNA_W15_BOT	1	1	1	1	1 (1)
164_TOT_eDNA_W18_TOP	1	1	1	0	1 (0)
164_TOT_eDNA_W21_TOP	1	1	1	1	2 (1)

Table 4 (attached separately). The frequency of occurrence of all detected families. Numbers correspond to the number of taxa belonging to those families in those samples.

METHODS

DNA from each filter was extracted using a commercial DNA extraction kit with a protocol modified to increase DNA yields. An extraction blank was also processed for the extraction batch. DNA was purified to remove PCR inhibitors using a commercial purification kit.

Comment: DNA yields were as expected.

Purified DNAs were amplified with PCR for a hypervariable region of the 16S rRNA gene to target mammals as part of the eDNA survey - Mammal workflow. Our standard analysis includes 12 replicate PCRs per sample.

All PCRs were performed in the presence of both a negative control and a positive control sample (a mock community with a known composition). Amplification success was determined by gel electrophoresis.

Comment: PCR reactions were successful for 39 of 40 samples. Electrophoresis bands were strong and of the expected size. Sample '164_TOT_eDNA_W15_TOP' failed to amplify despite troubleshooting steps. Overall, 4-12 successful PCRs replicates were obtained for each of the 39 samples submitted for sequencing. No bands were observed on electrophoresis gels for the extraction blank or negative controls.

PCR replicates were pooled and purified, and sequencing adapters were added. Success was determined by gel electrophoresis.

Comment: All samples were successfully indexed, electrophoresis bands were strong and of the expected size. No repeat reactions were necessary.

Amplicons were purified and checked by gel electrophoresis, these were then quantified using a Qubit high sensitivity kit according to the manufacturer's protocol.

Comment: All amplicons were successfully purified.

All purified index PCRs were pooled into a final library with equal concentrations. The final library was sequenced using an Illumina MiSeq V3 kit at 10.5 pM with a 20% PhiX spike in.

Sequence data were processed using a custom bioinformatics workflow for quality filtering, OTU clustering, and taxonomic assignment.

Comment: Both negative and positive controls were as expected. Very few sequences were discarded prior to dereplication, which is indicative of high-quality data with minimal PCR and sequencing errors. 30 of 39 sequenced samples did not produce any target reads. A total of 243,386 high-quality sequences, including 243,386 target sequences, were included in the final dataset.

Consensus taxonomic assignments were made for each OTU using sequence similarity searches against the NCBI nt (GenBank) reference database. Assignments were made to the lowest possible taxonomic level where there was consistency in the matches. Conflicts were flagged and resolved manually. Minimum similarity thresholds of 99%, 97%, and 95% were used for species-, genus- and higher-level assignments respectively. In cases where there were equally good matches to multiple

species, public records from GBIF were used to assess which were most likely to be present in the United Kingdom. Higher-level taxonomic identifications or multiple potential identifications were reported in cases that could not be resolved in this way.

The OTU table was then filtered to remove low abundance OTUs from each sample (<0.05% or <10 reads, whichever is the greater threshold for the sample). Unidentified, non-target, and common contaminant sequences were then removed.

Note that unidentified or misidentified taxa can result from incomplete or incorrect reference databases, and taxa may be missed due to low quality DNA, environmental contaminants, or the dominance of other species in the sample.

Please note that the abundance of taxa cannot be directly inferred from the proportion of total sequence reads. While the proportion of sequence reads is a consequence of abundance, it is also impacted by biomass, activity, surface area, condition, distance from the physical sample, primer bias, and species-specific variation in the genome.

Table 5 (attached separately). Sample information table.

END OF REPORT

Report issued by: Ben Jones

Contact: team@naturemetrics.co.uk

GLOSSARY

adapter	short, artificially synthesised nucleotide sequence which attaches to the ends of the target DNA or RNA sequences prior to sequencing. They are typically used to aid in attachment of the target sequence to other functional molecules/sequences.
amplicon	A DNA sequence which is the product of PCR amplification.
bioinformatics	An interface between genetics, computational biology, statistics, and programming in which DNA or other biological data is processed, analysed and integrated into research or communications.
bioinformatics workflow	Refers to a data processing workflow that takes the raw sequence data from high-throughput sequencing (often 20 million sequences or more) and transforms it into usable ecological data. Key steps for metabarcoding workflows include quality filtering, trimming, merging paired ends, removal of sequencing errors such as chimeras, clustering of similar sequences into molecular Operational Taxonomic Units, and matching one sequence from each cluster against a reference database. The output is a OTU-by-sample table showing how many sequences from each sample were assigned to each OTU.
BMWP	Short for biological monitoring working party, an index that can be used to measure water quality by scoring the presence of aquatic invertebrate indicator taxa. The index is reliant on taxa that are less tolerant of polluted water bodies (e.g. Ephemeroptera, Plecoptera, Trichoptera).
BOLD	Barcode Of Life Database; a specialised database of eukaryote COI reference sequences.
contaminant sequences	The sensitivity of high-throughput sequencing of eDNA means that contamination is always a concern that needs to be minimised. The sources of contamination are threefold: Natural - Examples of natural contaminants include: frequent visitors to site, faecal discharge from predators, livestock, wastewater, and fishing bait. This type of contamination is typically unavoidable and very difficult to quantify. Sequences of this type are typically flagged and conservatively removed from the sequencing output. Typical contaminant species include cow, pig, dog, cat, sheep, etc. Sampling - Human contamination of sampling equipment can reduce the efficiency of the sequencing. This type of

	contamination can be minimised by stringent contamination protocols, such as PPE. Laboratory - Residual DNA can contaminate other samples processed at the same time in other labs. At NatureMetrics this is mitigated by a designated eDNA laboratory, strict decontamination procedures, negative controls, and good laboratory practices.
dereplication	The identification of unique sequences so that only one copy of each sequence is reported.
eBioAtlas	A global partnership between IUCN and NatureMetrics to map the world's biodiversity using DNA from water samples as a foundation for the Global Biodiversity Framework and to enable IUCN Red List Assessments.
eDNA	Short for 'environmental DNA'. Refers to DNA deposited in the environment through excretion, shedding, mucous secretions, saliva etc. This can be collected in environmental samples (e.g. water, sediment) and used to identify the organisms that it originated from. eDNA in water is broken down by environmental processes over a period of days to weeks. It can travel some distance from the point at which it was released from the organism, particularly in running water. eDNA is sampled in low concentrations and can be degraded (i.e. broken into short fragments), which limits the analysis options.
extraction blank	A DNA extraction with no sample added to assess potential contamination during the DNA extraction process.
gel electrophoresis	The process in which DNA is separated according to size and electrical charge via an electric current, while in a gel. The process is used to confirm the successful amplification of a specifically sized fragment of DNA.
high-throughput sequencing	Technology developed in the 2000s that produces millions of sequences in parallel. Enables thousands of different organisms from a mixture of species to be sequenced at once, so community DNA can be sequenced. Various different technologies exist to do this, but the most commonly used platform is Illumina's MiSeq. Also known as Next-Generation Sequencing (NGS) or parallel sequencing.
inhibitors/inhibition	Naturally-occurring chemicals/compounds that cause DNA amplification to fail, potentially resulting in false negative results. Common inhibitors include tannins, humic acids and other organic compounds. Inhibitors can be overcome by either diluting the DNA (and the inhibitors) or by additional cleaning of the DNA, but

dilution carries the risk of reducing the DNA concentration below the limits of detection. At NatureMetrics, inhibition is removed using a commercial purification kit.

invasive

Invasive species are defined using GRIIS (Global Register of Introduced and Invasive Species) which is a checklist of Introduced and Invasive species for each country. The IUCN describes an Introduced species as a species outside of its natural range and dispersal potential, and an Invasive species as an introduced species which becomes established in a habitat, is an agent of change or threatens native biological diversity.

IUCN Red List

The IUCN (International Union for the Conservation of Nature) is a global union of government and civil organisations that disseminates information to assist conservation. The IUCN Red List of Threatened Species is an inventory of the conservation status of over 100,000 species worldwide. The Red List evaluates data such as population trends, geographic range and the number of mature individuals in order to categorise species based on their extinction risk:

Extinct (EX) - No individual of this species remains alive.

Extinct in the Wild (EW) - Surviving individuals are only found in captivity.

Critically Endangered (CE) - species faces an extremely high risk of extinction in the wild. e.g. Population size estimated at fewer than 50 mature individuals.

Endangered (EN) - species faces a very high risk of extinction in the wild. e.g. Population size estimated at fewer than 250 mature individuals.

Vulnerable (VU) - species faces a high risk of extinction in the wild. e.g. Population size estimated at fewer than 10,000 mature individuals and declining.

Near Threatened (NT) - species is below the threshold for any of the threatened categories (CE, E, V) but is close to this threshold or is expected to pass it in the near future.

Least Concern (LC) - species is not currently close to qualifying for any of the other categories. This includes widespread and abundant species.

Data Deficient (DD) - There is currently insufficient data available to make an assessment of extinction risk. This is not a threat category - when more data becomes available the species may be recategorised as threatened.

Jaccard similarity index

This index is a calculation that compares two samples to see which taxa are shared and which are distinct. The higher the percentage,

	the more similar two samples are in their community composition.
metabarcoding	Refers to identification of species assemblages from community DNA using barcode genes. PCR is carried out with non-specific primers, followed by high-throughput sequencing and bioinformatics processing. Can identify hundreds of species in each sample, and 100+ different samples can be processed in parallel to reduce sequencing cost.
NCBI nt	National Centre for Biotechnology Information nucleotide database; a general reference database.
negative control	Used to determine whether PCR reactions are contaminated.
NMDS	Non-metric multidimensional scaling (NMDS) is a method that allows visualisation of the similarity of each sample to one another. The dissimilarity between each sample is calculated, taking into account shared taxa (Jaccard similarity index), and then configured into a 2D ordinal space that allows the similarity-based relationship between each sample to be plotted. Samples which are closer together are more similar to one another in terms of community composition, while samples which are further apart are less similar. This type of clustering analysis allows you to see if certain types of samples, for example, those from a particular habitat type, are more clustered together and therefore more similar to one another compared to other groups.
nucleotide	An individual unit of genetic material which, when strung together constitutes a DNA (or RNA) strand/sequence.
OTU	Operational Taxonomic Unit; similar sequences are clustered into OTUs at a defined similarity threshold. OTUs are approximately equivalent to species and are treated as such in our analyses. Species-level taxonomic assignments may or may not be possible, depending on the availability of reference sequences and the similarity between closely related species in the amplified marker. It may be possible to refine the taxonomic assignment for an OTU later as more sequences are added to reference databases.
PCR	Polymerase Chain Reaction; a process by which millions of copies of a particular DNA segment are produced through a series of heating and cooling steps. Known as an 'amplification' process. One of the most common processes in molecular biology and a precursor to most sequencing-based analyses.

positive control	Used to determine whether the PCR is working correctly.
primers	Short sections of synthesised DNA that bind to either end of the DNA segment to be amplified by PCR. Can be designed to be totally specific to a particular species (so that only that species' DNA will be amplified from a community DNA sample), or to be very general so that a wide range of species' DNA will be amplified. Good design of primers is one of the critical factors in DNA-based monitoring.
rarefaction curve	A plot showing the number of taxa as a function of the sequencing depth (number of reads). Rarefaction curves grow rapidly at first as common species are found then reach a plateau as only the rarest species remain to be detected. Rarefaction curves can provide an indication as to whether the species being studied have been comprehensively sampled.
rarefy	A normalisation technique which transforms the data to remove biases associated with uneven sampling depth (number of reads) across samples. The sampling depth of each sample is standardised to a specified number of reads (usually that of the sample with the lowest depth) by random resampling.
reference databases	Over time, the DNA sequences of many species have been compiled into publicly accessible databases by scientists from around the world. These databases serve as a reference against which unknown sequences can be queried to obtain a species identification. The most commonly accessed database is NCBI, which is maintained by the US National Institute of Health. Anyone can search for DNA sequences at https://www.ncbi.nlm.nih.gov.
richness	The total number of taxa within a sample.
rRNA	Ribosomal RNA.
SAC species	Typically the presence of these species potentially elevates the conservation status of a site to a Special Area of Conservation (SAC). Special Areas of Conservation (SACs) are strictly protected sites designated under the EC Habitats Directive.
sequence(s)	A DNA sequence is made up of four nucleotide bases represented by the letters A, T, C & G. The precise order of these letters is used to compare genetic similarity among individuals or species and to identify species using reference databases. In high-throughput sequencing analyses (e.g. metabarcoding), many identical copies of the same sequence are obtained for each species in the sample. The number of copies obtained per species is known as the number of sequence reads, and this is often -

	although not always - related to the relative abundance of the species.
SILVA	SILVA is a database of small (16S/18S, SSU) and large subunit (23S/28S, LSU) ribosomal RNA sequences for all three domains of life (Bacteria, Archaea and Eukarya).
taxon (s.) / taxa (pl.)	Strictly, a taxonomic group. Here we use the term to describe groups of DNA sequences (OTUs) that are equivalent to species. We do not use the term species because we are unable to assign complete identifications to all of the groups at this time due to gaps in the available reference databases.
taxonomy	The branch of science concerned with classification of organisms. species (s./pl.) - A group of genetically similar organisms that show a high degree of overall similarity in many independent characteristics. Related species are grouped together into progressively larger taxonomic units, from genus to kingdom. Homo sapiens (human) is an example of a species. genus (s.) / genera (pl.) - A group of closely related species. Each genus can include one or more species. Homo is an example of a genus. family (s.) / families (pl.) - A group of closely related genera. Homo sapiens is in the Family Hominidae (great apes). order (s.) / orders (pl.) - A group of closely related families. Homo sapiens is in the Order Primates. class (s.) / classes (pl.) - A group of closely related orders. Homo sapiens is in the Class Mammalia. phylum (s.) / phyla (pl.) - A group of closely related classes. Homo sapiens is in the Phylum Chordata.
UKBAP species	UK Biodiversity Action Plan species have been identified as being the most threatened and requiring conservation action under the UK Biodiversity Action Plan.
UNITE	A ribosomal RNA database for identification of fungi.

FISH METABARCODING **RESULTS**

Order number:	SO01060
Report number:	NM-MWJ503
Company:	APEM Ltd
Contact:	Chris Ashelby
Project:	Total Energies - West of Orkney Wind farm
Sample type:	NatureMetrics eDNA disk filter
Date of report:	27-Jan-2023
Number of samples:	40

Thank you for sending your samples for analysis by NatureMetrics. Your samples have been metabarcoded following our eDNA survey - Fish workflow. **A taxon-by-sample table of your samples is attached to this report (NM-MWJ503.SO01060.Fish.xlsx)**. Each row in the table represents one taxon (OTU), shown with the lowest possible taxonomic assignment based on currently available reference data. Each column represents a sample, showing the percentage of sequence reads per detected OTU (Table 1) and the number of sequence reads per detected OTU (Table 2) in that sample. Care should be taken in interpreting the numbers in terms of relative species abundance, but a high sequence proportion can be interpreted as lending greater confidence to a detection. This report contains biodiversity information that may be sensitive, particularly with respect to endangered or protected species. It is the responsibility of the client to ensure that due consideration is given to the data and that the information is shared in a responsible way.

Here we present an overview of the key results, followed by a more detailed report that starts with the taxonomic composition of the samples followed by a more detailed look at the steps taken to extract, amplify, sequence, and analyse your DNA. A glossary for terms in bold is provided at the end of the report to define key terms used within the report.

OVERVIEW OF YOUR RESULTS

- A total of 34 taxa were detected.
- Average taxon richness was 6.08 and ranged from 2 to 14.
- Most abundant sequences: Atlantic mackerel (*Scomber scombrus*).
- Most commonly detected taxa: Atlantic mackerel (*Scomber scombrus*), poor cod (*Trisopterus minutus*) and haddock (*Melanogrammus aeglefinus*).
- Species of note: Atlantic cod (*Gadus morhua* - Vulnerable), haddock (*Melanogrammus aeglefinus* - Vulnerable) and Atlantic horse mackerel (*Trachurus trachurus* - Vulnerable).
- Fish sequence data were obtained from 38 of 40 eDNA sequences.

FULL REPORT

Sample composition

A total of 34 taxa were detected (**Table 1** and **Table 2**). 82.4% (28 taxa) were at least 99% similar to a species in the global reference databases, and species names are suggested. The remaining taxa were identified to the lowest possible taxonomic level: 5.9% to genus (2 taxa), and the remainder to family (4 taxa). The taxa belong to 8 orders, 19 families, and 27 genera.

Species of note include the: Atlantic cod (*Gadus morhua* - Vulnerable), haddock (*Melanogrammus aeglefinus* - Vulnerable) and Atlantic horse mackerel (*Trachurus trachurus* - Vulnerable).

The average taxon richness was 6.08 and ranged from 2 ('164_TOT_eDNA_W14_BOT', '164_TOT_eDNA_W17_BOT', '164_TOT_eDNA_W17_TOP', '164_TOT_eDNA_W22_TOP') to 14 ('164_TOT_eDNA_W03_BOT'). The relative proportion of the sequences found in each of the samples is shown in **Figure 2**, **Figure 3**, **Table 1** and **Table 2** and the diversity is summarised in **Table 3** and **Table 4**.

Atlantic mackerel (*Scomber scombrus*), which accounted for 26.5% of the total sequence reads, was among the most abundant in terms of sequences. Among the most commonly detected species were Atlantic mackerel (*Scomber scombrus*), poor cod (*Trisopterus minutus*) and haddock (*Melanogrammus aeglefinus*) which were detected in 30, 30 and 19 samples, respectively.

High-quality fish sequence data were obtained for 37 of the 40 eDNA samples. eDNA metabarcoding of fish was not successful for '164_TOT_eDNA_W15_TOP' and '164_TOT_eDNA_W13_TOP', which failed to amplify despite troubleshooting. Sample '164_TOT_eDNA_W20_BOT' produced fewer than expected target reads; results for these samples are therefore considered tentative as they may not reflect the full range of fish species diversity in the sample(s). The report status of all samples is summarised in ***Table 5.***

All laboratory controls behaved as expected.

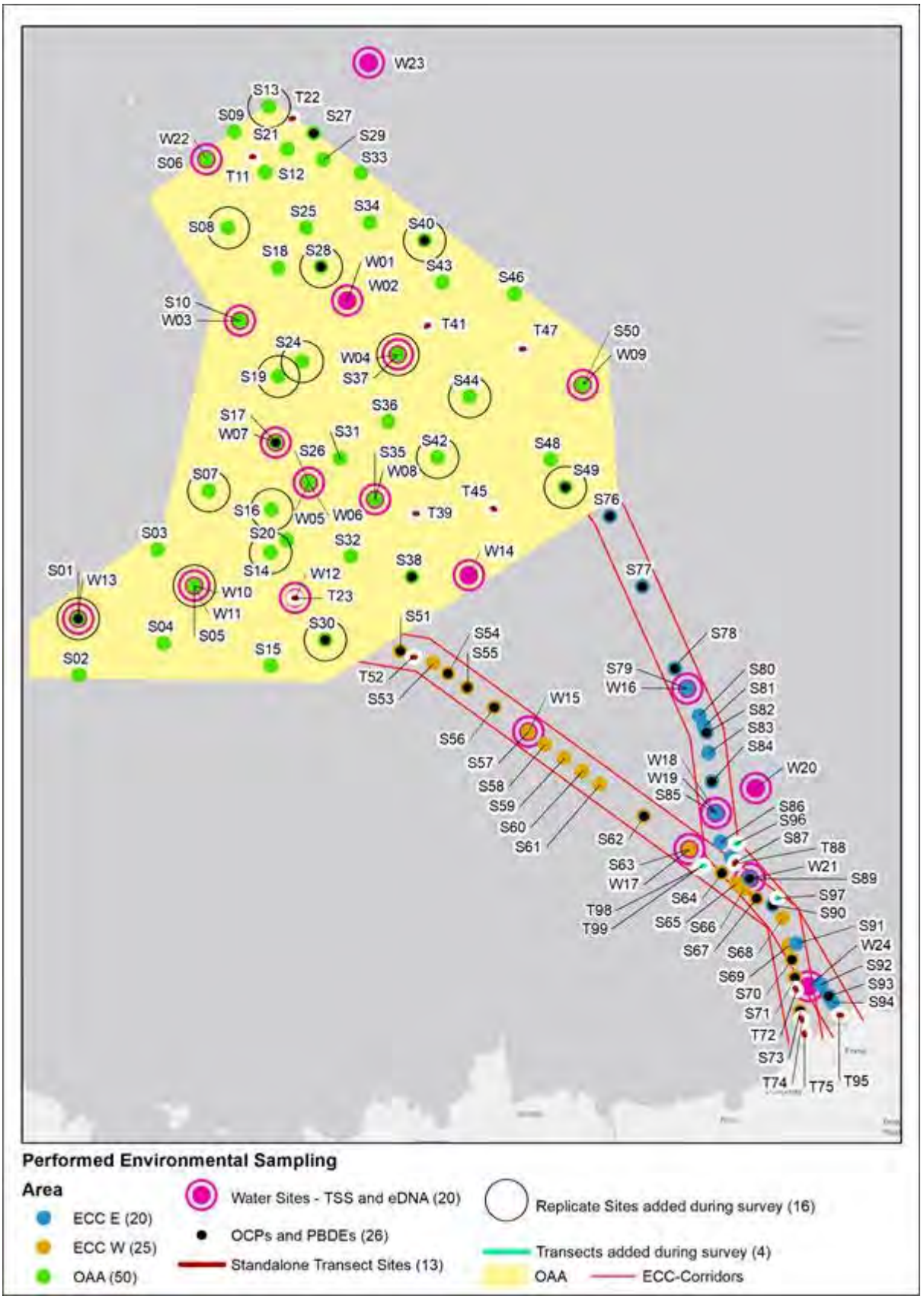

Figure 1. Sampling locations for West of Orkney offshore wind farm.

Table 1 (attached separately). Taxon-by-sample table by read proportion.

Table 2 (attached separately). Taxon-by-sample table by read count.

Figure 2 (attached separately). The proportion of the sequencing output allocated to the different taxa (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each taxon for that sample. The size of the bubble is relative to the number of sequences from all taxa detected in that sample.

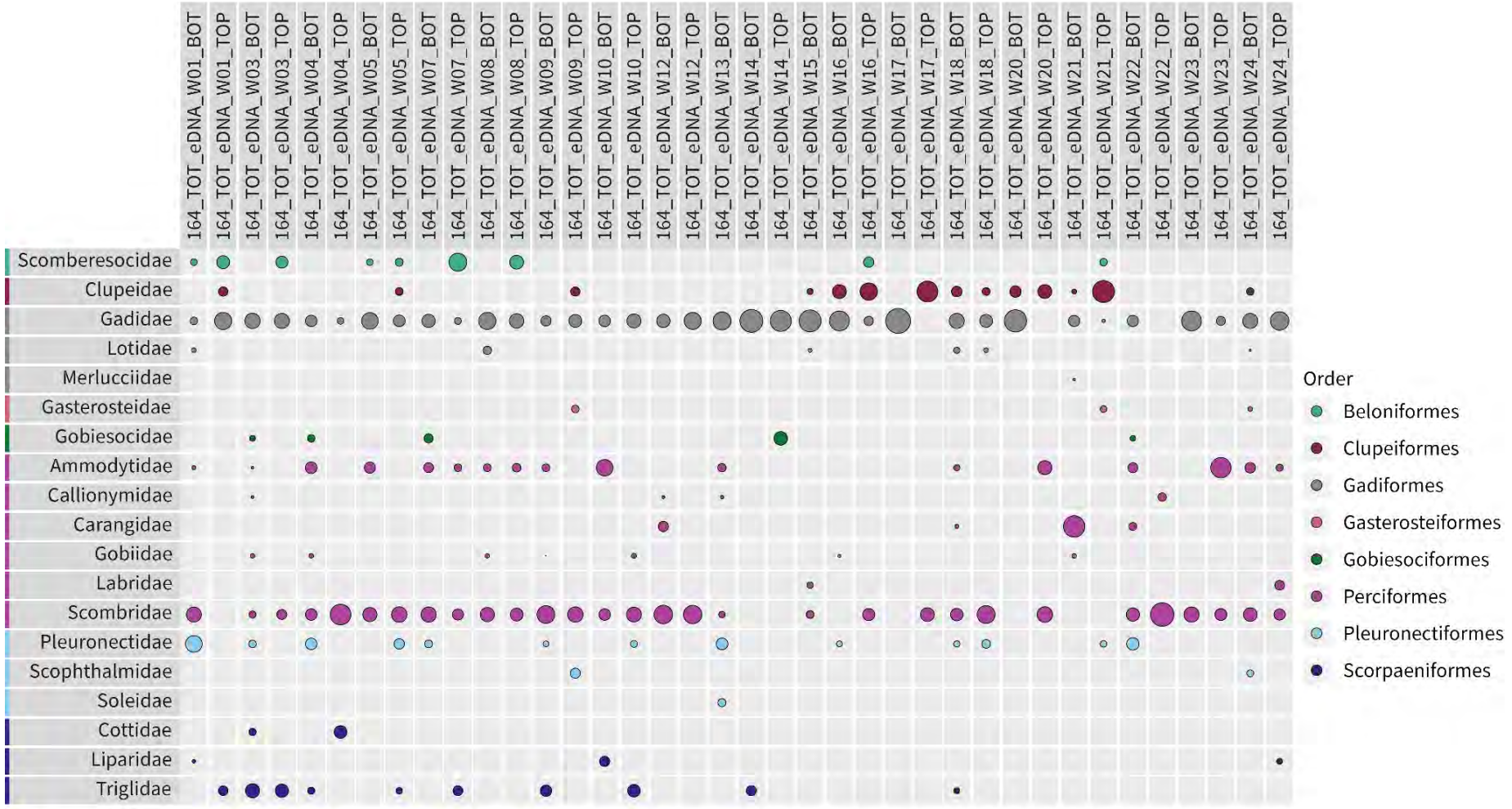

Figure 3. The proportion of the sequencing output allocated to the different orders (rows) within each sample (columns). Each bubble per sample represents the proportion of DNA for each order for that sample. The size of the bubble is relative to the number of sequences from all orders detected in that sample.

Table 3. Taxon richness among the samples.

Sample ID	Class	Order	Family	Genus	Taxa (Species)
164_TOT_eDNA_W01_BOT	1	5	7	6	8 (6)
164_TOT_eDNA_W01_TOP	1	4	4	3	5 (2)
164_TOT_eDNA_W03_BOT	1	5	9	10	14 (12)
164_TOT_eDNA_W03_TOP	1	4	4	3	5 (3)
164_TOT_eDNA_W04_BOT	1	5	7	6	8 (6)
164_TOT_eDNA_W04_TOP	1	3	3	3	3 (3)
164_TOT_eDNA_W05_BOT	1	3	4	4	6 (4)
164_TOT_eDNA_W05_TOP	1	6	6	8	10 (7)
164_TOT_eDNA_W07_BOT	1	4	5	5	6 (5)
164_TOT_eDNA_W07_TOP	1	4	5	2	5 (2)
164_TOT_eDNA_W08_BOT	1	2	5	5	6 (5)
164_TOT_eDNA_W08_TOP	1	3	4	3	5 (3)
164_TOT_eDNA_W09_BOT	1	4	6	7	8 (7)
164_TOT_eDNA_W09_TOP	1	5	5	6	6 (5)
164_TOT_eDNA_W10_BOT	1	3	4	4	6 (3)
164_TOT_eDNA_W10_TOP	1	4	5	6	8 (7)
164_TOT_eDNA_W12_BOT	1	2	4	5	6 (6)
164_TOT_eDNA_W12_TOP	1	2	2	3	4 (4)
164_TOT_eDNA_W13_BOT	1	3	6	7	8 (7)
164_TOT_eDNA_W14_BOT	1	2	2	1	2 (1)
164_TOT_eDNA_W14_TOP	1	2	2	4	4 (4)
164_TOT_eDNA_W15_BOT	1	3	5	7	8 (7)
164_TOT_eDNA_W16_BOT	1	4	4	6	7 (6)
164_TOT_eDNA_W16_TOP	1	4	4	3	4 (2)
164_TOT_eDNA_W17_BOT	1	1	1	2	2 (2)
164_TOT_eDNA_W17_TOP	1	2	2	2	2 (1)
164_TOT_eDNA_W18_BOT	1	5	8	9	11 (9)
164_TOT_eDNA_W18_TOP	1	4	5	6	6 (5)
164_TOT_eDNA_W20_BOT	1	2	2	3	4 (3)
164_TOT_eDNA_W20_TOP	1	2	3	3	3 (3)
164_TOT_eDNA_W21_BOT	1	3	5	9	9 (7)
164_TOT_eDNA_W21_TOP	1	5	5	4	5 (3)
164_TOT_eDNA_W22_BOT	1	4	6	10	11 (10)
164_TOT_eDNA_W22_TOP	1	1	2	2	2 (2)
164_TOT_eDNA_W23_BOT	1	2	2	2	3 (3)
164_TOT_eDNA_W23_TOP	1	2	3	3	4 (3)
164_TOT_eDNA_W24_BOT	1	5	7	9	10 (9)
164_TOT_eDNA_W24_TOP	1	3	5	6	7 (6)

Table 4 (attached separately). The frequency of occurrence of all detected families. Numbers correspond to the number of taxa belonging to those families in those samples.

METHODS

DNA from each filter was extracted using a commercial DNA extraction kit with a protocol modified to increase DNA yields. An extraction blank was also processed for the extraction batch. DNA was purified to remove PCR inhibitors using a commercial purification kit.

Comment: DNA yields were as expected.

Purified DNAs were amplified with PCR for a hypervariable region of the 12S rRNA gene to target fish as part of the eDNA survey - Fish workflow. Our standard analysis includes 12 replicate PCRs per sample.

All PCRs were performed in the presence of both a negative control and a positive control sample (a mock community with a known composition). Amplification success was determined by gel electrophoresis.

Comment: PCR reactions were successful for 38 of 40 samples. Electrophoresis bands were strong and of the expected size. '164_TOT_eDNA_W15_TOP' and '164_TOT_eDNA_W13_TOP' failed to amplify despite troubleshooting steps. Overall, 4-12 successful PCRs replicates were obtained for each of the 38 samples submitted for sequencing. No bands were observed on electrophoresis gels for the extraction blank or negative controls.

PCR replicates were pooled and purified, and sequencing adapters were added. Success was determined by gel electrophoresis.

Comment: All samples were successfully indexed, electrophoresis bands were strong and of the expected size. No repeat reactions were necessary.

Amplicons were purified and checked by gel electrophoresis, these were then quantified using a Qubit high sensitivity kit according to the manufacturer's protocol.

Comment: All amplicons were successfully purified.

All purified index PCRs were pooled into a final library with equal concentrations. The final library was sequenced using an Illumina MiSeq V3 kit at 10.5 pM with a 20% PhiX spike in.

Sequence data were processed using a custom bioinformatics workflow for quality filtering, OTU clustering, and taxonomic assignment.

Comment: Both negative and positive controls were as expected. Very few sequences were discarded prior to dereplication, which is indicative of high-quality data with minimal PCR and sequencing errors. A total of 1,093,885 high-quality sequences, including 1,093,885 target sequences, were included in the final dataset.

Consensus taxonomic assignments were made for each OTU using sequence similarity searches against the NCBI nt (GenBank) reference database. Assignments were made to the lowest possible taxonomic level where there was consistency in the matches. Conflicts were flagged and resolved manually. Minimum similarity thresholds of 99%, 97%, and 95% were used for species-, genus- and higher-level assignments respectively. In cases where there were equally good matches to multiple

species, public records from GBIF were used to assess which were most likely to be present in the United Kingdom. Higher-level taxonomic identifications or multiple potential identifications were reported in cases that could not be resolved in this way.

The OTU table was then filtered to remove low abundance OTUs from each sample (<0.05% or <10 reads, whichever is the greater threshold for the sample). Unidentified, non-target, and common contaminant sequences were then removed.

Note that unidentified or misidentified taxa can result from incomplete or incorrect reference databases, and taxa may be missed due to low quality DNA, environmental contaminants, or the dominance of other species in the sample.

Please note that the abundance of taxa cannot be directly inferred from the proportion of total sequence reads. While the proportion of sequence reads is a consequence of abundance, it is also impacted by biomass, activity, surface area, condition, distance from the physical sample, primer bias, and species-specific variation in the genome.

Table 5 (attached separately). Sample information table.

END OF REPORT

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GLOSSARY

adapter	short, artificially synthesised nucleotide sequence which attaches to the ends of the target DNA or RNA sequences prior to sequencing. They are typically used to aid in attachment of the target sequence to other functional molecules/sequences.
amplicon	A DNA sequence which is the product of PCR amplification.
bioinformatics	An interface between genetics, computational biology, statistics, and programming in which DNA or other biological data is processed, analysed and integrated into research or communications.
bioinformatics workflow	Refers to a data processing workflow that takes the raw sequence data from high-throughput sequencing (often 20 million sequences or more) and transforms it into usable ecological data. Key steps for metabarcoding workflows include quality filtering, trimming, merging paired ends, removal of sequencing errors such as chimeras, clustering of similar sequences into molecular Operational Taxonomic Units, and matching one sequence from each cluster against a reference database. The output is a OTU-by-sample table showing how many sequences from each sample were assigned to each OTU.
BMWP	Short for biological monitoring working party, an index that can be used to measure water quality by scoring the presence of aquatic invertebrate indicator taxa. The index is reliant on taxa that are less tolerant of polluted water bodies (e.g. Ephemeroptera, Plecoptera, Trichoptera).
BOLD	Barcode Of Life Database; a specialised database of eukaryote COI reference sequences.
contaminant sequences	The sensitivity of high-throughput sequencing of eDNA means that contamination is always a concern that needs to be minimised. The sources of contamination are threefold: Natural - Examples of natural contaminants include: frequent visitors to site, faecal discharge from predators, livestock, wastewater, and fishing bait. This type of contamination is typically unavoidable and very difficult to quantify. Sequences of this type are typically flagged and conservatively removed from the sequencing output. Typical contaminant species include cow, pig, dog, cat, sheep, etc.

	Sampling - Human contamination of sampling equipment can reduce the efficiency of the sequencing. This type of contamination can be minimised by stringent contamination protocols, such as PPE. Laboratory - Residual DNA can contaminate other samples processed at the same time in other labs. At NatureMetrics this is mitigated by a designated eDNA laboratory, strict decontamination procedures, negative controls, and good laboratory practices.
dereplication	The identification of unique sequences so that only one copy of each sequence is reported.
eBioAtlas	A global partnership between IUCN and NatureMetrics to map the world's biodiversity using DNA from water samples as a foundation for the Global Biodiversity Framework and to enable IUCN Red List Assessments.
eDNA	Short for 'environmental DNA'. Refers to DNA deposited in the environment through excretion, shedding, mucous secretions, saliva etc. This can be collected in environmental samples (e.g. water, sediment) and used to identify the organisms that it originated from. eDNA in water is broken down by environmental processes over a period of days to weeks. It can travel some distance from the point at which it was released from the organism, particularly in running water. eDNA is sampled in low concentrations and can be degraded (i.e. broken into short fragments), which limits the analysis options.
extraction blank	A DNA extraction with no sample added to assess potential contamination during the DNA extraction process.
gel electrophoresis	The process in which DNA is separated according to size and electrical charge via an electric current, while in a gel. The process is used to confirm the successful amplification of a specifically sized fragment of DNA.
high-throughput sequencing	Technology developed in the 2000s that produces millions of sequences in parallel. Enables thousands of different organisms from a mixture of species to be sequenced at once, so community DNA can be sequenced. Various different technologies exist to do this, but the most commonly used platform is Illumina's MiSeq. Also known as Next-Generation Sequencing (NGS) or parallel sequencing.
inhibitors/inhibition	Naturally-occurring chemicals/compounds that cause DNA amplification to fail, potentially resulting in false negative results.

Common inhibitors include tannins, humic acids and other organic compounds. Inhibitors can be overcome by either diluting the DNA (and the inhibitors) or by additional cleaning of the DNA, but dilution carries the risk of reducing the DNA concentration below the limits of detection. At NatureMetrics, inhibition is removed using a commercial purification kit.

invasive

Invasive species are defined using GRIIS (Global Register of Introduced and Invasive Species) which is a checklist of Introduced and Invasive species for each country. The IUCN describes an Introduced species as a species outside of its natural range and dispersal potential, and an Invasive species as an introduced species which becomes established in a habitat, is an agent of change or threatens native biological diversity.

IUCN Red List

The IUCN (International Union for the Conservation of Nature) is a global union of government and civil organisations that disseminates information to assist conservation. The IUCN Red List of Threatened Species is an inventory of the conservation status of over 100,000 species worldwide. The Red List evaluates data such as population trends, geographic range and the number of mature individuals in order to categorise species based on their extinction risk:

Extinct (EX) - No individual of this species remains alive.

Extinct in the Wild (EW) - Surviving individuals are only found in captivity.

Critically Endangered (CE) - species faces an extremely high risk of extinction in the wild. e.g. Population size estimated at fewer than 50 mature individuals.

Endangered (EN) - species faces a very high risk of extinction in the wild. e.g. Population size estimated at fewer than 250 mature individuals.

Vulnerable (VU) - species faces a high risk of extinction in the wild. e.g. Population size estimated at fewer than 10,000 mature individuals and declining.

Near Threatened (NT) - species is below the threshold for any of the threatened categories (CE, E, V) but is close to this threshold or is expected to pass it in the near future.

Least Concern (LC) - species is not currently close to qualifying for any of the other categories. This includes widespread and abundant species.

Data Deficient (DD) - There is currently insufficient data available to make an assessment of extinction risk. This is not a threat category - when more data becomes available the species may be

	recategorised as threatened.
Jaccard similarity index	This index is a calculation that compares two samples to see which taxa are shared and which are distinct. The higher the percentage, the more similar two samples are in their community composition.
metabarcoding	Refers to identification of species assemblages from community DNA using barcode genes. PCR is carried out with non-specific primers, followed by high-throughput sequencing and bioinformatics processing. Can identify hundreds of species in each sample, and 100+ different samples can be processed in parallel to reduce sequencing cost.
NCBI nt	National Centre for Biotechnology Information nucleotide database; a general reference database.
negative control	Used to determine whether PCR reactions are contaminated.
NMDS	Non-metric multidimensional scaling (NMDS) is a method that allows visualisation of the similarity of each sample to one another. The dissimilarity between each sample is calculated, taking into account shared taxa (Jaccard similarity index), and then configured into a 2D ordinal space that allows the similarity-based relationship between each sample to be plotted. Samples which are closer together are more similar to one another in terms of community composition, while samples which are further apart are less similar. This type of clustering analysis allows you to see if certain types of samples, for example, those from a particular habitat type, are more clustered together and therefore more similar to one another compared to other groups.
nucleotide	An individual unit of genetic material which, when strung together constitutes a DNA (or RNA) strand/sequence.
OTU	Operational Taxonomic Unit; similar sequences are clustered into OTUs at a defined similarity threshold. OTUs are approximately equivalent to species and are treated as such in our analyses. Species-level taxonomic assignments may or may not be possible, depending on the availability of reference sequences and the similarity between closely related species in the amplified marker. It may be possible to refine the taxonomic assignment for an OTU later as more sequences are added to reference databases.
PCR	Polymerase Chain Reaction; a process by which millions of copies of a particular DNA segment are produced through a series of

	heating and cooling steps. Known as an 'amplification' process. One of the most common processes in molecular biology and a precursor to most sequencing-based analyses.
positive control	Used to determine whether the PCR is working correctly.
primers	Short sections of synthesised DNA that bind to either end of the DNA segment to be amplified by PCR. Can be designed to be totally specific to a particular species (so that only that species' DNA will be amplified from a community DNA sample), or to be very general so that a wide range of species' DNA will be amplified. Good design of primers is one of the critical factors in DNA-based monitoring.
rarefaction curve	A plot showing the number of taxa as a function of the sequencing depth (number of reads). Rarefaction curves grow rapidly at first as common species are found then reach a plateau as only the rarest species remain to be detected. Rarefaction curves can provide an indication as to whether the species being studied have been comprehensively sampled.
rarefy	A normalisation technique which transforms the data to remove biases associated with uneven sampling depth (number of reads) across samples. The sampling depth of each sample is standardised to a specified number of reads (usually that of the sample with the lowest depth) by random resampling.
reference databases	Over time, the DNA sequences of many species have been compiled into publicly accessible databases by scientists from around the world. These databases serve as a reference against which unknown sequences can be queried to obtain a species identification. The most commonly accessed database is NCBI, which is maintained by the US National Institute of Health. Anyone can search for DNA sequences at https://www.ncbi.nlm.nih.gov.
richness	The total number of taxa within a sample.
rRNA	Ribosomal RNA.
SAC species	Typically the presence of these species potentially elevates the conservation status of a site to a Special Area of Conservation (SAC). Special Areas of Conservation (SACs) are strictly protected sites designated under the EC Habitats Directive.
sequence(s)	A DNA sequence is made up of four nucleotide bases represented by the letters A, T, C & G. The precise order of these letters is used to compare genetic similarity among individuals or species and to

	identify species using reference databases. In high-throughput sequencing analyses (e.g. metabarcoding), many identical copies of the same sequence are obtained for each species in the sample. The number of copies obtained per species is known as the number of sequence reads, and this is often - although not always - related to the relative abundance of the species.
SILVA	SILVA is a database of small (16S/18S, SSU) and large subunit (23S/28S, LSU) ribosomal RNA sequences for all three domains of life (Bacteria, Archaea and Eukarya).
taxon (s.) / taxa (pl.)	Strictly, a taxonomic group. Here we use the term to describe groups of DNA sequences (OTUs) that are equivalent to species. We do not use the term species because we are unable to assign complete identifications to all of the groups at this time due to gaps in the available reference databases.
taxonomy	The branch of science concerned with classification of organisms. species (s./pl.) - A group of genetically similar organisms that show a high degree of overall similarity in many independent characteristics. Related species are grouped together into progressively larger taxonomic units, from genus to kingdom. Homo sapiens (human) is an example of a species. genus (s.) / genera (pl.) - A group of closely related species. Each genus can include one or more species. Homo is an example of a genus. family (s.) / families (pl.) - A group of closely related genera. Homo sapiens is in the Family Hominidae (great apes). order (s.) / orders (pl.) - A group of closely related families. Homo sapiens is in the Order Primates. class (s.) / classes (pl.) - A group of closely related orders. Homo sapiens is in the Class Mammalia. phylum (s.) / phyla (pl.) - A group of closely related classes. Homo sapiens is in the Phylum Chordata.
UKBAP species	UK Biodiversity Action Plan species have been identified as being the most threatened and requiring conservation action under the UK Biodiversity Action Plan.
UNITE	A ribosomal RNA database for identification of fungi.