



JNCC/Cefas Partnership Report Series

Report No. 9

The Development of Monitoring Options for UK MPAs: Fladen Grounds R&D Case Study

Murray, J., Jenkins, C., Eggleton, J., Whomersley, P., Robson, L., Flavell, B. & Hinchen, H.

March 2016

© JNCC, Cefas 2015

ISSN 2051-6711

The Development of Monitoring Options for UK MPAs: Fladen Grounds R&D Case Study

Murray, J., Jenkins, C., Eggleton, J., Whomersley, P., Robson, L., Flavell, B. & Hinchen, H.

March 2016

© JNCC, Cefas, 2016

ISSN 2051-6711

For further information please contact:

Joint Nature Conservation Committee Monkstone House City Road Peterborough PE1 1JY http://jncc.defra.gov.uk

This report should be cited as:

Murray, J., Jenkins, C., Eggleton., J., Whomersley, P., Robson, L., Flavell, B. & Hinchen, H. 2016. The development of monitoring options for UK MPAs: Fladen Grounds R&D case study. *JNCC/Cefas Partnership Report*, **No. 9**

This report is compliant with the JNCC Evidence Quality Assurance Policy <u>http://jncc.defra.gov.uk/page-6675</u> and was peer reviewed by two independent experts and the JNCC project team.

Summary

In July 2014, 30 Nature Conservation Marine Protected Areas (NCMPAs) were designated in the seas around Scotland, of which 13 are located beyond 12 nautical miles. Introduced under the Marine (Scotland) Act (2010) for inshore waters, and the Marine and Coastal Access Act (2009) for offshore waters, NCMPAs have been introduced to ensure the full range of nationally important features in Scotland's waters are represented in the MPA network.

The Central Fladen NCMPA in the Northern North Sea has been designated for the protection of the burrowed mud feature, including both the seapen and burrowing megafauna in circalittoral fine mud, and tall seapen components, and for the sub-glacial tunnel valley representative of the Fladen Deeps Key Geodiversity Area. Burrowed mud habitats, such as those found within the Fladen grounds, are classified as moderately sensitive to shallow abrasion/penetration of the seabed, typically inflicted by bottom-contact fishing gears (Brooks *et al* 2013).

JNCC and Cefas completed a survey in March 2014 to collect evidence to support development of monitoring options, specifically for the Central Fladen NCMPA and, more generally, for offshore mud habitats. The survey was designed to achieve multiple objectives, which are broadly described below:

- 1. Acquire data to comprise the 'Before' sampling event in a Before, After, Control, Impact (BACI) design (Type 3 monitoring) by surveying similarly sized areas inside (impact) and outside (control) a proposed Management Scenario Area in the Central Fladen NCMPA.
- 2. Acquire data on the benthic community characteristic of burrowed mud in the Fladen Grounds across a fishing pressure gradient determined using surface abrasion gridded data (Type 2 monitoring).
- 3. Collect benthic contaminant samples across the survey area to augment the information available regarding the level of organic and heavy metal contaminants in the sediment.
- 4. Establish the first datapoint in a time-series study across the burrowed mud habitat within the Fladen Grounds (Type 1 monitoring).

The study aimed to investigate the effects of abrasive pressure inflicted by demersal fishing on the benthic communities across the greater Fladen Ground area through the testing of a number of biodiversity indices in a Type 2 monitoring approach. The indices found to be most appropriate for detecting meaningful change in habitat range, extent and condition in relation to increasing abrasion pressure would then be used to explore a potential management scenario within and around the Central Fladen MPA.

Assemblage metrics and indices were selected with consideration for those being developed as part of Marine Strategy Framework Directive (MSFD) requirements and were tested in the context of Type 2 monitoring: change along an abrasive pressure gradient. Environmental variables such as sediment type and geochemistry, as well as aspects of community ecology such as infaunal and epifaunal species and assemblage metrics, Biological Traits Analysis, bivalve size frequency and *Nephrops* burrow densities were tested using data collected from the 2014 Fladen Grounds survey.

Assemblage metrics were explored in relation to high- and low-abrasion pressure due to fisheries. In the high abrasion pressure boxes, a slightly higher silt/clay fraction was recorded and organic carbon and nitrogen and inorganic phosphate and total phosphate content were significantly higher. Analysis of variance identified a significant difference

between in infaunal assemblages between pressure blocks for a number of community metrics and when exploring biological trait composition, there were notable differences for several of the trait categories between abrasion pressure boxes suggesting that the communities are functioning differently in response to abrasion. Those within the high -abrasion areas contain higher relative abundances of burrow dwellers and subsurface deposit feeders, whereas surface deposit feeders and tube dwellers tended to be higher within the low pressure areas. However, due to inclement weather during the survey, the number and spatial spread of samples collected was reduced and it was therefore not feasible to draw broad conclusions on how infaunal community univariate metrics were responding to abrasive pressure across the whole Fladen Grounds.

More data were available for epifaunal analysis allowing the testing of univariate metric responses of epifaunal assemblages across a gradient of abrasive pressure. Regression analysis showed that epifaunal communities are being modified by abrasive pressure, and generally speaking, with increasing abrasive pressure, the number of epifaunal individuals was reduced although species diversity increased. Seapen abundance and density varied across the identified pressure gradient with *Pennatula phosphorea* demonstrating a negative relationship with increasing abrasive pressure whilst *Virgularia mirabilis* showed no discernible relationship.

The metrics tested in the Type 2 monitoring approach were used to provide the "Before" stage of a Type 3 BACI experimental approach around a management scenario area within and around the Central Fladen MPA. Sampling, using a random stratified sampling design, was conducted in a 'Control Area' (unimpacted by fishing) and an 'Impact Area' (which in the future management scenario would be closed to fishing) which is at present hypothetically being subjected to the same influence of abrasion. As expected, no significant differences were found when comparing univariate metrics on infaunal assemblages collected at Control and Impact stations. However, when comparing the epifaunal assemblages, significant differences in taxon richness, number of individuals and Hill's diversity were found despite multivariate analysis suggesting that assemblages from two treatments did overlap. Differences in the distribution of seapens across the experimental study area go some way to explain the differences in epifaunal univariate metrics.

The present investigation provides baseline empirical data that could be used towards a range of biodiversity indicators in relation to areas of high- and low-abrasion pressure at the Fladen Grounds. Knowledge acquired from this study will be used to inform future monitoring of the area and contribute to monitoring options for offshore mud habitats more generally.

Contents

1. Int	troductio	on	4				
1.1	Proje	ct background	4				
1.2	1.2 Specific aims and objectives						
1.3	Centr	al Fladen NCMPA	5				
1	1.3.1	Rationale for site designation	5				
1.4	Flade	n Grounds habitat types at the UK scale	6				
1.5	Over	view of the Fladen Grounds	7				
1	1.5.1	Site boundary	7				
1	1.5.2	Conservation objectives	8				
1.6	Relev	ant pressures to site features	9				
2. Ty	pe 1 mo	nitoring - Spatial and temporal data	11				
2.1	Introd	luction	11				
2.2	Metho	ods	11				
2	2.2.1	Spatial patterns in particle size distribution	11				
2	2.2.2	Spatial patterns in sediment geochemistry	11				
2	2.2.3	Spatial distribution of seapens and burrowing megafauna	12				
2	2.2.4	Contaminants analysis	12				
2.3	Resu	lts	13				
2	2.3.1	Spatial patterns in particle size distribution	13				
2	2.3.2	Spatial patterns in sediment geochemistry	15				
2	2.3.3	Spatial distribution of seapens and burrowing megafauna	19				
2	2.3.4	Contaminants analysis	23				
2.4	Discu	ssion and conclusions	24				
3. Ту	pe 2 mo	nitoring - Sensitivity of mud habitats to abrasive pressure	26				
3.1	Introd	luction	26				
3	3.1.1	Abrasive pressure gradient	26				
3	3.1.2	Environmental characteristics	26				
3	3.1.3	Infaunal species and assemblage metrics	27				
3	3.1.4	Biological Traits Analysis (BTA)	27				
3	3.1.5	Bivalve size frequency	27				
3	3.1.6	Epifaunal species and assemblage metrics	28				
3	3.1.7	Nephrops burrow density	29				
3.2	Metho	ods	29				
3	3.2.1 Grounds	Analysis of historical spatial and temporal abrasion pressure at the Fladen 29					
3	3.2.2	Survey design	29				
3	3.2.3	Sample acquisition	32				

3.2.	4 Sample processing	32
3.2.	5 Data analysis	33
3.3 F	Results	38
3.3.	1 Temporal and spatial patterns in abrasive pressure across the Fladen Gro	unds 38
3.3.	2 Environmental characteristics	42
3.3.	3 Infauna assemblages	45
3.3.	Biological traits analysis: Functional composition	48
3.3.	5 Biological traits analysis: Community vulnerability	53
3.3.	6 Bivalve size frequency distribution	55
3.3.	7 Epifauna community analysis	57
3.3.	8 Nephrops burrow distribution and density	59
3.4 E	Discussion	64
3.4.	1 Environmental characteristics	64
3.4.	2 Infaunal assemblage metrics	65
3.4.	Biological traits analysis: Community function and vulnerability	65
3.4.	4 Bivalve size structure	66
3.4.	5 Epifaunal assemblage metrics	67
3.4.	6 Seapen abundance and density	67
3.4.	7 Nephrops burrow density and distribution	68
3.5 5	ummary and conclusions	68
4. Type : NCMPA	3 monitoring - BACI study within the south east extent of the Central Fladen	70
4.1 l	ntroduction	70
4.2 N	1ethods	70
4.2.	1 Survey design	70
4.2.	2 Sample acquisition and processing	71
4.2.	3 Data analysis	71
4.3 F	Results	72
4.3.	1 Distribution of environmental characteristics	72
4.3.	2 Distribution of infaunal communities in 2014	73
4.3.	3 Distribution of epifaunal communities in 2014	76
4.3.	4 Temporal comparisons of infaunal and epifaunal communities	80
4.4 C	Discussion and conclusions	86
5. Concl	usions and recommendations	88
5.1 T	ype 1 monitoring – spatial and temporal data	88
5.2 1	ype 2 monitoring – sensitivity to abrasive pressure	88
5.3 T	ype 3 monitoring – BACI study	90
Referen	ces	91
Appendi	x 1: Sample acquisition	97

A1. 1 Sample acquisition
A1. 2 Seabed imagery
A1. 3 Global Positioning System (GPS)
A1. 4 Data QA/QC
Appendix 2: Contaminants analysis
A2.1. Sediment metals analysis
A2. 2. Sediment PAH analysis
A2.3. Sediment organohalogens analysis
A2.4. QA/QC
Appendix 3: Infaunal data: abrasion pressure study 102
A3.1. SIMPER results for the comparison of infaunal assemblages
A3. 2. Biological trait composition based on biomass 105
Appendix 4: BACI Study 109
A4.1. SIMPER output based for infaunal (square root transformed abundance data) for the Control and Impact areas within the Central Fladen NCMPA (2014 only)
A4.2 SIMPER Infauna results analysed by SIMPROF group 113
A4.3 SIMPER results comparing presence/absence data of epifaunal species for Control and Impact areas within Central Fladen NCMPA (2014 only)
A4.4. SIMPER output for infauna based on presence/absence data (2013 and 2014)115
A4.5 SIMPER output results comparing 2013 and 2014 epifaunal presence/absence data

1 Introduction

1.1 Project background

UK Governments have a requirement to monitor biodiversity across UK waters in order to fulfil their national and international obligations for marine biodiversity assessment and management. To address this, the UK Marine Biodiversity Monitoring R&D Programme (UK MBMP), a partnership between the Joint Nature Conservation Committee (JNCC), the Statutory Nature Conservation Bodies (SNCBs) and the UK Marine Monitoring and Assessment Strategy (UKMMAS) community, was formed to develop options for an integrated monitoring scheme for all marine biodiversity across all UK waters. The overall aim of this scheme is to collect the evidence necessary to fulfil marine biodiversity obligations and provide timely and effective advice for marine management. The sheer scale and potential cost of monitoring all biodiversity across UK waters necessitated the development of an overarching strategy to ensure that monitoring is prioritised effectively and robust monitoring data are collected (JNCC 2014).

The overarching strategy recognised that monitoring can be carried out to fulfil two broad objectives; first to identify the need for management measures, and second to identify whether management measures have been effective. Once the broad objectives have been clarified, then monitoring approaches can be developed to address the specific requirements of the geographical area in question. The strategy categorised monitoring approaches into three 'types of monitoring' which are described below. It should be noted that at this stage in the development of the UK MBMP, these approaches are used to detect change, but do not aim to assess present conditions against established thresholds or targets. At this point, the focus of monitoring activities is to develop robust indicators, investigate relationships, and establish datasets, against which future data may be compared to identify change.

Sentinel monitoring of long-term trends (Type 1 monitoring) – Objective: to measure rate and direction of long-term change.

This type of monitoring provides the context to distinguish directional trends from short-scale variability in space and time by representing variability across space at any one time and documenting changes over time.

To achieve this objective efficiently, a long-term commitment to regular and consistent data collection is necessary; this means dedicated time-series monitoring must be established in identifying trends because it is far superior to any combination of independent ad-hoc studies.

Operational monitoring of pressure-state relationships (Type 2 monitoring) -

Objective: to measure state and relate observed change to possible causes. This objective complements monitoring long-term trends and is best suited to explore the likely impacts of pressures on habitats and species and identify emerging problems. It leads to setting of hypotheses about processes underlying observed patterns.

It relies on finding relationships between observed changes in biodiversity and observed variability in pressures and environmental factors. It provides inference, but it is not proof of, cause and effect. The spatial and temporal scale for this type of monitoring activity will require careful consideration of the reality on the ground to ensure inference will be reliable; for example, inference will be poor in situations where the presence of a pressure is consistently correlated to the presence of an environmental driver (e.g. a specific depth stratum).

Investigative monitoring to determine management needs and effectiveness (Type 3 monitoring) – Objective: to investigate the cause of change.

This monitoring type provides evidence of causality. It complements the above types by testing specific hypothesis through targeted manipulative studies. The design and statistical approach that can be used in these cases gives confidence in identifying cause and effect. It is best suited to test state/pressure relationships and the efficacy of management measures.

Sampling strategies and methods for seabed habitat surveys, both within and outside Marine Protected Areas (MPAs), are under development as part of the UK MBMP. In 2014, two case study monitoring surveys were undertaken to test the developing monitoring concepts, sampling designs, monitoring methods and metrics/indicators for detecting meaningful change. These monitoring R&D surveys visited the Fladen Grounds Nature Conservation MPA (NCMPA) and the Dogger Bank Site of Community Importance (SCI). There will clearly be site- and feature-specific requirements for monitoring to detect change in the range, extent and condition of different habitat types which are being impacted by different human pressures. However, the overarching concepts that underpin the purpose and approach to undertaking effective monitoring will be relevant across all habitat types. This R&D report therefore forms part of the evidence base for the development of monitoring options for benthic habitats as part of the UK MBMP.

This report describes the findings of a dedicated survey undertaken to collect evidence to support the development of monitoring options specifically for the Central Fladen NCMPA, and ultimately, to provide options which could be applied to other offshore mud habitats.

1.2 Specific aims and objectives

The objectives of the survey (listed in order of priority) were to:

- Conduct a BACI study (Type 3 monitoring) by surveying similarly sized areas inside (impact) and outside (control) a proposed Management Scenario Area in the Central Fladen NCMPA. Since the Management Scenario Area is still at the proposed stage, the survey would cover the 'Before' aspect of the Before, After Control, Impact (BACI) design.
- 2. Acquire data on the benthic community characteristic of burrowed mud in the Fladen Grounds across a fishing pressure gradient determined using surface abrasion gridded data (Type 2 monitoring).
- Collect sediment contaminant samples across the survey area (including a sample in close proximity to a live production well – Claymore oil platform) to augment the information available regarding the level of organic and heavy metal contaminants in the sediment and sediment samples for particle size analysis to help explain changes in biota.
- 4. Establish a time-series study across the burrowed mud habitat within the Fladen Grounds involving a combined camera and grab sampling campaign (150 stations randomly selected) (Type 1 monitoring).

1.3 Central Fladen NCMPA

1.3.1 Rationale for site designation

The Marine (Scotland) Act 2010 and the UK Marine and Coastal Access Act 2009 gave powers to Scottish Ministers to designate Nature Conservation MPAs in Scotland's seas as part of a range of measures to manage and protect Scotland's seas for current and future generations. JNCC and Scottish Natural Heritage (SNH) applied the <u>Site Selection</u>

<u>Guidelines</u> to identify NCMPAs that, together with existing protected areas, will form a network of MPAs. The process will also help Scotland meet its contribution to UK commitments under international conventions and legislation such as the Convention on Biological Diversity, and the OSPAR Convention for an ecologically coherent network of MPAs.

NCMPAs were identified for a range of different features, including habitats, low-mobility species, mobile species, large-scale features and geodiversity features (i.e. geological and geomorphological seabed features such as iceberg ploughmarks (Brooks *et al 2013*). The features for which NCMPAs were initially identified are termed 'MPA search features,' and once designated are termed 'protected features'.

Burrowed mud was considered as a NCMPA search feature and is comprised of the following component biotopes and species:

- 'Seapens and burrowing megafauna in circalittoral fine mud' (Biotope code: SS.SMu.CFiMu.SpnMeg).
- 'Seapens, including [*Funiculina quadrangularis*], and burrowing megafauna in undisturbed circalittoral fine mud' (Biotope code: SS.SMu.CFiMu.SpnMeg.Fun; a sub-component of the above biotope).
- Tall seapen (Funiculina quadrangularis).
- Fireworks anemone (Pachycerianthus multiplicatus).
- Mud burrowing amphipod (Maera loveni).

1.4 Fladen Grounds habitat types at the UK scale

The Fladen Grounds was selected as a case study for the development of monitoring options for offshore mud habitats. This type of habitat is included on the Oslo and Paris Commission (OSPAR) list of Threatened and/or Declining species and habitats (OSPAR 2008) as the habitat type 'sea-pen and burrowing megafauna communities' and on the Scottish Priority Marine Features list as 'burrowed mud' (Scottish Government 2013). 'Burrowed mud' consists of fine mud, sandy mud and muddy sand at waters depths from approximately 10-500m. At the UK scale, burrowed mud is found mainly in deep waters below 50m, such as Irish Sea and North Sea offshore waters. However, relatively shallow, inshore records also occur, and an estimated 95% of records of inshore burrowed mud are located in Scotland (Tyler-Walters et al 2012), notably in sheltered basins along Scotland's fjordic coastline. Patchy burrowed mud deposits are also present in deep water off the west coast, along the edge of the Continental Shelf (where sand is also observed) and to the south of Rockall Bank. Funiculina guadrangularis (tall seapen) and the fireworks anemone Pachycerianthus multiplicatus are encountered most frequently in undisturbed muddy sediments on the west and south-west coasts of Scotland (Greathead et al 2007; Allan et al 2012). Due to their relatively restricted distribution in UK waters, these two species are considered to be of conservation importance as species rather than as part of a habitat type.

The muds are heavily bioturbated by burrowing megafauna, such as the commercially important Norway lobster *Nephrops norvegicus* (hereafter referred to as *Nephrops*) with burrows and mounds typically forming a prominent feature of the sediment surface. Burrowing megafauna are important bioturbators of the sediment they inhabit, increasing the structural complexity and depth of oxygen penetration into the sediments, enhancing the survival of other species and increasing biodiversity in what would otherwise be a low diversity habitat (Widdicombe *et al* 2004). The burrows also provide a source of refuge for smaller invertebrates and fish (Hughes 1998).

1.5 Overview of the Fladen Grounds

The Fladen Grounds is a large area of muddy sediment in the Northern North Sea, located to the north-east of Scotland, and covering an area of nearly 30,000km². The muddy sediment is heavily bioturbated in some areas by burrowing species such as *Nephrops* and mud shrimp. The burrowing activity of these species plays an important role in supporting life in the area; the constant churning of the mud releases nutrients and helps to mix oxygen into the mud. Longer-lasting burrows also provide shelter to other marine life from predators. Several different types of seapen can also be found anchored in the muddy seabed, typically *Virgularia mirabilis* (slender seapen) and *Pennatula phosphorea* (phosphorescent seapen). This type of habitat is included on the OSPAR list of Threatened and/or Declining species and habitats (OSPAR 2008) as the habitat type 'sea-pen and burrowing megafauna communities' and on the Scottish Priority Marine Features list as 'burrowed mud' (Scottish Government 2013).

The Fladen Grounds region in the North Sea is now the biggest *Nephrops* fishery in the world (Ungfors *et al* 2013). Due to the distance between the Fladen Grounds and the nearest landing port, the fishery is restricted to multi-day trips by larger boats than those found in more inshore grounds. Assessments of the Fladen Grounds fishery for *Nephrops* are performed by Marine Scotland Science using underwater TV fishery-independent stock surveys. These <u>Nephrops fisheries stock assessments</u> provide another source of evidence for verifying the burrowed mud habitat. Data on substratum type and the presence of characterising epifauna are recorded and these data have been processed according to Marine Scotland Science's semi-quantitative ROCA abundance scale (Allan *et al* 2012).

1.5.1 Site boundary

Three options of possible MPAs (pMPA) were identified for the representation of the burrowed mud search feature within the Fladen Grounds, which included the Central Fladen, South-east Fladen, and Western Fladen pMPAs (see Figure 1). The three options were considered to be ecologically equivalent for the representation of the seapens and burrowing megafauna component of burrowed mud. However, Central Fladen was the only site that included records of the tall seapen sub-component/species. As such, the representation of burrowed mud within the Fladen Grounds could be achieved by either taking forward the Central Fladen pMPA in its entirety, or taking forward only the tall seapen 'Core' part of Central Fladen together with either the South-east Fladen or Western Fladen pMPA options. Views on the three options were sought as part of the public consultation on NCMPAs in summer 2013, and having considered the feedback, Scottish Ministers designated the entirety of the Central Fladen NCMPA in July 2014 (which merged the Core area and wider Central Fladen boundary). The South-east Fladen and Western Fladen pMPAs are no longer being considered.

The Central Fladen NCMPA has also been shaped to include a sub-glacial tunnel-valley geodiversity feature on the seafloor, representative of an area of geomorphological interest known as the Fladen Deeps. In places, the tunnel-valley is up to 150m deep, 4km wide and 40km long, and is likely to have been formed by pressurised melt-water flowing beneath the ice sheet. The sub-glacial tunnel-valley geodiversity feature is scientifically important as it holds potentially valuable evidence about past changes in the extent and geometry of the last British-Irish Ice Sheet (Brooks *et al* 2013).

Further information about Central Fladen NCMPA is available on the JNCC website: <u>http://jncc.defra.gov.uk/page-6476</u>



Figure 1. Location of the three options identified for the representation of burrowed mud within the wider Fladen Grounds, which included the Central Fladen NCMPA (and Central Fladen Core area), along with the South-east and Western Fladen pMPAs. Only the Central Fladen NCMPA (and Central Fladen Core area) was designated in July 2014; the South-east Fladen and Western Fladen pMPAs are no longer under consideration.

1.5.2 Conservation objectives

Conservation objectives set out the desired state for the protected feature(s) of an MPA. The <u>Management handbook</u> for NCMPAs states that "Conservation objectives for NCMPAs will normally be to conserve the feature in the first instance, acknowledging where there is uncertainty regarding feature condition. In cases where uncertainty exists this will be recorded and reviewed as further knowledge and evidence emerges."

As there is no direct evidence of damage to any of the protected features within the Central Fladen NCMPA (although activities are known to occur to which the protected features will be exposed), the conservation objective of 'conserve in favourable condition', was set, noting that there is uncertainty in feature condition.

The conservation objectives for the protected features of the Central Fladen NCMPA are:

Subject to natural change, **conserve the burrowed mud feature in favourable condition**, such that:

- its extent is stable or increasing; and
- its structures and functions, its quality, and the composition of its characteristic biological communities are such as to ensure that it is in a condition which is healthy and not deteriorating.

Subject to natural change, **conserve the geomorphological interest feature in favourable condition**, such that:

- its extent, component elements and integrity are maintained;
- its structure and functioning are unimpaired; and
- its surface remains sufficiently un-obscured for the purposes of determining the conditions in the points above.

2 Relevant pressures to site features

A pressure is generally defined as the mechanism through which an activity interacts with the marine environment, and these pressures can be physical, chemical or biological. Pressures result from a variety of different human activities, and the intensity of the pressures can differ between activities. Marine features are often sensitive to the effects of pressures which can in some cases adversely affect their condition, especially if they are unable to, or are very slow to, recover from damage. However, the sensitivity of given receptors depends on a number of different factors.

The aim of the UK MBMP is to detect pressures and gather an understanding of how these pressures are impacting marine habitats. However, the overall result of the monitoring work will be to identify appropriate management measures and test their effectiveness for the activity, rather than the pressures. Marine pressures are mitigated through the use of management measures and as such, in some cases, pressures will not occur from an activity due to the use of these mitigation measures.

A list of pressures occurring within the North-East Atlantic marine environment and associated definitions was formally agreed by the OSPAR Intersessional Correspondence Group on Cumulative Effects (ICG-C) (OSPAR 2011). These pressures can be 'mapped' via geospatial data layers in order to investigate where, spatially, marine habitats and species are being exposed to a pressure. Mapping pressures has been identified as a key process in supporting the implementation of the risk-based approach to the Marine Biodiversity Monitoring R&D Programme (JNCC 2011a), and to facilitate this process, JNCC used the ICG-C pressures list to prioritise pressures impacting benthic habitats within UK waters (JNCC 2011b). The highest priority pressures on seabed habitats were considered to be 'removal of target and non-target species' as a result of fishing activities and 'physical habitat damage'. Physical habitat damage can be split into a number of sub-types, but the sub-type Penetration and/or disturbance of the substrate below the surface of the seabed, including abrasion' (referred to here simply as 'abrasion') was ranked as one of the highest priority pressures. This is predominately due to the relatively large spatial footprint arising from demersal fishing activities operating across the area (JNCC 2011b), which is one of the key activities causing abrasion pressure.

Abrasion pressure is defined in the OSPAR ICG-C pressure list as:

The disturbance of sediments where there is limited or no loss of substrate from the system. This pressure is associated with activities such as anchoring, taking of sediment/geological cores, cone penetration tests, cable burial (ploughing or jetting), propeller wash from vessels, certain fishing activities, e.g. scallop dredging, beam trawling.

Abrasion relates to the damage of the seabed surface layers (typically up to 50cm depth). Activities associated with abrasion can cover relatively large spatial areas and include: fishing with towed demersal trawls (fish & shellfish); bio-prospecting such as harvesting of biogenic features such as maerl beds where, after extraction,

conditions for re-colonisation remain suitable or relatively localised activities including: seaweed harvesting, recreation, potting, aquaculture.

A further step in implementing a risk-based approach is the ability to assess the sensitivity of marine features to pressures to which they are exposed. As part of the process of establishing a UK network of MPAs, Defra led on work through the contract "Marine Protected Areas - gathering/developing and accessing the data for the planning of a network of Marine Conservation Zones, MB0102" to develop a matrix linking the sensitivity of marine features to anthropogenic pressures. This work was adapted to reflect additional evidence required in Scottish seas through the Marine Scotland's Features Activities Sensitivities Tool (FEAST). The Features Activities Sensitivities Tool enables users to explore what is known about feature sensitivity to pressures and the marine activities that can cause them.

As one of the key activities occurring within the region of the Fladen Grounds is demersal trawling from the *Nephrops* fishery, abrasion (surface and sub-surface) was identified as a priority pressure to inform the development of management and monitoring options for the Central Fladen NCMPA. Burrowed mud is considered to have medium sensitivity to abrasion (surface and sub-surface) caused by demersal trawling based on the FEAST tool, and within the area it has high exposure to the activity causing the pressure.

The tunnel valley geodiversity feature is considered to have a low sensitivity to the pressures associated with activities taking place within the MPA based on the FEAST tool. This was based on the fact that this is a relict feature typically characterised by relatively erosion-resistant geology. As such it has been defined as having no resilience but high resistance. It is therefore not considered to be a significant risk to the feature achieving its conservation objective and so the feature will not be considered further in the context of management discussions.

3 Type 1 monitoring - Spatial and temporal data

3.1 Introduction

Monitoring long-term change and attributing that change to natural or anthropogenic sources is a key aim of Type 1 monitoring within the UK Marine Biodiversity Monitoring R&D Programme. To achieve this, there is a requirement to build a temporal dataset of environmental variables and human pressures relevant to the features of interest so that any biological change can be detected in the context these variables and pressures.

Although a Type 1 monitoring survey design was not a specific objective of the 2014 dedicated survey, data acquired, in combination with the 2013 JNCC-Cefas survey (Eggleton *et al* 2013), could be used to provide a spatial and temporal (two year) overview of the Fladen Grounds and contribute to the establishment of a temporal dataset of environmental variables. In this section, data from 2013 and 2014 were combined to explore sedimentary patterns across the site as well as the distribution of seapens. In addition, patterns in sediment geochemistry and sedimentary contaminants from the 2014 dataset are presented.

3.2 Methods

3.2.1 Spatial patterns in particle size distribution

Sediment samples collected at the Fladen Grounds in 2013 and 2014 (see Appendix 1 for detailed sample acquisition methods) were analysed at half phi intervals using a combination of laser diffraction (<1mm fraction) and dry sieving techniques (>1mm) as described in National Marine Biological Analytical Quality Control Scheme PSA guidance (Mason 2011). Mean, sorting, skewness and kurtosis were also calculated for all samples and each sample was classified according to one of four EUNIS sediment classes as defined by Long (2006). Gradistat software (Blott & Pye 2001) was then used to produce particle size distribution (PSD) statistics. Full-resolution PSD data for all sediment samples from both 2013 and 2014 were grouped using Entropy, a non-hierarchical clustering method that groups large matrices of PSD datasets into a limited and easy to handle number of groups. EntropyMax is a MS Windows-based software package that groups large matrices of PSD datasets into a finite number of groups (Stewart *et al* 2009).

3.2.2 Spatial patterns in sediment geochemistry

For analysis of total organic carbon (TOC) and nitrogen, sediment samples were freezedried, any material >2mm was removed and the remainder ground before being sent for analysis. Inorganic carbon (carbonates) was removed using a sulphurous acid digest and TOC measured using a Carlo Erba EA1108 Elemental analyser. Quality assurance procedures included analysis of three repeats completed for one in ten samples. Limits of detection are for organic carbon <0.02% and nitrogen <0.002%, and measurement limits are calculated as ten times detection limit.

Analysis of sediment total phosphate, inorganic phosphate and organic phosphate was completed. The method for extraction of phosphate in inorganic and total fractions from marine sediment is based on Aspila *et al* (1976) with the subsequent analysis of the extract based on that of Hansen and Grasshoff (1983). During total phosphate analysis, a subsample of sediment was removed and baked to release organic phosphate, followed by hydrochloric acid digest to extract the phosphorous. The solvent was separated from the solids using a centrifuge. The centrifuge extracts were analysed using a San++ Continuous Flow Analyzer (© Skalar Analytical B.V.). The method is based on that of Hansen and

Grasshoff (1983), consisting of a multi-stage digestion process converting dissolved phosphates to heptamolybdate-reactive orthophosphate prior to photometric measurements. The procedure for analysis of inorganic phosphate was the same as for total phosphate, except the sample was not baked so no organic phosphorous was released during the hydrochloric acid digest. Organic phosphate was calculated by subtracting inorganic phosphate from total phosphate.

3.2.3 Spatial distribution of seapens and burrowing megafauna

Sixty-nine video tows from 2013 and 160 from 2014 were analysed following guidance developed by Cefas and JNCC for the acquisition and processing of video and still images (Coggan *et al* 2007). Each video tow was analysed and faunal counts and SACFOR scores for all species, including the three species of seapen *P. phosphorea, V. mirabilis* and *F. quadrangularis,* were recorded. Identification of anthropogenic activities such as the presence of trawl scars, fishing gears and litter was also documented at each station. Video tow data from 2014 was also analysed for the frequency of *Nephrops* burrows following standard methods used for underwater television surveys (UWTV) to assess *Nephrops* populations in UK and Irish waters (ICES 2007). Two fractions of all video tows (each representing 10% of the total tows) were selected for a second count to ensure data quality.

Density per square metre of the three species of seapen, *P phosphorea, V. mirabilis* and *F. quadrangularis*, was calculated using counts within a fixed field of view from video data from both 2013 and 2014 surveys. *Nephrops* burrow density per m² was calculated using 2014 data only, but values are presented alongside average burrow density data collected by Marine Scotland during 2008-2010 *Nephrops* TV surveys and provided by JNCC.

3.2.4 Contaminants analysis

Analysis of sediment for metals, poly-aromatic hydrocarbons (PAHs) and organohalogens was performed using samples collected at five stations across the Fladen Grounds (Figure 2). Full details on the analytical protocols used are provided in Appendix 2: Contaminants analysis. All contaminants laboratory analyses were carried out under full analytical quality control procedures

The measured environmental concentration for each contaminant was calculated to allow for comparisons with Background Assessment Concentrations (BACs), developed by OSPAR and Effects Range Low (ERL) and Effects Range Median (ERM) concentrations which were developed for the US EPA, and are stored on a large database of sediment toxicity and benthic community information (Long and McDonald 1998). The ERL and ERM methodology derives Sediment Quality Guidelines (SQGs) representing, respectively, the 10th and 50th percentiles of the effects dataset. This approach is a reasonably conservative one, and has been partially validated using North American field data. Concentrations below the ERL rarely cause adverse effects in marine organisms.



Figure 2. Location of stations where sediment sampling for contaminant samples was undertaken at the Fladen Ground on the 2014 surveys.

3.3 Results

3.3.1 Spatial patterns in particle size distribution

Four sediment groups were determined in the best group output from Entropy using sediment samples from both 2013 and 2014 surveys. The optimum number of clusters is achieved when the Calinski–Harabasz (C–H) statistic is at its maximum (Orpin and Kostylev, 2006). In addition to this statistic, expert judgement was employed and that meant that in some cases, where groups were sufficiently similar, they were considered to be the same group and suffixed with an 'a' or a 'b' to show original grouping. Sediment characteristics and profiles for each of these final groups (three groups, group 1 being split into a and b) are given in Table 1.

Table 1. Sediment group characteristics as determined from EntropyMax cluster analysis of the 2013 and 2014 full resolution particle size distribution data.

			MODE:		SORTING	(%) proportion						
Group	Sample Type	Sediment description	1 (µm)	2 (µm):	Methods of moments Logarithmic (\$)	Gravel	Silt/ clay			Sand		
								Very coarse	Coarse	Medium	Fine	Very fine
1a	Bimodal, Very Poorly Sorted	Sandy Mud	53.3	9.4	1.78	0.00	83.04	0.01	0.10	0.32	1.30	15.23
1b	Bimodal, Poorly Sorted	Sandy Mud	75.4	9.4	1.87	0.08	51.50	0.05	0.62	1.22	10.24	36.30
2a	Bimodal, Very Poorly Sorted	Muddy Sand	150.9	9.4	2.03	0.22	37.59	0.07	0.86	3.37	31.61	26.29
3a	Unimodal, Very Poorly Sorted	Slightly Gravelly Muddy Sand	213.4		2.33	2.48	23.85	0.51	6.88	19.95	31.91	14.42

Based on this analysis, the Fladen Grounds can broadly be described as being a sandy mud substrate (Figure 3). The south east extent of the Central Fladen NCMPA (Figure 3) exhibits the highest sediment heterogeneity with groups 2a - muddy sand and 3a - slightly gravelly muddy sand, defining this area. Samples allocated to group 1b - muddy sand, extend into the rest of the Central Fladen NCMPA area and to the south east extent of the Central Fladen NCMPA boundary.



Figure 3. Spatial distribution of sediment cluster groups for all sediment samples collected at the Fladen Grounds (including both 2013 and 2014 surveys).

3.3.2 Spatial patterns in sediment geochemistry

The spatial distribution of geochemical values (organic carbon, organic nitrogen, total phosphate, inorganic phosphate and organic phosphate) measured in sediment samples from the 2014 survey at the Fladen Grounds are presented in Figure 4 to Figure 8. Low levels of organic carbon (% m/m) (Figure 4) were found in sediment samples collected from within the south east portion of Central Fladen NCMPA with the exception of a single higher recording. The remaining variables were generally distributed evenly across the stations surveyed in 2014. The mean and range of values for organic carbon (% m/m), organic nitrogen (% m/m), total phosphate (% by mass), inorganic phosphate (% by mass) and organic phosphate (% by mass) are presented in Table 2.

Table 2. Mean and range of geochemical value Fladen Grounds.	ues measured in se	diment samples from the 2014 survey at the
	N /	

	Mean	Rang	ge
		High	Low
Organic carbon (% m/m)	0.56	2.10	0.1
Organic nitrogen (% m/m)	0.09	0.28	0.04
Total phosphate (% by mass)	0.04	0.098	0.022
Inorganic phosphate (% by mass)	0.04	0.088	0.018
Organic phosphate (% by mass)	0.01	0.052	0



Figure 4. Spatial distribution of organic carbon (% m/m) in sediment samples collected at the Fladen Grounds in 2014.



Figure 5. Spatial distribution of organic nitrogen (% m/m) in sediment samples collected at the Fladen Grounds in 2014.



Figure 6. Spatial distribution of total phosphate (% by mass) in sediment samples collected at the Fladen Grounds in 2014.



Figure 7. Spatial distribution of inorganic phosphate (% by mass) in sediment samples collected at the Fladen Grounds in 2014.



Figure 8. Spatial distribution of organic phosphate (% by mass) in sediment samples collected at the Fladen Grounds in 2014.

The ratio of organic carbon to organic nitrogen (i.e. the C:N ratio) for all sediment samples were between 4:1 and 10:1. The organic nitrogen to phosphate (N: P) molar mass ratios were greater than 16:1 although two samples contained relatively low N: P ratios which could be caused by biological contamination of the sample, or may indicate physical turnover of sediments.

3.3.3 Spatial distribution of seapens and burrowing megafauna

In this study, higher densities of both *Pennatula. phosphorea* (Figure 9) and *Virgularia. mirabilis* (Figure 10) were recorded in 2014 than in 2013, with total counts for a single video tow reaching 1,851 individuals of *P. phosphorea* and 1,461 individuals of *V. mirabilis. F. quadrangularis* was only observed in the south east extent of the Central Fladen NCMPA in 2014, and in an area of coarser sediment outside the western boundary of the site in video tows taken in 2013 (Figure 11). Inside the Central Fladen NCMPA, the number of *F. quadrangularis* reached 0.32 individuals per m² (82 individuals in a 257m video tow). Central Fladen NCMPA exhibited much lower densities of *P. phosphorea* and *V. mirabilis* when compared to other areas surveyed within the Fladen Grounds. The highest densities of *P. phosphorea* per m² were observed in the low abrasion pressure boxes reaching up to 7.85 individuals per m² (1,851 individuals in a 236m video tow). The highest densities of *V. mirabilis* were recorded in video tows on the western boundary of Central Fladen NCMPA where densities reached 6.1 per m² (1,388 individuals in a 277m video tow). Summary information on the spatial distribution of seapens is presented in Table 3. It should be noted that different areas and sampling strategies were employed in 2013 compared to 2014.



Figure 9. Density per m^2 of *Pennatula phosphorea* (dark blue dots) and absence (burgundy dots) across the Fladen Grounds in 2013, and in 2014 (sky blue dots – present, pink dots - absent). All dots represent the start of line position for a single video tow. Inset map shows density per m^2 of *P. phosphorea* observed in the south east of Central Fladen NCMPA.



Figure 10. Density per m^2 of *Virgularia mirabilis* (dark green dots) and absence (burgundy dots) across the Fladen Grounds in 2013 and in 2014 (light green dots – present, pink dots - absent). All dots represent the start of line position for a single video tow. Inset map shows density per m^2 of *V. mirabilis* observed in the south east of Central Fladen NCMPA.



Figure 11. Density per m^2 of *Funiculina quadrangularis* (burnt orange) and absence (burgundy dots) across the Fladen Grounds in 2013 and in 2014 surveys (orange dots – present, pink dots - absent. All dots represent the start of line position for a single video tow. Inset map shows density per m^2 of *F. quadrangularis* observed in the south east of Central Fladen NCMPA.

		2013 (<i>n</i> =69)	2014 (<i>n</i> =160)
	Mean count per tow	9.8	202
Ponnatula	Range of counts per tow	2-53	1-1851
rennatula	Mean density per tow	0.01	1.09
phosphorea	Range of densities per tow	0.01-0.04	0.01-7.86
	Percent of tows with 0 observations	19%	3%
	Mean count per tow	6.8	111
Virgularia	Range of counts per tow	2-49	1-1461
virguiaria	Mean density per tow	0.01	0.53
miabilis	Range of densities per tow	0.01-0.03	0.01-6.10
	Percent of tows with 0 observations	21%	8%
	Mean count per tow	2.5	0.84
Eunioulino	Range of counts per tow	4-98	1-82
runiculina	Mean density per tow	0.001	0.004
quadrangularis	Range of densities per tow	0.01-0.03	0.01-0.32
	Percent of tows with 0 observations	91%	95%

Table 3. Summary information of counts and densities of seapens; P. phosphorea V. mirabilis F. quadrangularis from video tow data collected in 2013 and 2014.

Nephrops burrow densities per m² calculated using data collected in 2014 by JNCC and Cefas, and data from years 2008 to 2010 collected during Marine Scotland's *Nephrops* TV surveys, are presented in Figure 12. In 2014, the highest burrow densities reached 0.31 per m² or 66 burrows in a 211m video tow. Values from the historical data (2008-2010) were slightly higher with densities reaching 0.43 per m² in 2008; however, most historical stations where located in the northern extent of the Central Fladen NCMPA which was not surveyed in 2014. Summary information on the spatial distribution of *Nephrops* burrows is presented in Table 4.



Figure 12. *Nephrops* burrow density per m² across the Fladen Grounds from the 2014 surveys (yellow) and *Nephrops* TV surveys between 2008 and 2010 (gold). Inset map shows density per m² of *Nephrops* burrows observed in the south east of Central Fladen NCMPA.

Table 4. Summary information of burrow counts and densities per m² across the Fladen Grounds calculated from video tow data collected in 2014. Nephrops burrow density information only is presented for the year 2008, 2009 and 2010.

	2008	2009	2010	2014
Mean count	-	-	-	202
Range of counts	-	-	-	1-1851
Mean density	0.3	0.23	0.25	1.09
Range of densities	0.2-0.43	0.09-0.31	0.23-0.5	0.01-7.86
Percent of tows with 0 observations	-	-	-	3%

3.3.4 Contaminants analysis

Analysis of sediment for metals, polyaromatic hydrocarbons (PAHs) and polychlorinated biphenyles (PCBs) are presented in Table 5 and Table 6 respectively. Of the metals analysed, chromium was found in higher concentrations than the OSPAR background levels at three of the five stations. Nickel was recorded in higher than OSPAR Background Assessment Concentrations (BACs) levels at one of the five stations, but the quantities of nickel at all of the five stations exceeded theEffects Range Low (ERL) levels which suggest possible toxicity for the species found at those sites. Only Benzo [g,h,i] perylene of the PAHs was found in higher concentrations that the ERL levels at one station while all concentrations of PCBs were below the presented acceptable levels.

Table 5. OSPAR BAC, ERL and ERM values provided by the OSPAR Commission (2009) and US EPA (2013) and results of metal analysis from the five stations sampled for sediment contaminants. Samples with levels above the OSPAR BAC are shown in red.

	OSPAR BAC (µg	ERL (ua a ⁻	ERM (µa a ⁻¹	CA05	CA37	7B05	1B02	CLMR01
Metals	g ⁻¹ dw)	¹ dw)	dw)					
Chromium (Cr)	81	81	370	99.14	74.23	93.4	71.68	93.31
Nickel (Ni)	36	20.9	51.6	32.13	26.04	28.54	25.89	37.29
Copper (Cu)	27	34	270	12.69	8.79	9.69	7.76	13.09
Zinc (Zn)	122	150	410	55.47	39.93	43.5	36.6	60.23
Arsenic (As)	25	8.2	70	10.22	6.31	5.3	5.56	10.86
Cadmium (Cd)	0.31	1.2	9.6	0.3	<0.2	0.32	0.24	0.3
Lead (Pb)	38	46.7	218	26.2	19.56	17.07	16.85	24.74
Mercury (Hg)	0.07	0.15	0.71	<0.225	<0.239	<0.226	<0.225	<0.209

Table 6. EAC, ERL and ERM values provided by the OSPAR Commission (2009) and US EPA (2013) and results of PAH and PCB analysis from the five stations sampled for sediment contaminants. Samples with levels above the ERL levels are shown in red.

	EAC	ERL	ERM	CA05	CA37	7B05	1B02	CLMR01
	(µg g⁻¹ dw)	(µg g⁻¹ dw)	(µg g⁻¹ dw)					
PAH								
Naphthalene	NA	160	2100	2.601	15.450	2.963	5.667	2.017
Fluorene	NA	19	540	1.081	4.993	1.429	2.463	0.811
Phenanthrene	NA	240	1500	6.433	31.916	8.343	14.204	4.851
Anthracene	NA	85	1100	0.658	3.977	0.791	1.333	0.421
Pyrene	NA	665	260	4.743	19.407	5.262	10.315	2.988
Benz[a]anthracene	NA	261	1600	2.909	13.158	4.549	5.574	2.119
Chrysene	NA	384	2800	3.358	13.968	3.319	6.204	2.120
Benzo[a]pyrene	NA	430	1600	8.924	33.196	9.007	17.315	4.993
Indeno[1,2,3-	NA	240	NA					
cd]pyrene				35.834	130.738	34.783	65.333	19.670
Benzo[g,h,i]perylene	NA	85	NA	23.345	87.360	22.865	43.759	13.223
PCB								
CB#28	<0.2	<0.2	<0.2	<0.2	<0.2	1.7	NA	NA
CB#52	<0.2	<0.2	<0.2	<0.2	<0.2	2.7	NA	NA
CB#101	<0.2	<0.2	<0.2	<0.2	<0.2	3	NA	NA
CB#118	<0.2	<0.2	<0.2	<0.2	<0.2	0.6	NA	NA
CB#153	<0.2	<0.2	<0.2	<0.2	<0.2	40	NA	NA
CB#138	<0.2	<0.2	<0.2	<0.2	<0.2	7.9	NA	NA
CB#180	<0.2	<0.2	<0.2	<0.2	<0.2	12	NA	NA

3.4 Discussion and conclusions

The Fladen Grounds is predominantly an area of sandy mud but with greater sediment heterogeneity in the south east part of the Central Fladen NCMPA, and a 'muddy' area in the centre and south east of the Fladen Grounds. These findings support previous observations from *Nephrops* stock assessment surveys that the softest sediments are located in the centre and south east of the grounds where *Nephrops* abundances have been found to be greatest (Ungfors *et al* 2013). The increased sandy gravelly sediment fraction observed within the south east part of the Central Fladen NCMPA was also associated with a lower density of *Nephrops* burrows while the sandy mud sediment found within the north west of the NCMPA boundary have higher burrow densities. This is unsurprising given that *Nephrops* fisheries occur on muddy bottoms that have a specific silt and clay content, a key requirement for their burrowing behaviours (Ungfors *et al* 2013).

Lower densities of both *P. phosphorea* and *V. mirabilis* were also observed in the coarser sediments of the south east extent of the Central Fladen NCMPA and in some tows, these species were absent. Conversely, this was one of only two areas across the Fladen Grounds wider area that *F. quadrangularis* was observed. These findings are particularly interesting when considering the results of a study by Greathead *et al* (2014) who modelled the potential distribution of the three seapen species on the west coast of Scotland. The authors found *V. mirabilis to* have the widest modelled geographical extent with the highest tolerance to gravel content, a sedimentary characteristic that was much less favourable for *P. phosphorea* and *F. quadrangularis*. However, highest densities of *V. mirabilis* in this study were associated with an area of sandy mud along the central northern boundary of the NCMPA.

Sediment contaminant data presented here have been compared to OSPAR guidelines. However, status and temporal trends are traditionally assessed by comparing values recorded in the most-recent monitoring year to the available assessment criteria for background levels. If the most-recent monitoring year is significantly below the BAC then concentrations are said to be 'at background'. No formal assessment of status can be made when there is only one or two years of data and temporal trends can only be interpreted when at least five years of data are available. With this in mind, it is not possible to draw broad conclusions regarding the levels of contamination at the Fladen Grounds, but from the five samples analysed, the levels of PCBs and PAHs are at or below OSPAR background levels. Only the metals chromium, nickel and cadmium are present in concentrations that may have an effect on the biological communities present. In order to fully understand aspects of sedimentary biogeochemistry and contaminant effects at the Fladen Grounds, a dedicated survey would be required.

4 Type 2 monitoring - Sensitivity of mud habitats to abrasive pressure

4.1 Introduction

As discussed in Section 1.1, a key objective of the 2014 Fladen Ground survey was to calculate a number of biodiversity metrics for the potential detection of meaningful change in habitat range, extent and condition in relation to increasing abrasion pressure. The metrics that were found to respond best to pressure at the site could then be applied to each of the three types of monitoring; Type 1 monitoring: rate and direction of change in the long-term (time-series); Type 2 monitoring: change along a pressure gradient; and Type 3: cause and effect through experimental design.

For the 2014 Fladen Grounds survey, metrics were selected with consideration for the relevant biodiversity indicators that are being developed as part of Marine Strategy Framework Directive (MSFD) requirements. When developed, the MSFD indicators will fit into a series of descriptors and associated criteria, as presented in the Commission Decision on Good Environmental Status (GES) (European Commission 2010) and many of these will be sampled through the UK Marine Biodiversity Monitoring R&D Programme's (UK MBMP) integrated monitoring scheme. It is intended that the outputs of the MSFD indicator development will, in turn, support an assessment of whether GES is being achieved across UK waters. Each metric selected for the Fladen Grounds survey had relevance to one or more of the MSFD Commission Decision Criterion with most relating to the ability to detect change in benthic community condition (Descriptor 1.6 "Habitat condition").

While the range of biodiversity metrics chosen for testing at the Fladen Grounds were developed with MSFD descriptors in mind, they were also devised based on the habitat type and MPA protected features present at the site. In this section, the metrics chosen have been tested in the context of Type 2 monitoring: to detect a trend along gradient of abrasion pressure. Environmental variables such as sediment type and geochemistry, as well as aspects of community ecology such as infaunal and epifaunal species and assemblage metrics, Biological Traits Analysis, bivalve size frequency and *Nephrops* burrow densities were tested using data collected from the 2014 Fladen Grounds survey.

4.2 Abrasive pressure gradient

JNCC has produced a recommended method for the creation of a standard UK-wide geodata 'layer' showing the intensity of abrasion on substrate caused by human activities (currently the layer accounts only for fisheries abrasion), focusing on the area beyond 12 nautical miles (nm) from the coast (Church *et al* 2015). It is intended that this method and the metrics and parameters used (fishing gear types and associated trawl widths and speeds) could be adopted as common approaches to aid comparison between studies in the future. In this study, the JNCC method was used to identify areas for survey along a categorical abrasion pressure gradient at the Fladen Grounds and ultimately, to test if the method was sensitive enough to detect a biological change with varying abrasive pressure.

4.2.1 Environmental characteristics

Trawling has been linked to a number of state changes in the physical aspects of the sea bed including alterations to sea bed morphology, sediment re-suspension, increased bottom water turbidity and altered nutrient cycling (Percival *et al* 2003; Tillin *et al* 2006; Pusceddu *et al* 2014). The continuous turnover of the upper most sediment layers has been associated with a decrease in organic carbon turnover rates (Pusceddu *et al* 2014) and mixing of

organic matter and oxygen-rich surface waters with newly exposed anoxic sediments (Percival *et al* 2003). The collection of particle size data and measurement of sediment organic carbon and nitrogen and nitrate: phosphate ratios were used to test these environmental metrics, both to relate physical changes of the sea bed to differing abrasion pressure, as well as provide environmental context for any observed differences in faunal assemblages.

4.2.2 Infaunal species and assemblage metrics

In addition to the physical changes to soft sediment habitats that chronic trawling activities can have, there has also been a strong link to changes in infaunal benthic communities (Jennings *et al* 2001; Hinz *et al* 2009). In the study by Hinz *et al* (2009), which explored the impacts of otter trawling of a *Nephrops* fishery, significant negative responses were found for all the univariate descriptors explored (abundance, biomass and species richness). In the present study, infaunal species and assemblage metrics, including univariate and multivariate analyses were selected to compare the burrowed mud protected feature in areas of low abrasion with areas of high abrasion. These metrics could also be used to support indicator development for Criterion 1.6 of the Commission Decision on GES, "Habitat Condition", in particular the supporting indicators 1.6.1 "Condition of the typical species and communities and 1.6.2 "Relative abundance and/or biomass, as appropriate".

4.2.3 Biological Traits Analysis (BTA)

Information on the physical and behavioural attributes of faunal communities can give an indication of habitat condition, as the functions of benthic organisms are crucial to ecosystem function and the regulation of ecosystem processes in the marine environment (Snelgrove 1998). These functions are determined by species' biological traits. Biological traits such as life history, morphology and ecology reflect both their sensitivity to given pressures and their function as part of the ecosystem (Frid *et al* 2000).

The need to understand how human impacts affect the functioning of the ecosystem has led to an increase in the use of biological traits as a proxy for ecological function in marine research (Tillin et al 2006; De Juan et al 2007; Barrio Froján et al 2011; De Juan and Demestre 2012: Paganelli et al 2012: Wan Hussin et al 2012: Fleddum et al 2013: Bolam et al 2014). Biological Traits Analysis has advantages over species identification-based approaches by avoiding constraints imposed by biogeography: Geographically separate communities that occupy similar substrates may have few species in common but may function in a similar way and provide the same goods and services to the local ecosystem. Bremner et al (2006) identified 28 biological traits as key indicators of ecosystem functioning at a potential SAC. In reality, information on all traits for the majority of species is limited. Researchers have therefore focused on those traits that are most relevant to the specific questions they wish to address. With respect to impacts of seabed disturbance, traits such as size, longevity, body design, living habit, living location (position in sediment) feeding type, mobility and reproductive strategies have been frequently used in analysis (see Tillin and Tyler-Walters 2014, and references herein). For the purposes of this report we have used a subset of eight traits, identified by Tillin and Tyler-Walters as influencing the sensitivity of benthic macroinvertebrates to physical damage (abrasion, subsurface penetration and disturbance), to explore how they respond to different levels of abrasive pressure.

4.2.4 Bivalve size frequency

Bivalves can play a pivotal role in ecosystem functioning through their contribution to benthic-pelagic coupling, nutrient regeneration and their facilitation of surrounding

communities (Norkko *et al* 2013). However, larger benthic species, including bivalves, generally suffer greater mortalities following a trawl event compared to smaller species because they are crushed by the path of the net or are caught in the net and subsequently discarded (Jennings *et al* 2001). As part of MSFD indicator development, a candidate indicator titled "Size-frequency distribution of bivalves or other sensitive/indicator species" was proposed to measure the number and/or biomass of bivalve individuals per size class. This indicator has been identified by OSPAR's Intersessional Correspondence Group on the Coordination of Biodiversity Assessment and Monitoring (ICG-COBAM) to meet Criterion 6.2 of the Commission Decision on GES, "Condition of benthic community", in particular the supporting indicator 6.2.3 "Proportion of biomass/number of individuals above specified length/size".

This candidate indicator was based on a proposal from HELCOM for a soft-bottom bivalve size-structure indicator for physical disturbance or hypoxia. The basis for the indicator is that benthic communities typically consist of a mixture of long-living and short-living species. The short-living species are usually small with low individual biomass, whilst the long-living species can reach much bigger sizes and higher individual biomass. Under natural conditions, populations of large species consist of different size-classes representing different age-groups. The natural balance between both 1) the large and small species within the community and 2) the large and small specimens within the population of a single species can be affected by anthropogenic influences such as physical disturbance, e.g. caused by bottom trawling or sediment extraction (Basset et al 2012, Hiddink et al 2006, Pearson and Rosenberg 1978; Tyler-Walters et al 2009). The proposed metric is a comparison of the current population structure with a (theoretical) natural population structure resulting in a value for the 'degree of naturalness' of the population structure. It should be noted however that bivalves are well known for having 'good' spat fall years and other years of poor recruitment which may make determining what the size-frequency histogram for a particular geographical area should look like problematic.

The objectives of the 2014 survey of the Fladen Grounds included the collection of data for the proposed bivalve size-frequency indicator due to the substrata in the area representing a suitable habitat for bivalves, and the ability to compare the results of the data analysis across the abrasion pressure gradient. Furthermore, data on bivalves was collected during the previous 2013 JNCC-Cefas survey to the Fladen Grounds, and this therefore presented an opportunity to compare data collected across two years.

4.2.5 Epifaunal species and assemblage metrics

Epifaunal species, which are defined as those animals living on the surface of the seabed, suffer mortalities following trawling due to the direct impact of the trawl passing over, or to subsequent discarding since they have no commercial value (Jennings *et al* 2001; Hinz *et al* 2009). Abundance and species richness metrics were selected to explore variation in epifaunal assemblages present in the burrowed mud protected feature across the pressure gradient in areas of high, low and intermediate abrasion pressure. One of the key epifaunal species associated with the burrowed mud protected feature is seapens. Physical disturbance from abrasion pressure may have modifying effects on seapen distribution and as a result, they have been identified as an important indicator for the quality of mud habitats and their associated communities (Macdonald *et al* 1996). As such, abundance and densities per m² of the three species of seapen found in the Fladen Grounds area, *P. phosphorea, V. mirabilis* and *F. quadrangularis*, were selected as a discrete biological metric for testing.

4.2.6 *Nephrops* burrow density

As discussed previously, the Fladen Grounds is a key *Nephrops* fishery in the North Sea. *Nephrops* burrows are also considered a key feature of the burrowed mud protected feature alongside the presence of seapens. Burrow density was therefore considered to be an important metric to explore along the abrasion pressure gradient.

4.3 Methods

4.3.1 Analysis of historical spatial and temporal abrasion pressure at the Fladen Grounds

Annual abrasion data layers were created for the wider Fladen Grounds between the years 2009 and 2013 according to the JNCC abrasion pressure method (Church *et al* 2015). The JNCC method reports using a 0.05 decimal degrees (dd) grid size to aggregate ping information, however, for this investigation a grid with a resolution of 0.039 dd was used.

A spatial and temporal assessment of abrasion pressure at the Fladen Grounds was carried out to provide historical context to the newly acquired empirical data. Annual change from year to year was explored by subtracting one year from the previous year using the raster calculator function in ArcGIS v10.1. Similarly, several further spatial maps, including; mean abrasive effort, range and standard deviation were created to explore the variability in abrasive pressure across the Fladen region over the five years of available data.

Abrasion layers were aggregated in several temporal combinations to explore how this may affect the exploratory power of abrasive pressure with relation to community composition in the Fladen Grounds region. Four alternative pressure metrics for abrasion were explored and are presented below with their abbreviations in brackets (used from this point forward);

- UK and non-UK combined VMS for all benthic gear types between 2012 and the survey commencement in 2014 (SWA1214);
- UK and non-UK combined VMS for subsurface benthic gear types between 2012 and the survey commencement in 2014 (SWASUB1214);
- UK and non-UK combined VMS for all benthic gear types between 2013 and the survey commencement in 2014 (SWA1314). Along with;
- UK and non-UK combined VMS for subsurface benthic gear types between 2013 and the survey commencement in 2014 (SWASUB1314).

It should be noted that for the purposes of survey planning and design, abrasive pressure values forming the gradient were based on annual fishing effort (swept area (m²)) for 2012 and 2013 data only.

4.3.2 Survey design

Abrasive pressure values based on swept area from 2012 only (calculated according to JNCC method Church *et al* 2015) alongside species abundance values were used for a power analysis to provide an indication of how many samples would be required to achieve an acceptable level of power (>85%) in detecting a 30% change in species abundance. This was considered to be the minimum power acceptable in terms of potential to achieve statistical significance with the sample analysis.

To determine the pressure gradient, a non-parametric Mann Kendall test was used to account for abundances for a particular pressure class. For a particular value within that class, the number of abundance values in higher pressure classes that are greater than (assigned +1) or less than it (assigned -1) are counted. This was summed for all points in the

simulated data set, with the exception of the highest pressure class. If the p value for this statistic was less than 0.05 then the gradient is assumed to have been detected. The power was calculated as the proportion of the 1,000 simulations that detected the gradient (Table 7).

 Table 7. Results of the power analysis to estimate the number of stations (N) required within each pressure gradient box.

Ň	5	7	8	9	10	11	12
Power	0.54	0.72	0.76	0.81	0.87	0.91	0.92

Absolute abrasion scores were placed into a categorisation (seven categories in all) and two sampling cells were selected from each, giving a total of fourteen treatments. Ten randomly assigned benthic samples were to be taken within each treatment (1A & B – 7A & B) and can be seen in (Figure 13). This sampling effort was deemed appropriate based on a power analysis (Table 7) which looked at the variance in abundance of three infaunal species, from data collected in 2012. These three species were selected as they had demonstrated sensitivity in abundance to the pressure gradient, and responded in a negative relationship with increasing pressure.



Figure 13. Planned Fladen fishing pressure survey locations, as per the cruise specific 'Plan of action' prior to the survey. Paired boxes have similar calculated surface abrasion (per unit swept area) with Box 1 having the lowest and Box 7 having the highest.

Due to inclement weather experienced during the survey and adjustments to the VMS data interpretation, changes were made to the sampling design. These changes were reported to the relevant personnel in JNCC and Cefas prior to commencing data collection. The final fishing pressure values used are presented in Table 8 and the final boxes sampled shown in Figure 14.

 Table 8. Fishing pressure cell values calculated from VMS data for UK and non-UK fishing vessels between

 January 2013 and pre-survey 2014.

Cell code	Fishing pressure (swept area km²)	Pressure category
N1B	0.05556	Low
N1A	1.359368	Low
7B	11.202131	Intermediate
1B	17.55449	Intermediate
2A	27.542	Intermediate
6A	34.4932	Intermediate
N6A	91.269696	High
N6B	53.267224	High



Figure 14. Location of surveyed abrasion pressure boxes from the 2014 survey showing two High pressure boxes (N6A, N6B), two Low pressure boxes (N1A, N1B) and four Intermediate pressure boxes (7B, 2A, 6A, 1B). Both infauna and epifauna were collected in Low and High abrasion pressure boxes, only epifauna was sampled for Intermediate pressure boxes.

Annual abrasive effort, calculated as swept area (m²), for these sampled sites is presented in Figure 15. From the plot it can be assumed that Low abrasion pressure boxes (N1A and N1B) were consistently un-fished, whilst High abrasion pressure boxes (N6A and N6B) were consistently exposed to a greater level of fishing activity.


Figure 15. Annual fishing effort (swept area (m²)) for 2012 and 2013 for the four surveyed abrasion pressure boxes at the Fladen Grounds. N1A and N1B are Low pressure and N6A and N6B are High pressure boxes.

4.3.3 Sample acquisition

Samples of the seabed were collected aboard the RV *Cefas Endeavour* (survey code: CEND05/14). Technical specifications of the survey equipment used are provided in the cruise report (McIlwaine 2015). Due to inclement weather during survey, it was not possible to collect the full suite of groundtruth data. In total, 10 Day grab samples were collected from within two High abrasion pressure boxes (N6a, N6b) and two Low abrasion pressure boxes (N1a, N1b) and ten video tows were acquired from two High abrasion pressure boxes (N6a, N6b), two Low abrasion pressure boxes (N1a, N1b) and four Intermediate abrasion pressure boxes (2A, 1B, 6A and 7B). Camera tows were prioritised as the most appropriate method to collect data on the benthic community characteristic of burrowed mud, namely seapens. Detailed sample acquisition methods are provided in Appendix 1: Sample acquisition.

4.3.4 Sample processing

4.3.4.1 Environmental characteristics

Sediment samples from the 40 Day grab deployments were analysed for particle size distribution as described in Section 3.2.1 and grouped in sediment groups using Entropy non-hierarchical clustering (Stewart *et al* 2009). In addition, sediment samples were also analysed for total organic carbon and nitrogen, total phosphate, inorganic phosphate and organic phosphate as detailed in Section 3.3.2.

4.3.4.2 Infauna: species and assemblage analysis

Infaunal processing of the Day grab samples followed standard laboratory practices. Results were checked following the recommendations of the NMBAQC scheme (Worsfold *et al* 2010). Taxa were identified to highest taxonomic resolution and weighed.

4.3.4.3 Bivalve size frequency distribution

Morphometric measurements for individual bivalves of all species present were taken. Metrics recorded for each individual included: 1) maximum valve width (posterior to anterior, dorsal up in mm) per individual; or 2) maximum valve height (umbo to ventral maxima in mm) per individual; as well as whole body biomass (blotted wet weight in g) per individual. The valve measurement recorded (valve width or height) was determined by the value which was greater (e.g. *Abra* spp. was measured using metric 1 while *Pecten maximus* was measured using metric 2). The valve measurement applied was used consistently for a given species throughout the analysis. As well as the bivalve measurements taken of individuals present in the abrasion pressure samples, measurements from samples collected in 2013 (survey: CEND01/13X) and the BACI experimental study in 2014 (survey: CEND05/14) were recorded to provide additional data points to create species size frequency distribution plots across the Fladen Grounds.

4.3.4.4 Epifauna: species and assemblage analysis

Eighty video tows and 910 still images from eight abrasion pressure boxes (two High, two Low and four Intermediate) were processed following guidance documents developed by Cefas and the JNCC for the acquisition and processing of video and still images (Coggan *et al* 2007). Each video transect was split into one minute segments and faunal counts and SACFOR scores were determined. Identification of anthropogenic activities such as the presence of trawl scars, fishing gears and litter was recorded at each station. In addition, any observations of potentially damaged seapens (e.g. broken or lying flat on the sea bed) were noted. A description of seapen condition and a time stamp of when the individual was seen were recorded.

4.3.4.5 Nephrops burrow density and distribution

Additional processing of each video tow was conducted to count the number of *Nephrops* burrows. *Nephrops* burrows were counted for each one minute video segment following methods used in annual UWTV *Nephrops* stock assessment surveys. Two sets (10% of all tows each) of video tows were selected for a second count to ensure data quality.

4.3.5 Data analysis

4.3.5.1 Infaunal univariate data analysis

Univariate metrics calculated for each sample included total number of individuals (N), total number of taxa (taxon richness) (S), Hill's (1973) taxon diversity index (N1) and total wet weight biomass (B). Boxplots were produced in SigmaPlot[®] for each of the metrics. Smoothed semi-variogram plots for infaunal univariate metrics and each abrasion pressure box were produced to test for spatial auto-correlation. The plots confirmed independence of the boxes tested, however, while analysis may be able to demonstrate that certain abrasion pressure boxes of the same abrasion pressure category elsewhere in the site. A one-way ANOVA was used to explore the relationship between these univariate metrics (with the exception of biomass) and the four levels of abrasion pressure (two classed as Low and two classed as High pressure). An unpaired t-test was performed on the univariate metrics for samples classed according to Low or High pressure.

4.3.5.2 Biological Traits Analysis

Eight biological trait categories with 35 modalities were selected to investigate the functional composition and vulnerability of benthic infaunal communities within the High and Low abrasion pressure boxes (Table 9). Trait information was extracted from a pre-existing and pre-coded trait database developed within Cefas. Where trait information for a species was absent from the database, information was sourced from published literature and internet searches. Where no information could be found, the traits from conspecifics or closely related taxa were assigned. This allowed all taxa to be included in the analysis.

 Table 9. Eight trait categories and their associated modalities, adapted from Tillin and Tyler-Walters (2014) and used in the present study.

Trait category	Modality
Sediment position	Surface
	0-5cm
	6-10cm
	>10cm
Feeding type	Suspension
	Surface deposit
	Subsurface deposit
	Predator
	Scavenger/Opportunist
Maximum size	>10mm
(from literature)	11-20mm
	21-50mm
	51-100mm
	101-200mm
	201-500mm
Flowibility	>500mm
Flexibility	None
	1000000000000000000000000000000000000
Ero gility	High (>45)
гадішу	Intermediate
	Pobust
Living babit	Attached
Living habit	Front
	Burrow dwelling
Longevity	<1 vear
Longovity	1-3 years
	3-10 years
	>10 vears
Mobility	Sessile
	Low mobility (creep, crawl, climb)
	Active burrower
	Swimmer

Trait modalities were assigned to individual taxa using a 'fuzzy coding' approach (Chevene *et al* 1994) according to the extent to which they displayed the modalities of each trait. Fuzzy coding allows taxa to exhibit categories to different degrees, avoiding the obligate assignment of a taxon to a single category that can lead to inaccurate characterisation of biological or ecological taxa profiles (Usseglio-Polatera *et al* 2000). In order to classify a taxon according to its affinity for more than one modality within a trait, each modality was given a score between 0 and 3, where 0 conveys that the taxon has no affinity for that modality, 1 or 2 express partial affinity and 3 indicates total and exclusive affinity for that modality (Bolam *et al* 2014). In reality, certain traits such as sediment position, feeding type

and longevity were predominantly expressed as partial affinities for most taxa. This reflected 1) variability of the trait within a particular taxon, 2) variability in the trait for a taxon from different published sources, and 3) variability displayed between different species within a genus. In contrast, entries for other traits, e.g. morphology, mobility and size, were often represented by a total affinity for one particular modality.

When all taxa had been coded for the species by traits matrix, the codes were converted to proportions (i.e. *affinity* scores) for each taxon so that the total for each taxa x trait = 1. For example, for the trait 'feeding mode', *Amphiura* was assigned a '3' for suspension feeding, a '3' for surface-deposit feeding and a zero for the remaining categories; this was subsequently standardised to 0.5 and 0.5, respectively. Two different analytical methods were then applied to the traits matrix to:

- compare the modality composition of each trait category (weighted by abundance or biomass) between High and Low abrasion pressure. This will highlight if there are any differences in community function when subjected to different pressures. This method makes no assumption of how vulnerable a trait modality is to demersal trawling; and
- calculate the perceived vulnerability of communities within the High and Low abrasion pressure boxes using all eight trait categories as per Tillin and Tyler Walters, 2014, and using a reduced number of traits (five) following the approach of de Juan and Demestre (2012). This method makes a predetermined assumption regarding the perceived vulnerability of each trait modality to demersal trawling.

Detailed methodologies for the two methods are provided below:

Method1: Functional composition

The standardised species by traits matrix was weighted by abundance or biomass to produce two station by trait matrices. Modality composition of each of the eight traits was investigated by converting the station by trait matrices to percentage composition (for each trait category and station). This allowed a direct comparison of trait composition between stations and will give an indication of potential differences in infaunal community function. Maps of each trait category for both abundance and biomass weighted trait data were created using ArcGIS v10.1.

Method 2: Community vulnerability

Community vulnerability was assessed using the standardised species by trait matrix, following methods detailed in De Juan and Demestre, 2012, whereby each modality within a trait category is scored according to its perceived vulnerability to trawling, as follows:

- 0 = Not affected by trawling or advantageous to trawling
- 1 = Low vulnerability to trawling
- 2 = Moderate vulnerability to trawling
- 3 = High vulnerability to trawling

For instance, surface-dwelling organisms were classified as highly vulnerable to trawling and therefore their trait affinity score would be multiplied by 3. Smaller organisms are assumed to suffer lower mortality as they are pushed aside by the pressure wave in front of the fishing gear (Gilkinson *et al* 1998) and therefore were assigned a lower score. Organisms living greater than 10cm deep in the sediment were assumed to be unaffected by trawling and therefore their trait affinity score was multiplied by 0.

Vulnerability scores were assigned to each of the modalities (Table 11) using the eight trait categories as shown in Table 10:

Score	Position in sediment	Feeding type	Mobility	Size (mm)	Flexibility	Fragility	Living habit	Longevity (years)
0	>10cm	Scavenger/ opportunist	Swimmer	<20		Robust		<1
1	6-10cm	Subsurface deposit/ predator	Active burrower	21-50	>45°		Burrow dwelling/ Tube dwelling	1-3
2	0-5cm	Surface deposit	Low mobility	51- 100	10-45°	Intermediate	Free- living	3-10
3	Surface	Suspension	Sessile	>100	None	Fragile	Attached/ Erect	>10

Table 10. Trait modalities in relation to their vulnerability to trawling scores (eight traits).

The trait affinity scores for each species were then weighted by the vulnerability scores to produce a species by trait vulnerability matrix as in Table 11.

 Table 11. Example calculation of trait vulnerability scores for five different species.

Species by trai	it affinit	t y											
Score	3	2	1	0	3	2	1	1	0	3	2	1	0
Position			Feeding				Mobility						
Example species	Surf	0- 5cm	6- 10cm	>10cm	Susp	SurDep	Sub surf dep	Pred	Scav Opp	Ses	Crawl	Bur	Swim
Pennatula	0.50	0.17	0.17	0.17	1.00	0.00	0.00	0.00	0.00	1.00	0.00	0.00	0.00
Glycera	0.00	0.33	0.33	0.33	0.00	0.00	0.00	0.50	0.50	0.00	0.00	1.00	0.00
Ampelisca	0.50	0.50	0.00	0.00	0.50	0.50	0.00	0.00	0.00	0.00	0.50	0.25	0.25
Brissopsis	0.00	1.00	0.00	0.00	0.00	0.50	0.50	0.00	0.00	1.00	0.00	0.00	0.00
Limatula	1.00	0.00	0.00	0.00	1.00	0.00	0.00	0.00	0.00	0.50	0.00	0.00	0.50

T

		_
		1
		1
		1
/	<	~
	\sim	

Species by trai	it vulne	rability											
Pennatula	1.5	0.33	0.17	0	3	0	0	0	0	3	0	0	0
Glycera	0	0.67	0.33	0	0	0	0	0.5	0	0	0	1	0
Ampelisca	1.5	1	0	0	1.5	1	0	0	0	0	1	0.25	0
Brissopsis	0	2	0	0	0	1	0.5	0	0	3	0	0	0
Limatula	3	0	0	0	3	0	0	0	0	1.5	0	0	0

The scores for each taxon were summed across the eight traits. The maximum possible score was 24, representing taxa most vulnerable to trawling. The abundances of the taxa with the same total score within a station were summed. This resulted in a vulnerability scores by abundance matrix based on eight traits.

To determine how species vulnerability scores could be affected by choice of trait used in the analysis, we recalculated community vulnerability using five trait categories: sediment position, feeding type, mobility and size, plus an additional trait category, termed by de Juan and Demestre as 'Fragility'. This latter category differs from the Fragility category detailed in Tillin and Tyler Waters (2014), which was purely based on how easily an organism can be damaged by physical impact. By combining information on fragility, flexibility, regeneration potential, protection and some aspects of morphology, the category now incorporates information an organism's ability to withstand and recover from physical impact. Hence, taxa classed as the most robust are those that have hard shell, or are vermiform, or can regenerate, whilst those classed as fragile are taxa which have a fragile shell or structure. Each trait modality was assigned a vulnerability to trawling score (Table 12).

Score	Position in sediment	Feeding type	Mobility	Size (mm)	Fragility
0	>10cm	Scavenger/	Swimmer	<20	Hardshell/ vermiform/
		opportunist			regeneration
1	6-10cm	Subsurface deposit/	Active	21-50	Strong/flexible
		predator	burrower		
2	0-5cm	Surface deposit	Low mobility	51-100	No protection
3	Surface	Suspension	Sessile	>100	Fragile shell/structure

 Table 12. Traits modalities in relation to their vulnerability to trawling scores (5 traits).

The species by trait matrix (five trait categories) was weighted by the trait vulnerability to trawling scores as previously and the scores summed for each taxon across the five traits. The highest score possible using five traits was 15, again representing the most vulnerable taxa. As previously, the abundances of the taxa with the same total score within a station were summed. This resulted in a vulnerability score by abundance matrix based on five traits.

The vulnerability scores (using five traits) were grouped into five Functional Indicator groups used by de Juan and Demestre, 2012, where G1 comprised taxa with lowest scores (low vulnerability to trawling) and G5 comprised taxa with highest scores (highest vulnerability to trawling). This grouping methodology was also adapted to accommodate the vulnerability scores using eight traits (Table 13).

Functional group	Score (8 traits)	Score (5 traits)	Vulnerability
G5	21-24	14-15	Highest
G4	17-20	11-13	High
G3	12-16	8-10	Moderate
G2	8-11	5-7	Low
G1	0-7	0-4	Lowest

Table 13. Functional indicator groups assigned to the scores calculated using 8 traits and 5 traits.

For each matrix, the relative abundance of each of the five Functional Indicator groups was calculated for each of the abrasion pressure boxes.

4.3.5.3 Epifaunal species and assemblage analysis

Univariate metrics calculated for each sample included total number of individuals (N), total number of taxa (S), and Hill's (1973) taxon diversity index (N1). Boxplots were produced in SigmaPlot[®] for each of the metrics. Smooth semi-variograms were used to test for spatial auto-correlation between abrasion pressure boxes confirming independence of the specific boxes surveyed. Relationships between abrasion pressure and univariate metrics for the eight abrasion pressure boxes (10 samples within a box N= 80) were modelled using linear regression analysis in the statistical package R. The relationship between densities per m² of

the three species of seapen, *P phosphorea, V. mirabilis* and *F. quadrangularis* and *Nephrops* burrows and abrasion pressure were also modelled using linear regression.

4.4 Results

4.4.1 Temporal and spatial patterns in abrasive pressure across the Fladen Grounds

Figure 16 presents annual surface abrasion effort, calculated as swept area (m²), across the Fladen Grounds for the years of and between 2009 and 2013. Abrasion pressure was found to be variable across the area with effort being targeted in different locations from year to year. In 2011 there was a decline in effort in the area to the south east, and outside the boundary of the Central Fladen NCMPA. However, in 2010, 2012 and 2013 this same area (located in the centre of the Fladen Grounds maps below) appears to be a main focus for the fishery. Within the south east extent of the Central Fladen NCMPA there has been relatively low fishing effort over the time periods presented when compared with the wider fishing effort across the rest of the NCMPA.

Change in annual effort per abrasion grid cell was calculated (Figure 17) and illustrates the variable nature of the fishery from year to year. Relative effort within the central Fladen Grounds, to the south east extent of the NCMPA, shows a decrease between the years 2010 and 2011 and again, between 2012 and 2013. In these years, the effort appears to be redistributed to areas north and south of the core fishery (outside of the south east of Central Fladen NCMPA), and is reflected by a decrease in these areas when abrasion values are increasing in the central Fladen Grounds region (2009-2010 and 2011-2012).

Mean abrasion scores were calculated for the years 2009 to 2013 (Figure 18). It can be seen that, though the fishing effort varies on an annual basis, effort across the Fladen Grounds is in fact relatively stable over longer time periods with the greatest effort being established within the central area of the Fladen Grounds. Results of calculating the standard deviation and range of these abrasion cells across the five years (Figure 19 and Figure 20 respectively) suggest that over longer time periods, the fishery using the Fladen Grounds has a relatively stable effort, and while it does deviate from its mean effort, it is also within a relatively stable range.



Figure 16. Annual surface abrasion pressure scores across the greater Fladen Grounds area for the years between 2009 and 2013. Abrasion scores are calculated as the absolute area, in m², of seabed that has potentially been in contact with a benthic trawl within each 0.05deg cell.



Figure 17. Annual change in surface abrasion pressure scores across the greater Fladen Grounds area for the years between 2009 and 2013. Annual change has been calculated by subtracting surface abrasion values for a given year from the subsequent year. Positive values present an increase in effort within a cell, whilst negative values are demonstrative of a decrease.



Figure 18. Mean surface abrasion pressure scores across the greater Fladen Grounds area calculated for data from 2009-2013.



Figure 19. Range of surface abrasion pressure scores across the greater Fladen Grounds area calculated for data from 2009-2013.



Figure 20. Standard deviation of surface abrasion pressure scores across the greater Fladen Grounds area calculated for data from 2009-2013.

4.4.2 Environmental characteristics

Although four sediment groupings were determined when analysing samples from both 2013 and 2014 surveys (see Section 3.3.1), only three were identified when grouping sediment samples from the 2014 survey alone (Table 14). The Entropy-based sediment groupings for 2014 data and the relative proportions of the major sediment fractions (gravel, sand and silt/clay) for each station located within the Low and High abrasion pressure boxes are presented in Figure 21 and Figure 22 respectively. All stations sampled in the High abrasion boxes were categorised as group 1a (slightly gravelly sandy mud) and showed a high proportion of silt/ clay when comparing the relative proportions of the major sediment fractions. Only two stations in each of the Low abrasion boxes were not grouped into 1a. In each box there was one sample in the 2a group and one sample in the 3a category. Samples in the Low abrasion boxes had a higher proportion of sand than those in the High abrasion boxes.

			MC		SORTING Methods of		(%) Proportion						
Group	Sample Type	Sediment description	MC	JDE.	Logarithmic (φ)	Gravel	Silt/ clay	Sand					
			1 (µm)	2 (µm)				Very coarse	Coarse	Medium	Fine	Very fine	
1a	Bimodal, Poorly Sorted	Slightly Gravelly Sandy Mud	75.4	9.4	1.86	0.10	53.82	0.06	0.01	0.24	9.40	36.37	
2a	Bimodal, Very Poorly Sorted	Slightly Gravelly Muddy Sand	150.9	9.4	2.03	0.17	37.33	0.11	1.06	2.71	30.91	27.71	
3a	Unimodal, Very Poorly Sorted	Slightly Gravelly Muddy Sand	213.4		2.42	3.13	23.97	0.66	6.80	18.30	32.41	14.72	

 Table 14. Particle size distribution groups based on 2014 samples only.



Figure 21. Spatial distribution of particle size groups for sediment samples collected in the high and low abrasion boxes at the Fladen Ground. Sediment groups calculated using 2014 data only.



Figure 22. Proportions (as %) of gravel, sand and silt/clay from PSA samples collected in the high and low abrasion boxes at the Fladen Ground.

The mean values for organic carbon, nitrogen and phosphate and total and inorganic phosphate presented in Table 14, are consistently higher in samples from the High abrasion pressure boxes and significantly higher (p<0.01) for organic carbon, organic nitrogen inorganic phosphate and total phosphate.

Table 15. Mean value, standard deviation (SD) and associated P values of organic carbon, nitrogen and phosphate and total and inorganic phosphate in sediment samples collected from the low and high abrasion boxes at the Fladen Grounds in 2014.

	Low abra	asion	High abra	asion	P value
	Mean value	SD	Mean value	SD	
Organic carbon	0.610	0.0896	0.729	0.0899	<0.01
Organic nitrogen	0.102	0.013	0.112	0.010	<0.01
Organic phosphate	0.006	0.003	0.008	0.003	0.131
Inorganic phosphate	0.040	0.005	0.046	0.002	<0.01
Total phosphate	0.047	0.005	0.054	0.004	<0.01

4.4.3 Infauna assemblages

Infaunal metrics including total number of taxa, total number of species, Hill's diversity and biomass are presented in Figure 23 and show that the total number of individuals and total number of taxa were higher in Low abrasion pressure box samples compared to High abrasion pressure box samples. Variability in number of individuals and abundance within the boxes was determined by calculating the % coefficient of variation (Table 16). The % coefficient of variation was greater in the high abrasion pressure boxes for both metrics. To further explore these patterns between the four surveyed abrasion pressure boxes, a one-way ANOVA was carried out (Table 17). ANOVA was used to test the variability between blocks of a similar categorical nature (i.e. Low vs. Low or High vs. High abrasive pressure), as well as between those that were dissimilar (Low vs. High). The null hypothesis for this test is that there is no difference in the mean infaunal metric being tested between the surveyed abrasion boxes.

Results from this test show that there is a statistical difference in the univariate parameters tested between all four boxes. Total number of taxa (S) between the Low pressure sites (N1A and N1B) was not significantly different from each other and, similarly, total number of individuals (N) between the High pressure sites (N6A and N6B) were not statistically distinct from one another. Tests between the High and Low pressure boxes consistently demonstrated statistically significant differences between the total number of taxa and total number of individuals. However, Hill's diversity did not present a consistently significant differences between the two abrasion pressure categories and no statistically significant differences in biomass were observed between any of the abrasion pressure boxes tested.



Figure 23. Box and whisker plots of the univariate metrics (total number of individuals (N), total number of taxa (S), Hills diversity (N1) and biomass (g)) for grab samples collected High and Low abrasion pressure boxes. Black dots represent outliers.

Table 16. Mean, standard deviation and % coefficient of variation for the univariate metrics 'Number of Taxa' and 'Number of individuals' for each of the abrasion pressure boxes.

				Mean		
	Mean Taxa	SD	%CV	Abundance	SD	%CV
N1A	49	5.142416207	10.49472695	200.9	28.04144	13.95791
N1B	45.3	5.173651193	11.42086356	130.3	36.90845	28.32574
N6A	33.1	5.329165038	16.10019649	93.4	48.37182	51.78996
N6B	25.7	5.10881591	19.87866113	84.8	48.58944	57.29886

Table 17. One-way ANOVA p-value results for univariate response metrics, with green cells representing those tests that were found to be significant at the 1% level. Where; S - Number species, N - abundance, N1 - Hills diversity and B - biomass (g).

	S	Ν	N1	В
N1A vs N1B	0.0194	0.0001	0.0033	0.9762
N1A vs N6A	<0.0001	<0.0001	0.1375	0.7598
N1A vs N6B	<0.0001	<0.0001	0.0107	0.6975
N1B vs N6A	<0.0001	0.0080	0.1112	0.8124
N1B vs N6B	<0.0001	0.0010	<0.0001	0.7063
N6A vs N6B	0.0033	0.4663	0.0002	0.5439

An unpaired t-test was carried out to compare univariate metrics for the two categorical treatments of High and Low abrasive pressure, where n=20 for each treatment (Table 18). Results show a statistically significant difference between treatments for number of species and abundance. All other univariate indices were not significantly different between the High and Low treatments.

Table 18. Unpaired t-test p-value results for univariate metrics, with green cells representing those tests that were found to be significant at the 1% level. Where; S – Number species, N – abundance, N1– Hill diversity and B – biomass (g).

	S	Ν	N1	В
H vs L	<0.0001	<0.0001	0.0234	0.898

Infaunal species present in each of the four abrasion pressure boxes were grouped to major taxonomic group (e.g. crustacea, polychaeta, mollusca) to explore the relative proportions of species in each group and to test the hypothesis that increasing abrasion can lead to a change in the structure of functional groups (e.g. removal of large bivalves and dominance of polychaetes). Despite the higher number of species and abundances in the low abrasion boxes when considering univariate metrics, the relative proportions of species to the major phyla were consistent between all four abrasion boxes. In all four boxes, polychaetes constituted most of the infauna with 54%, 55%, 53% and 53% for N1A, N1B, N6A and N6B respectively. Crustacea contributed 23% and 17% for the Low abrasion boxes and 22% for both High abrasion boxes and molluscs contributed 14% and 15% for Low and 11% and 14% for High.

Multivariate analysis of infaunal communities between abrasion pressure boxes was also explored. Figure 24 shows a clear difference between the infaunal communities found in the Low abrasion boxes and those found in the High abrasion boxes. However, the replicate boxes within the High and Low abrasion pressure areas also cluster together. SIMPER analysis revealed high within-treatment (High/Low abrasion) variability (average similarity for samples within the Low abrasion pressure boxes = 51.58%, and High abrasion pressure boxes = 56.32%). Average dissimilarity between the two treatments was 71.42% with 25 species accounting for 50% of the dissimilarity and 106 species accounting for 90% of the dissimilarity.

Benthic community variability within the pressure boxes was calculated on the Bray-Curtis resemblance matrix using the Multivariate dispersion (MVDISP) routine in Primer. This routine calculates the dispersion of samples within each treatment group, with larger values indicating greater within-group dispersion or variability (Table 19).

 Table 19. Index of multivariate dispersion calculations indicating community variability with the Low and High abrasion pressure boxes.

Pressure box	Dispersion value
N1A	1.01
N1B	1.497
N6A	0.841
N6B	0.652

The results show that communities within the Low abrasion pressure boxes show higher variability than communities within the High abrasion pressure boxes.

The polychaete worms, *Galathowenia oculata, Eclysippe vanelli, Paramphinome jeffreysii* and *Diplocirrus glaucus,* along with the bivalve mollusc *Mendicula ferruginosa* were present in high abundances within the Low abrasion pressure boxes, whilst the bivalve mollusc *Thyasira equalis,* cumacean *Leucon nasica* and the polychaete worm *Heteromastus filiformis* were present in higher abundances in the High abrasion pressure boxes. Thirty-five species contributing to the dissimilarity between treatments were absent from the High abrasion. The full SIMPER results for the dissimilarity between treatments are presented in Appendix 3.





4.4.4 Biological traits analysis: Functional composition

The relative abundance of each of the eight trait categories is presented in Figure 27 to Figure 33. Differences in trait composition were apparent for some but not all traits between the High and Low abrasion pressure boxes. When weighted by abundance (Figure 25),

maximum size (Figure 26), feeding type (Figure 29), flexibility (Figure 30) and living habit (Figure 32) showed the most notable differences between abrasion categories.

These apparent differences were primarily due to significantly higher abundances of the polychaetes Paramphinome jeffreysii, Diplocirrus glaucus, Galathowenia oculata and Eclysippe vanelli within the Low abrasion pressure boxes. The combined abundances of these four species account for 43% of the total abundance within the Low abrasion pressure boxes compared to only 4% of the total abundance in the High abrasion pressure boxes. These four species are classed as highly flexible, highly fragile, living within the top 5cm of sediment, having a life-span of between 1 and 10 years and growing to a maximum size of between 11 and 50mm. Their feeding modes and living habits differ, but none are considered subsurface deposit feeders or burrow dwellers. G. oculata and E. vanelli are tube-dwellers, accounting for over 60% of the total abundance attributed to this modality within the Low abrasion pressure boxes. Despite this dominance the remaining 40% still account for twice the number of tube-building individuals sampled from within the High abrasion pressure boxes. Absolute abundances of burrow dwellers and subsurface deposit feeders were higher within the High abrasion pressure boxes in comparison with the Low abrasion pressure boxes. Significantly higher abundances of the brittle star Amphiura chiajei and the polychaete Heteromastus filiformis within the High abrasion pressure boxes were primarily accountable for the higher total abundance of burrow-dwellers. Whilst H.filiformis and Thyasira equalis were primarily responsible for the higher abundances of sub-surface deposit feeders.

No clear differences in trait composition were apparent between Low and High abrasion pressure areas when the traits matrix was weighted by biomass due to the presence of one or two heavy organisms which dominated the biomass at some stations (Appendix 3).



Figure 25. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for position in the sediment.



Figure 26. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for maximum size.



Figure 27. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for mobility.



Figure 28. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for longevity.



Figure 29. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for feeding type.



Figure 30. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for flexibility.



Figure 31. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for fragility.



Figure 32. Trait modality proportions weighted by abundance for low abrasion cells and high abrasion cells for living habit.

4.4.5 Biological traits analysis: Community vulnerability

Taxa were classified into four of the five functional indicator groups using the eight trait approach (Figure 33). None of the taxa were classified into G1, which represents the lowest vulnerability to trawling. The majority of taxa were classified as moderately vulnerable to trawling (G3) within all abrasion pressure boxes. Within the Low abrasion pressure boxes, G3 accounted for 74-78% of taxa, whereas within the High abrasion pressure boxes 69-71% of all taxa were classified within this group. Using eight traits, *Pennatula phosphorea* was the only taxon classified as the most vulnerable to trawling (G5) within the Low abrasion pressure boxes. This species was absent from both highly abraded boxes. Group 4 was represented by a higher percentage of taxa (8-14%) in the High abrasion pressure boxes than in the Low abrasion pressure boxes (4-6%) due to the classification of the brittlestar *Amphiura chiajei* within this group. Total abundance of this species within the High abrasion pressure boxes.

All functional indicator groups were represented using the five trait approach (Figure 34). G1 and G2, accounted for over 50% of the relative abundance in the Low abrasion pressure boxes and over 30% in the High abrasion pressure boxes. G3 was represented by 37-40% of taxa within the Low abrasion pressure boxes and 51-62% of taxa within the High abrasion pressure boxes. The percentage of taxa classified as highly vulnerable to trawling (G4) was slightly higher for the High abrasion pressure boxes (~2%) in comparison with the Low (~1%). Polychaetes dominated G1-3, whereas crustaceans (amphipods and cumaceans) were found only in G1 and G2. With the exception of *Cuspidaria obesa, Nucula sulcata* (classified into G2) and *Tellimya ferruginosa* (classified into G4), bivalves were classified as moderately vulnerable to trawling (G3), whereas gastropods (with the exception of *Philine*) were allocated to G2. In addition to *P. phosphorea*, individuals identified only as Cnidaria

were also classified as having the highest vulnerability to trawling using the five trait approach. Both of these taxa were absent from the High abrasion pressure boxes.

Taxa with the highest vulnerability to trawling accounted for only a small percentage of the total abundance using either the five or eight trait approach. Sabellid worms (e.g. *Euchone, Perkinsiana rubra*), echinoderms (e.g. *Brissopsis lyrifera* and *Echinocardium flavescens*) and the tube anemone *Cerianthus lloydii* were classified as highly vulnerable to trawling using both methods. *Euchone* and *P. rubra* were each present only as a single occurrence at stations N1A10 and N1B03 respectively within the Low abrasion pressure boxes (sediment group 1a). *B. lyrifera* and *C. lloydii* were present in low but comparable abundances within High and Low abrasion pressure boxes (all except one station was allocated to sediment group 1a), whereas *E. flavescens* was present only within the High abrasion pressure boxes (all four stations allocated to sediment group 1a).



Pressure box

Figure 33. Relative abundance of each functional indicator groups (based on 8 traits) within boxes subjected to High (N6A &B) or Low abrasion pressure (N1A &B).



Figure 34. Relative abundance of each functional indicator groups (based on 5 traits) within boxes subjected to High (N6A &B) or Low abrasion pressure (N1A &B).

4.4.6 Bivalve size frequency distribution

Maximum bivalve measurements (mm) and biomass (g wwt) for all species in 2013 and 2014 samples were collated to identify which species were suitable for size frequency distribution analysis (Table 21). Only species with at least 40 individuals (n = 5) were used for size frequency distribution plots due to the requirement for a good representation of measurements across the size range: *Abra nitida, Arctica islandica, Axinulus croulinensis, Mendicula ferruginosa* and *Thyasira equalis* (Figure 35). Most individuals of *A. nitida, A. croulinensis, M. ferruginosa* and *T. equalis* were generally in the middle of the observed size range. *A. islandica,* however, has a greater number of individuals in the smaller size class of less than 5mm as well as a second peak in the largest size class of greater than 20mm.

Comparisons of species presence and absence in grab samples from the High and Low abrasion pressure boxes revealed that four species; *Ennucula tenuis*, *Nucula sulcata*, *Tellimya ferruginosa* and *Timoclea ovata* were present in grab samples collected from the Low abrasion pressure boxes but absent from samples collected from the High abrasion pressure boxes. Where a species was found in both High and Low abrasion pressure samples, a series of measurements including number of individuals measured, mean maximum valve length (mm), standard deviation of maximum valve length, range of valve length observed, mean biomass (g wwt), and the standard deviation of biomass were calculated (Table 18). In addition, these metrics were also calculated for all individuals measured across the Fladen Grounds (including 2013 and 2014 but excluding the pressure gradient samples) to provide a whole site baseline.

	Total No. individuals	No. Stations			Abra	sion
Bivalve speceis	measured	present	2013	2014	Low	High
Abra nitida	78	57	✓	\checkmark	\checkmark	✓
Arctica islandica	42	29	✓	✓	✓	✓
Astarte sulcata	26	17	✓	✓	Х	Х
Axinulus croulinensis	406	122	✓	✓	✓	✓
Decipula tenella	11	8	✓	х	Х	Х
Ennucula tenuis*	14	12	х	\checkmark	✓	Х
Jupiteria minuta	14	7	✓	х	Х	Х
Mendicula ferruginosa	836	143	✓	✓	✓	✓
Nucula sulcata *	20	13	х	\checkmark	✓	Х
Nuculana minuta	11	9	х	\checkmark	Х	Х
Tellimya ferruginosa *	75	37	х	\checkmark	✓	Х
Thyasira equalis	843	120	✓	✓	✓	✓
Thyasira obsoleta	34	22	✓	✓	Х	Х
Timoclea ovata*	15	15	✓	✓	✓	Х

Table 20. Bivalve species present in grab samples. (Bold denotes species taken forward for further analysis in relation to abrasion pressure, * denotes species which were present in Low abrasion pressure samples but not High abrasion pressure samples).



Figure 35. Size frequency distribution plots using the maximum value length measurement (mm 2 d. P.) for bivalve species present in grab samples from the 2013 and 2014 surveys of the Fladen Grounds (only species with at least 40 individual measurements are presented).

Table 21. Comparison of bivalve size metrics from all individuals measured across the Fladen Grounds (including 2013 and 2014 but excluding the pressure gradient samples) and individuals from both the High and Low abrasion pressure boxes. Green shading denotes significant differences in valve measurements when comparing samples from High and Low abrasion pressure boxes.

Species	Sample area	Mean valve length (mm)	Std dev valve length (mm)	Range valve length (mm) Low to High	Mean biomass (g wwt)	Std dev biomass (g wwt)
Abra nitida	All	5.8670	0.0417	2.74, 12.07	0.0195	0.0417
	High	10.5567	0.0336	9.1, 12.6	0.0736	0.0336
	Low	4.8464	0.0149	2.8, 9	0.0118	0.0149
Arctica	All	6.7172	14.2662	1.44, 81.73	2.3424	14.2662
islandica	High	1.8150			0.0014	
	Low	1.8750			0.0016	
Axinulus	All	1.5860	0.0009	1.12, 3.9	0.0005	0.0009
croulinensis	High	1.4493	0.0012	1.2, 1.95	0.0006	0.0012
	Low	1.5007	0.0004	1.3, 1.75	0.0004	0.0004
Mendicula	All	2.0473	0.0016	1.39, 3.27	0.0028	0.0016
ferruginosa	High	1.8894	0.0011	1.4, 2.3	0.0021	0.0011
	Low	2.0139	0.0014	1.4, 2.5	0.0030	0.0014
Thyasira	All	3.9000	0.0051	0.23, 8.4	0.0078	0.0051
equalis	High	2.5849	0.0038	1.25, 4.4	0.0045	0.0038
	Low	1.7727	0.0003	1.72, 2.1	0.0003	0.0003

Results from the t-test were used to test the null hypothesis that the mean of maximum valve length measured for a species in both the High- or Low-abrasion pressure samples were not significantly different. Only *M. ferruginosa* had significantly larger individuals in the Low abrasion pressure samples when compared with those measured in the High-abrasion pressure samples (P value = 0.0018). No significant difference in valve measurements was found for either *A. croulinensis* (P value = 0.226) or *T. equalis* (P value = 1.656) when comparing High- and Low-abrasion pressure boxes, however, samples from the High boxes had significantly larger individuals of *A. nitida* (P value = 0.0091) than those measured in the Low boxes.

4.4.7 Epifauna community analysis

Univariate metrics (Abundance, number of species and Hills diversity) for each of the eight epifaunal sample stations was plotted against the four potential pressure scores (calculated as per Section 3.2.1). Results from each of the four linear regressions are presented in Table 22 for the coefficient of determination (R^2) and associated p-values. All R^2 values were calculated using the Pearson correlation co-efficient. Total number of individuals (S), total number of species (N) and Hills diversity (N1) are presented in Figure 36.

In light of the additional sampling points for epifaunal communities along the abrasion pressure gradient, it was possible to conduct a linear regression to explore univariate metric responses of epifaunal communities to different levels of abrasive pressure. Linear regression demonstrated that the identified pressure gradient at the Fladen Grounds was a poor fit for the prediction of number of species and the relationship was not statistically significant at the 1% level. Though predictions were better for abundance than for number of species, and also statistically significant, the R² values were still low and cannot be presumed a good fit. Hills diversity appears to present a much better fit, with R² values over 0.55 in all cases, and was statistically significant at the 1% level. Diversity measures demonstrated a positive relationship with abrasion, except for abundance which had a negative relationship with increasing abrasive pressure. The positive relationship between

Hill number and abrasion demonstrates an assemblage moving closer towards a composition whereby no one species dominates the total abundance.



Figure 36. Box and whisker plots of the univariate metrics (total number of individuals (N), total number of taxa (S) and Hills diversity (N1) for epifaunal assemblages identified from video tows collected in High (N6A, N6B), Low (N1A, N1B) and Intermediate (1B, 2A, 6A, 7B) abrasion pressure boxes. Black dots represent outliers.

Table 22. R^2 and p-values for linear regression of epifaunal univariate response metrics against the four variations of abrasion scoring. Where: swa1214 – combined VMS for the years 2012-14, swa1314 - combined VMS for the years 2013-14, swasub1214- combined VMS for subsurface gears only for the years 2012-14 and swasub1314- – combined VMS for subsurface gears only for the years 2013-14

	S		Ν		N1	
	R^2	p-value	R^2	p-value	R ²	p-value
swa1214	0.0083	0.4228	0.1674	0.0002	0.5538	<0.0001
swa1314	0.0067	0.4714	0.1875	0.0001	0.6031	<0.0001
swasub1214	0.0010	0.7859	0.2087	<0.0001	0.5942	<0.0001
swasub1314	0.0009	0.7961	0.2159	<0.0001	0.6083	<0.0001

Number of individuals of two seapen species, *Pennatula phosphorea* and *Virgularia mirabilis* were calculated from video tow data. Seapen abundance was normalised per tow and also converted to a density score, number of seapens per m². Both species were plotted against linear regression models to explore whether increasing abrasive pressure was affecting species abundance and density values.

Figure 37 shows the regression outputs for *P. phosphorea* density per m^2 . The density of *P. phosphorea* decreased with increasing abrasion pressure, a relationship that demonstrates a relatively good model fit (R^2 values around 0.4 across the four abrasion pressure scores). It should be noted that although this relationship is statistically significant, only 40% of the variance is explained by the regression (Table 23). It should also be noted that when using multiple years worth of VMS data there was a positive influence on the model fit to the data.

Figure 38 shows the regression outputs for *V. mirabilis* density per m^2 . The density of *V. mirabilis* was found to increase with higher abrasion pressure in a positive relationship. However, the low R^2 values suggest that the linear model is a poor representation of the environmental conditions influencing the *V. mirabilis* community (Table 23). Similar to *P. phosphorea*, increasing the time period over which abrasion data was collated had a positive influence on the model's fit to the *V. mirabilis* data.

	<i>P. phosphorea</i> density per m ²				V. mirabilis density per m			
	DF	R^2	F-stat	P value	DF	R^2	F-stat	P value
swa1214	78	0.4537	64.78	7.58E-12	78	0.1525	14.03	0.000342
swa1314	78	0.4114	54.53	1.45E-10	78	0.08173	6.943	0.01015
swasub1214	78	0.4547	65.03	7.07E-12	78	0.09425	8.117	0.005606
swasub1314	78	0.389	49.66	6.42E-10	78	0.03988	3.24	0.07572

 Table 23. R² values and P values associated with linear regression models for density per m² of P. phosphorea and V. mirabilis calculated from video tows.

4.4.8 Nephrops burrow distribution and density

Video tow data were also analysed for the number of *Nephrops* burrows per tow. As for seapens, the abundance of *Nephrops* burrows was normalised per tow, and a density value was calculated to show number of *Nephrops* burrows per m² (Figure 39) and associated R² values and P values (Table 24). There is no significant relationship between *Nephrops* burrow density and abrasive pressure.

		Nephrops burrow density per m ²					
	DF	R^2	F-stat	P value			
swa1214	77	0.006294	0.4877	0.487			
swa1314	77	0.008392	0.6517	0.422			
swasub1214	77	0.004036	0.3121	0.578			
swasub1314	77	0.006291	0.4875	0.4872			

Table 24. R² values and P values associated with linear regression models for density per m² of *Nephrops* burrows calculated from video tows.



Figure 37. Linear regression for *P. phosphorea* density per m² calculated from video tows.



Figure 38. Linear regression for *V. mirabilis* density per m² calculated from video tows.



Figure 39. Linear regression for *Nephrops* burrow density per m² calculated from video tows.

4.5 Discussion

To assess the possible effects of abrasion pressure on habitats and their communities, there is a requirement to provide accurate data on the distribution and intensity of fishing activities associated with the pressure (Denderen *et al* 2014). In this study, fishing effort was assigned to the different areas of the Fladen Grounds by aggregating VMS 'pings' to a regular grid as described in the JNCC method (Church *et al* 2015) to produce an effort score (abrasion value in this instance). Using the scores for each grid cell, abrasive pressure across the Fladen Grounds was assessed and found to be variable with effort occurring in different areas from year to year. While the Central Fladen NCMPA is positioned in an area of relatively high abrasion pressure in 2010, 2012 and 2013, the south east extent of the NCMPA site is subject to a relatively low fishing effort when compared with the rest of the MPA. Fishing effort in this instance has been expressed as annual averages and does not take into consideration seasonal variability in fishing activity highlighting some of the difficulties encountered when attempting to make links between pressure and state. This may be an important limitation to consider when attempting to link fishing activity with biological processes which often exhibit a strong seasonal variability (Denderen *et al* 2014).

The 'effort scores' were also used to inform the selection of abrasion pressure boxes for testing the possible effects of fishing activity on habitats and benthic communities. However, the Church *et al* (2015) method assumes the even distribution of fishing effort within a given grid cell to produce the effort score which is subsequently assigned to any sampling unit that falls within that given cell. The assumption that effort is homogeneously distributed in all instances is not necessarily a true one and effort can be localised within cells due to varying habitat types, obstructions and depth. It is possible that this could lead to increased variability within sample blocks and as a result, the true effects of fishing activity on faunal communities may be masked. Therefore, when allocating fishing effort values for analytical purposes in the future, it is essential that the variability in fishing effort within a cell is considered during the interpretation of results.

4.5.1 Environmental characteristics

The continual scraping and re-suspension of surface sediments caused by trawling has been linked to an increase in the silt/ clay fraction in the superficial layers of the seabed (Jennings and Kaiser 1998; Trimmer *et al* 2005). Indeed, the present study found that samples collected in the High-abrasion pressure boxes exhibited slightly higher silt/clay fractions when compared to sediment samples from the Low abrasion boxes. However, the boxes surveyed were spatially separated, so it is not possible to conclude here if those differences are indicative of trawling activity or are the result of the presence of different habitats in the different abrasion pressure boxes.

While modifications to particle size distribution of sediments is reasonably well understood, the impacts of trawling on sediment biogeochemistry are still relatively unknown. Trimmer *et al* (2005) measured a number of sediment characteristics including: organic matter (OM) and sediment metabolism, oxygen uptake, denitrification, sulphate reduction and sediment-water nutrient exchange, along gradients of trawling activity in the southern North Sea. The authors found the biogeochemical processes in the upper layers of sediment, both oxic and suboxic, seemed unaffected by trawling in the long-term. However, Jennings *et al* (2001b) suggested that re-suspension of sediment following trawling could lead to nutrient release and enhancement of primary production over a large scale. In this study, organic carbon and nitrogen and inorganic phosphate and total phosphate content were higher in High abrasion when compared to the Low abrasion pressure boxes. A robust interpretation of these results is not possible in the absence of higher resolution data on when the area was last impacted and therefore if the nature of the disturbance is acute or chronic. An experimental approach to better understand the response to both acute and chronic disturbance on biogeochemical

processes would provide a context against which point samples of biota could then be compared. This may elucidate if it is the action of trawling that is disturbing the Redox Potential Discontinuity directly or if it is the removal of biota which is having an effect.

4.5.2 Infaunal assemblage metrics

Due to the inclement weather on survey, analysis of variance based on univariate metrics of infauna was only possible for two abrasion pressure categories (High and Low). As a result of this reduced sampling effort and spatial spread of stations, it was not feasible to draw broad conclusions on how infaunal community univariate metrics were responding to increasing abrasive pressure across the whole site. However, an unpaired T-test between the high and low abrasion treatments demonstrated significant differences between them. Tests between the categories demonstrated statistical differences in number of species and number of individuals, with lower values of these metrics observed from the high pressure areas. These results support the findings by Hinz *et al* (2009) that reported reduced infaunal abundances and species richness with increasing otter trawling activity at a *Nephrops* fishing ground in the Irish Sea. Some studies have also reported a community shift towards one dominated by polychaetes (Jennings *et al* 2001a). However, it was found that polychaete richness and abundances were higher within the Low abrasion pressure box and that when considering the community as a whole, both the low and high abrasions boxes exhibit the same relative proportions of major phyla.

Variability within univariate metrics for the Low and High pressure boxes was also observed, with species number and abundance showing greater variability within the High-abrasion pressure boxes. However, community variability, as assessed through multivariate analyses, showed increased variability within the Low-abrasion pressure boxes. Multivariate analysis also revealed significant differences in community composition between samples taken from within the high and low abrasion boxes. Thirty five species (contributing to the dissimilarity between treatments) were absent from samples taken from within the High-abrasion boxes compared to five from within the Low-abrasion boxes. The significant reduction and increased variability in species richness and abundances within the High-abrasion box are consistent with what is expected from trawling pressure impacts. However, additional replicates of high and low pressure boxes would give greater confidence to these findings by increasing the power to detect differences between the two pressure categories, and to also increase our understanding of the sources of variability at this site.

4.5.3 Biological traits analysis: Community function and vulnerability

Differences in trait composition were observed for communities subjected to Low- and Highabrasion pressure. These differences were primarily due to significantly elevated abundances of four polychaete species within the Low-abrasion pressure boxes. Increased abundances of polychaetes have previously been attributed to high levels of disturbance but also to higher levels of fine-grained sediment (Bergman & van Santbrink 2000; Kaiser et al 2000), however Jennings et al 2001b reported decreases in polychaete biomass at high levels of disturbance, whilst at moderate levels, polychaete abundances proliferated. The decreases seen by Jennings et al 2001b, were attributed to unsustainable rates of direct mortality from contact with the gear or indirect mortality from exposure to predators or scavengers. Within the abrasion pressure boxes at the Fladen grounds, abundances of tubedwellers were significantly lower within the high abrasion boxes. Kaiser and Spencer (1996) found that tube-building worms were amongst the most sensitive taxa affected by trawl disturbance. Trawl disturbance has also been linked to increases in predators/scavengers, however higher abundances of predator/scavengers were associated with the Low-abrasion pressure boxes primarily due to high abundances of Paramphinome jeffreysii. The significant reduction in the abundance of four polychaete species within the highly abraded boxes,

together with an increase in burrow dwelling individuals indicates the communities are functioning differently to those subjected to lower abrasion pressure. When combined with the infaunal assemblage results our findings support those of Kaiser and Jennings, 1996, who found a reduction in tube-dwelling organisms and those that live on or within the surface sediments in areas subjected to physical disturbance.

When weighted by biomass, trait composition was highly variable both within and between the different levels of fishing pressure. Large species, such as *Echinocardium flavescens* and *Brissopsis lyrifera* tended to dominate the biomass in some samples (both in the Highand Low-abrasion pressure boxes), even when only one individual was present. Larger organisms that occupy the top most layers of the sediment would be more appropriately sampled using scientific trawls and their trait composition will be better represented using this method.

The choice and number of traits used to assess the vulnerability of benthic communities to trawling has a significant effect on the resulting functional indicator groups. When using eight traits, Group 1 (lowest vulnerability to trawling) was completely absent from the data. The majority of species were also allocated to one group (G3) Indicating the potential sensitivity of species to trawling pressure. When assumptions were made concerning the ability of some species to regenerate or that vermiform species are likely to be unaffected by trawling despite their fragile structure (de Juan and Demestre 2012), some species were re-classified as having low vulnerability to trawling and are represented in both the high- and low-fishing pressure areas. Both methods highlight that infaunal communities within both the High- and Low-abrasion boxes are primarily composed of low to moderately vulnerable taxa with only minor representation of highly vulnerable species.

4.5.4 Bivalve size structure

Bivalve size structure was explored as one of OSPAR's candidate MSFD indicators for physical disturbance or hypoxia as large and long-lived species often play key roles in the functioning of the benthic communities (Darr et al 2013). In the current study, the presence, abundance and size of infaunal bivalves were explored to assess their potential as a metric for detecting the effects of increasing abrasion pressure. Four species, E. tenuis, N. sulcata, T. ferruginosa and T. ovata were present in Low-abrasion samples but absent from Highabrasion samples. These are small species, but described as solid in structure and are associated with sandy or muddy sand sediments. Thyasira equalis was the only species found in higher numbers in grab samples taken from within the High-abrasion boxes. The average size of individuals of this species was also, on average, larger in samples taken from within the high abrasion boxes (max. valve length of 2.01mm) than in those from the Low-abrasion boxes (max. valve length of 1.8mm), although not significantly so. Thy as iridae are known for their extensive burrowing behaviour and *T. equalis* in particular has been found to thrive in deeper waters preferring sediments with low organic content (Rozemarijn et al 2011). Its ability to suspension feed and obtain nutrients from chemo-autotrophic bacteria in the gills may also provide an ecological advantage in the dynamic environment associated with higher abrasion pressure which could explain high numbers of the species.

Maximum lengths of all other bivalve species selected for size structure analysis had equally small maximum lengths. Small bivalves, occupying the surface layers of the sediment are most likely to be washed away by pressure waves caused by fishing gear with the potential for re-establishment upon settlement out of the wave. The only species present within the study area large enough to be impacted by fishing gear, was the ocean quahog, *Arctica islandica*. Maximum valve length recorded for this species was 81mm, however only one individual was found in a sample collected from within the high abrasion area and one in the low abrasion boxes. Both of these individuals measured less than 2mm in length. Bivalves used to test this metric were collected using a Day grab which provides a point sample

covering an area of 0.1m² (to a depth of approximately 15cm). In order to effectively quantify the abundance and size of any large and long-lived species which are likely to be found in low abundances, a grab sample is not the most appropriate gear. This is supported by a study by Witbaard & Bergman (2003) in which a Triple D dredge revealed densities of *A. islandica* peaking at 28,600 individuals per 100m² at the Fladen Grounds. Therefore, to further test if this indicator can show a change in relation to physical abrasion pressure, other methods of data collection, designed for sampling the larger, more sparsely distributed fauna need to be explored including exploring the posibibility of sampling the bycatch from commercial trawlers.

4.5.5 Epifaunal assemblage metrics

The linear regression analysis shows that epifaunal communities are being modified by abrasive pressure in some univariate metrics but not others. Number of species did not show a significant relationship with increasing abrasive pressure and the linear model was a poor fit. However, other community metrics did show a significant relationship with reasonable model fits to a linear regression. In general, increasing abrasive pressure appears to be reducing the number of epifaunal individuals whilst increasing species diversity. Hinz *et al* (2009) found that epifaunal biomass was unaffected by increasing trawling activity but that the catch was dominated by large specimens of *Asterias rubens* that were responding to an increase in food availability. Further work to investigate not only the total number and abundance of epifaunal species, but also the biomass of individual species, would provide additional information on epifaunal community structure and function. The collection of epifaunal samples using a beam trawl would permit this additional quantitative analyses to be carried out. Scientific beam trawl samples also provide a sampling gear that operates at an appropriate scale to study the effects of fishing (over 10s of m²) by integrating the patch effects that are unavoidable with point sampling (Jennings *et al* 2001a).

4.5.6 Seapen abundance and density

As part of the epifaunal data analysis it was possible to explore how seapen abundance and density varied across the identified pressure gradient. Physical disturbance from abrasion pressure may have modifying effects on seapen distribution and as a result, they have been identified as an important indicator for the quality of mud habitats and their associated communities (Macdonald *et al* 1996).

Linear regression analysis of Pennatula phosphorea and Virgularia mirabilis against the different pressure levels produced conflicting results. P. phosphorea demonstrated a negative relationship with increasing abrasive pressure whilst V. mirabilis showed no significant relationship. P. phosphorea is a smaller species, approximately 40cm in length, and more compact than the whip-like V. Mirabilis, which grows to around 60cm (Hayward and Ryland 1990). P. phosphorea is also only capable of partially retracting back into the sediment, unlike V. mirabilis, which has the ability to fully retract into the sediment using its large muscular peduncle (Hoare and Wilson, 1977 cited in Greathead et al., 2014). P. phosphorea has also been found to be less tolerant of sediment variability (Jones et al 2000). It is possible, therefore, that V. miribilis is more resilient to community level impacts caused by habitat disturbance, either by its ability to retract into the sediment, which limits its exposure to damage, or by its ability to populate a wider ecological niche in terms of sediment characteristics. This is supported by a recent study by Greathead et al (2014) where authors found that V. miribilis displayed the widest population distribution in modelled extents and was more tolerant to a broader range of environmental conditions than P. phosphorea or F. quadrangularis. A greater understanding of how fishing pressure affects the distribution of the three species of seapen is required. While Troffe et al (2005) found that seapens had a low catchability in beam trawls, a targeted study to explore the impact of
fishing on the three species of seapen would provide robust evidence to understand how the morphological and functional characteristics of *P. phosphorea* and *V. mirabilis* are affected by trawling and which species may be most appropriate as a biological indicator for burrowed mud habitats. However, the current analysis suggests that the density of the seapen *P. phosphorea* at the Fladen Grounds is a good indicator of the impacts associated with increasing abrasive pressure.

4.5.7 Nephrops burrow density and distribution

Densities of *Nephrops* burrows per m² were calculated from video tows collected in 2014. In addition, average burrow density data collected by Marine Scotland Science for *Nephrops* fisheries stock assessments during 2008-2010 were provided by JNCC. The highest densities of *Nephrops* burrows reached 0.31 per m² or 66 burrows in a 211m video tow, recorded in 2014. Analyses carried out to assess if there was any relationship between abrasive pressure and number of *Nephrops* burrows showed no relationship between level of abrasive pressure and number of burrows. As discussed with a number of the other metrics, it is possible that the way the pressure scores were allocated to the individual grid cells or the lack of temporal resolution of fishing effort within the region may mask a trend and thus this metric cannot be fully defined here. Consideration must also be given to the sediments present and their suitability for *Nephrops* and other burrowing megafauna.

4.6 Summary and conclusions

The results presented in this section describe the environmental and biological characteristics of benthic communities in areas subjected to different levels of abrasion pressure. Those under higher abrasion pressure exhibited a greater proportion of the silt/ clay fraction with increased organic carbon, organic nitrogen, total phosphate and inorganic phosphate contents. Reduced species numbers and lower abundances of both infauna and epifauna species were found in High-abrasion pressure samples. Species which have been considered vulnerable to trawling were particularly affected. Significantly reduced abundances of tube dwellers and the seapen *P. phosphorea* were most affected by increased abrasion. Surprisingly, *V. mirabilis* was found in increasing numbers and densities of *Nephrops* burrows were seemingly unaffected in areas of higher abrasion.

Distribution and abundances of seapens observed in video tows was a key component driving differences in epifaunal assemblages. Seapens are conspicuous and easily identified using video techniques. However, the response to abrasion from the remaining epifaunal assemblage was less clear from video tow data. Use of scientific beam trawls could provide a better indication of impacts on species diversity and changes in the biomass and size composition of key functional groups. For example, biomass based biological trait analysis would provide data on the presence of opportunist, mobile scavengers which may populate a recently disturbed area following a disturbance.

Infaunal and epifaunal assemblages were richer both in terms of the number of species present and their abundances in Low abrasion pressure boxes. Interestingly, infauna in both treatments was dominated by four polychaete species. Proliferation of smaller infaunal species has been previously attributed to moderate levels of trawling (Jennings *et al* 2001b). Species with life cycles that proliferate quickly, particularly those classified as meiofaunal, are of future interest as they have the capacity to rapidly populate trawled areas and process the carbon and nitrogen released following re-suspension.

The analyses conducted in this study have highlighted some interesting relationships between the infauna and abrasive pressure, however, it is clear that how the faunal assemblage composition responds to changes in abrasive pressure is not as straightforward as initially anticipated. Continued investigation would enable such relationships to be more clearly understood.

5 Type 3 monitoring - BACI study within the south east extent of the Central Fladen NCMPA

5.1 Introduction

The primary objective of the Type 3 monitoring survey design was to establish a 'Before' time point of a Before-After-Control-Impact (BACI) experimental design. The area chosen for this part of the survey was located in the south east extent of the Central Fladen NCMPA as one of the proposed areas of the NCMPA for management measures. In order to assess the effectiveness of any measures put in place, samples were taken from inside and outside the proposed closed area. These samples represent the "Before" time point of the BACI design and are intended to detect any changes over time between a "Control" area that would remain fished and the 'Impact' area where fishing is likely to cease. In this context, 'Impact' refers to the effect that a management scenario or closure would have on the seabed habitat and associated benthic species and does not reflect any assumption about the direction of change. 'Control' and 'Impact' sampling will allow any effects of fisheries management actions to be discerned from natural variability, stochastic events, and underlying trends in the larger area.

The abrasion pressure gradient study in Section 3 identified and tested a series of metrics for detecting a biological response to increasing abrasion effort. In this section, the tested metrics have been used to address the objectives of the BACI paired design and provide the 'Before' dataset on which to base any subsequent monitoring. In addition, spatially relevant data points from the 2013 JNCC-Cefas survey (Eggleton *et al* 2013) are also used in the analysis to provide a between year comparison of infaunal and epifaunal assemblages and to begin integrating data from multiple years.

5.2 Methods

5.2.1 Survey design

Power analyses were conducted on infaunal abundance data collected from the 2013 JNCC-Cefas survey at the Fladen Grounds to inform the design of the BACI survey. These analyses identified that 40 samples per treatment would achieve an acceptable level of power (>80%) to achieve statistical significance in detecting a 30% change in species number, and a 40% change in species abundance. Collecting more samples to detect lower levels of change (i.e. 10% or 20%) was not considered feasible given the time available and competing sampling priorities, as the power analysis results indicated that many more samples would be required to confidently increase detection of change. Therefore, 40 sample stations, which were restricted to mud and sandy mud habitats as predicted by a modelled sediment map (EU Seamap), were assigned randomly within each treatment; Control area and Impact area (impact area defined as the area under a management scenario) (Figure 40).



Figure 40. The Before-After-Control-Impact (BACI) survey design showing the location of all 80 target sampling stations (40 stations per treatment) at the Central Fladen NCMPA.

5.2.2 Sample acquisition and processing

The survey was conducted from the RV *Cefas Endeavour* during the 2014 survey of the Fladen Grounds (survey code: CEND05/14). Technical specifications of the survey equipment used are given in the cruise report (McIlwaine 2015). In total, 36 Day grab samples were collected from within the Impact area. The remaining four samples within this area were acquired using a mini-Hamon grab for macrofauna and PSA and a Shipek grab for sediment geochemistry samples due to the coarser nature of the sediment at these stations. Thirty-nine of the 40 samples from the Control area were collected using the Day grab and only one required the combination of mini-Hamon grab and Shipek grab. In addition, video tows at each of the 80 stations were collected using the camera sledge. Detailed sample acquisition methods are provided in Appendix 1.

Sediment samples from the 80 grab deployments were analysed for particle size distribution (PSD) and sediment organics as detailed in Section 3.2.1. Entropy non-hierarchical clustering (Stewart *et al* 2009) was used to group samples based on PSD from the BACI study with those collected from the abrasion pressure sediment samples to provide sediment groupings for the 2014 dataset. Infaunal processing of grab samples and epifaunal processing of video and stills were completed as described in Section 4.3.4.

5.2.3 Data analysis

Univariate metrics calculated for each sample included total number of individuals (N), total number of taxa (S), Hill's (1973) taxon diversity index (N1) and total wet weight biomass (B) (biomass was calculated for infauna data only). Boxplots were produced in SigmaPlot[®] for each of the metrics. When analysing epifaunal assemblages between years, presence and absence data were used. Analysis of epifauna data from 2014 only was conducted using a

species abundance matrix as the level of species identification within a sampling year was consistent and therefore comparable. The non-parametric Mann-Whitney U test was used to test each of the univariate metrics, both between years (2013 and 2014) and between samples in the Control area and Impact area when using 2014 data only.

5.3 Results

5.3.1 Distribution of environmental characteristics

All three of the sediment groupings identified when categorising 2014 PSD data were present across the south east extent of the Central Fladen NCMPA (Figure 41). However, sediment group 1a (slightly gravelly sandy mud) was present only in three discrete patches within the Control area. The Impact area, which lies within the boundary of the NCMPA, was found to be coarser in nature comprising of groups 2a, very poorly sorted slightly gravelly muddy sand with a bimodal distribution and group 3a, which is very poorly sorted slightly gravelly muddy sand but with a unimodal distribution. Figure 42 shows the relative proportions of the major sediment fractions (gravel, sand and silt/clay) for each station located within the Control area and Impact area. Gravel was present only in the centre of the Impact area corresponding with group 3a. Group 1a sediment samples located in three patches within the Control area also exhibited the highest proportion of silt/ clay with 50% or more of the total sample.



Figure 41. Spatial distribution of particle size distribution groups for sediment samples collected in the Control area and Impact area at the Fladen Grounds. Sediment groups calculated using 2014 data only.



Figure 42. Proportions (as %) of gravel, sand and silt/clay from PSA samples collected in the Control area and Impact area at the Fladen Grounds.

The mean values for organic carbon, nitrogen and phosphate and total and inorganic phosphate presented in Table 25, are consistently higher in samples from the Control area and significantly higher (p<0.01) for organic nitrogen.

Table 25. Mean content and associated p values of organic carbon, nitrogen and phosphate and total and
inorganic phosphate in sediment samples collected from the within the Control area and Impact area at the
Fladen Grounds in 2014.

	Control		Imp	P value	
	Mean value	SD	Mean value	SD	
Organic carbon	0.477	0.141	0.413	0.144	0.0209
Organic nitrogen	0.082	0.017	0.074	0.008	<0.01
Organic phosphate	0.006	0.003	0.005	0.002	0.320
Inorganic phosphate	0.033	0.006	0.030	0.004	0.2830
Total phosphate	0.038	0.008	0.035	0.004	0.172

5.3.2 Distribution of infaunal communities in 2014

Figure 43 shows the variability of four univariate metrics for samples taken from within the Control and Impact areas of the Central Fladen NCMPA. No significant differences (p<0.01) were found between the two treatments (Control and Impact) for any of the metrics calculated. Higher than average biomass values were observed in three samples taken from within the Impact area. These values were due to the presence of three large specimens of the ocean quahog, *Arctica islandica*, the largest of which was 92g total wet weight.



Figure 43. Box and whisker plots of the univariate metrics (total number of individuals (N), total number of taxa (S), Hills diversity (N1) and biomass (g)) for the Control and Impact areas within the Central Fladen NCMPA. Black dots represent outliers.

Multivariate analysis of square root transformed data from the 2014 survey suggests similarities in infaunal assemblage composition between a subset of the Control and Impact area samples (Figure 44). There is greater variability in the impact samples and while the control and impact samples do overlap, there is not total overlap showing some separation. However, the high 2-dimensional stress value of the nMDS (0.19) indicates that the MDS solution is a relatively poor goodness of fit and SIMPER analysis confirms this high within-treatment variability (Impact = 41.75%, Control = 39.43%). Species accounting for 50% of variability within each treatment are shown in Table 26. Thirty three species account for 50% of the dissimilarity between treatments (average dissimilarity = 61.02%) (See Appendix A1.2 for SIMPER output).



Figure 44. nMDS ordination of samples taken from within the Central Fladen NCMPA, displayed according to treatment.

Table 26. Ranked Species contributing to within treatment (Control and Impact areas) similarity (50%) for samples taken within the Central Fladen NCMPA in 2014.

Control		Impact			
Species	% contribution	Species	% contribution		
Thyasira equalis	8.10	Spiophanes kroyeri	7.05		
Mendicula ferruginosa	6.99	Galathowenia oculata	6.91		
Heteromastus filiformis	5.96	Eclysippe vanelli	6.87		
Spiophanes kroyeri	5.76	Mendicula ferruginosa	6.15		
Axinulus croulinensis	5.41	Nephasoma minutum	5.98		
Eclysippe vanelli	5.29	Paramphinome jeffreysii	5.39		
Abyssoninoe hibernica	4.78	Axinulus croulinensis	4.64		
Nephasoma minutum	4.28	Eudorella emarginata	3.98		
Eudorella emarginata	4.28	Abyssoninoe hibernica	3.82		

The SIMPROF routine was used to determine significantly different groups of species at the 5% significance level. The nMDS ordination in Figure 45 shows eight significantly different groups and four samples thath were classed as outliers as they did not cluster with any one group. Samples within group I are the most numerous and consist of samples from within both the Impact and Control areas. However SIMPER analysis shows that the within-group similarity for group I is relatively low (47.72%), as are the average similarity values for the remaining groups (see Appendix A4.1 for SIMPER output).

Figure 46 shows the same nMDS displayed according to sediment groups. Samples within SIMPROF group's f and h corresponded well with sediment group 1b (sandy mud). Both groups also contained higher average abundances of the bivalve *Thyasira equalis* than any other group. The majority of samples were classified as sediment group 2a or 3a, both of which are composed of muddy sands.



Figure 45. nMDS ordination of samples taken from within the Central Fladen NCMPA, displayed according to SIMPROF group (p=0.05).



Figure 46. nMDS ordination of samples taken from within the Central Fladen NCMPA, displayed according to sediment group.

5.3.3 Distribution of epifaunal communities in 2014

Figure 47shows the variability of the univariate metrics; total number of taxa (S), total number of individuals (N) and Hills diversity (N1) identified in video tows taken from within the Control and Impact areas located in the south east extent of the Central Fladen NCMPA. Significant differences (p<0.01) were found between the two treatments (Control and Impact) for all of the three metrics calculated.



Figure 47. Box and whisker plots of the univariate metrics (total number of individuals (N), total number of taxa (S) and Hills diversity (N1) for epifaunal samples taken in the Control and Impact areas within the Central Fladen NCMPA.

An nMDS ordination was produced using epifaunal presence/absence transformed data and displayed according to treatment (Control area/Impact area) (Figure 48). High 2-dimensional stress value indicates a poor goodness of fit between samples and high within-treatment variability was observed for both the Control samples (average similarity = 56.19%) and the Impact samples (average similarity = 62.95) in SIMPER analysis.



Figure 48. nMDS ordination of epifaunal data (collected using video) from within the Central Fladen NCMPA in 2014 displayed according to treatment (Control/ Impact).

SIMPER analysis also showed that the seapen *V. mirabilis* was present in significantly higher numbers of samples in the Control area (98% of samples) than in the Impact area (83% of samples) (Figure 49), whilst *P. phosphorea* was present in a similar number of samples within both treatments (Control = 95%, Impact = 93%) (Figure 50). *F. quadrangularis* was present in only 5% of samples in the control area, and 15% of samples in the Impact area (Figure 51). The SIMPER results comparing the differences between treatments can be found in Appendix A4.2.



Figure 49. Count of *Virgularia mirabilis* within the Control area and Impact area in the south east of Central Fladen NCMPA.



Figure 50. Count of *Pennatula phosphorea* within the Control area and Impact area in the south east of Central Fladen NCMPA.



Figure 51. Count of *Funiculina quadrangularis* within the Control area and Impact area in the south east of Central Fladen NCMPA.

5.3.4 Temporal comparisons of infaunal and epifaunal communities

The location of samples collected from within the south east extent of the Central Fladen NCMPA in 2013 and 2014 and which have been used for temporal comparisons of infaunal assemblages are shown in Figure 52.



Figure 52. The location of all 80 target sampling stations (40 stations per treatment) in the south east extent of the Central Fladen NCMPA for the 2014 Before-After-Control-Impact (BACI) survey design and the location of grab stations visited during the 2013 survey included in the analysis.

Figure 53 shows the variability of four univariate metrics that were calculated using infaunal abundance matrices from 2013 and 2014. No significant differences (p<0.01) were found between years for any of the metrics calculated. Higher than average biomass values were observed in five samples taken in 2014 and in two samples from 2013 and can be attributed to the presence of large specimens of the ocean quahog, *Arctica islandica*.



Figure 53. Box and whisker plots of the univariate metrics (total number of individuals (N), total number of taxa (S), Hills diversity (N1) and biomass (g)) for samples collected in 2013 and 2014 within the south east extent of the Central Fladen NCMPA only.

Non-metric Multidimensional Scaling (nMDS) ordinations comparing data from 2013 and 2014 within the south east extent of Central Fladen NCMPA were produced for both presence/absence transformed infaunal data (Figure 54), and square-root transformed abundance data (Figure 55). Samples generally clustered according to sampling year when considering both presence/absence and species abundance data. SIMPER analysis showed that community variability across the samples was high within both years (average similarity = 42.20% in 2013 and 39.78% in 2014).

Table 27 shows the species contributing to 50% of the within-year similarity using the square-root transformed abundance data. Six species characterising the top 50% of the similarity between the 2013 samples were also having the largest influence in characterising the communities in 2014. *Lanice conchilega* was absent from 2014 samples but present within 72% of samples (see Appendix A4.4 for SIMPER outputs based on presence/absence) in 2013, whilst *Nephasoma minutum* was absent from 2013 samples but present within 71% of samples in 2014. Considerably lower abundances of *Ampharete falcata* were found in samples taken in 2014 (in 3% of samples taken in 2014 compared to 88% of samples in 2013). These three species, together with 31 additional species contributed to 50% of the community dissimilarity between years. No further analyses, using both datasets to describe spatial patterns in communities within the Central Fladen NCMPA were undertaken due to the strong separation by year.



Figure 54. nMDS ordination of presence/absence data from 2013 and 2014 from grab samples taken within the Central Fladen NCMPA.



Figure 55. nMDS ordination of abundance data from 2013 and 2014 from grab samples taken within the Central Fladen NCMPA.

Table 27. Species contributing to within year similarity (50%) for samples taken within the Central Fladen NCMPA in 2013 and 2014. Species highlighted in grey were only present in one year.

2013		2014			
Species	% contribution	Species	% contribution		
Spiophanes kroyeri	8.11	Mendicula ferruginosa	6.69		
Mendicula ferruginosa	6.47	Spiophanes kroyeri	6.54		
Paramphinome jeffreysii	4.47	Eclysippe vanelli	6.12		
Ampharete falcata	4.96	Thyasira equalis	5.59		
Galathowenia oculata	4.78	Nephasoma minutum	5.22		
Terebellides stroemi	4.64	Galathowenia oculata	5.15		
Axinulus croulinensis	3.81	Axinulus croulinensis	5.09		
Dipolydora	3.76	Abyssoninoe hibernica	4.38		
Lanice conchilega	3.64	Eudorella emarginata	4.20		
Eclysippe vanelli	3.58	Paramphinome jeffreysii	3.84		
Thyasira equalis	2.42				

The location of video samples, collected from within the south east extent of the Central Fladen NCMPA in 2013 and 2014, which have been used for temporal comparisons of infaunal communities are shown in Figure 56.



Figure 56. The location of all 80 target sampling stations (40 stations per treatment) for the 2014 Before-After-Control-Impact (BACI) survey design and stations from the 2014 sensitivity to abrasion pressure study (intermediate abrasion) and 2013 survey which fall within the south east extent of the Central Fladen NCMPA and were used for temporal comparisons.

Figure 57 shows the variability of total number of taxa (S) identified in video tows taken from within south east extent of the Central Fladen NCMPA in 2013 and 2014. A significant difference (p = 0.035) was found when comparing the mean total number of taxa per standardised video tow recorded between the two years.



Figure 57. Box and whisker plot of total number of taxa (S) for calculated from presence/ absence epifauna data identified in video tows from 2013 and 2014 within the Central Fladen NCMPA. Black dots represent outliers.

Figure 58 shows an nMDS ordination of the epifaunal data (presence/absence) collected in 2013 and 2014 within the south east extent of Central Fladen NCMPA. The 2-dimensional stress value is high suggesting a relatively poor goodness of fit between samples. SIMPROF analysis showed high community variability within both years (average similarity was 48.96% in 2013 and 58.20% in 2014). Differences in species presence/absence are presented in Appendix A4.5. Six of the 36 species accounting for 90% of the dissimilarity between years were present in 2013 but absent from 2014. Conversely, seven species present in 2014 were absent in 2013.



Figure 58. nMDS ordination of epifaunal data (collected using video) from within the Central Fladen NCMPA in 2013 and 2014.

5.4 Discussion and conclusions

The results presented in this section provide the 'Before' stage of the BACI experimental design with both the Control and Impact areas hypothetically being subjected to the same influence of abrasion. This explains why no significant differences were found when comparing univariate metrics on infaunal assemblages collected at Control and Impact stations in 2014 only, and when clustering by treatment in multivariate analysis. Following the collection of the 'Before' dataset at the BACI study site, it is important to note that sediment samples classified as 1a, sandy mud, were found to be located in the Control treatment only. As sediment composition is a well known driver of benthic assemblage distribution, it would be difficult to attribute any future changes to the structure and functioning of these communities to the cessation of abrasion pressure in the Impact area or to differences in sediment type present between the Control and Impact areas.

Conversely, when comparing epifaunal assemblages, significant differences in number of taxa, number of individuals and Hill's diversity were found despite multivariate analysis suggesting that assemblages from the two treatments did overlap. Differences in the distribution of seapens across the experimental study area partly explains these observed differences. *V. mirabilis* was present in significantly higher numbers in the Control area when compared to the Impact area, whilst *P. phosphorea* was present in a similar number of samples within both treatments. *F. quadrangularis* on the other hand was present in only 5% of samples in the Control area, and 15% of samples in the Impact area. These distribution patterns of the three species of seapen appear to have a relationship with sediment. Highest numbers of *V. mirabilis* were recorded where the sediment group 1a, sandy mud, was located. Group 1a was only allocated to samples in the Control area of the design with no representation within the Impact area. *F. quadrangularis* on the other hand was found in association with coarser sediments where gravel was present in the sediment fraction. Further analysis using existing datasets on seapen abundance and associated environmental conditions could be used to formally test the relationship between the three

species of seapen and sediment composition would allow more robust predications on their potential distribution across the wider Fladen Grounds and indeed in mud habitats more generally. This would be particularly important if the implementation of an Impact area in a management scenario is specifically tasked with the protection of the Tall seapen (*Funiculina quadrangularis*). In this case, the Control area may have been better placed to the south west of the Central Fladen NCMPA where in 2013, densities of *F. quadrangularis* were observed in comparable numbers to those found in the south east extent of the Central Fladen NCMPA in both 2013 and 2014.

Data points from 2013 were included in this study to provide a first step in bringing together temporal (two years') data at the Fladen Grounds and contribute to the development of the Type 1 monitoring approach. While there were no significant differences between the infaunal univariate metrics studied, multivariate analysis showed the clustering by year of the assemblages. A number of factors could be contributing to these between year differences including seasonality, notably possible differences in the prevalence and intensity of winter storms (2013 survey was conducted in January, 2014 survey was conducted in March), the human factor associated with the identification process for infauna, or the fundamental differences in the design and objectives of the two surveys. Still, the presentation of two years of data at the site highlights some key questions which should be addressed when developing Type 1 monitoring approaches and building temporal datasets.

6 Conclusions and recommendations

This report presents results from analyses that were conducted to address Type 1, 2 and 3 monitoring approaches (see below for distinctions between the types) using data collected at the Fladen Grounds from the 2014 JNCC-Cefas survey. From these results, a series of conclusions and general recommendations are presented for each of the three types of monitoring.

6.1 Type 1 monitoring – spatial and temporal data

Lower densities of the seapens *P. phosphorea* and *V. mirabilis* were observed in coarser sediments at the Fladen Grounds, while the distribution of *F. quadrangularis* was seemingly restricted to areas exhibiting a coarser sediment fraction. These findings disagree with research conducted on seapens on the West Coast of Scotland where *V. mirabilis* was found to have the highest tolerance to gravel content, sediment which was less favourable for *P. phosphorea* and *F. quadrangularis*.

1. Further work to formally test the relationship between the three species of seapen and sediment composition would allow more robust predictions on their potential distribution across the wider Fladen Grounds and indeed in mud habitats more generally.

Limited sampling of sediment contaminants was undertaken in this survey as it was a secondary priority to abrasion pressure. Therefore, no assessment of sediment contamination was undertaken during this study.

2. In order to fully understand aspects of sedimentary biogeochemistry and contaminant effects at the Fladen Grounds, a dedicated survey would be required. Repeat survey would allow temporal trends to be assessed.

Data from 2013 and 2014 were compiled to create a temporal (two years') data set and to contribute to the development of baseline data set essential for Type 1 monitoring. During the compilation of these data, a number of between-year differences were observed. These could be attributed to a number of factors which include seasonality and storms (2013 survey was conducted in January, 2014 survey was conducted in March), the human factor associated with the identification process for infauna, or the fundamental differences in the design and objectives of the two surveys.

3. When planning surveys to contribute to Type 1 monitoring approaches and the creation of temporal datasets, consideration must be given to consistencies during survey planning. These include time of year, the methods used and the quality control procedures for the acquisition and processing of samples, and the design of the survey approach.

6.2 Type 2 monitoring – sensitivity to abrasive pressure

The JNCC abrasion pressure method used to inform the abrasion pressure gradient at the Fladen Grounds is expressed as annual averages and assumes even distribution of fishing effort within a given grid cell. Therefore the method does not take into consideration seasonal variability in fishing activity and assumes that effort is homogeneously distributed within a cell. This is not necessarily true as effort can be localised within cells due to varying habitat types, seabed obstructions and water depth.

4. Future studies may wish to consider variability in fishing effort within a cell by looking at how 'pings' are distributed within the area, as well as considering seasonality in fisheries, especially when attempting to make links between pressure and state.

Organic carbon and nitrogen and inorganic phosphate and total phosphate content were higher in High-abrasion areas when compared to the Low-abrasion pressure areas. However, a robust interpretation of these results was not possible in the absence of higher resolution data on when the area was last impacted, and therefore if the disturbance is acute or chronic.

5. An experimental approach to better understand the biogeochemical response to both acute and chronic disturbance would provide a context against which infaunal point samples could then be compared to elucidate if any changes are due to the physical action of trawling or the removal of bioturbators.

Statistical analysis of Infaunal data revealed significant differences in number of species, number of individuals and Hill's diversity with lower values of these metrics observed in samples from the high pressure areas. While it was concluded that there appears to be an infaunal response to increased abrasion pressure within the four experimental boxes studied, these findings cannot be applied to say something about abrasion pressure across the wider Fladen Grounds. An **increased sampling effort along a gradient**, such as the one planned for the 2014 survey, would allow conclusions to be made on a greater geographical scale than the boxes studied here.

Only small bivalve species were available for size frequency distribution analysis as a result of samples being collected using a $0.1m^2$ Day grab. A Day grab provides a point sample covering an area of $0.1m^2$ (to a depth of approximately 15cm).

6. To quantify the abundance and size of any large and long-lived species which are likely to be found in low abundances, an alternative gear such as the Triple D dredge which is designed for sampling the larger, more sparsely distributed fauna should be considered.

The results of this study suggest that the trait composition of infauna subjected to Low-and High-abrasion pressure are different.

7. Future work could consider using biological trait analysis to calculate productivity and or bioturbation potential of the benthos. Productivity may be particularly informative when considered alongside the investigation of biogeochemical responses to the acute and chronic effects of trawling.

Increasing abrasive pressure appeared to reduce the number of epifaunal individuals whilst increasing species diversity.

8. Further work to investigate not only the total number and abundance of epifauna but also the biomass of individual species would facilitate a greater understanding of change in epifaunal community structure and function. The collection of epifaunal samples using a beam trawl would permit these additional quantitative analyses to be carried out as they operate at an appropriate scale to study the effects of fishing (over 10s of m²) by integrating the patch effects which are unavoidable with point sampling.

Current analysis suggests that the density of the seapen *P. phosphorea* at the Fladen Grounds is a good indicator of the impacts associated with increasing abrasive pressure.

The number of *P. phosphorea* appeared to decrease with increasing abrasive pressure whilst *V. mirabilis* showed no discernible relationship. The morphology and ecology of *P. phosphorea* may mean it is less resilient to habitat disturbance when compared with *V. mirabilis*.

9. A greater understanding of how fishing pressure affects the distribution of the three species of seapen is required to increase our understanding of how the morphological and functional characteristics of seapens are affected by trawling. This would enable the development of a biological indicator, based on seapens, for abrasion pressure on burrowed mud habitats.

6.3 Type 3 monitoring – BACI study

This study was designed to provide the "Before" stage of the Before, After, Control, Impact (BACI) experimental design with both the Control and Impact areas hypothetically being subjected to the same influence of abrasion. As the results describe the faunal communities before any management measures are put in place, it was expected that no significant differences would be found when comparing infaunal assemblages collected at Control and Impact stations. However, epifaunal assemblages were found to be different driven mainly by the distribution of seapens. It was suggested that the differences in seapen distribution could be linked to sediment composition as one sediment group was found only in the Control treatment. Recommendation 1 and 8 would aid the interpretation of data on seapen density and its link to sediment characteristics and response to the cessation of fishing.

10. Clear guidance on what feature of conservation importance the 'Impact' area is designed to protect should be established before a BACI experiment is designed.

For example, if the implementation of an Impact area in a management scenario is specifically tasked with the protection of the Tall seapen (*Funiculina quadrangularis*), the Control area may have been better placed to the south west of the Central Fladen NCMPA where in 2013, *F. quadrangularis* was observed in comparable numbers.

References

AGER, O. & WILDING, C. 2009. *Funiculina quadrangularis*. The tall seapen. Marine Life Information Network: Biology and Sensitivity Key Information Sub-programme [on-line]. Plymouth: Marine Biological Association of the United Kingdom. [cited 03/09/2012]. Available from: http://www.marlin.ac.uk/specieshabitats.php?speciesID=3353>.

ASPILA, K.I., AGEMAIN, H. & CHAU, A.S.Y. 1976. A semi-automated method for the determination of inorganic, organic and total phosphate in sediments. *Analyst*, **101**(1200): 187-197.

BASSET. A., BARBONE, E., BORJA, A., BRUCET, S., PINNA, M., QUINTANA, X.D, REIZOPOULOU, S., ROSATI, I. & SIMBOURA, N. 2012. A benthic macroinvertebrate size spectra index for implementing the Water Framework Directive in coastal lagoons in Mediterranean and Black Sea ecoregions. *Ecological Indicators*, **12**, 72-83.

BERGMAN, M.J.N. & VAN SANTBRINK, J.W. 2000. Mortality in megafaunal benthic populations caused by trawl fisheries on the Dutch continental shelf in the North Sea in 1994. ICES *Journal of Marine Science*, **57**, 1321-1331.

BLOTT, S.J. & PYE, K. 2001. GRADISTAT: a grain size distribution and statistics package for the analysis of unconsolidated sediments. *Earth Surface Processes and Landforms*, **26**, 1237-1248.

BOLAM, S.G., COGGAN, R.C., EGGLETON, J.E., DIESING, M. & STEPHENS, D., 2014. Sensitivity of macrobenthic secondary production to trawling in the English sector of the Greater North Sea: a biological traits approach. *Journal of Sea Research*, **85**, 162–177.

BREMNER, J., ROGERS, S. I., & FRID, C. L. J. 2006. Matching biological traits to environmental conditions in marine benthic ecosystems. *Journal of Marine Systems*, **60** (3), 302-316.

BROOKS, A.J. 2013. Assessing the sensitivity of geodiversity features in Scotland's seas to pressures associated with human activities. Scottish Natural Heritage Commissioned Report No. 590.

BROOKS, A.J. KENYON, N.H. LESLIE, A., LONG, D. & GORDON, J.E. 2013. Characterising Scotland's marine environment to define search locations for new Marine Protected Areas. Part 2: The identification of key geodiversity areas in Scottish waters. Scottish Natural Heritage Commissioned Report No. 432.

CHANIOTIS, P.D., CRAWFORD-AVIS, O.T., CUNNINGHAM, S., GILLHAM, K., TOBIN, D. & LINWOOD, M. 2011. *Identifying locations considered to be least damaged/more natural in Scotland's seas.* Report produced by the Joint Nature Conservation Committee, Scottish Natural Heritage and Marine Scotland for the Scottish Marine Protected Areas Project.

CHEVENE, F., DOLÉDEC, S. & CHESSEL, D. 1994. A fuzzy coding approach for the analysis of long-term ecological data. *Freshwater Biology*,**31**, 295-309.

CHURCH N.J., JOHNSON G.E., EASSOM A., TOBIN D., EDWARDS D., CAMERON A. & WEBB, K.E. 2015. JNCC Recommended Pressure Mapping Methodology 1. Abrasion: Methods paper for creating a geo-data layer for the pressure 'Physical Damage (Reversible Change) - Penetration and/or disturbance of the substrate below the surface of the seabed, including abrasion'. *JNCC report*, No. 515, JNCC, Peterborough.

COGGAN, R., MITCHELL, A., WHITE, J. & GOLDING, N. 2007. Recommended operating guidelines (ROG) for underwater video and photographic imaging techniques. www.searchmesh.net/pdf/GMHM3_Video_ROG.pdf

CONNER, D.W., ALLEN, J.H., GOLDING, N., HOWELL, K.L., LIEBERKNECHT, L.M., NORTHERN, K.O. & REKER, J.B. 2004. The Marine Habitat Classification for Britain and Ireland Version 04.05 JNCC, Peterborough, ISBN 1 861 07561 8 (internet version) jncc.defra.gov.uk/MarineHabitatClassification.

DARR, A., KORPINEN, S., WESTERBOM, M. & NYGARD, H. 2013. Population structure of long-lived macrozoobenthic species. HELCOM Core Indicator Report. Online. 14/10/2014 http://www.helcom.fi/Core%20Indicators/HELCOM- CoreIndicator Population structure of long-lived macrozoobenthic species.pdf

DENDEREN, P. D., HINTZEN, N. T., VAN KOOTEN, T., & RIJNSDORP, A. D. 2014. Temporal aggregation of bottom trawling and its implication for the impact on the benthic ecosystem. *ICES Journal of Marine Science: Journal du Conseil*, fsu183.

DE JUAN, S., THRUSH, S. F., & DEMESTRE, M. 2007. Functional changes as indicators of trawling disturbance on a benthic community located in a fishing ground (NW Mediterranean Sea). *Marine Ecology Progress Series*, **334**, 117-129.

DE JUAN, S. & DEMESTRE, M. 2012. A Trawl Disturbance Indicator to quantify large scale fishing impact on benthic ecosystems. *Ecological Indicators*, **18**, 183-190.

EGGLETON, J., JENKINS, C. & SCHINAIA, S. 2013. Offshore seabed survey of the Fladen Grounds Scottish possible MPAs – Final Report. CEFAS Report C5973.

EUROPEAN COMISSION. 2010. 2010/477/EU: Commission Decision of 1 September 2010 on criteria and methodological standards on good environmental status of marine waters (notified under document (2010) 5956) Text with EEA relevance.

FLEDDUM, A., ATKINSON, L. J., FIELD, J. G. & SHIN, P. 2013. Changes in biological traits of macro-benthic communities subjected to different intensities of demersal trawling along the west coast of southern Africa. *Journal of the Marine Biological Association of the United Kingdom*, **93** (08), 2027-2038.

FRID, C., ROGERS, S., NICHOLSON, M., ELLIS, J. & FREEMAN, S. 2000. Using biological characteristics to develop new indices of ecosystem health. Mini-symposium on Defining the role of ICES in supporting biodiversity conservation. pp:1 – 23.

FROJAN, C. R. B., COOPER, K. M., BREMNER, J., DEFEW, E. C., HUSSIN, W. M. W. & PATERSON, D. M. 2011. Assessing the recovery of functional diversity after sustained sediment screening at an aggregate dredging site in the North Sea.Estuarine, *Coastal and Shelf Science*, **92** (3), 358-366.

GILKINSON, K., PAULIN, M., HURLEY, S. & SCHWINGHAMER, P. 1998. Impacts of trawl door scouring on infaunal bivalves: results of a physical trawl door model/dense sand interaction. *Journal of Experimental Marine Biology and Ecology*, **224** (2), 291-312.

GREATHEAD, C.F., DONNAN, D.W., MAIR, J.M. & SAUNDERS, G.R. 2007. The sea pens *Virgularia mirabilis, Pennatula phosphorea* and *Funiculina quadrangularis*: distribution and conservation issues in Scottish waters. *Journal of the Marine Biological Association*, **87**; 1095-1103.

GREATHEAD, C., DEMAIN, D., DOBBY, H., ALLAN, L. & WEETMAN, A. 2011. Quantitative analysis of the distribution and abundance of the burrowing megafauna and large epifauna community in the Fladen fishing ground, northern North Sea. *Scottish Marine and Freshwater Science or Marine Scotland Science Report*, **2**, (2).

GREATHEAD, C., GONZALEZ-IRUSTA, J. M., CLARKE, J. BOULCOTT, P., BLACKADDER, L., WEETMAN, A., & WRIGHT, P. J. 2014. Environmental requirements for three sea pen species: relevance to distribution and conservation. *ICES Journal of Marine Science*: Journal du Conseil, fsu129.

HANSEN, H.P. & GRASSHOFF, K. 1983. Automated chemical analysis. In: K. Grasshoff, M. Ehrhardt and K. Kremling (Editors), *Methods of Seawater Analysis*, Second Edition. Weinheim: Verlag Chemie pp. 347-379.

HAYWARD, P. J. & RYLAND, J. S. 1990. The marine fauna of the British Isles and northwest Europe. 2 vols. Oxford, Clarendon Press.

HIDDIINK, J.G., JENNINGS, S. & KAISER, M.J. 2006. Indicators of the ecological impact of bottom-trawl disturbance on seabed communities. *Ecosystems*, **9**, 1190-1199.

HINZ, H., PRIETO, V. & KAISER, M. J. 2009. Trawl disturbance on benthic communities: chronic effects and experimental predictions. *Ecological Applications*, **19** (3), 761-773.

HUGHES, D.J. 1998. Sea pens and burrowing megafauna: An overview of dynamics and sensitivity characteristics for conservation and management of marine SACs. Report prepared for SAMS UK Marine SACs Project, 105 pp.

HUSSIN, W. R. W., COOPER, K. M., FROJAN, C. R. B., DEFEW, E. C. & PATERSON, D. M. 2012. Impacts of physical disturbance on the recovery of a macrofaunal community: a comparative analysis using traditional and novel approaches. *Ecological Indicators*, **12** (1), 37-45.

ICES. 2007. Workshop on the use of UWTV surveys for determining abundance in Nephrops stocks throughout European waters. ICES Document CM 2007/ACFM: 14. 310 pp.

JENNINGS, S. & KAISER, M.J. 1998. The effects of fishing on marine ecosystems. *Advances in Marine Biology*, **34**, 201-352.

JENNINGS, S., DINMORE, T.A., DUPLISEA, D.E., WARR, K.J. & LANCASTER, J.E. 2001. Trawling disturbance can modify benthic production processes. *Journal of Animal Ecology*, **70**, 459-475.

JNCC. 2014. The UK marine biodiversity monitoring strategy. Version 2.2. 1-19, INPRESS.

JONES, A. J., HISCOCK, K. & CONNOR, D.W. 2000. Marine habitat reviews: A summary of ecological requirements and sensitivity characteristics for the conservation and management of marine SACs. JNCC Report, JNCC, Peterborough, pp 178.

KAISER, M.J., RAMSAY, K., RICHARDSON, C.A., SPENCE, F.E. & BRAND, A.R. 2000. Chronic fishing disturbance has changed shelf sea benthic community structure. *Journal of Animal Ecology*, **69**, 494-503.

KAISER, M.J. & SPENCER, B.E. 1996. The effects of beam trawl disturbance on infaunal communities in different habitats. *Journal of Animal Ecology*, **65**, 348-358.

LONG. D. 2006. BGS detailed explanation of seabed sediment modified Folk classification. www.searchmesh.net

MACDONALD, D. S. LITTLE, M., ENO, N. C. & HISCOCK, K. 1996. Disturbance of benthic species by fishing activities: a sensitivity index. Aquatic Conservation: *Marine and Freshwater Ecosystems*, **6** (4), 257-268.

ALLAN, L., DEMAIN, D., WEETMAN, A., DOBBY, H. & MCLAY, A. 2012. Data Mining of the Nephrops Survey Database to Support the Scottish MPA Project. Scottish Marine and Freshwater Science, 03/09. 35 pp. http://www.gov.scot/Publications/2012/12/4074 (June 2015).

MARINE SCOTLAND SCIENCE. MSS. 2011b. *Scottish sea fisheries statistics 2010. A national statistics publication for Scotland*. The Scottish Government, ISBN: 978-1-78045-362-0. Available from <<u>http://www.scotland.gov.uk/Resource/Doc/357661/0120860.pdf</u>>

MASON, C. 2011. NMBAQC's Best Practice Guidance. Particle Size Analysis (PSA) for Supporting Biological Analysis. National Marine Biological AQC Coordinating Committee, 72pp, December 2011. <u>https://data.gov.uk/data/.../2f6d1e8d-0f09-4222-a8bd-52212c60f5f</u>.

MCLLWAINE, P. 2015. Fladen Nature Conservation Marine Protected Area Survey Report. pp 93. Unpublished report for JNCC.

NORKKO, A., VILLNAS, A., NORKKO, J., VALANKO, S., & PILDITCH, C. 2013. Size matters: implications of the loss of large individuals for ecosystem function. *Scientific reports*, **3**, Article Number 2646.

ORPIN, A.R. & KOSTYLEV, V.E. 2006. Towards a statistically valid method of textural sea floor characterization of benthic habitats. *Marine Geology*, **225**, 209-222.

PAGANELLI, D., MARCHINI, A. & OCCHIPINTI-AMBROGI, A. 2012. Functional structure of marine benthic assemblages using Biological Traits Analysis (BTA): a study along the Emilia-Romagna coastline (Italy, North-West Adriatic Sea). *Estuarine, Coastal and Shelf Science*, **96**, 245-256.

PEARSON, T. H. & ROSENBERG, R. 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. *oceanography marine biology annual review*, **16**, 229-311.

PERCIVAL, P., FRID, C. & UPSTILL-GODDARD, R. 2005. The impact of trawling on benthic nutrient dynamics in the North Sea: implications of laboratory experiments. In American Fisheries Society Symposium Amercian Fisheries Society, (Vol. 41, p. 491).

PUSCEDDU, A., BIANCHELLI, S., MARTIN, J., PUIG, P., PALANQUES, A., MASQUE, P. & DANOVARO, R. 2014. Chronic and intensive bottom trawling impairs deep-sea biodiversity and ecosystem functioning. *Proceedings of the National Academy of Sciences*, **111** (24), 8861-8866.

ROZEMARIJIN, K., SCHANDER, C., KONGSRUF, J, A. & WILLASSEN, E. 2011. Ecology of twelve species of Thyasiridae (Mollusca: Bivalvia). *Marine pollution bulletin*, **62** (4), 786-791.

SCOTTISH GOVERNMENT. 2014. Marine Protected Areas and Burrowed Mud Position Paper [online] Available at: <u>http://www.gov.scot/resource/0038/00389464.doc</u> SNELGROVE, P. V., GRASSLE, J. P. & BUTMAN, C. A. 1998. Sediment choice by settling larvae of the bivalve, Spisula solidissima (Dillwyn), in flow and still water. *Journal of Experimental Marine Biology and Ecology*, **231** (2), 171-190.

STEWART, L.K., KOSTYLEV, V.E. & ORPIN, A.R. 2009. Windows-based software for optimising entropy-based groupings of textural data. *Computers & Geosciences*, **35**, 1552-1556.

TILLIN, H. M., HIDDINK, J. G., JENNINGS, S. & KAISER, M. J. 2006. Chronic bottom trawling alters the functional composition of benthic invertebrate communities on a sea-basin scale. *Marine Ecology Progress Series*, **318**, 31-45.

TILLIN, H. & TYLER-WALTERS, H. 2014. Assessing the sensitivity of subtidal sedimentary habitats to pressures associated with marine activities: Phase 2 Report – Literature review and sensitivity assessments for ecological groups for circalittoral and offshore Level 5 biotopes. *JNCC Report*, No. 512B, JNCC, Peterborough.

TRIMMER, M., PETERSEN, J., SIVYER, D.B., MILLS, C., YOUNG, E. & PARKER, E.R. 2005. Impact of long-term benthic trawl disturbance on sediment sorting and biogeochemistry in the Southern North Sea. *Marine Ecology Progress Series*, **298**, 79-94.

TROFFE, P.M., LEVINGS, C.D., PIERCEY, G.B.E., & KEONG, V. 2005. Fishing gear effects and ecology of the sea whip (*Halipteris willemoesi* (Cnidaria: Octocorallia: Pennatulacea)) in British Columbia, Canada: preliminary observations. *Aquatic Conservation: Marine and Freshwater Ecosystems*, **15** (5), 523-533.

TYLER-WALTERS, H., ROGERS, S. I., MARSHALL, C.E. & HISCOCK, K. 2009. A method to assess the sensitivity of sedimentary communities to fishing activities. *Aquatic Conservation: Marine and Freshwater Ecosystems*, **19**, 285–300.

TYLER-WALTERS, H., JAMES, B. (EDS.), WILDING, C., DURKIN, O., LACEY, C., PHILPOTT, E., ADAMS, L., CHANIOTIS, P.D., WILKES, P.T.V., SEELEY, R., NEILLY, M., DARGIE, J. & CRAWFORD-AVIS, O.T. 2012. Descriptions of Marine Protected Area (MPA) search features. A report produced by MarLIN (Marine Life Information Network), SMRU Ltd., Scottish Natural Heritage and the Joint Nature Conservation Committee, for the Scottish Marine Protected Areas Project.

UNGFORS, A., BELL, E., JOHNSON, M. L., COWING, D., DOBSON, N. C., BUBLITZ, R. & SANDELL, J. 2013. *Nephrops* Fisheries in European Waters. *Advances in marine biology*, **64**, 247-314.

USSEGLIO-POLATERA, P., BOURNAUD, M., RICHOUX, P. & TACHET, H. 2000. Biomonitoring through biological traits of benthic macroinvertebrates: how to use species trait databases? *Hydrobiological* 422/423: 153-162.

WIDDICOMBE, S., AUSTEN, M. C., KENDALL, M. A., OLSGARD, F., SCHAANNING, M. T., DASHFIELD, S. L. & NEEDHAM, H. R. 2004. Importance of bioturbators for biodiversity maintenance: indirect effects of fishing disturbance. *Marine Ecology Progress Series* **275**, 1-10.

WITBAARD, R. & BERGMAN, M. J. N. 2003. The distribution and population structure of the bivalve *Arctica islandica* in the North Sea: what possible factors are involved? *Journal of Sea Research*, **50** (1), 11-25.

WORSFOLD, T.M., HALL, D.J. & O'REILLY, M. 2010. Guidelines for processing marine macrobenthic invertebrate samples: a processing requirements protocol version 1 (June 2010). Unicomarine Report NMBAQCMbPRP to the NMBAQC Committee. 33 pp. [Accessed 18/12/2014].

Appendix 1: Sample acquisition

A1. 1 Sample acquisition

The primary sediment sampling gear was a 0.1 m^2 Day grab, which is ideally suited for the muddy and sandy sediments expected in the area. Where the Day grab was unsuccessful, a mini Hamon grab (0.1m^2) fitted with a video camera and lighting was available to collect sediment samples for infauna community analysis. A Shipek grab was also used to specifically collect sediment samples for contaminants analysis. For the abrasion pressure gradient study, the Day grab was the only gear deployed to collect sediment samples. All grab systems were deployed from the side gantry.

Samples were collected from within a 100m diameter 'bull ring' around the target sampling location. On recovery of the grab, a photograph of the sample was taken (*in situ* for the Day and Shipek grabs and following transfer to a large container for the mini Hamon grab). The sampling depth (Day and Shipek grab) or volume (mini Hamon grab) was estimated before a sediment subsample was collected for particle size distribution analysis (PSA) using either a 3cm diameter core (Day and Shipek grab) or a 125ml scoop (mini Hamon grab). Sediment sub samples for PSA were placed in labelled containers and frozen immediately.

The remaining sample was decanted into a plastic box and transferred to the sample processing area on board the vessel if an infaunal sample was required. Benthic fauna were collected by washing the sample with sea-water over a 1mm mesh sieve. The retained >1 mm fraction was transferred to a labelled container and preserved in buffered 4% formaldehyde for later analysis ashore. A visual assessment was made of the sediment type sampled by the grab and noted on the field records.

A separate deployment of either the Day or Shipek grab was used to collect a 2cm deep surface scrape of sediment for analysis of sediment organic carbon and nitrogen (Sediment OCN) and nitrate:phosphate (N:P) or a suite of organic and heavy metal contaminants. In addition, five samples for heavy metal and organohaline contaminant analysis were collected; four stations from across the BACI and abrasion pressure survey areas and one additional station in close proximity to the Claymore oil platform. The equipment used to collect contaminant samples was rinsed in Hexane solvent to prevent cross contamination between samples prior to use at contaminant stations.

A1. 2 Seabed imagery

Video and still images were acquired using a camera sledge (CS) system. Set-up and operation followed the MESH 'Recommended Operating Guidelines (ROG) for underwater video and photographic imaging techniques' (Coggan *et al* 2007). Video was recorded simultaneously to a Sony GV-HD700 DV tape recorder and a computer hard drive. A video overlay was used to provide station metadata, time and position (of the vessel's GPS antenna) onto the recorded video image.

Camera tows lasted a minimum of 10 minutes with the sledge being towed at $\sim 0.5 - 0.7$ knots through the 100m diameter target bullring. Stills images were captured at regular one minute intervals with additional opportunistic images taken if specific features of interest were encountered during towing. Field notes were made during each camera deployment, noting station and sample metadata, real-time observations of substrate and taxa.

A1. 3 Global Positioning System (GPS)

Position fixes were recorded using the Tower Navigation software (Tower Software Ltd) on the RV Cefas Endeavour. This software records, as a minimum, the geographic position of the sampling equipment based on its location on-board. For grab sampling, the system applies offsets to calculate the position of the side gantry, whereas for camera deployments calculations are made to provide the stern gantry position. The actual position of the gear on the seabed was recorded using an Ultra Short Base Length (USBL) positioning beacon. In strong tides and deep water an offset of up to ~10 metres may occur. Comparison of the USBL position and the gantry position will allow for the accuracy to be determined.

Positions for grab samples were also recorded manually (manual fixes) at the instant the grab contacted the seabed.

During video tows, fixes were recorded every five seconds between the start and end of each tow. Still images were matched to the nearest fix recorded. The corrected positional data for the still images provides geo-referenced still images.

A1. 4 Data QA/QC

For quality control and quality assurance of the data, all activities in the field were performed according to the recommendations in the following documents:

- Biological Monitoring: General Guidelines for Quality Assurance¹
- Quality Assurance in Marine Biological Monitoring²
- Recommended operating guidelines for underwater video and photographic imaging techniques³

¹ Reference URL: <u>http://www.marbef.org/qa/documents/PKG85.pdf</u>

² Reference URL: <u>http://www.nmbaqcs.org/qa-standards/qa-in-marine-biological-monitoring.aspx</u>

³ Reference URL: <u>http://www.searchmesh.net/PDF/GMHM3_Video_ROG.pdf</u>

Appendix 2: Contaminants analysis

A2.1. Sediment metals analysis

For metal analysis of, the fine fraction (<63 µm), was digested in a mixture of hydrofluoric, hydrochloric and nitric acids using enclosed vessel microwave (MarsXpress Microwave Reaction System, CEM Ltd, Buckingham, UK). Typically, approximately 0.2g of sample was weighed out and pre-digested overnight in a mixture of nitric acid, hydrochloric acid and hydrofluoric (Aristar grade 69%, VWR, Leicestershire, UK). The excess of HF was complexed with the addition of a saturated boric acid solution. The digest was then further diluted prior to analysis by inductively coupled plasma-mass spectrometry (ICP-MS, Agilent 7500ce, Agilent Technologies, Waldbronn, Germany) and by inductively coupled plasma-optical emission spectroscopy (ICP-OES, Thermo iCAP 6500 Duo, Thermo Scientific, Hemel Hempstead, UK). Quantification of trace elements was performed by external calibration using 8 calibration levels (0-500ng/ml).

A2. 2. Sediment PAH analysis

For the analysis of PAHs in sediment samples, each homogenised wet sediment sample was spiked with an analytical surrogate consisting of a suite of deuterated PAHs (naphthalene-d8, acenaphthylene-d8, anthracene-d10, dibenzothiophene-d8, pyrene-d10, benzo[a]anthracene-d12, benzo[a]pyrene-d12 and dibenz[a,h]anthracene-d14) and extracted by alkaline saponification in methanolic potassium hydroxide followed by liquid/liquid solvent extraction using glass-distilled grade pentane and drying of the extracts with sodium sulphate. The total hydrocarbon concentration in the extracts was determined by means of ultra-violet fluorescence spectrometry as a screen of the level of contamination. Sub-samples of wet sediment were dried to measure total solids percentage, which was used to enable the calculation of PAH concentrations on a dry weight (dw) basis.

An aliquot of each sediment extract was concentrated and passed through a short alumina chromatography column and eluted in dichloromethane:pentane [1:1] in order to clean-up the extract. The cleaned up extract was concentrated to 1ml and analysed for a suite of parent and alkylated PAHs by gas chromatography-mass spectrometry in electron impact ionization mode (GC-MS) using a 6890 GC coupled to a 5975 MSD (Agilent Technologies, Walbron, Germany) in synchronous multiple ion detection/full scan, monitoring the molecular ions of each compound or group (the latter in the case of the alkylated PAH) determined. Aliguots of the extracts (2µl) were analysed using a DB-5.625 (30m x 0.25mm x 0.25µm) cross linked fused silica capillary column coated with 95 % dimethyl 5 % diphenyl polysiloxane (J&W Scientific, Folsom, CA, USA). The carrier gas was helium at a constant flow of 1 ml/min. Injection was in pulsed splitless mode with an injector temperature of 300 °C. The injection was made with the column at 60°C and following injection the oven temperature was held at 60 °C for 2 min and subsequently raised to 310°C at a rate of 5 °C/min where it was held for 10 min giving a standard run time of 62 min. The GC was directly coupled to the MS detector via a transfer line heated to 310 °C. The mass spectrometer was operated in SIM/full scan mode and scans were from 35-325 Daltons. Quantification for PAHs was performed using surrogate standards and 5 calibration levels (range 25 - 500 ng/ml).

A2.3. Sediment organohalogens analysis

Analysis of the sediment for organohalogens was undertaken by air drying and sieving (<2mm) sediment samples in a controlled environment. 10 g of dried sediment were mixed with sodium sulphate, transferred to a glass Soxhlet thimble and topped with 1cm of sodium sulphate. The samples were subjected to Soxhlet extraction using acetone: n-hexane 1:1

(v:v) for ~6 hours. Prior to extraction, the samples for BDE209 analysis were spiked with 13C12-BDE209. Sulphur residues were removed at this stage with copper filings.

Sediment extracts for hexabromocyclododecane (HBCDD) analysis were spiked with an analytical surrogate (consisting of d18- α -, d18- β -, d18- γ -HBCDD and 13C12-TBBPA) and cleaned up by high resolution gel permeation chromatography (HRGPC), using a series 1100 HPLC system (Agilent Technologies, Waldbronn, Germany), followed by acid silica chromatography. The HRGPC columns used were two Envirogel TM columns (Waters Corporation, Milford, MA, USA; 150 mm x 19 mm i.d.and 300 mm x 19 mm i.d.), connected in series and protected by an Envirogel TM guard column (Law et al 2006). For the acid silica chromatography step, concentrated HRGPC fractions were eluted through 8 g of 45% acid silica with 25 ml of 1:1 dichloromethane:hexane (Harrad et al 2009). Three diastereoisomers, α -, β - and y-HBCDD, and tetrabromobisphenol-A (TBBP-A) were determined using ultra performance liquid chromatography (UPLC; Acquity, Waters) tandem mass spectrometry (TQ MS/MS; Xevo TQ MS, Waters). Compound separation was achieved on a BEH C18 UPLC column (1.7µm, 2.1 x 50mm; Waters) using a gradient program from 30%:70% to 0.1%:99.9% 2mM ammonium acetate buffered water:acetonitrile. Quantitation for HBCDD and TBBP-A was performed using isotope dilution and 8 calibration levels (range 0.5 - 200 ng/ml).

An aliquot of the sediment extracts was cleaned up and fractionated using alumina (5% deactivated) and silica (3% deactivated) columns, respectively. The silica column fractionation results in two fractions, the first fraction containing polychlorinated biphenyls (PCBs) and BDE209, the second fraction containing polybrominated diphenylethers (PBDEs),. The final GC-ready fractions were spiked with PCB53 (for PCBs) and PCB200 (for PBDE analysis) and made up to a final volume of 1ml.

PCB concentrations in sediment samples were determined with an Agilent 6890 GC with μ ECD (Agilent Technologies, Waldbronn, Germany). The separation of analytes was performed on a 50.0 m × 200 μ m, 0.33- μ m-film-thickness DB-5 capillary column (J&W). The carrier and ECD make-up gas were hydrogen (32.2 psi constant pressure, initial velocity 50 cm/s) and argon/methane (95:5), respectively. The initial oven temperature was 90°C, held for 2.00min, then increased to 165°C at 15°C/min, to 285°C at 2°C/min, and finally held for 23 min. The injector temperature and detector temperature was 270°C and 300°C, respectively. A 1- μ l extract was injected in splitless mode with a purge time of 2 min. The PCB standard solutions contained the following 27 compounds in iso-octane: Hexachlorobenzene; p,p'-DDE; CB101; CB105; CB110; CB118; CB128; CB138; CB141; CB149; CB151; CB153; CB156; CB158; CB170; CB18; CB180; CB183; CB187; CB194; CB28; CB31; CB44; CB47; CB49; CB52; CB66, together with the internal standard CB53. Quantitation was performed using internal standards and 7 calibration levels (range 0.5 – 100ng/ml).

PBDE congeners were determined by gas chromatography-mass spectrometry in electron capture negative ionization (GC-MS-ECNI) (De Boer *et al* 2001) with an Agilent 6890 GC with 5973 MS (Agilent Technologies, Waldbronn, Germany) in negative chemical ionisation (NCI) mode. The separation of analytes was performed on a 50.0 m × 250 μ m, 0.25- μ m-film-thickness DB-5 capillary column (J&W). The carrier gas was helium (30 psi constant pressure, average velocity 40 cm/s) and the reagent gas was methane (40 psi). The initial oven temperature was 90°C, held for 2.00min, then increased to 200°C at 30°C/min, to 295°C at 2.5°C/min, and finally held for 31.3 min. The injector temperature and detector temperature was 270°C and 200°C, respectively. A 2- μ l extract was injected in splitless mode with a purge time of 2 min. Quantitation for PBDEs was performed using internal standards and 8 calibration levels (range 0.1 – 50 ng/ml). The PBDE standard solutions contained the following 11 compounds in iso-octane: BDE17; BDE28; BDE47; BDE66; BDE100; BDE99; BDE85; BDE154; BDE153; BDE138; BDE138; BDE183; together with the internal

standard CB200. BDE209 concentrations were determined with an Agilent 6890 GC with 5973 MS (Agilent Technologies, Waldbronn, Germany) in NCI mode using 13C12-BDE209 as internal standard. The separation of analytes was performed on a 15.0 m x 250 μ m, 0.1- μ m-film-thickness DB-1 capillary column (J&W). The carrier gas was helium (1.3ml/min constant flow, average velocity 59 cm/s) and the reagent gas was methane (40 psi). The initial oven temperature was 90°C, held for 1.00min, then increased to 200°C at 25°C/min, to 295°C at 10°C/min, and finally held for 20 min. The injector temperature and detector temperature was 250°C and 200°C, respectively. A 2- μ I extract was injected in pulsed splitless mode with a 20psi pulse until 1 min and a purge time of 2 min. Quantitation of BDE209 was performed using an internal standard and 7 calibration levels (range 0.5 – 500 ng/ml).

A2.4. QA/QC

To ensure quality control (QC) for contaminants analysis, the laboratory biannually participates in the Quasimeme (Quality Assurance (QA) of Information for Marine Environmental Monitoring in Europe) proficiency testing scheme as External Quality Assurance. For Internal Quality Assurance, all analyses were carried out under full analytical quality control procedures that included the analysis of certified reference material(s) and a procedural blank sample with every batch samples analysed so that the day-to-day performance of the methods could be assessed. If levels of target analytes in the samples were outside of the range of the instrument calibration, extracts were diluted to be within range and re-analysed. Reference materials used were NIST-1944 (New Jersey Harbour sediment; National Institute of Standards and Technology, Gaithersburg, USA), PACS-2, (marine sediment, National Research Council Canada, Ontario, Canada) and TH2 (harbour sediment, Environment Canada, National Water Research Institute, Ontario, Canada). The results obtained for the reference materials were plotted as Shewhart guality control charts for each compound or trace element determined. The charts had previously been created by the repeated analysis of the above certified reference materials in the Cefas Lowestoft Laboratory using the North West Analytical Quality Analyst software[™] (Northwest Analytical Inc., USA). Warning and control limits had been defined for the charts as 2σ and $3\sigma - 2x$ and 3x the standard deviation from the mean for each compound or trace element, respectively. The results obtained for all samples were within the limits set by the control charts.

Appendix 3: Infaunal data: abrasion pressure study

A3.1. SIMPER results for the comparison of infaunal assemblages

Groups Low & High

Average dissimilarity = 71.42

	Group Low	Group High				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Galathowenia oculata	4.36	0.47	3.21	1.79	4.5	4.5
Eclysippe vanelli	4.05	0.49	2.95	3.31	4.13	8.63
Thyasira equalis	0.62	3.54	2.45	2.76	3.43	12.06
Paramphinome jeffreysii	3.56	0.62	2.41	1.68	3.37	15.43
Diplocirrus glaucus	3.58	1.08	2.3	2.26	3.22	18.65
Leucon nasica	0.1	2.12	1.69	2.83	2.36	21.02
Heteromastus filiformis	0.58	2.52	1.67	1.76	2.34	23.36
Paramphitrite birulai	1.81	0	1.52	2.04	2.12	25.49
Mendicula ferruginosa	3.15	1.37	1.49	1.91	2.09	27.57
Praxillella affinis	1.82	0	1.48	1.67	2.07	29.64
Orbinia norvegica	0.15	1.51	1.17	1.97	1.64	31.28
NEMERTEA	1.99	0.7	1.16	1.65	1.62	32.91
Eriopisa elongata	0.17	1.53	1.15	1.52	1.61	34.52
Apseudes spinosus	0	1.35	1.12	1.93	1.57	36.08
Amphiura chiajei	1.59	2.41	1.11	1.36	1.55	37.63
Falcidens crossotus	1.22	0.05	0.99	1.5	1.39	39.02
Arcturella dilatata	1.25	0.27	0.94	1.45	1.32	40.34
Amphiura filiformis	1.51	0.75	0.94	1.4	1.32	41.66
Terebellides stroemi	0.85	1.72	0.92	1.43	1.29	42.95
Myriochele	1.13	0.12	0.92	1	1.28	44.24
Levinsenia gracilis	0.32	1.27	0.91	1.48	1.27	45.51
Aricidea catherinae	1.08	0	0.88	1.22	1.23	46.74
Eudorella emarginata	0.6	1.32	0.86	1.42	1.2	47.94
Lumbrineris cingulata	1.95	1.17	0.84	1.32	1.18	49.12
Spiophanes kroyeri	1.05	1.79	0.78	1.27	1.09	50.21
Nephtys hystricis	0.25	0.96	0.74	1.22	1.04	51.25
Harpinia antennaria	0.84	1.61	0.74	1.28	1.04	52.29
Urothoe elegans	0.88	0	0.74	1.23	1.03	53.33
Notomastus	0.84	0	0.7	1.23	0.98	54.31
Ampharete lindstroemi	0.84	0	0.7	1.38	0.98	55.28
Byblis gaimardii	0	0.8	0.66	0.79	0.93	56.21
Nucula sulcata	0.77	0	0.63	1.3	0.88	57.09
Peresiella clymenoides	0.77	0	0.63	1.16	0.88	57.97
Axinulus croulinensis	0.89	1.06	0.62	1.25	0.87	58.84
Tellimya ferruginosa	0.34	0.7	0.61	1.06	0.86	59.69
Abyssoninoe hibernica	2.03	1.88	0.6	1.29	0.84	60.54

Laonice sarsi	0.79	0.61	0.59	1.17	0.83	61.36
Trichobranchus roseus	0.62	0.46	0.57	1.09	0.8	62.16
Clymenura	0.69	0.05	0.57	0.92	0.79	62.96
Pholoe baltica (sensu						
Petersen)	0.68	0.1	0.55	1.16	0.76	63.72
Nephasoma minutum	0.62	0.15	0.54	0.87	0.75	64.47
Prionospio dubia	0.64	0	0.52	0.85	0.72	65.2
Glycera alba	0.51	0.43	0.5	1.02	0.7	65.9
Anobothrus gracilis	0.61	0.12	0.5	0.96	0.7	66.6
Pista cristata	0.56	0.15	0.48	1	0.67	67.26
Goniada maculata	0.55	0.27	0.48	0.98	0.67	67.93
Pholoe pallida	0.46	0.41	0.47	0.98	0.66	68.59
Polycirrus	0.51	0.15	0.46	0.87	0.65	69.24
Scoloplos armiger	0.37	0.39	0.45	0.9	0.63	69.87
Chone	0.52	0.3	0.43	0.99	0.61	70.47
Ampelisca typica	0.49	0	0.4	0.85	0.57	71.04
Spiophanes bombyx	0.34	0.31	0.4	0.82	0.56	71.6
Maera loveni	0.12	0.42	0.39	0.66	0.55	72.14
Nephtys hombergii	0.45	0	0.39	0.89	0.55	72.69
Ophelina norvegica	0	0.45	0.37	0.71	0.52	73.21
Abra nitida	0.42	0.15	0.37	0.85	0.52	73.73
Scolelepis korsuni	0.3	0.26	0.37	0.81	0.51	74.24
Amphipholis squamata	0.39	0.09	0.36	0.6	0.5	74.74
Orbinia kupfferi	0.42	0	0.35	0.8	0.49	75.23
Ampelisca tenuicornis	0.4	0	0.33	0.8	0.47	75.7
Nephtys paradoxa	0	0.41	0.33	0.62	0.47	76.17
Owenia fusiformis	0.41	0	0.32	0.7	0.45	76.62
Lysippe sexcirrata	0.39	0	0.32	0.71	0.45	77.07
Ampelisca gibba	0.37	0	0.32	0.55	0.44	77.51
Brissopsis lyrifera	0.27	0.22	0.32	0.73	0.44	77.96
Hyalinoecia tubicola	0.37	0	0.31	0.62	0.43	78.38
Timoclea ovata	0.35	0	0.29	0.73	0.4	78.79
Scutopus ventrolineatus	0.27	0.1	0.27	0.64	0.38	79.17
Glyphohesione klatti	0.34	0	0.27	0.64	0.38	79.55
Eudorella truncatula	0.32	0	0.27	0.62	0.38	79.93
Westwoodilla (Type A)	0.25	0.15	0.27	0.69	0.38	80.3
Harpinia laevis	0.31	0	0.27	0.56	0.37	80.68
Rhodine	0.32	0	0.27	0.65	0.37	81.05
Panthalis oerstedi	0.05	0.3	0.26	0.68	0.37	81.42
Acidostoma neglectum	0.1	0.24	0.25	0.57	0.35	81.77
Thyasiridae	0.27	0	0.24	0.56	0.33	82.11
Natatolana borealis	0.24	0.1	0.24	0.58	0.33	82.44
Amphictene auricoma	0.17	0.15	0.22	0.58	0.31	82.75
Sphaerodorum gracilis	0.27	0	0.22	0.57	0.31	83.05
Ampelisca	0.2	0.1	0.22	0.59	0.3	83.36
Phoronis	0.27	0	0.22	0.47	0.3	83.66
		-	-			
Pennatula phosphorea	0.25	0	0.21	0.57	0.3	83.96
--------------------------	------	------	------	------	------	-------
Nicippe tumida	0.2	0.1	0.21	0.59	0.3	84.26
Ceratocephale loveni	0.1	0.2	0.21	0.59	0.3	84.56
Campylaspis costata	0.2	0.1	0.21	0.59	0.3	84.86
Labidoplax digitata	0.25	0	0.2	0.57	0.29	85.14
Echinocardium flavescens	0	0.24	0.19	0.48	0.27	85.42
Euclymene oerstedii	0.15	0.1	0.19	0.45	0.27	85.68
Ditrupa arietina	0.17	0.1	0.19	0.53	0.27	85.95
Prionospio cirrifera	0.2	0.05	0.19	0.54	0.27	86.22
Harpinia crenulata	0.22	0	0.19	0.48	0.26	86.48
Iphinoe serrata	0.24	0	0.19	0.47	0.26	86.74
Ampelisca macrocephala	0.1	0.15	0.18	0.53	0.26	86.99
Myrtea spinifera	0.2	0	0.18	0.5	0.25	87.25
Gnathiidae (female)	0.15	0.1	0.18	0.53	0.25	87.5
Paradoneis armata	0.22	0	0.17	0.49	0.24	87.74
Philine	0.1	0.12	0.17	0.46	0.24	87.99
Polynoidae	0.2	0	0.17	0.5	0.24	88.22
GASTROPODA	0.07	0.15	0.17	0.48	0.24	88.46
Ophelina	0.2	0	0.17	0.5	0.23	88.7
Streblosoma intestinalis	0.07	0.15	0.17	0.47	0.23	88.93
Euclymene droebachiensis	0.1	0.1	0.16	0.37	0.23	89.16
Cylichna cylindracea	0.2	0	0.16	0.5	0.23	89.39
Dipolydora coeca	0.2	0	0.16	0.5	0.22	89.61
Thysanocardia procera	0.15	0.05	0.15	0.47	0.21	89.83
Prionospio	0.19	0	0.15	0.41	0.21	90.03

A3. 2. Biological trait composition based on biomass Trait modality proportions weighted by biomass for Low and High abrasion pressure cells

















Appendix 4: BACI Study

A4.1. SIMPER output based for infaunal (square root transformed abundance data) for the Control and Impact areas within the Central Fladen NCMPA (2014 only)

Groups BACI Control & BACI Impact

Average dissimilarity = 61.02

	Group BACI Control	Group BACI Impact				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Nephasoma minutum	2.29	3.08	2.65	1.2	4.35	4.35
Galathowenia oculata	1.69	2.64	1.89	1.3	3.11	7.46
Thyasira equalis	2.32	1.61	1.59	1.18	2.6	10.05
Paramphinome						
jeffreysii	1.15	2.05	1.38	1.34	2.26	12.31
Eclysippe vanelli	1.64	2.37	1.25	1.17	2.05	14.36
filiformic	1 75	0.00	1 24	1 22	2.04	16.4
ninormus Spienbanes kroveri	1.75	0.99	1.24	1.52	2.04	10.4
	1.81	2.17	1.05	1.11	1.72	10.12
Spiophanes bombyx	1.05	1.26	1.01	1.22	1.65	19.77
Axinulus croulinensis	1.55	1.//	1	1.08	1.64	21.41
Mendicula ferruginosa	1.95	2.04	0.98	1.03	1.6	23.02
Lumbrineris cingulata	1.27	1.4	0.96	1.21	1.58	24.59
Abyssoninoe hibernica	1.51	1.46	0.96	1.14	1.58	26.17
Eudorella emarginata	1.32	1.51	0.92	1.2	1.5	27.67
Diplocirrus glaucus	1.1	0.92	0.91	1.16	1.49	29.16
Amphiura chiajei	1.09	0.76	0.9	1.23	1.48	30.64
Harpinia antennaria	0.82	0.81	0.85	1.13	1.39	32.02
Notomastus	0.86	1.14	0.84	1.2	1.38	33.4
NEMERTEA	1.22	1.12	0.81	1.14	1.32	34.73
Laonice sarsi	0.82	1.02	0.76	1.14	1.25	35.97
Trichobranchus roseus	0.72	0.95	0.75	1.16	1.22	37.19
Terebellides stroemi	1.11	1.11	0.73	1.11	1.2	38.4
Byblis gaimardii	0.59	0.61	0.73	0.96	1.19	39.59
Prionospio cirrifera	0.38	0.68	0.67	1.03	1.1	40.69
Urothoe elegans	0.32	0.72	0.67	1	1.09	41.78
Anobothrus gracilis	0.42	0.59	0.62	0.98	1.01	42.79
Glycera lapidum	0.3	0.63	0.61	1.03	1.01	43.8
Polycirrus	0.39	0.63	0.61	1	1	44.79
Glycera alba	0.54	0.44	0.58	0.96	0.94	45.74
Nephtys hystricis	0.5	0.31	0.57	0.91	0.93	46.67
Leucon nasica	0.52	0.22	0.55	0.85	0.9	47.57
Amphictene auricoma	0.41	0.45	0.54	0.91	0.88	48.45
Orbinia norvegica	0.43	0.25	0.54	0.75	0.88	49.33
Tellimya ferruginosa	0.27	0.45	0.53	0.78	0.86	50.19

Amphiura filiformis	0.28	0.44	0.52	0.79	0.85	51.05
Phoronis	0.36	0.42	0.52	0.85	0.85	51.9
Dipolydora coeca	0.32	0.45	0.51	0.87	0.84	52.73
Abra nitida	0.49	0.23	0.5	0.86	0.82	53.56
Haploops tubicola	0.18	0.5	0.5	0.88	0.82	54.38
Apseudes spinosus	0.43	0.24	0.48	0.66	0.79	55.17
Hyalinoecia tubicola	0.11	0.38	0.46	0.62	0.76	55.93
Scolelepis korsuni	0.27	0.36	0.45	0.77	0.74	56.68
Sige fusigera	0.31	0.35	0.45	0.8	0.74	57.42
Maera loveni	0.3	0.29	0.45	0.6	0.74	58.15
Myriochele	0.16	0.36	0.42	0.56	0.69	58.85
Aricidea catherinae	0.15	0.39	0.42	0.7	0.68	59.53
Thyasira obsoleta	0.21	0.3	0.4	0.65	0.66	60.19
Levinsenia gracilis	0.37	0.12	0.4	0.66	0.66	60.85
Brissopsis lyrifera	0.16	0.38	0.39	0.77	0.65	61.49
Pennatula phosphorea	0.29	0.23	0.39	0.68	0.64	62.14
Poecilochaetus						
serpens	0.14	0.35	0.39	0.73	0.64	62.77
Nephtys paradoxa	0.3	0.21	0.38	0.69	0.63	63.4
Scopelocheirus hopei	0.14	0.29	0.37	0.26	0.61	64.01
Anaitides groenlandica	0.25	0.26	0.37	0.73	0.6	64.61
Golfingia vulgaris	0.15	0.28	0.37	0.64	0.6	65.21
Augeneria	0.12	0.35	0.36	0.69	0.6	65.81
Ampharete lindstroemi	0.07	0.35	0.36	0.68	0.58	66.39
Echinocardium	0.40		0.05	0.74	0.50	
flavescens	0.19	0.3	0.35	0.71	0.58	66.98
Arcturella dilatata	0.21	0.28	0.35	0.69	0.58	67.55
Edwardslidae	0.23	0.26	0.35	0./1	0.57	68.12
Owenia fusiformis	0.16	0.26	0.34	0.5	0.56	68.69
Virgularia mirabilis	0.29	0.13	0.33	0.61	0.55	69.23
Lagis koreni	0.18	0.24	0.33	0.64	0.54	69.77
Gnathia oxyuraea	0.14	0.3	0.33	0.68	0.54	70.31
Orbinia kuptferi	0.26	0.17	0.32	0.67	0.52	70.84
Eudorella truncatula	0.09	0.26	0.32	0.5	0.52	/1.36
Dasybranchus	0.24	0.17	0.31	0.64	0.5	/1.86
Panthalis oerstedi	0.26	0.11	0.3	0.63	0.5	72.36
Chone	0.23	0.17	0.29	0.61	0.48	72.84
Lysippe sexcirrata	0.13	0.26	0.29	0.65	0.48	/3.32
Amaeana trilobata	0.19	0.16	0.29	0.54	0.48	/3.8
droebachiensis	0.13	0 24	0.29	0.6	0 48	74 28
Pista cristata	0.15	0.24	0.23	0.0	0.40	74.20
Astarte sulcata	0.19	0.18	0.20	0.05	0.10	75 18
Amphipholis squamata	0.15	0.10	0.27	0.50	0.45	75 62
lupiteria minuta	0.06	0.15	0.27	0.54	0.45 0 44	76.07
Phtisica marina	0.06	0.22	0.26	0.54	0.4 4 በ	76 5
Frionisa elongata	0.00	0.24	0.20	0.54	0.43	76 92
Enopisa ciongata	0.2	0.14	0.20	0.57	0.72	10.55

Cerianthus lloydii	0.08	0.24	0.25	0.58	0.41	77.34
Nuculoma tenuis	0.14	0.15	0.24	0.53	0.39	77.73
Gnathiidae (female)	0.09	0.21	0.24	0.56	0.39	78.12
Ophelina acuminata	0.03	0.24	0.23	0.54	0.37	78.49
Scoloplos armiger	0.17	0.06	0.22	0.42	0.36	78.85
Sthenelais limicola	0.14	0.11	0.22	0.47	0.36	79.21
Scoletoma	0.21	0.05	0.22	0.53	0.35	79.57
Falcidens crossotus	0.13	0.18	0.22	0.57	0.35	79.92
Rhodine	0.1	0.16	0.21	0.52	0.34	80.26
Streblosoma						
intestinalis	0.08	0.19	0.2	0.52	0.33	80.59
Westwoodilla caecula	0.11	0.14	0.2	0.48	0.32	80.91
Jasmineira	0.09	0.15	0.19	0.45	0.31	81.23
Ampelisca						
macrocephala	0.16	0.08	0.19	0.49	0.31	81.54
Chaetoderma						
nitidulum	0.1	0.13	0.19	0.48	0.31	81.85
Aphelochaeta	0.08	0.16	0.19	0.49	0.31	82.15
Brachydiastylis resima	0.16	0.08	0.19	0.49	0.3	82.46
Paramphitrite birulai	0.15	0.08	0.18	0.49	0.3	82.76
Diastyloides biplicatus	0.03	0.19	0.18	0.47	0.29	83.05
Harpinia crenulata	0.13	0.05	0.17	0.42	0.28	83.33
Streblosoma	0.05	0.17	0.17	0.42	0.28	83.61
Tmetonyx cicada	0.03	0.17	0.16	0.28	0.26	83.87
Typhlotanais						
aequiremis	0.13	0.08	0.16	0.42	0.26	84.13
Scolelepis tridentata	0.08	0.11	0.15	0.42	0.25	84.38
Philine	0.08	0.1	0.15	0.42	0.24	84.62
Goniada maculata Hippomedon	0.1	0.06	0.15	0.39	0.24	84.86
denticulatus	0.09	0.08	0.14	0.38	0.23	85.09
Nicippe tumida	0.1	0.07	0.14	0.39	0.23	85.32
Ampelisca gibba	0.1	0.08	0.14	0.43	0.23	85.56
Ceratocephale loveni	0.15	0	0.14	0.41	0.23	85.79
Astropecten irregularis	0.05	0.11	0.14	0.39	0.23	86.01
Euchone	0.03	0.13	0.13	0.4	0.21	86.23
Nothria conchylega	0	0.13	0.13	0.32	0.21	86.44
Golfingia	0.09	0.06	0.13	0.3	0.21	86.65
Polynoidae	0.1	0.05	0.12	0.4	0.2	86.85
Labidoplax buskii	0.1	0.03	0.12	0.35	0.2	87.04
Clymenura	0.03	0.11	0.12	0.36	0.2	87.24
Scalibregma celticum	0.03	0.11	0.12	0.36	0.2	87 / 3
Aoridae (female)	0.05	0.13	0.12	0.30	0.10	87.62
Cirratulus caudatus	0.03	0.15	0.12	0.37	0.15	07.02 97.91
Cirratulus caudatus Caballidaa	0.05	0.1	0.12	0.51	0.19	07.01
Sabelliude Eulima hilinaata		0.14	0.12	0.50	0.19	00 10
	0.05	0.09	0.11	0.30	0.18	00.19
i firacia phaseolina	0.09	0.05	0.11	0.36	0.18	88.37

Maldanidae	0.1	0.03	0.11	0.36	0.17	88.54
Harpinia laevis	0.06	0.08	0.1	0.36	0.17	88.71
Peresiella clymenoides	0.04	0.08	0.1	0.32	0.17	88.88
Anaitides rosea	0.05	0.08	0.1	0.36	0.17	89.05
Acidostoma neglectum	0.08	0.03	0.1	0.29	0.17	89.22
Timoclea ovata	0.08	0.05	0.1	0.35	0.17	89.38
Ophelina norvegica	0.08	0.03	0.1	0.32	0.16	89.55
Natatolana borealis	0.05	0.08	0.1	0.36	0.16	89.71
Jasmineira caudata	0.07	0.05	0.1	0.31	0.16	89.88
Ophelina						
cylindricaudata	0.05	0.05	0.1	0.29	0.16	90.04

A4.2 SIMPER Infauna results analysed by SIMPROF group

	Group							
Species	а	d	f	g	h	j	k	I
Thyasira equalis			3.62		3.21		2.03	2.08
Galathowenia oculata		2.45				3.16	2.57	2.56
Mendicula ferruginosa			2.12		1.41	2.5	1.98	2.26
Eclysippe vanelli				2.3		2.8	1.77	2.34
Spiophanes kroyeri			2.37	2				2.36
Paramphinome								
jeffreysii				1.49		2.81		2.07
Axinulus croulinensis						2.65	1.68	1.93
Spiophanes bombyx	1.37			2.02		1.87		
Abyssoninoe hibernica				1.14	1.83			1.83
Lumbrineris cingulata				1.38	1.06		1.77	
Nephasoma minutum								3.63
Hyalinoecia tubicola	1.57	1.57						
NEMERTEA					1.37		1.6	
Heteromastus								
filiformis					1.92			
Amphictene auricoma						1.87		
Eudorella emarginata								1.68
Jupiteria minuta		1.41						
Orbinia norvegica					1.31			
Glycera lapidum		1						

A4.3 SIMPER results comparing presence/absence data of epifaunal species for Control and Impact areas within Central Fladen NCMPA (2014 only)

Groups BACI Control & BACI Impactt

Average dissimilarity = 43.33

	Group BACI	Group BACI				
	control	inpacti	Av.Dis	Diss/S	Contrib	Cum.
Species	Av.Abund	Av.Abund	S	D	%	%
Actinauge richardi	0.27	0.58	2.46	1.05	5.68	5.68
Neptunea antiqua	0.22	0.55	2.45	1.02	5.66	11.34
Hydroid/bryozoan turf	0.54	0.88	2.37	0.92	5.47	16.81
Pectinidae	0.46	0.58	2.36	0.99	5.46	22.27
Astropecten irregularis	0.51	0.63	2.31	0.97	5.33	27.6
Sabellidae tubes	0.61	0.88	2.09	0.83	4.82	32.42
Spirontocaris						
liljeborgii	0.07	0.43	1.96	0.86	4.52	36.94
Porifera	0.1	0.4	1.87	0.83	4.31	41.25
Paguridae	0.68	0.88	1.86	0.74	4.29	45.54
Flabellum	0.22	0.35	1.84	0.83	4.24	49.78
Suberites ficus	0.29	0.2	1.67	0.76	3.86	53.64
Asterias rubens	0.22	0.28	1.67	0.76	3.86	57.51
Hormathia digitata	0.17	0.28	1.59	0.72	3.68	61.18
Nephrops norvegicus	0.22	0.08	1.23	0.58	2.83	64.01
Bolocera tuediae	0.8	0.98	1.16	0.51	2.68	66.69
Actinostola callosa	0.05	0.18	0.92	0.5	2.12	68.82
Asteronyx loveni	0.07	0.15	0.9	0.49	2.08	70.89
Virgularia mirabilis	0.98	0.83	0.89	0.48	2.05	72.94
Funiculina						
quadrangularis	0.05	0.15	0.82	0.47	1.9	74.84
Brachyura	0.07	0.13	0.81	0.46	1.88	76.72
Urticina eques	0.07	0.13	0.79	0.46	1.83	78.55
Edwardsiidae	0.12	0.05	0.79	0.43	1.82	80.37
Cerianthus lloydii	0.12	0.05	0.74	0.43	1.72	82.08
Pennatula phosphorea	0.95	0.93	0.54	0.36	1.24	83.33
Ophiura	0.02	0.1	0.5	0.36	1.15	84.47
Lithodes maja	0	0.1	0.44	0.33	1.02	85.49
Euphausiidae	0.07	0.03	0.42	0.32	0.98	86.47
Hippasteria phrygiana	0.05	0.05	0.41	0.32	0.95	87.42
Myxicola	0.1	0	0.41	0.33	0.94	88.36
Bivalvia	0.02	0.05	0.36	0.28	0.82	89.18
Scaphander lignarius	0	0.08	0.33	0.28	0.76	89.93
Caridea	0.05	0.03	0.33	0.27	0.75	90.69

A4.4. SIMPER output for infauna based on presence/absence data (2013 and 2014)

Resemblance: S17 Bray Curtis similarity

Cut off for low contributions: 50.00%

Group 2013

Average similarity: 45.56

Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Spiophanes kroyeri	1	2.6	6.61	5.7	5.7
Mendicula					
ferruginosa	0.96	2.34	3.06	5.14	10.84
Galathowenia oculata	0.92	2.15	2.17	4.72	15.55
Terebellides stroemi	0.92	2.12	2.2	4.65	20.2
Ampharete falcata	0.88	1.94	1.76	4.26	24.46
Paramphinome					
jeffreysii	0.88	1.93	1.76	4.24	28.7
Dipolydora	0.84	1.84	1.49	4.05	32.75
Axinulus croulinensis	0.84	1.84	1.49	4.04	36.79
Eudorella emarginata	0.8	1.61	1.28	3.53	40.32
Abyssoninoe					
hibernica	0.76	1.47	1.13	3.22	43.54
Harpinia antennaria	0.72	1.27	1	2.79	46.33
Eclysippe vanelli	0.72	1.27	1	2.78	49.11
Lanice conchilega	0.72	1.25	1	2.74	51.86

Group 2014

Average similarity: 42.43

Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Spiophanes kroyeri	0.91	2.24	2.06	5.28	5.28
Mendicula					
ferruginosa	0.91	2.22	2.07	5.23	10.52
Eclysippe vanelli	0.9	2.16	1.91	5.1	15.61
Axinulus croulinensis	0.84	1.86	1.46	4.39	20.01
Eudorella emarginata	0.83	1.84	1.35	4.33	24.33
Abyssoninoe					
hibernica	0.83	1.8	1.39	4.24	28.58
Thyasira equalis	0.8	1.79	1.25	4.21	32.79
Terebellides stroemi	0.79	1.69	1.21	3.99	36.78
Lumbrineris cingulata	0.78	1.61	1.18	3.8	40.58
NEMERTEA	0.76	1.56	1.13	3.67	44.25
Paramphinome					
jeffreysii	0.76	1.5	1.13	3.54	47.79
Heteromastus					
filiformis	0.74	1.48	1.05	3.48	51.27

Groups 2013 & 2014

Average dissimilarity = 61.32

	Group 2013	Group 2014				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ampharete falcata	0.88	0.03	1.16	2.23	1.89	1.89
Lanice conchilega	0.72	0	0.95	1.5	1.55	3.43
Nephasoma minutum	0	0.71	0.93	1.51	1.52	4.96
Dipolydora	0.84	0.31	0.88	1.23	1.43	6.39
Amaeana trilobata	0.68	0.14	0.86	1.25	1.4	7.78
Cerianthus lloydii	0.56	0.15	0.73	1.04	1.19	8.97
Nephtys hystricis	0.52	0.34	0.69	0.98	1.12	10.1
Phtisica marina	0.52	0.14	0.69	0.99	1.12	11.21
Amphiura chiajei	0.52	0.6	0.68	0.96	1.11	12.32
Glycera lapidum	0.52	0.4	0.68	0.97	1.11	13.43
Anobothrus gracilis Trichobranchus	0.48	0.4	0.67	0.95	1.09	14.52
roseus	0.56	0.64	0.67	0.93	1.09	15.61
Harpinia antennaria	0.72	0.55	0.66	0.92	1.08	16.69
Spiophanes bombyx	0.56	0.7	0.66	0.92	1.08	17.77
Abra nitida	0.48	0.31	0.66	0.96	1.07	18.85
Laonice	0.56	0.69	0.66	0.92	1.07	19.92
Scolelepis	0.44	0.36	0.66	0.94	1.07	20.99
Phoronis	0.44	0.34	0.65	0.93	1.06	22.04
Notomastus	0.6	0.68	0.64	0.9	1.05	23.1
Diplocirrus glaucus	0.68	0.64	0.63	0.88	1.03	24.12
Westwoodilla caecula	0.48	0.11	0.63	0.94	1.03	25.15
Polycirrus	0.28	0.44	0.63	0.92	1.03	26.18
Urothoe elegans	0.32	0.43	0.63	0.92	1.02	27.2
Byblis gaimardii	0.16	0.44	0.61	0.89	0.99	28.2
Glycera alba	0.16	0.43	0.61	0.87	0.99	29.19
NEMERTEA	0.64	0.76	0.61	0.84	0.99	30.17
Lumbrineris cingulata Heteromastus	0.64	0.78	0.58	0.83	0.95	31.12
filiformis	0.68	0.74	0.57	0.81	0.94	32.06
Leucon nasica	0.32	0.29	0.57	0.84	0.92	32.98
Amphiura filiformis	0.32	0.26	0.55	0.82	0.9	33.88
Amphictene auricoma	0.2	0.36	0.55	0.83	0.9	34.78
Thyasira equalis	0.68	0.8	0.55	0.78	0.9	35.68
Prionospio cirrifera	0.12	0.4	0.54	0.84	0.89	36.56
Arcturella	0.32	0.21	0.53	0.79	0.87	37.43
Aricidea	0.32	0.24	0.52	0.81	0.85	38.27
Owenia fusiformis	0.36	0.14	0.51	0.79	0.84	39.11
Polynoidae	0.36	0.08	0.51	0.76	0.84	39.94
Chone	0.32	0.18	0.51	0.77	0.83	40.78
Lagis koreni	0.32	0.19	0.5	0.78	0.81	41.59

The development of monitoring options at UK MPAs: Fladen Ground

Brissopsis lyrifera	0.24	0.25	0.5	0.75	0.81	42.4
Gnathia	0.28	0.2	0.49	0.74	0.81	43.2
Hyalinoecia tubicola	0.28	0.19	0.49	0.72	0.8	44
Haploops tenuis	0.36	0	0.49	0.73	0.79	44.8
Nephtys paradoxa	0.24	0.23	0.48	0.73	0.78	45.57
Scoletoma	0.28	0.15	0.47	0.71	0.77	46.35
Eclysippe vanelli	0.72	0.9	0.47	0.67	0.77	47.12
Abyssoninoe						
hibernica	0.76	0.83	0.47	0.68	0.77	47.89
Pista cristata	0.28	0.18	0.46	0.73	0.75	48.64
Paramphinome						
jeffreysii	0.88	0.76	0.45	0.64	0.74	49.38
Apseudes spinosus	0.2	0.25	0.45	0.72	0.74	50.12

A4.5 SIMPER output results comparing 2013 and 2014 epifaunal presence/absence data

Groups 2013 & 2014

Average dissimilarity = 57.02

	Group 2013	Group 2014				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Spirontocaris liljeborgii	0.74	0.22	2.78	1.26	4.88	4.88
Asteroidea	0.65	0.01	2.74	1.3	4.81	9.69
Astropecten irregularis	0.09	0.58	2.64	1.05	4.62	14.31
Unidentified Hydrozoa/Bryozoa	0.48	0.68	2.35	0.96	4.13	18.44
Pectinidae	0.57	0.48	2.29	0.96	4.01	22.45
Virgularia mirabilis	0.52	0.91	2.28	0.9	4.01	26.46
Polychaeta tubes	0.78	0.65	2.03	0.81	3.55	30.01
Actinauge	0.17	0.41	1.89	0.85	3.31	33.32
Porifera	0.35	0.24	1.85	0.82	3.25	36.58
Crangon	0.43	0.01	1.84	0.85	3.23	39.81
Paguridae	0.74	0.78	1.82	0.72	3.2	43.01
Buccinum undatum	0.39	0.01	1.79	0.75	3.14	46.14
Neptunea antiqua	0	0.39	1.65	0.76	2.9	49.04
Arctica islandica	0.35	0.01	1.57	0.68	2.76	51.8
Aequipecten opercularis	0.35	0	1.45	0.71	2.54	54.34
Aporrhais	0.3	0.01	1.44	0.65	2.53	56.88
Anthozoa	0.26	0.02	1.33	0.57	2.32	59.2
Suberites ficus	0	0.29	1.28	0.62	2.24	61.44
Crustacea	0.3	0.02	1.23	0.66	2.16	63.61
Asterias rubens	0.04	0.26	1.19	0.6	2.09	65.69
Asteronyx loveni	0.22	0.1	1.08	0.6	1.89	67.58
Bolocera tuediae	0.91	0.89	1.05	0.45	1.84	69.42
Funiculina quadrangularis	0.22	0.09	1.05	0.59	1.83	71.25
Dendrophylliidae	0.26	0	1.04	0.59	1.83	73.08
Gracilechinus acutus	0.83	0.98	1.02	0.45	1.79	74.88
Flabellum	0	0.25	1.01	0.56	1.77	76.64
Nephrops norvegicus	0.09	0.16	0.99	0.51	1.73	78.37
Sabella pavonina	0.22	0	0.95	0.51	1.67	80.04
Hormathia digitata	0	0.22	0.93	0.5	1.63	81.67
Pennatula phosphorea	0.87	0.95	0.9	0.42	1.57	83.24
Bivalvia	0.13	0.03	0.73	0.42	1.29	84.53
Majidae	0.17	0	0.66	0.45	1.16	85.69
Cerianthus lloydii	0	0.13	0.6	0.37	1.06	86.75
Pycnogonida	0.13	0	0.54	0.38	0.94	87.69
Polymastia	0.13	0	0.49	0.38	0.86	88.55
Brachyura	0	0.11	0.48	0.34	0.84	89.39
Actinostola callosa	0	0.1	0.4	0.32	0.71	90.09







JNCC/Cefas Partnership Report Series. *The Development of Monitoring Options for UK MPAs: Fladen Grounds R&D Case Study*, **No. 9**. Murray, J., Jenkins, C., Eggleton, J., Whomersley, P., Robson, L., Flavell, B. & Hinchen, H. March 2016. ISSN 2051-6711