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Marine Strategy Framework Directive Indicators for Biogenic Reefs formed by Modiolus modiolus, Mytilus edulis and Sabellaria spinulosa Part 1: Defining and validating the indicators

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This project report has been reviewed by a Project Steering Group consisting of experts from the Statutory Nature Conservation Bodies.

Executive summary

Under the Marine Strategy Framework Directive (MSFD) (Directive 2008/56/EC) Article 10, Member States are required to establish targets and indicators for the Descriptors of Good Environmental Status (GES). European and national interpretation of Descriptor 1 on Biological Diversity provides a strong steer towards indicators of biogenic structures or 'reefs'. These types of habitat are typically threatened and / or declining, are considered biodiversity 'hotspots', and are often subject to conservation management. In this report we consider how indicators would best work for habitats formed by the horse mussel (*Modiolus modiolus*), the blue mussel (*Mytilus edulis*) and the ross worm (*Sabellaria spinulosa*). We also consider what anthropogenic pressures the indicators would respond to. How these indicators would be made operational (deployment strategy, timing, resourcing, etc) is the subject of further work.

Heriot Watt University in association with Pelagica and Salacia Marine reviewed and analysed the available extant data from survey and monitoring programmes for horse mussels, blue mussels and ross worm habitats. The available data was drawn largely from work undertaken by Heriot Watt University, Scottish Natural Heritage, Natural Resources Wales, Queen's University Belfast, the Inshore Fisheries and Conservation Authorities and from studies that were part of the Regional Environmental Characterisation programme. The present analysis involved accessing thousands of records, scientific samples and images of biogenic habitats. The project also involved undertaking specific methodological trials on a horse mussel bed and running a practitioners workshop in Birmingham for 14 experts involved in blue mussel stock assessment.

Overall, the project considered methods used to assess the density and community composition of horse mussels, blue mussels and ross worms. The different monitoring and survey methods were evaluated to test the responses of the ensuing metrics to inherent temporal and spatial variation as well as the known response to anthropogenic pressures. Sources of variance were evaluated and the statistical power to detect change was tested. Recommendations for monitoring methods and metrics for horse mussel, blue mussel and ross worm dominated habitats are therefore made in this report, as are recommendations for further research and development. Detailed Procedural Guidelines for recommended monitoring methods are captured in the Appendices.

Summary of monitoring recommendations for horse mussel (*Modiolus modiolus*) habitats

- Quality standards and procedures for *M. modiolus* indicators should be scoped in order to make indicators fully operational;
- Since Water Framework Directive (WFD) multimetric indicators use a component of diversity, a trial examining the performance of the Infaunal Quality Index (IQI) (and perhaps others such as the AZTI Marine Biotic Index (AMBI)) should be investigated for possible alignment of monitoring under the WFD and MSFD;
- The effects of re-suspended sediments (from demersal fishing plumes etc) on *M. modiolus* density and community diversity should be investigated as an important pressure that is not yet accounted for in monitoring and management.

Density

- Diving surveyors should use quadrats and cell frequency counts to estimate the density of *M. modiolus*. This method is no more time consuming than any other *in situ* method but has lower inter-surveyor variability and greater statistical power to detect change than any other method. Spatial and temporal variation in beds is detectable and the method responds to physical anthropogenic pressures such as trawling;
- Imagery from towed digital stills should be used for density monitoring of *M. modiolus* beds that are too deep, large or otherwise inaccessible for divers. Again, quantification using cell frequency produces the least inter-surveyor variability and the highest statistical power to detect change. At some sites the problem of epifauna such as brittlestars obscuring *M. modiolus* may be mitigated by counting the soft coral colonies *Alcyonium digitatum* as a proxy because, in dense beds, soft corals only attach to the underlying live *M. modiolus*. This proxy is not universally applicable and should therefore be used as a last resort.

Community

- Diving surveyors should undertake quality controlled and effort limited Phase II 'samples' to monitor the community associated with *M. modiolus* reefs. Diversity indices (Shannon-Wiener diversity (H') and Pielou's Eveness (J)) derived from these methods are consistently high whereas the community itself shows high degrees of variation in composition. Diversity indices should therefore be used as the monitoring metrics and these respond to temporal and spatial variation. Impact case studies showed that these metrics also respond to anthropogenic pressures such as trawling.
 - An alternative approach would be for the divers to photographically record the same quadrats used in density studies (above) and subsequently derive diversity indices from these (analogous to the next recommended method). The relative power of *in situ* Phase II recording vs diver imagery from quadrats should be evaluated.
 - Clump sampling would provide greater community detail, greater spatial resolution and lower variance but the additional expense of sample work-up probably means it should be considered only as an infrequent ground-truthing method or if community diversity indices are subsequently found to respond to diffuse impacts such as water quality;
- High resolution towed still cameras should be used to monitor community diversity indices for extensive or deep *M. modiolus* beds where diving is impractical or comparatively too expensive. Similar epifaunal species are recorded from stills and Drop down video (DDV) but stills provide more consistent and higher quality imagery. Impact case studies showed that the diversity indices respond to anthropogenic pressures such as trawling but detection has not been demonstrated using towed still cameras.
 - Still images from towed gear need to be systematically evaluated over time in response to anthropogenic pressures before this method can be fully recommended;

Summary of monitoring recommendations for blue mussel (Mytilus edulis) habitats

 Monitoring of *M. edulis* density indicators should focus on stable (> 2 year) beds that are biogenic structures¹. Mussel spat (<10mm) should be excluded;

¹ The biotope '*Mytilus edulis* beds on reduced salinity infralittoral rock' for example, is not appropriate for Descriptor 1 biodiversity monitoring

 Overarching survey (aerial photography or side scan) is needed to target monitoring on mussel beds, and account for inter-annual variation in bed area and distribution within systems.

Density

- The MarinX ('Dutch wand') stock survey method is recommended for monitoring intertidal mussel density. The method is sensitive to spatial, temporal variation and extractive activities and is statistically robust. Systematic parallel monitoring of pressures and environmental parameters will be necessary for data interpretation.
 - Mussel coverage may be a cost effective proxy for density and would require less sample effort. However, field trials are required to establish if this is advantageous in an operational context;
- For subtidal beds density estimations should be obtained from high definition images from freshwater lens cameras (as tested for *Sabellaria spinulosa*). The method is expected to be sensitive in the same ways as intertidal *M. edulis* density methods. Percentage cover of *M. edulis* in subtidal beds may also be a cost effective proxy for density.

Community

- The MarinX walk survey method should be adapted to incorporate taxonomic analyses of pooled core samples for intertidal beds. The cores would provide both mussel density and community metrics. Standard sampling protocols for intertidal sediments can be incorporated for this purpose (see Dalkin & Barnett 2001). Based on the other studies in this report, it is likely that diversity indices such as Shannon-Wiener diversity (H'), total abundance of individuals (N) and Pielou's Evenness (J) will be appropriate metrics.
 - Field trials are needed to examine variation in community composition and biodiversity metrics in *M. edulis* beds; evaluate whether these indices are responsive and how they might be measured within an existing survey protocols;
- There is a gap in information for community indicators for subtidal mussel beds. Day, long-armed Van Veen or small (0.1m²) Hamon grabs are probably appropriate for obtaining quantitative samples. Remote imaging methodologies may also be able to capture potential density and community data. However, before field trials are initiated, consideration will need to be given to the likelihood that subtidal *Mytilus edulis* beds are sufficiently widespread features to warrant inclusion in a national monitoring programme.

Summary of monitoring recommendations for ross worm (Sabellaria spinulosa) habitats

- A precautionary, cost-effective, tiered approach to monitoring *S. spinulosa* reefs is recommended:
 - High resolution acoustic data is required to ensure sampling is properly stratified in target areas;
 - Seabed imagery is then recommended for widespread, cost-effective monitoring of density and community diversity;
 - Quantitative grab sampling is then recommended to verify the assessment based on the imagery;
 - The verification from grab sampling could itself be tiered with more rapid, costeffective *S. spinulosa* counts and *P. longicornis* counts in the first instance. More time consuming analysis of the fauna should be reported at a later date or if concerns are raised;

- Reef areas defined by high resolution acoustic data are considered the best way to identify the presence of a reef for sampling. Further work to classify *S. spinulosa* reef habitats using acoustic methods has been identified here as a key research priority for the advancement of MSFD indicator development because the published literature and data examined in this study indicate that the density of *S. spinulosa* can be as high as 13,000 individuals m⁻²;
- Seabed imagery is proposed as the most favoured monitoring method. Density estimates from seabed imagery are correlated with density estimates from grab sampling anyway and have the added advantage of not being as damaging. There is also a density diversity relationship in *S. spinulosa* reefs. However, direct assessment of species associated with *S. spinulosa* reefs is nevertheless important because compositional changes could be early warning signs of stress. Power analyses, indicating the level of sampling effort required were undertaken and indicate that monitoring with seabed imagery would be cost-effective but probably prohibitively expensive if soley based on grab sampling.
- Shannon Wiener's diversity (H') and Pielou's Evenness (J') are the most effective community metrics [diversity indices] to monitor because they are subjected to the lowest variance. The number of species (S), total abundance (N), species richness (d') and Shannon Wiener's diversity (H') increase with *S. spinulosa* reefs habitat. Pielou's Evenness, a measure of the equitability of species abundance, declined in reefs and in some cases no differences were detected. Incorporating a measure of species equitability / evenness is recommended in monitoring because it will reveal important information about the developmental stage of the reef.
- *S. spinulosa* density measures and *P. longicornis* abundance both represent potential proxy measures for the rapid assessment of reef diversity because both species increase in abundance as diversity increases. However, it is recommended that these rapid assessment measures are used to compliment full species counts from grab samples rather than to replace them entirely because compositional changes in the community may constitute an early sign of stress.
- Statutory Nature Conservation Body (SNCB) advice on the restriction of grab sampling in *S. spinulosa* reefs should be revisited in light of evidence in the present work of their likely rapid recovery and the exceedingly small scale of damage from the monitoring recommendations.
- The evidence available to assess *S. spinulosa* density and community diversity responses to anthropogenic pressures is insufficient and therefore it is not possible at this time to fully validate MSFD indicators of *S. spinulosa* reef condition. An indicator validation case study is recommended using Thanet offshore wind farm as an unimpacted / low physical impact site. With appropriate pressure data from demersal fisheries there is good reason to believe a study of this type will be successful. Research into the response of *S. spinulosa* reefs to anthropogenic disturbance (especially demersal fisheries) has been identified as a key research requirement.
- There are opportunities to investigate trait-based indicators and condition indexes (e.g. AZTI Marine Biotic Index (AMBI)). Such indicators may provide further insight into the ecological functioning of these reef habitats and may allow for a more holistic approach to monitoring.

It is likely that our ability to monitor biogenic reef habitats remotely will improve in the future as sampling technologies, such as drones, underwater camera systems, Remotely Operated Vehicles (ROVs), Automatic Unmanned Vehicles (AUVs) and acoustic systems continue to advance. Monitoring programmes should be adjusted to make the most of new technology as it becomes available.

Glossary

AFBI	Agri-Food and Biosciences Institute		
AFDW	Ash-Free Dry Weight		
AGDS	Acoustic Ground Discrimination Systems		
AMBI	AZTI Marine Biotic Index		
AUV	Automatic Unmanned Vehicle		
BACI	Before-After-Control-Impact		
BAP	Biodiversity Action Plan		
BCD	Below Chart Datum		
BIM	Bórd Iascaigh Mhara		
BSL	Below Sea Level		
CBD	Convention on Biological Diversity		
CCW	Countryside Council for Wales (now Natural Resources Wales)		
CTD	Conductivity, Tempertature, Depth		
CV	Co-efficient of Variance		
DDC	Drop down camera		
DDV	Drop down video		
Defra	Department for Environment, Food and Rural Affairs		
DF	Degrees of Freedom		
DOENI	Department of Environment Northern Ireland		
EAARL	Experimental Advanced Airborne Research LiDAR		
EEZ	Exclusive Economic Zone		
EIA	Environmental Impact Assessment		
EMS	Electronic Monitoring System		
EU	European Union		
EUBS	European Union Biodiversity Strategy		
EUNIS	European Nature Information System		
FCC	Freshwater Curtain Camera		
FOCI	Feature of Conservation Importance		
GAM	Generalized Additive Model		
GES	Good Environmental Status		
GEcS	Good Ecological Status		
GIS	Geographical Information System		
GLM	Generalized Linear Model		
GLMM	Generalized Linear Mixed Model		
GPS	Geo-Positioning System		
HAP	Habitat Action Plan		
HCI	Habitat of Community Interest		
HPI	Habitat of Principal Importance		
HWU	Heriot Watt University		
IDW	Inverse Distance Weighting		
IFCA	Inshore Fisheries and Conservation Authority		
IQI	Infaunal Quality Index		

JNCC	Joint Nature Conservation Committee	
MarLIN	Marine Life Information Network	
MALSF	Marine Aggregate Levy Sustainability Fund	
MCZ	Marine Conservation Zone	
MDS	Multi-dimensional Scaling	
MNCR	Marine Nature Conservation Review	
ММО	Marine Management Organisation	
MPA	Marine Protected Area	
MS	Mean Squares	
MSFD	Marine Strategy Framework Directive	
NASA	National Aeronautics Space Administration	
NMBAQC	National Marine Biological Analytical Quality Control	
NMHC	National Marine Habitat Classification	
NE	Natural England	
NERC (Act)	Natural Environment and Rural Communities	
NRA	National Rivers Authority	
NRW	Natural Resources Wales	
OSPAR	Oslo and Paris Convention for the Protection of the Marine Environment of the North East Atlantic	
PERMANOVA	Permutational Multivariate Analysis of Variance	
PG	Procedural Guidance	
PMF	Priority Marine Feature	
PRIMER	Plymouth Routines for Multivariate Analysis in Ecology	
PSD	Particle Size Distribution	
QA	Quality Assurance	
QC	Quality Control	
QUB	Queen's University Belfast	
REC	Regional Environmental Characterisation	
ROV	Remote Operated Vehicle	
SAC	Special Area for Conservation	
SCI	Site of Community Importance	
SIMPER	Similarity of Percentages	
SNCB	Statutory Nature Conservation Body	
SNH	Scottish Natural Heritage	
SPA	Special Protection Area	
SPUE	Sightings per Unit Effort	
SS	Sums of Squares	
SSSI	Site of Special Scientific Interest	
TAC	Total Allowable Catch	
ТМАР	Tri-lateral Monitoring and Assessment Programme (Wadden Sea)	
VMS	Vessel Monitoring System	
WFD	Water Framework Directive	

Contents

Glossai	ſy	7
1.Genei	ral introduction	1
1.1 Po	licy context	1
1.1.1	Identifying GES targets and indicators for benthic habitats in the UI	К3
1.1.2	Legislation and policy relevant to biogenic reefs	5
1.2 Aiı	ms of the present work	7
1.2.1	Objectives	7
	· · · · · · · · · · · · · · · · · · ·	
2. i ne d	evelopment of Descriptor 1 (Biological Dive	rsity)
indicato	ors for <i>Modiolus modiolus</i> reefs	9
2.1 Int	roduction	9
2.1.1	Legislation relevant to Modiolus modiolus reefs	11
2.1.2	Modiolus modiolus beds in the context of the MSFD	
2.2 De	tecting and sampling <i>M. modiolus</i> reefs	17
2.2.1	In situ methods	
2.2.2	Remote methods	
2.3 Ev	idence base	23
2.4 Me	ethods for indicator definition	
2.4.1	Density indicators	
2.4.2	Community Indicators	
2.5 Re	sults and Discussion	35
2.5.1	Density indicators	35
2.5.2	Community indicators	63
2.6 Va	lidation of Modiolus modiolus reef indicators	96
2.6.1	Sensitivity of Modiolus modiolus to anthropogenic pressures	
2.6.2	Impact case study	
2.6.3	Key findings and research needs	105
3.The d	evelopment of Descriptor 1 (Biological Dive	rsity)
indicate	ors for <i>Mvtilus edulis</i> reefs	108
3.1 Inf	roduction	108
311	Legislation relevant to Mytilus equilis beds/reefs	109
312	Mytilus edulis reef in the context of the MSED	
32 De	finition of <i>Mytilus</i> edulis bed indicators	119
321	Fvidence base	119
322	Density indicators	120
323	Community indicators	158
3.3 Va	lidation of <i>Mytilus edulis</i> bed indicators	167
331	Sensitivity of <i>Mytilus edulis</i> to anthropogenic disturbance	167
4.The d	evelopment of Descriptor 1 (Biological Dive	rsity)
indicate	ors for Sabellaria spinulosa reefs	171
4.1 Inf	roduction	171

4.1.1	Legislation relevant to Sabellaria spinulosa reefs	171
4.1.2	Sabellaria spinulosa reef in the context of the MSFD	172
4.1.3	Sabellaria spinulosa biotopes	176
4.2 Ev	idence base	178
4.2.1	Study sites	181
4.2.2	Remote sampling data	187
4.2.3	In situ sampling data	191
4.2.4	Geographical grouping	192
4.3 Me	ethods of detecting and sampling Sabellaria spinulosa reef	194
4.3.1	Method efficiency and suitability for MSFD monitoring	194
4.4 Re	elationship between the density of Sabellaria spinulosa and the assoc	iated
macrofa	unal diversity	203
4.4.1	Relationship between the density of Sabellaria spinulosa and the diversit	y of
species	s associated with the reef	203
4.5 Sa	bellaria spinulosa reef density indicators	209
4.5.1	The range of <i>S. spinulosa</i> densities found within reefs	209
4.5.2	Seabed imagery vs grab sampling for measuring S. spinulosa density	210
4.5.3	Sampling required to detect changes in the densities of S. Spinulosa	214
4.6 Sa	bellaria spinulosa reef community indicators	217
4.6.1	Fauna associated with S. spinulosa reefs in different locations and on dif	ferent
sedime	nt types	217
4.6.2	Suitability of seabed imagery for acquiring diversity indices of the fauna	
associa	ated with <i>S. spinulosa</i> reefs	238
4.6.3	Sampling required to detect changes in the diversity of S. spinulosa biog	enic
reefs	240	
4.7 Va	lidation of Sabellaria spinulosa reef indicators	243
4.7.1	Sensitivity of <i>S. spinulosa</i> reefs to anthropogenic disturbance	244
4.7.2	Case study to validate the response of potential Descriptor 1 indicators to	0
anthrop	oogenic disturbance: East Coast REC	249
4.8 Ke	ey findings and research needs	253
5.Refer	ences	. 257
C Lint -	f Annondio o	070
O.LIST O	r Appendices	. 210

1. General introduction

1.1 Policy context

The Marine Strategy Framework Directive (MSFD) (Directive 2008/56/EC) was formally adopted by the European Union in July 2008. It forms the environmental pillar of the EU's Integrated European Maritime Policy and compliments the economic and social aspects of this policy.

The MSFD outlines a legislative framework for an ecosystem-based approach to the management of human activities that supports the sustainable use of marine goods and services. The overarching goal of the Directive is to achieve 'Good Environmental Status' (GES) by 2020 across Europe's marine environment.

In order to achieve GES in a coherent and strategic manner, the Directive establishes four European Marine Regions (Article 4), based on geographical and environmental criteria. The North East Atlantic Marine Region is divided into four sub-regions, with UK coastal waters lying in two of these (the Greater North Sea and the Celtic Seas, see Figure 1.1). Each Member State is required to develop a marine strategy for their waters (Exclusive Economic Zones (EEZ) or extended Continental Shelf areas), in co-ordination with other countries within the same marine region or sub-region. This co-ordination is to be achieved through the Regional Seas Conventions, which for the UK is the OSPAR Convention for the Protection of the Marine Environment of the North-East Atlantic (www.ospar.org).

For the North-East Atlantic region, outer boundaries are indicated for the sub-regions listed in the Directive, without addressing the remaining parts of the overall OSPAR marine region (e.g. waters in the Iceland Sea, Norwegian Sea and Barents Sea; Figure 1.1).



Figure 1.1 Final draft map of MSFD marine regions and sub-regions. All EEZ boundaries shown are indicative only and are subject to an on-going consultation with Member States. The areas currently shown follow the boundaries of EEZ or other maritime zones where Member States exercise sovereign rights or jurisdiction (such as fisheries zones).

Marine strategies will be developed by Member States to protect and conserve the marine environment, prevent its deterioration, and, where practicable, restore marine ecosystems in areas where they have been adversely affected. Although the strategies should be specific to the waters of the Member State, they should also reflect the overall perspective of the marine region or sub-region because GES will be assessed at the sub-regional scale. The marine strategies to be developed by each Member State will contain:

- An initial assessment of the current environmental status of that Member State's marine waters;
- A determination of what GES means for those waters;
- Targets and indicators designed to show whether a Member State is achieving GES;
- A monitoring programme to measure progress towards GES;
- A programme of measures designed to achieve or maintain GES;

The Directive (2008/56/EC) does not describe a specific programme of measures that Member States should adopt to achieve GES, except for the establishment of Marine Protected Areas (MPAs). However, the Directive outlines 11 high-level Descriptors of GES in Annex 1 of the Directive. These are:

- 1. Biological diversity is maintained. The quality and occurrence of habitats and the distribution and abundance of species are in line with prevailing physiographic, geographic and climatic conditions.
- 2. Non-indigenous species introduced by human activities are at levels that do not adversely alter the ecosystems.
- 3. Populations of commercially exploited fish and shellfish are within safe biological limits, exhibiting a population age and size distribution that is indicative of a healthy stock.
- 4. All elements of the marine food webs, to the extent that they are known, occur at normal abundance and diversity and levels capable of ensuring the long-term abundance of the species and the retention of their full reproductive capacity.
- 5. Human-induced eutrophication is minimised, especially adverse effects thereof, such as losses in biodiversity, ecosystem degradation, harmful algal blooms and oxygen deficiency in bottom waters.
- 6. Sea-floor integrity is at a level that ensures that the structure and functions of the ecosystems are safeguarded and benthic ecosystems, in particular, are not adversely affected.
- 7. Permanent alteration of hydrographical conditions does not adversely affect marine ecosystems.
- 8. Concentrations of contaminants are at levels not giving rise to pollution effects.
- 9. Contaminants in fish and other seafood for human consumption do not exceed levels established by Community legislation or other relevant standards.
- 10. Properties and quantities of marine litter do not cause harm to the coastal and marine environment.
- 11. Introduction of energy, including underwater noise, is at levels that do not adversely affect the marine environment.

Under Article 10 of the Directive there is a requirement for each Member State to establish targets and indicators for each of the Descriptors by July 2012, designed to guide progress towards achieving GES and taking account of the continuing application of relevant existing environmental targets laid down at a national, community and international level in respect of the same waters. The Commission Decision of September 2010 on criteria and methodological standards on Good Environmental Status of marine waters (2010/477/EU) describes the criteria and indicators for each MSFD Descriptor for which Member States must develop suitable operational indicators and targets. See Box 1.1 for further background information and policy context on MSFD requirements for the development of targets and indicators.

Box 1.1 Sources of further background information and policy context.

Commission Decision of 1 September 2010 on criteria and methodological standards on good environmental status of marine waters (2010/477/EU)

(<u>http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2010:232:0014:0024:EN:PDF</u>) Describes the high level criteria and indicators identified by the Commission which underpin each of the MSFD descriptors and identifies the aspects for which operational indicators and targets must be developed.

Task Group 1 report on biological diversity (2010)

Cochrane, S.K.J., D.W. Connor, P. Nilsson, I. Mitchell, J. Reker, J. Franco, V. Valavanis, S. Moncheva, J. Ekebom, K. Nygaard, R. Serrão Santos, I. Naberhaus, T. Packeiser, W. van de Bund and A.C. Cardoso 2010. Marine Strategy Framework Directive. Guidance on the interpretation and application of Descriptor 1: Biological diversity. Report by Task Group 1 on Biological diversity for the European Commission's Joint Research Centre, Ispra, Italy. (http://www.ices.dk/projects/MSFD/TG1final.pdf)

Expert guidance produced for the Commission by ICES and the JRC in order to inform the production of the Commission Decision 2010/477/EU for Descriptor 1 on biodiversity.

OSPAR MSFD Advice manual on biodiversity (2012)

OSPAR, 2012. MSFD advice manual on biodiversity. Approaches to determining good environmental status, setting of environmental targets and selecting indicators for Marine Strategy Framework Directive Descriptors 1, 2, 4 and 6.

(<u>http://www.ospar.org/documents/dbase/publications/p00581_advice%20document%20d1_d2_d4_d6_biodiversity.pdf</u>)

Advice manual to guide OSPAR Contracting Parties in producing a regionally coordinated approach to MSFD implementation for the four biodiversity descriptors.

1.1.1 Identifying GES targets and indicators for benthic habitats in the UK

The UK Department for Environment, Food and Rural Affairs (Defra), on behalf of the Devolved Administrations (DAs), requested that the UK Healthy and Biologically Diverse Seas Evidence Group (HBDSEG²) develop options for GES targets and indicators for the three biodiversity descriptors, specifically Descriptors 1, 4 and 6 listed above. In 2011, HBDSEG produced advice to Government on these targets and indicators (Moffat *et al* 2011), drawing, where possible, on existing targets and indicators in use under other Directives and Conventions. The HBDSEG advice used the European Commission Decision of September 2010 on criteria and methodological standards on Good Environmental Status of marine waters (2010/477/EU) as a basis for structuring the targets and indicators that are relevant to benthic habitats.

The focus for this project is on Commission indicator 1.6.1 - *Condition of the typical species and communities* (but clearly there are links to other Commission indicators e.g. 1.5.1 and 1.5.2). In this case, the Commission indicator 1.6.1 - *Condition of the typical species and communities* - is very broadly described and in reality encompasses a number of operational indicators which will actually be monitored to determine community condition. An indicator is considered to be a variable which supplies information on other variables that are difficult to access and can be used to take a decision. Indicators enable us to understand a complex system and distil it into its most important aspects (Cochrane *et al* 2010).

² The Healthy and Biologically Diverse Seas Evidence Group (HBDSEG) of the UK Marine Monitoring and Assessment Strategy (UKMMAS) is responsible for coordinating and implementing monitoring and observation programmes, covering marine ecosystem health and biodiversity processes.

Table 1.1 Descriptors, criteria and indicators from Commission Decision 2010/477/EU for which advice on targets and indicators was provided for benthic habitats.

Descriptor	Criterion	Indicator
1 (Biological diversity)	1.4 Hobitot distribution	1.4.1 Distributional range
		1.4.2 Distributional pattern
	1 E Llabitat avtant	1.5.1 Habitat area
		1.5.2 Habitat volume, where relevant
	1.6 Habitat condition	1.6.1 Condition of the typical species and communities
		1.6.2 Relative abundance and/or biomass, as appropriate
		1.6.3 Physical, hydrological and chemical conditions
6 (Seafloor integrity)	6.1 Physical damage, having regard to substrate characteristics	6.1.1 Type, abundance, biomass and areal extent of relevant biogenic substrate
		6.1.2 Extent of the seabed significantly affected by human activities for the different substrate types
		6.2.1 Presence of particularly sensitive and/or tolerant species
	6.2 Condition of benthic community	6.2.2 Multi-metric indices assessing benthic community condition and functionality, such as species diversity and richness, proportion of opportunistic to sensitive species
		6.2.3 Proportion of biomass or number of individuals in the macrobenthos above some specified length/size
		6.2.4 Parameters describing the characteristics (shape, slope and intercept) of the size spectrum of the benthic community

Many of the targets and indicators proposed by HBDSEG, especially for benthic habitats are, however, not yet defined, validated or operational. In this context, the term 'defined' means that the indicator scope, scale and metrics to be measured have been identified. The term 'validated' means that the indicator has been tested to demonstrate that it actually works i.e. it can detect an impact that is known to be occurring, it is responding to the pressure of interest and it is possible to measure the change. This validation step requires data. Subsequently, an indicator becomes 'operational' when appropriate monitoring, quality standards and a process for disseminating the results have been put in place (Moffat *et al* 2011). In order to incorporate proposed benthic habitat indicators into the next MSFD reporting round, they need to be fully operational by 2014 so that they can be included in the future monitoring programme. In order to achieve this goal, considerable research and development work is needed in order to firstly define and validate the indicators before suitable monitoring is put in place. A research and development work programme has therefore been developed by HBDSEG which will make priority indicators operational for the MSFD biodiversity descriptors.

The potential biodiversity indicators identified for rock and biogenic reef habitats (Moffat *et al* 2011) were put forward by an expert sub-group of HBDSEG which began its discussions in 2011. The suite of indicators identified through this process was further refined and prioritised and it was agreed that biogenic reef indicators required research and development work.

Biogenic reefs are often the most species rich types of benthic habitat (e.g. Trigg *et al* 2011) and are singled-out for protective measures by a range of legislative and policy drivers. However, biogenic reefs are highly spatially and temporally variable (e.g. Lindenbaum *et al* 2008), occur within a wide range of environmental conditions, and data are collected using a variety of methods across UK waters without a detailed understanding of impacts. Ultimately, this means that there is work to be done to establish best practice for methods and national assessments.

1.1.2 Legislation and policy relevant to biogenic reefs

i. European Habitats Directive (92/43/EEC)

In 1992 the European Union adopted the Habitats Directive (Council Directive 92/43/EEC on the conservation of natural habitats and of wild fauna and flora) through which it meets its obligations as a signatory of the Bern Convention on the Conservation of European Wildlife and Natural Habitats. The main aim of the Habitats Directive is to promote the maintenance of biodiversity by taking measures to maintain and restore natural habitats and wild species at a Favourable Conservation Status, introducing robust protection for those habitats and species of European importance. The Habitats Directive was the first statutory driver to advocate the precautionary approach: permitting projects that have ascertained no adverse effect on the integrity of protected sites (although there are provisions for projects with overriding public interest). The Habitats Directive was initially applied out to UK territorial waters (12 nm) but following a legal challenge by Greenpeace this was extended to cover the whole of the UK Continental Shelf.

ii. OSPAR Convention

The OSPAR Convention for the Protection of the Marine Environment of the North East Atlantic was adopted in 1992 combining and updating the 1972 Oslo Convention on dumping waste at sea and the 1974 Paris Convention on land-based sources of marine pollution. The OSPAR Convention aims to provide a comprehensive and simplified approach to addressing all sources of pollution which might affect the maritime area, as well as matters relating to the protection of the marine environment. It is through this commitment that international and regional OSPAR Marine Protected Areas (MPAs) are designated. The overarching aim of the OSPAR Convention is as follows:

"Our mission is to conserve marine ecosystems and safeguard human health in the North-East Atlantic by preventing and eliminating pollution; by protecting the marine environment from the adverse effects of human activities; and by contributing to the sustainable use of the seas."

Although the OSPAR Convention was adopted in 1992, it was not until 2000 that the UK sanctioned Annex V on the protection and conservation of ecosystems and biological diversity of the maritime area. The OSPAR Biodiversity Strategy is made up of four elements:

- a. Ecological quality objectives: in support of the ecosystem approach to the management of human activities a pilot on ecological quality objectives for the North Sea has been undertaken. Consideration is now being given to extending ecological quality objectives to other OSPAR sub-regions.
- b. Species and habitats: assessments are made of species and habitats that are threatened or declining and programmes and measures are developed for their protection.
- c. Marine Protected Areas: an ecologically coherent network of well-managed marine protected areas is being created. This includes novel work on Marine Protected Areas in areas beyond national jurisdiction.
- d. Human activities: the human activities in the OSPAR maritime area which may adversely affect it are being assessed and programmes and measures to safeguard against such harm are being developed.

iii. Country Biodiversity Strategies

The Biodiversity Action Plan (BAP) (BRIG 2007) was the UK Government response to the Rio Convention on Biological Diversity (CBD) signed in 1992. It described the UK's biological resources as well as detailed plans for the protection of these resources. The establishment of devolved administrations in Scotland, Wales and Northern Ireland in 1998 led the four countries to develop their own country strategies for biodiversity and the environment, allowing conservation approaches to differ according to the different environments and priorities within the countries.

In 2007 a shared vision for UK biodiversity conservation was adopted by the devolved administrations and the UK government, and is described in 'Conserving Biodiversity – the UK Approach' (UK Biodiversity Partnership 2007). This document reflected the new key drivers for conservation action since the UK BAP was created, including the EU Gothenberg agreement in 2001 to halt the loss of biodiversity by 2010, and the findings of the Millennium Ecosystem Assessment (2005). Additionally, it outlined the need for the four countries to work together to meet shared challenges and achieve common goals, and described the requirements for future work at a UK level.

The 'UK Post-2010 Biodiversity Framework' (JNCC and Defra 2012) now succeeds the UK BAP and 'Conserving Biodiversity – the UK Approach', and is the result of a change in strategic thinking following the publication of the CBD's 'Strategic Plan for Biodiversity 2011–2020' and its 20 'Aichi targets', at Nagoya, Japan in October 2010, and the launch of the new EU Biodiversity Strategy (EUBS) in May 2011. The framework demonstrates how the work of the four countries and the UK contributes to achieving the 'Aichi targets', and identifies the activities required to complement the country biodiversity strategies in achieving the targets.

iv. Marine Strategy Framework Directive (2008/56/EC)

The Marine Strategy Framework Directive (MSFD) was adopted in June 2008 and it is concerned primarily with preserving the general health of European marine habitats and the biodiversity associated with them.

Biogenic reefs are suitable GES targets for Descriptors 1 (Biological diversity) and 6 (Seafloor integrity) under the MSFD (Cochrane *et al* 2010). Because *M. modiolus*, *M. edulis* and *S. spinulosa* biogenic reefs / beds are identified under Community (EU Habitats Directive) and International (OSPAR) legislation they are considered a 'special' habitat as defined in Table 1 of Annex III of the MSFD.

v. Marine and Coastal Access Act (2009)

The Marine and Coastal Access Act 2009 received royal assent on 12 November 2009 and introduced a new framework for managing the many demands placed on the sea, improving marine conservation and opening up access for the public to the English coast.

Provisions are made in Part 5 of the Act for designation and protection through a new type of marine protected area, called Marine Conservation Zones (MCZs). MCZs will exist alongside European Marine Sites (SACs and SPAs) to form a marine protected areas network.

vi. Marine (Scotland) Act 2010

The Marine (Scotland) Act, which was introduced to Scottish Parliament on the 29th April 2009 and gained Royal Assent on 10th March 2010, provides the legal mechanism to help

ensure clean, healthy, safe, productive and biologically diverse marine and coastal environments, managed to meet the long term needs of both nature and people, by putting in place a new system for improved management and protection of the marine and coastal environment.

The Marine (Scotland) Act 2010 introduces new powers relating to functions and activities in the Scottish marine area, including provisions enabling Scottish Ministers to designate three types of Marine Protected Area (MPA) across Scottish territorial waters: Nature Conservation MPAs – for the conservation of Scotland's most important marine biodiversity and geodiversity features; Historic MPAs – for the protection of historically important marine sites such as wrecks or national monuments; and Research/Demonstration MPAs – to demonstrate or research new methods of managing Scotland's marine environment. Scottish Ministers also have devolved responsibility under the UK Marine and Coastal Access Act 2009 for the designation of MPAs for the conservation of important marine biodiversity and geodiversity out to 200 nautical miles.

1.2 Aims of the present work

The HBDSEG rock and biogenic reef expert group identified two relevant and complementary biogenic reef indicators for possible use under MSFD Descriptor 1 (specifically, to address Commission Decision indicator 1.6.1 on condition of the typical species and communities; see Moffat *et al* 2011). These indicators are:

- a. Density of biogenic reef forming species
- b. Adundance of associated species on biogenic reefs

The aim of the present work (using largely extant data) was to define and validate the indicators for biogenic reef habitats formed by the horse mussel *Modiolus modiolus*, the blue mussel *Mytilus edulis* and the ross worm *Sabellaria spinulosa* as identified by Cochrane *et al* (2010). These habitats occur down to 200m water depth and could (if appropriate) be included in the UK's MSFD monitoring programme to be established by 2014³. Although biogenic reefs formed by other organisms such as *Ostrea edulis*, *Serpula vermicularis*, *Limaria hians* and *Lophelia pertusa* exist in the UK these reefs are not listed by Cochrane *et al* (2010) or are too deep (>200m) to be considered in the present work.

1.2.1 Objectives

The following objectives have been formulated with the intention of identifying sampling methods and associated metrics suitable for use in monitoring the density of reef building organisms and the abundance of associated reef species. Since these two aspects of biogenic reef ecology may in some cases be related, the relationship between the density of reef building taxa and associated macrofaunal diversity has also been explored with a view to identifying indicators that would act as good proxies for both.

i. Methods of detecting / sampling biogenic reef

- **a.** Investigate the methods currently being employed to sample biogenic reefs in the UK and assess their efficiency and suitability for MSFD monitoring;
- **b.** Identify potential sources of error / variance in biogenic reef sampling procedures.

³ It is anticipated that biogenic reef habitat types which occur below 200m water depth will be covered by future work planned as part of the UK Marine Biodiversity Monitoring Programme and as our understanding of these deep-sea communities improves.

ii. Relationship between the density of reef building taxa and the associated macrofaunal diversity

a. Examine the relationship between density of the reef building taxa, environmental factors, and the macrofaunal diversity associated with the reef.

iii. Density indicators

- **a.** Establish the natural range of densities (of reef forming taxa) found within biogenic reefs;
- **b.** Establish the suitability of remote and *in situ* sampling techniques for acquiring measures or proxies for density;
- **c.** Determine the level of sampling required to adequately detect changes in densities.

iv. Diversity Indicators

- **a.** Establish whether certain fauna are typically associated with biogenic reefs in different locations and on different sediment types;
- **b.** Establish the suitability of *in situ* and remote sampling techniques for acquiring measures or proxies for the diversity of fauna associated with biogenic reefs;
- **c.** Determine the level of sampling required to adequately detect changes in the diversity of fauna associated with biogenic reefs.

v. Indicator validation

a. Where possible, describe the response of the indicators to known anthropogenic pressures using extant data.

2. The development of Descriptor 1 (Biological Diversity) indicators for *Modiolus modiolus* reefs

2.1 Introduction

The horse mussel *Modiolus modiolus* (Linnaeus 1758) is a large mussel species widespread in the North Atlantic. Under certain conditions *M. modiolus* aggregates to form infaunal and semi-infaunal beds that vary in structure, extent and density, from scattered clumps (Elsäßer *et al* 2013; Moore *et al* 2013; Roberts *et al* 2011) to dense beds and elevated bioherms (Lindenbaum *et al* 2008; Wildish *et al* 1998). Horse mussel aggregations are complex, raised, tri-dimensional structures that support a relatively diverse faunal assemblage compared to surrounding areas (Fariñas-Franco *et al* 2013; Service & Magorrian 1997) therefore qualifying as biogenic reefs according to the Manual for Interpretation of European Union Habitats (European Commission 2013). It is important to distinguish between the bio-geographical distribution of *M. modiolus* as a species and the rich biogenic reefs it can create in suitable areas. The biogenic reefs formed by the horse mussel are far more restricted in their distribution than the species itself and these reefs become rarer in the northern and southern fringes of its geographical distribution (OSPAR 2009).

Fossil records suggest the current geographical range of *M. modiolus* has contracted considerably over the past 20,000 years. During the warmer late Pleistocene and early Holocene (8,000 BP) *M. modiolus* was present as far north as the Norwegian islands of Svalbard (Salvigsen 2002) where it is currently extinct (Berge *et al* 2005). To the south, *M. modiolus* communities were common in Southern Spain and Morocco as the species was part of the 'boreal invasion' of the Eastern Atlantic and Mediterranean that took place during the much colder conditions of the last glacial period (75,000 – 10,000 BP; Cortés-Sánchez 2008; Cortés-Sánchez *et al* 2011; Taviani & Bouchet 1991). Therefore, the adaptation of *M. modiolus* to cold water environments (see Lesser & Kruse 2004) is reflected in its current distribution restricted to the circumboreal in the Arctic, Pacific and Atlantic oceans (Figure 2.1; OBIS 2012).

In Western Europe *M. modiolus* ranges from the White Sea (Solyanko *et al* 2011) to Brittany and although it is recorded from the Kategatt (Dinesen & Ockelmann 2005) and the Oresund (Goransson & Karlsson 1998) in Sweden and Denmark, it is absent from the Baltic proper. It is often recorded from coastal areas of Norway (Haug *et al* 2004; Julshamn *et al* 2008), off the Faeroes (Dinesen & Ockelmann 2005) and Iceland (Ragnarsson & Burgos 2012).

In the UK, *M. modiolus* reefs, varying in their extension and condition, are found in Northern Ireland, west and north-east mainland Scotland, Orkney and Shetland, and north Wales (Comely 1978; Fariñas-Franco *et al* 2013; Hirst *et al* 2012; Mair *et al* 2000; Moore *et al* 2011; Rees *et al* 2008). They are also recorded off the Isle of Man (Bradshaw *et al* 2002; Jasim & Brand 1989; Veale *et al* 2000).



Figure 2.1 World distribution of the horse mussel *Modiolus modiolus* (Linnaeus 1758). The areas in grey indicate current distributional range extrapolated from confirmed published records. The map does not imply a continuum, being an overestimation of the distribution of *M. modiolus* within the continental shelf areas where it has been recorded (adapted from Fariñas-Franco 2012).

As a habitat, *M. modiolus* reefs correspond to four *M. modiolus* biotope types under the EUNIS classification system (<u>www.eunis.eea.europa.eu</u>; Table 2.1).

Table 2.1 *Modiolus modiolus* biotopes listed in the EUNIS classification and their correspondence with biotopes within the National Marine Habitat Classification (NMHC) of Britain and Ireland (Connor et al 2004).

EUNIS code	EUNIS Description	Equivalent NMHC code	Brief description
A5.621	<i>Modiolus modiolus</i> beds with hydroids and red seaweeds on tide-swept circalittoral mixed substrata.	SS.SBR.SMus.ModT	Modiolus modiolus beds on mixed substrata (cobbles, pebbles and coarse muddy sediments) in moderately strong currents or wave exposed areas, typically on the open coast but also in tide-swept channels of marine inlets.
A5.622	<i>Modiolus modiolus</i> beds on open coast circalittoral mixed sediment.	SS.SBR.SMus.ModMx	Muddy gravels and coarse sands in deeper water of continental seas may contain venerid bivalves with beds of <i>Modiolus modiolus</i> .
A5.623	Modiolus modiolus beds with fine hydroids and large solitary ascidians on very sheltered circalittoral mixed substrata.	SS.SBR.SMus.ModHAs	Beds or scattered clumps of <i>Modiolus modiolus</i> in generally sheltered conditions with only slight tidal movement. Typically occurs in sea lochs and the Shetland voes.
A5.624	<i>Modiolus modiolus</i> beds with <i>Chlamys varia</i> , sponges, hydroids and bryozoans on slightly tide-swept very sheltered circalittoral mixed substrata.	SS.SBR.SMus.ModCvar	Dense <i>Modiolus modiolus</i> beds, covered by hydroids and bryozoans, on soft gravelly, shelly mud with pebbles in areas of slight or moderate tidal currents. The variable scallop <i>Chlamys varia</i> is frequently found in large numbers amongst the <i>Modiolus modiolus</i> shells.

Due to their protected status and designation in UK MPAs, *M. modiolus* reefs need to be adequately monitored to ensure management measures deliver their Favourable Conservation Status (FCS). Furthermore, inclusion as a 'special' habitat type under the MSFD (Cochrane *et al* 2010) requires monitoring to assess status and inform wider management measures. *M. modiolus*, as a species is probably more resistant to direct physical impact than *M. modiolus* biotopes which are widely regarded as being sensitive to anthropogenic pressures, particularly physical damage from mobile fishing gear (Kenchington *et al* 2006; Service & Magorrian 1997; Strain *et al* 2012).

M. modiolus is characterised by fast somatic growth during the first 3-4 years, delaying maturity until reaching the 30-40mm refuge size. This strategy is probably an adaptation that has evolved as a result of heavy predation of the juvenile stages (Seed & Brown 1975). *M. modiolus* is also a long lived species with irregular recruitment and a strong affinity to settling amongst con-specifics (Fariñas-Franco *et al* 2013; Seed & Brown 1975). Therefore, as a long-lived, slow-growing k-strategist with an irregular reproductive cycle, *M. modiolus* is likely to be very susceptible to abrasion and other physical pressures (see Section 2.6). For example, bed fragmentation and decreases in parental stock resulting from direct impacts on the reefs may lead to Allee effects⁴, endangering the self-sustainability of some horse mussel beds as suggested for Strangford Lough by Elsäßer *et al* (2013) and Roberts *et al* (2011).

2.1.1 Legislation relevant to Modiolus modiolus reefs

Modiolus modiolus reefs are habitats of high conservation importance (Holt *et al* 1998; Tyler-Walters 2007). Similarly to other reef building bivalves (Coen & Grizzle 2007) *M. modiolus* are key to the functioning of benthic ecosystems, increasing habitat complexity and biodiversity and contributing to nutrient recycling and de-nitrification processes as well as substratum stabilization (Navarro & Thompson 1997; Ojeda & Dearborn 1989; Rees *et al* 2008; Witman 1985). *M. modiolus* reefs are also potential nursery grounds for commercial species (Holt *et al* 1998; OSPAR 2009; Roberts *et al* 2011). *M. modiolus* reef habitat (and not the species *per se*) is specifically listed as an OSPAR threatened or declining habitat (and therefore an MSFD 'special' habitat type) due to strong evidence of decline and threat within its European range as well as its importance in marine ecosystem functioning (OSPAR 2003). The OSPAR status for *M. modiolus* reefs is reflected in several national and European legislative instruments (Table 2.2).

Legislative Instrument	Mechanism for Protection
European Habitats Directive 1992	Special Areas of Conservation (SACs)
OSPAR Convention 1992	OSPAR Marine Protected Areas (MPAs)
Nature Conservation (Scotland) Act 2004	Scotland's Biodiversity Strategy
Natural Environment and Rural Communities Act 2006	England's Biodiversity Strategy Environment Strategy for Wales
Wildlife and Natural Environment Act (Northern Ireland) 2011	Northern Ireland's Biodiversity Strategy
Marine and Coastal Act 2009	Marine Conservation Zones (MCZ)
Marine (Scotland) Act 2010	Nature Conservation Marine Protected Areas (MPAs)
Marine Strategy Framework Directive 2008	"Good Environmental Status" targets

Table 2.2 Table summarising the legislative instruments used to protect *Modiolus modiolus* reefs in the UK.

⁴ Odum (1975, p.126) referred to the reduced reproductive or survival fitness of under-crowded populations as 'Allee effects'. Allee effects can also be described as the 'positive' relationship between any component of individual fitness and either numbers or density of conspecifics' (Stephens 1999, p.189)

i. European Habitats Directive (92/43/EEC)

Although not specifically listed as Priority Habitats, aggregations of *M. modiolus* qualify as Habitats of Community Interest (HCI) under two habitat types listed in Annex I of the EU Habitats Directive 92/43/EEC: 1160 (Large Shallow Inlets and Bays) and 1170 (Reefs). In the UK, the presence of *M. modiolus* reefs is explicitly listed as one of the qualifying sub-features chosen for the designation of the following Special Areas of Conservation (SACs):

- Strangford Lough (Northern Ireland) UK0016618: presence of *M. modiolus* reefs in the central and northern parts of the Lough. The climax association of *M. modiolus* and *Chlamys varia* occurs in soft substratum in very sheltered conditions and it is rare in the UK.
- Loch Creran (Scotland): M. modiolus reefs occurring in the upper basin of the loch.
- Pen Llŷn a'r Sarnau (Wales): reefs on the north coast of Pen Llŷn are dominated by *M. modiolus.*

Although they are not the primary reason for their designation, *M. modiolus* reefs or beds are present in other SACs: Loch Duich, Long and Alsh and Loch Laxford in western Scotland; Sullom Voe in Shetland (Mair *et al* 2010a; Marine Bio-images 2007; Moore *et al* 2011; 2013). Sparse *M. modiolus* were also recorded in The Wash and Humber SACs in England during the assessment of diversity indicators for *Sabellaria spinulosa* reefs (Figure 2.2), confirming predictions of suitable range for the habitat by Gormley *et al* (2013).



Figure 2.2 Sparse clumps of *Modiolus modiolus* with abundant bryozoan coverage in the Humber Regional Environmental Characterisation (REC) area. This image was collected as part of the Humber REC surveys (<u>http://www.cefas.defra.gov.uk/alsf.aspx</u>) and assessed as part of the indicator definition process for *S. spinulosa* reefs (Chapter 4 of above report).

ii. OSPAR Convention

The inclusion of *M. modiolus* beds on the OSPAR List of threatened and/or declining species and habitats was a result of the application of criteria for habitat sensitivity, ecological significance and overall decline. The impact caused by bottom trawls and dredges on horse mussel beds has been widely discussed (see Bradshaw *et al* 2002; Roberts *et al* 2004; Strain *et al* 2012) and is specifically mentioned as one of the reasons for their inclusion (OSPAR 2008). The sensitivity to such human pressures is greatly exacerbated by the life history of the species, i.e. poor recruitment, irregular gametogenic cycle and probably genetic depression; see for example Halanych & Vodoti (2013).

As a result of the inclusion of *M. modiolus* beds in the OSPAR List of threatened and/or declining species and habitats, a full review of the distribution of the habitat in the OSPAR marine regions, its condition and known sensitivity to anthropogenic and natural pressures and recommendations of conservation or restoration measures was published (OSPAR 2009; Figure 2.3).



Figure 2.3 Distribution of *M. modiolus* beds in territorial waters within OSPAR regions (map and annotations by E.I.S. Rees, School of Ocean Sciences, University of Wales).

iii. Country Biodiversity Strategies

In England and Wales *M. modiolus* beds are listed as Habitats of Principal Importance (HPI) under Section 41 of the Natural Environment and Rural Communities (NERC) Act 2006 (<u>http://www.naturalengland.org.uk</u>). In Northern Ireland a *M. modiolus* Habitat Action Plan is

currently under implementation (DOENI 2005). The Scottish Biodiversity List was published to satisfy <u>Section 2(4) of The Nature Conservation (Scotland) Act 2004</u>. The list includes horse mussel beds under the marine section of the list with the following comments: (i) conservation action is needed; (ii) avoid negative impacts; (iii) threatened species under OSPAR; (iv) legally protected under Annex I of the European Habitats Directive; (v) listed on the UK BAP; and (vi) important habitat for supporting marine plants and animals.

iv. Marine and Coastal Access Act (2009)

Provisions are made in Part 5 of the Act for designation and protection through Marine Conservation Zones (MCZs) alongside existing European Marine Sites (SACs and SPAs), to form a MPA network. *M. modiolus* beds are identified as a priority habitat for protection in the "Ecological Network Guidance" both as the Broad Scale Habitat - subtidal biogenic reefs A5.6 - and as a Habitat Feature of Conservation Importance (FOCI).

v. Marine (Scotland) Act 2010

M. modiolus biotopes designated as Priority Marine Features (PMFs) and MPA search features include all four EUNIS biotope types (<u>http://www.snh.gov.uk/docs/B1064114.pdf</u>; Moore & James 2011).

vi. Marine Strategy Framework Directive (2008/56/EC)

The Marine Strategy Framework Directive was adopted in June 2008 and it is concerned primarily with preserving the general health of European marine habitats and the biodiversity associated with them. As with *S. spinulosa* and *M. edulis* reefs, *M. modiolus* beds are considered a 'special' habitat as defined in Table 1 of Annex III of the MSFD due to their inclusion on the OSPAR threatened and/or declining habitats list. Biogenic structures formed by *M. modiolus* are therefore potentially suitable for the development of GES targets for Descriptor 1 (Biological diversity) and 6 (Seafloor integrity) under the MSFD (Cochrane *et al* 2010).

2.1.2 Modiolus modiolus beds in the context of the MSFD

In the UK, *M. modiolus* can be found as scattered, isolated individuals or forming semiinfaunal and infaunal beds (Figure 2.4). According to Holt *et al* (1998) only two types of *M. modiolus* aggregations satisfy the requirements of dimension, elevation above the seafloor and distinctiveness of the substratum created to be truly regarded as biogenic reefs (Figure 2.4):

- Semi-infaunal reefs or beds of varying degree of elevation. These represent the majority of the *M. modiolus* structures currently found in Scotland (Mair *et al* 2000; Moore *et al* 2006) and Northern Ireland (Roberts *et al* 2011). They consist of accumulations of shell, sediment and pseudofaeces with patchy or continuous clumps of large semi-buried adults and smaller specimens living as epifauna or amongst the byssi. In Wales, these semi-infaunal *M. modiolus* bioherms rise more than 1m above the lag gravels where they are found (Lindenbaum *et al* 2008).
- Infaunal reefs consisting of extensive, high relief aggregations of M. modiolus living deeply buried in gravels and subjected to very strong current regimes. In the Bay of Fundy (Canada), extensive horse mussel bioherms rise up to 3m above the seafloor (Wildish 2009). Infaunal reefs elevated more than 1m above the seabed have also been recorded off the Pen Llŷn in North Wales (Lindenbaum *et al* 2008). Epifaunal aggregations of *M. modiolus* are not considered biogenic reefs as they may consist only

of isolated individuals, relics from an old reef or they might not be substantial in size, have enough elevation or do not create a distinct substratum.

The Interpretation Manual of European Union Habitats defines reefs as 'solid and soft bottoms, which arise from the seafloor in the sublittoral and littoral zone. Reefs may support a zonation of benthic communities of algae and animal species as well as concretions and corallogenic concretions'. The definition for biogenic reefs clearly identifies bivalve mussel beds as biogenic reefs: '[biogenic reefs are] concretions, encrustations, corallogenic concretions and bivalve mussel beds originating from dead or living animals. i.e. biogenic hard bottoms which supply habitats for epibiotic species' (European Commission 2013). However, aggregations of *M. modiolus* regardless of density offer a hard substratum for encrusting and sessile species to settle while biodeposits (faeces and pseudofaeces) attract an array of infaunal and crevice fauna, thus significantly increasing biodiversity in species-poor substrata dominated by gravels or mud (Navarro & Thompson 1997; Rees *et al* 2008; Witman 1985). Recent experiments in Strangford Lough also confirmed a significant increase in species diversity and richness in translocated patches of live *M. modiolus* compared to control areas without them (Fariñas-Franco *et al* 2013).

The habitat engineering effect by *M. modiolus* occurs even at low mussel densities. For example, remnant impacted beds in Strangford Lough have densities of less than 20 mussels m⁻² and yet still host communities with well over 100 different taxa (Roberts *et al* 2011). However, due to the variation in structure and extent, from scattered clumps to extensive, dense beds and elevated reefs, what constitutes a bed in terms of extent and density is not clearly defined in the MSFD context. According to OSPAR (2009), patches covering more than 10m² with more than 30% cover represent the lower limits to classify an aggregation of *M. modiolus* as a 'reef'. The OSPAR Case Report for *M. modiolus* beds (2008) also follows Rees' criteria and specifically refers to beds of 30% cover or more. Reef elevation is not specifically mentioned in the OSPAR reports while Holt *et al* (1998) suggest that non-elevated structures (i.e. not arising from the seafloor) are likely to be structurally and functionally indistinguishable from elevated reefs.

One of the aims of this work is to identify the variation in density and associated community diversity for populations of *M. modiolus* across its UK range. Therefore all existing data for aggregations of *M. modiolus* were interrogated regardless of whether or not they conformed to the arbitrary 30% cover criteria (with the exception of records of single individuals). For example, the majority of remnant beds in Strangford Lough and most beds in the west of Scotland cover less than 10% of the seafloor in some areas however they remain host to diverse biotic assemblages and probably contribute to the functioning of their respective ecosystems in a similar way than do denser, more extensive beds.



Figure 2.4 Aggregations of *Modiolus modiolus* form beds that qualify as biogenic reefs under the EU MSFD. Top: Extensive bed of infaunal and semi-infaunal horse mussels in gravelly substratum (North of Point of Ayre, Isle of Man. Photo: Rohan Holt, NRW). Bottom: Dispersed remnant clumps on muddy substratum and shell fragment (Round Island Pinnacle, Strangford Lough, Northern Ireland. Photo: Jose M. Fariñas-Franco).

2.2 Detecting and sampling *M. modiolus* reefs

2.2.1 In situ methods

i. Quadrat counts and cover estimations

Quadrats are a tool commonly used in ecological studies to obtain quantitative ecological data including community diversity and abundance of conspicuous taxa. In temperate subtidal ecology quadrat sizes are usually less than 1m²; 0.25 and 0.0625m² are the most widely used sizes. Quadrats provide a standardised spatial sampling measure and facilitate robust statistical analyses of community or species change in time and space. The use of quadrats is also a cheap, non-destructive and versatile way of ensuring repeatability across surveys. Quadrats have been used to survey both *M. modiolus* densities and the associated reef community (Mair *et al* 2010a; Marine Bio-images 2007; Moore *et al* 2012; Sanderson *et al* 2008). Methods for determining density of *M. modiolus* using 0.25m² quadrats (Figure 2.5) include:

a. Cross-hair estimates (e.g. Mair et al 2000)

A 0.5m x 0.5m quadrat divided by strings or wires placed every 10cm produces a total of 16 string intersects or cross-hairs. Upon placing the quadrat onto the seafloor the diver counts only the intersections directly above live mussels thus a 100% cover is obtained when all 16 intersections are positive for *M. modiolus*. The technique is aimed at speeding-up the counting process to allow divers to cover a larger area in a single dive. The rationale behind this method is that a diver will count intersects faster than having to count all the mussels in the quadrat.

b. Cell frequency counts (e.g. Emu Ltd 2006)

A cross-stringed quadrat with 16 intersections contains a total of 25 individual square cells. Cell frequency cover estimations involve counting the cells where *M. modiolus* is present. If the same mussel appears in more than one cell, all cells are counted. The maximum cover of 100% is attained if 25 cells contain *M. modiolus*.

c. Total counts (e.g. Mair et al 2000; Marine Bio-images 2007)

The diver clears the quadrat of obscuring flora and epifauna (i.e. red algae; brittlestars) and counts all mussels present within the quadrat frame.

d. Clearance quadrats

The diver removes all the *M. modiolus* present within the quadrat. Although this is probably the most accurate counting method it is also a far more time-consuming and destructive one. It also increases the workload for the diver, reduces visibility and, although accurate, it is usually applied to population structure analyses. It can be used to calibrate estimations obtained from cell frequency/cross string cover estimate methods. For example, Mair *et al* (2000) found that percentage cover calculated using the cross-string counts (counts divided by 16 intersections) was positively correlated with clearance counts (r=0.52; P<0.05) in Loch Creran (West Scotland) where there was a maximum density of 28 mussels m⁻² and cover of less than 20%.



Figure 2.5 0.25m² wired quadrat used in the field for *in situ M. modiolus* density and cover estimations (Photo © Richard Shucksmith).

ii. Video quadrats

This method has been used to target both the faunal assemblage and conspicuous taxa such as *M. modiolus* or *Alcyonium digitatum*. Although it allows for post-processing on return to the laboratory, it can be difficult to identify and count the mussels if ophiuroids or other epifauna/epiflora are abundant. The diver uses a high-definition video camera to systematically record each quadrat sub-cell until all 25 cells have been recorded (typically taking about 20 seconds). On return to the laboratory, frames of each cell are captured and stitched together using photo editing software to create a high-resolution mosaic of the benthic community under each quadrat. These high-resolution images allow for identification and enumeration of the taxa providing quantitative diversity data and counts of conspicuous species of interest (Figure 2.6). Fauna are then recorded to the highest taxonomic resolution possible and recording rules applied to reduce variability from cryptic organisms.

Densities of *M. modiolus* can be estimated by directly counting all mussels within the frame or applying the cell frequency method instead to establish percentage cover. Ophiuroids are common in some *M. modiolus* beds (e.g. Orkney, Pen Llŷn a'r Sarnau) thus hampering mussel counts. However, it is possible to use the conspicuous epibiont *A. digitatum* to confirm the presence of an underlying *M. modiolus* and thereby estimate densities (NRW unpublished data; and see also Sanderson *et al* 2008).

In situ video recording is adequate for extracting community diversity and richness information. It is nonetheless more time consuming than other density estimation methods and consequently of limited value in spatial *M. modiolus* density mapping surveys.



Figure 2.6 High definition image of a quadrat deployed on a *M. modiolus* reef (Pen Llŷn a'r Sarnau SAC, Wales). The image is constructed by stitching video footage captures using Adobe Photoshop®.

iii. Marine Nature Conservation Review (MNCR) Phase II surveys

MNCR type surveys (see Hiscock 1996; and Holt & Sanderson 2001) have been undertaken to determine presence and relative abundance of flora and fauna along 50m x 2m transects laid by divers. Abundance is estimated using the MNCR SACFOR abundance scale (<u>http://jncc.defra.gov.uk/page-2684</u>) which is based on percentage cover of encrusting and turf taxa and number of individuals or colonies per surface area for conspicuous sessile and mobile epifauna. The method could also be used as a quick, semi-quantitative method to establish if *M. modiolus* reef density and percentage cover vary over larger areas. Sometimes the method has been supplemented by video records of stringed quadrats deployed at regularly spaced positions along the transects for later analysis in the laboratory.

2.2.2 Remote methods

i. Remote imaging systems

Digital video and still photography cameras mounted on sledges and frames are remote surveying technologies commonly used to map benthic biotopes because they allow a fast, non-destructive, qualitative assessment of epifaunal benthic communities over wide spatial areas usually out of reach for divers using standard SCUBA equipment. The video footage provides information on substrate type, depth and fauna and flora. A semi-quantitative MNCR SACFOR abundance score can also be calculated for each taxa. The combined physical and biological characteristics of each distinct video transect are then matched

against the closest national biotope type as described by Connor *et al* (2004) or its EUNIS equivalent (<u>http://eunis.eea.europa.eu</u>). Drop down video (DDV) systems are more versatile than towed systems because surveyors have some level of control over the camera system by hauling or paying out the umbilical to avoid obstacles or investigate features of interest. A digital stills camera can also be mounted alongside the video system as a means to obtain high definition digital stills of the benthic communities amenable for quantitative analysis (Figure 2.7).



Figure 2.7 Drop down sledge video and camera system employed during MPA search feature and Priority Marine Feature (PMF) surveys undertaken by HWU with Marine Scotland Science off Noss Head, north-east Scotland (Hirst *et al* 2012; equipment loaned from University Marine Biological Station, Millport).

Remote imaging methods have been used to identify the presence and extent of *M. modiolus* reef biotopes in Scotland (Hirst *et al* 2012; Moore & Atkinson 2012). In Wales, *M. modiolus* reefs found in the Pen Llŷn ar Sarnau SAC have been regularly surveyed using DDV from 2005 to 2010 (Keenan *et al* 2010) following early trials (Holt *et al* 2001, Sotheran *et al* 2004). DDV footage was analysed in both Welsh and Scottish surveys to determine the dominant biotopes, using SACFOR abundances of the associated taxa including *M. modiolus*. Although the MNCR SACFOR scales are semi-quantitative in nature they can be converted into an equivalent ordinal numerical scale thus allowing for quantitative multivariate analysis using Primer (Clarke & Gorley 2006, Keenan *et al* 2010). Historical SACFOR abundance data from DDV surveys were also used to estimate change in densities of *M. modiolus, A. digitatum* and other conspicuous epifauna (Keenan *et al* 2010).

In Strangford Lough towed sledges mounted with video and still cameras were used to obtain quantitative estimates of species composition in *M. modiolus* beds damaged by

bottom trawling and dredging (Service & Magorrian 1997). These authors used a visual fast count technique whereby total species counts are weighted based on their expected frequency of occurrence. Each video was divided into equal time intervals that were treated as sample replicates thus allowing for balanced, replicated statistical analysis. Magorrian & Service (1998) also calculated percentage cover of *M. modiolus* and other conspicuous species from photograph stills and used it in correlation and multivariate community analyses.

Although remote imagery is useful for biotope mapping and broad semi-quantitative community analyses, the acquisition of accurate density estimates for horse mussels is difficult where high densities of epibiota, mostly ophiuroids (*Ophiothrix fragilis* in particular) and ascidians, are found covering the live mussels. Estimating if the mussels are alive or dead can also be problematic if the mantle is not visible (Emu Ltd 2006; Marine Bio-images 2007).

ii. Remote Operated Vehicles (ROVs)

The lighter Drop down camera (DDC) and Drop down video (DDV) systems can be manoeuvred to some extent by a topside operator. The main advantage of Remote Operated Vehicle (ROV) - mounted camera and video systems (see Figure 2.8 for an example) is their enhanced manoeuvrability compared to drop and towed cameras. ROV mounted cameras have been used to determine *M. modiolus* reef presence and extent by Roberts *et al* (2011) in Strangford Lough and Emu Ltd (2006) in Loch Alsh. Although ROVs can ground-truth the presence and extent of *M. modiolus* reefs estimated from acoustic maps (e.g. using multibeam, side-scan sonar), their operational capabilities are limited by environmental conditions, particularly turbidity and high current speeds (>1.5 knots according to CEFAS 2002). ROVs consequently have a poor track record of detecting or estimating density of *M. modiolus*. As with other remote imaging methods, quantitative assessment of community composition and *M. modiolus* density is prone to error under conditions of high abundance of epifauna (Emu Ltd 2006; Mair *et al* 2000; Moore *et al* 2006). The difficulties associated with determining if the mussels are alive or dead have also been reported as an additional source of error in ROV footage analysis (Marine Bio-Images 2007).



Figure 2.8 ROV Seaeye system used by HWU / Marine Scotland Science during the PMF and MPA search feature validation surveys carried out off the north-east coast of Scotland in 2011 (Hirst *et al* 2012).

iii. Grabs and dredges

Van Veen and Day grabs are usually employed to sample muddy to gravelly substrates and are of limited use for *M. modiolus* reef sampling because they do not achieve appropriate penetration of the reef and their jaws get jammed open (Hirst *et al* 2012). Larger Hamon grabs can be far more successful (E.I.S Rees, pers. comm.; Boyd *et al* 2006) but relatively large research vessels are required to deploy such gear. Benthic trawls are destructive remote sampling methods commonly employed to survey biogenic reefs formed by *S. spinulosa* (Limpenny *et al* 2010) and have also been the default sampling method in the past for faunistic and population structure studies in *M. modiolus* beds (Brown & Seed 1976; Kenchington *et al* 2007; Roberts 1975; Wildish *et al* 1998). Due to the sensitivity of this biotope to physical damage (Service & Magorrian 1997) and its protected status (e.g. OSPAR Priority Marine Habitat), destructive sampling gears are not regarded as suitable monitoring technologies for *M. modiolus* reefs and are not evaluated during the current work.

iv. Acoustic methods

Acoustic methods include (Figure 2.9):

a. Single beam Acoustic Ground Discrimination Systems (AGDS)

A vessel mounted single beam echosounder is used to emit a single acoustic beam which is reflected upon hitting the seafloor and received by a hull-mounted transducer. The method works on the basis that different substrata have different acoustic reflectance properties. The most common AGDS is the RoxAnnTM system. Ground-truthed RoxAnnTM has been previously used to detect and map *M. modiolus* beds in Strangford Lough by Magorrian *et al* (1995) and in the Pen Llŷn a'r Sarnau SAC (Lindenbaum *et al* 2008; Sanderson *et al* 2001). In the former, success was limited because the acoustic maps failed to differentiate between dead and live *M. modiolus*. In the Pen Llŷn surveys by Lindenbaum *et al* (2008) the level of agreement between AGDS and Side Scan Sonar acoustic maps was appropriate.

b. Swath side-scan sonar

The system uses a towed transponder which emits high frequency acoustic signals. Backscattered acoustic pulses are reflected back to the transducer and converted into high resolution maps of the seabed surface. Side-scan sonar maps can be used to determine extent of *M. modiolus* beds as well as being capable of showing scars and marks consistent with those left by mobile benthic fishing gear (Cook *et al* 2013).

c. Swath multibeam systems

Sonar transponders emit acoustic beams in a fan shape producing a wide swath of echo data, several times the depth of water below the hull. The results are high definition bathymetric maps which can reveal seabed features in fine detail (Lindenbaum *et al* 2008; Limpenny *et al* 2010). During the current work, multibeam bathymetric maps where used to map deep *M. modiolus* beds off Noss Head, in northwest Scotland suggesting these are the largest recorded *M. modiolus* beds in Scotland (ca. 4km²; Hirst *et al* 2012).

Remote sensing acoustic technologies are a cost-effective solution used to successfully detect, map and assess the structural integrity of *M. modiolus* beds. Lindenbaum *et al* (2008; Figure 2.9) suggested a swath system (side-scan sonar) with the ability to analytically differentiate acoustic backscatter combined with robust ground truthing would provide the best method to determine extent of *M. modiolus* beds. Such a system has recently been

tested off the Ards Peninsula (Hugh Edwards, Department of Environment Northern Ireland, pers comm. 2012) but it has not yet been possible to evaluate the efficacy in the present work. However, with the current technology, acoustic methods do not yield enough quantitative information to determine the density of live *M. modiolus* within beds and as a result are not considered as potential monitoring techniques in the present work on density and community indicators.



Figure 2.9 A) Seabed composition indicated by $RoxAnn^{TM}$ (Sanderson *et al* 2001); B) Side-scan sonar output showing edge of *M. modiolus* reef features (confirmed following ground-truthing), Pen Llŷn a'r Sarnau SAC bioherms (Sanderson *et al* 2001); C) Benthic features probably attributable to scallop dredging activities, Pen Llŷn bioherms (Lindenbaum unpublished); D) Multibeam bathymetry map representing seabed features off Noss Head, north-east Scotland (Hirst *et al* 2012). Ground-truthing surveys using towed cameras confirmed changes in pattern of seabed rugosity was consistent with the presence of abundant and superabundant *M. modiolus* beds.

2.3 Evidence base

The complete list of datasets consulted during the preparation of this report is displayed in Appendix 2.1. While some reports, published and unpublished, were solely used in background research to define ranges of distribution, coverage and diversity for *M. modiolus* biotopes, most sources contained data that were later accessed and incorporated into statistical, comparative analyses aiming to define and validate density and diversity indicators. A Marine Recorder Snapshot (June 2013) was consulted to help compile a

distribution map for *M. modiolus* biotopes across the UK and extract environmental metadata to support the statistical models (Figure 2.10).



Figure 2.10 Marine recorder locations with *M. modiolus* biotopes in the UK, the Isle of Man and Ireland according to the most recent Marine Recorder snapshot (June 2013). Names indicate areas where survey reports and/or datasets were consulted and obtained during the preparation of this report (Appendix 2.1). Records in St Georges Channel (southern Irish Sea) are known to be spurious (E.I.S. Rees pers comm. 2009).

A total of 37 published and unpublished reports were collated and reviewed during the evaluation process. Sources included Heriot Watt University (HWU), Natural Resources Wales (NRW), ERT Ltd., Marine Bio-images Ltd, Emu Ltd., Isle of Man Government, Marine Scotland and Queen's University Belfast (QUB). Data extracted for indicator definition and validation related analyses corresponded to 19 different locations distributed across the UK (Figure 2.11 & 2.12):

- Shetland, Scotland (Figure 2.12A): Busta Voe, Calback and Voxter Ness, Uyea Sound and Basta Voe. (HWU and ERT Ltd.);
- Orkney, Scotland (Figure 2.12B): Shapinsay, Gutter Sound and North Cava Source: HWU;
- Noss Head, Scotland (Figure 2.12B; HWU / Marine Scotland);
- West of Scotland (Figure 2.12C): Loch Linnhe (two locations, HWU), Loch Leven (HWU), Loch Alsh (two locations, separate surveys by HWU, Marine Bio-images Ltd., Emu Ltd.), Loch Creran (HWU);
- Wales (Figure 2.12D): Pen Llŷn *M. modiolus* bioherms (NRW);
- Strangford Lough, Northern Ireland (Figure 2.11; limited access to QUB data).
- Isle of Man (Figure 2.11; NRW; Isle of Man Government).

Whilst some datasets had to be manually extracted from existing reports (i.e. Bunker 1999), most were sourced in their original raw formats from HWU and NRW:

2.3.1. Data relevant to reef density indicator development:

- Raw *M. modiolus* cover estimates from Loch Creran (1999 and 2005; Mair *et al* 2000; Moore *et al* 2005; HWU unpublished 2008 data); Loch Alsh (Emu Ltd 2006; Mair *et al* 2000; Marine Bio-images 2007; Moore *et al* 2013); and Shetland (Mair *et al* 2000; 2010b).
- DDV footage from *M. modiolus* beds off Pen Llŷn was provided by NRW alongside existing reports describing temporal change in the abundance of *M. modiolus* and associated epifauna (Keenan *et al* 2010).
- Remote digital stills from Noss Head (~200 photographs) were obtained and examined to determine their usefulness in acquiring density indicators (Hirst *et al* 2012).

2.3.2 Data relevant to reef community indicator development:

- *In situ* MNCR Phase II data were obtained from a limited number of locations including Loch Alsh; Loch Creran; Orkney and Shetland in Scotland (see Table 2.3); and Pen Llŷn in Wales (Bunker 1999).
- Historical clump community data were obtained from 15 different locations including Shetland (Hirst *et al* 2013; Mair *et al* 2000); Orkney (Hirst *et al* unpublished); Loch Linnhe (Moore *et al* 2012); Loch Alsh (Emu Ltd 2006; Mair *et al* 2000; Moore *et al* 2013); Loch Creran (Mair *et al* 2000; Moore *et al* 2006); Strangford Lough (Roberts *et al* 2004; 2011); and Pen Llŷn (Bunker *et al* 1999; Rees *et al* 2008).
- SACFOR abundance estimations derived from DDV footage from Pen Llŷn, Orkney and Shetland were also accessed and analysed.

This project therefore provided a unique opportunity to jointly analyse historical *M. modiolus* reef datasets for the first time to establish spatial and temporal variations in *M. modiolus* reef density and community diversity to develop indicators of GES in the context of the MSFD.



Figure 2.11 Map showing survey locations in the UK where *M. modiolus* density and community data were sourced during the present indicator definition and validation process. Additonal data collected by Roberts *et al* (2011) and Fariñas-Franco (2012) were used in some indicator validation analyses for comparative purposes (E). A. Shetland; B. Orkney and north-east Scotland; C. western Scotland; D. north Wales; E. Strangford Lough; F. Isle of Man.


Figure 2.12 Maps showing position and type of survey points with data extracted and used to define and validate *M. modiolus* reef density and community indicators: A) Shetland; B) Orkney and north-east Scotland; C) west Scotland and D) north Wales. Red = clump removal samples; Purple = density estimates; Green = MNCR surveys; Yellow = remote surveys (DDV/DDC). Most clump collection sites overlap with MNCR survey records.

2.4 Methods for indicator definition

2.4.1 Density indicators

i. In situ methods

An extensive literature review was undertaken to establish temporal and spatial trends in *M. modiolus* density across UK study sites. Historical density data amenable to statistical analysis were available from Scottish survey reports (Emu Ltd 2006; Mair *et al* 2000; Marine Bio-images 2007; Moore *et al* 2006; 2012; 2013). Quantitative method comparisons across the UK were not possible because most surveys exclusively used the cross-hair cover estimation method (see Appendix 2.1). Temporal and spatial variation in percentage cover using this method was nonetheless evaluated by fitting Generalized Linear Mixed Models (GLMMs) with Poisson errors and *logit* link functions. Survey location and year were used as the fixed factors and quadrat position was used as the random factor. GLMMs are able to deal with unbalanced experimental designs while temporal and spatial pseudoreplication was accounted for by including quadrat as a random factor in the models (Venables & Dichmont 2004; Zuur *et al* 2009).

In order to estimate the scale of errors associated with different density estimation methods, as well as to quantify surveyor bias, it was necessary to undertake field trials. Existing, unpublished data obtained from comparative transect surveys carried out by HWU in Loch Creran in 2008 were supplemented with density data collated in May 2013 during field trials in Scapa Flow (Orkney) as part of the current project. Three in situ counting methods (crosshairs, cell frequency and total counts; Section 2.2.1) were tested in the field. The main aim was to determine their efficiency in detecting spatial and variability between surveyors within a relatively homogenous *M. modiolus* bed located north and west of Cava Island in Orkney (Figure 2.13). The location was temporarily marked by deploying a weighed shotline onto a pre-arranged position. In total 10 cross-stringed guadrats were randomly positioned by divers around the shotline. All quadrats were pegged to the seafloor to avoid error associated with them moving. Following a detailed briefing to explain counting methodologies, a team of eight scientific divers descended the shotline in pairs every 20 minutes and carried out the counts using each counting method, also noting the time needed for each method. Further to the counts, digital photographs were acquired before the divers started as a simulation of a drop down camera system. The aim was to compare density estimations obtained from remote systems (digital stills) and from the various in situ methods.

Spatial variation in percentage cover and density of *M. modiolus* was estimated by sampling seven stations randomly positioned across the estimated extent of the *M. modiolus* bed located north-west of North Cava. The area surveyed extended ca. 600m along the 20m depth contour line northwest of the island. The positions were stratified within an area thought to be *M. modiolus* bed based on side scan sonar and exploratory dives earlier in the year (HWU unpublished). The bed was thought to extend over a moderately large area north and west of Cava and as far as the Karlesrhue wreck. At each position divers descended a shotline in pairs, randomly positioning 10 replicate quadrats onto the seabed. Photographs of each quadrat were taken by one diver followed by *M. modiolus* counts using cross-string, cell frequency and total quadrat counts (Figure 2.14). All counts were timed to determine effort and cost effectiveness.



Figure 2.13. Location of survey area (A) and sampling stations (B) for *M. modiolus* density indicators in Scapa Flow, Orkney (May 2013).



Figure 2.14 Divers counting *M. modiolus* from cross-stringed quadrats during field trials in Scapa Flow, Orkney, in May 2013 (Photo © Richard Shucksmith).

All the statistical analyses were carried out using R (R Development Core Team 2013). Intersurveyor variability was investigated by fitting GLMMs with Poisson errors and *logit* link functions. Raw counts of *M. modiolus* were the continuous, dependent variable while surveyor ID and counting method were fixed factors in the model. Quadrat ID was included as a random factor to account for potential spatial autocorrelation (Bolker et al 2009). Best fit models were selected using a stepwise progression whereby the full saturated model is compared with less complex models followed by deleting non-significant factors. The minimal adequate model (i.e. the more parsimonious model) was checked to determine the homocedasticity and normality of residual errors (Crawley 2013). Tukey's post-hoc and padjusted values were carried out to identify where the significant differences lay. Regression analyses and Spearman correlation indexes were used to determine the relationship and correlation between the proxy measurements for cover against the total counts of mussels which were regarded as the true density value. Mean density and standard deviation obtained for each method were used in subsequent power analyses. These analyses aimed to determine the number of guadrat replicates needed in future surveys to detect different percentages of change in mean *M. modiolus* density at a significant level (α) of 0.05. The power (1-beta) used (that is the probability of rejecting the null hypothesis when it is false) was set at 0.8 and 0.95 (probability of type II errors 0.2 and 0.05).

Spatial variation in *M. modiolus* cover across the North Cava bed was estimated using GLMMs with binomial error distributions. By converting intersect and cell counts to relative frequencies (dividing the counts by the number of 16 or 25, respectively) the measurements were effectively standardised allowing for statistical comparisons of horse mussel cover using binomial error GLMMs.

Finally, coefficients of variance were calculated for each method tested when comparisons were possible. This index is calculated as the coefficient between the variance expressed as

standard deviation and the mean to allow comparison between samples with dissimilar means (e.g. Lindenbaum *et al* 2002).

ii. Remote methods

The usefulness and accuracy of remote imaging methods to capture density indicator metrics were also evaluated. Towed, high definition digital camera stills collated during Hirst et al's (2012) surveys at Noss Head, in north-east Scotland (see Figure 2.12B), had not been previously processed for either community or density indicators. Out of the total 11 stations with records for *M. modiolus*, seven were classified as *M. modiolus* beds. In total 57 high resolution (240dpi) photographs of *M. modiolus* beds located > 40m bsl) were collated using a sledge frame mounted Kongsberg camera (see Hirst et al 2013 for details). The high definition digital stills were used to evaluate density and percentage cover of the areas where *M. modiolus* was previously estimated as abundant or superabundant (SACFOR scores) following the analyses of the towed video footage. In total five replicate images were randomly chosen for each sampling station. The images were processed using the open source software Image J® (Schindelin et al 2012). The method chosen was similar to that described for film photographic stills by Service in Davies et al (2001) whereby a grid was laid over the picture and counts were made of the number of cells containing *M. modiolus*. Cross-hair intersects counts and total counts were also tested to evaluate the overall effectiveness of counting methods previously tested by divers in situ (Figure 2.15).



Figure 2.15 Noss Head *M. modiolus* bed high definition digital photograph analysed using Image J[®]. Note grid superimposed on the image to help during the counting process. Dots represent individual, live *M. modiolus* being counted.

Cell frequency and intersect count data were standardised to relative frequencies (percent cover) while total counts were converted to relative abundances (mussels per m²) based on the field of view of the drop down camera at each sampled station. Grid dimensions changed as the field of view area covered by the camera also changed, from 0.3 to 0.7m² approx. The variation in cover and density across the survey area and differences in estimates obtained from each method were investigated by means of GLMMs. Post-survey power analyses and the correlation/regression between the counting methods were calculated

using similar approaches to those described for the *in situ* estimations by divers. Correlation analyses were undertaken using non-parametric Spearman indexes. Bootstrapping was necessary to determine the variance, mean and confidence intervals for regression and correlation parameters due to the scarcity of data.

Power analyses were undertaken to determine the sampling effort needed to detect 10-40% change in mean density at significance alpha 0.05 in a dense bed of *M. modiolus* using Noss Head as a case study. Co-efficients of variance were calculated for each counting method to assess variability in density estimates using remote systems (e.g. Lindenbaum *et al* 2002). Towed video footage from 2004-2010 obtained by NRW from *M. modiolus* beds off Pen Llŷn, in North Wales, had already been processed by Keenan *et al* (2010). Their reports were evaluated and the results described in Section 2.4.2c.

In certain biotopes *M. modiolus* density might be estimated using a proxy indicator if there is a correlation between *M. modiolus* density and abundance of a conspicuous epifaunal organism attached to the mussel. A conspicuous proxy might be useful when horse mussels themselves become periodically obscured by ophuroids for example (see Sanderson et al 2008; Figure 2). Changes in cover of conspicuous proxy epifauna, however, could also be related to: a) natural seasonal changes, or; b) decreases following anthropogenic impact (Cook et al 2013; Roberts et al 2011) and substantial variation due to the former would make the indicator unhelpful. NRW had already investigated the use of *in situ* video guadrats using counts of Alcyonium digitatum as a proxy species based on the correlation described by Sanderson et al (2008). Data for A. digitatum and M. modiolus without A. digitatum from video footage of fixed quadrats positioned on *M. modiolus* beds off north Pen Llŷn, Wales and the Isle of Man were available from surveys undertaken by divers between 2004 and 2010. The raw data were investigated in the current study to determine whether changes in A. digitatum abundance were matched by those of live M. modiolus, and hence verify its suitability as a proxy for the density of *M. modiolus* (at least for dense beds with abundant *A. digitatum*). It could be hypothesised, based on evidence from side-scan sonar surveys, that for a protected area such as Pen Llŷn a'r Sarnau SAC where physical impact (e.g. demersal fishing) had been excluded, the relationship between A. digitatum and M. modiolus would be relatively stable over time. To test that hypothesis, numbers of A. digitatum and visible M. modiolus were used as dependent variable in Poisson GLMMs with logit link functions using taxa and year as fixed, categorical effects and quadrat as the random factor.

As with the *in situ* methods, all statistical analyses of remotely acquired data were carried out using R (R Development Core Team 2013).

2.4.2 Community Indicators

i. In-situ methods

a. Collection of *M. modiolus* clumps by divers

Replicate clumps were removed from *M. modiolus* beds surveyed in 15 different locations in the UK (Figures 2.11 & 2.12): a diver carefully removed a clump of *M. modiolus* of a size that can fit within a 5L plastic bucket. The bucket was sealed with a lid, placed inside a bag and recovered to the surface. On return to the laboratory the samples were sieved through a 0.5mm mesh and all taxa retained were identified to species level and counted. The only exception was for samples collected from the horse mussel beds in Pen Llŷn a'r Sarnau SAC, obtained from a 25 x 25cm area using a suction sampler (see Rees *et al* 2008). Although the sample size was different it was decided to include these data as an indication of the effect of sample size on community composition and associated univariate diversity, richness and evenness metrics.

b. In-situ MNCR surveys

Phase II MNCR type semi-quantitative SACFOR data obtained from historical dive surveys carried out at 15 different locations were used in the community indicator analyses (Figure 2.11).

ii. Remote methods

Data were obtained from DDV surveys carried out by NRW in Pen Llŷn a'r Sarnau SAC (nine stations) and by HWU in Shetland (22 stations), Noss Head (11) and Orkney (19). Community data were recorded in each case using SACFOR abundance scales.

iii. Statistical analyses

Taxonomic data used to define and validate community indicators were collected using slightly different sampling approaches by different organisations over a number of years thus requiring a careful process of data cleaning and standardisation (see list of data sources in Appendix 2.1). Quantitative species abundance data were standardised against the World Register of Marine Species (WoRMs; <u>http://www.marinespecies.org/</u>) to ensure taxonomic consistency across datasets. These data were also truncated to remove juveniles, which are often but not always recorded. Taxa such as cumaceans, ostracodes, nematodes and tubificid oligochaetes were removed from the analysis because their very high abundance in some surveys (e.g. HWU 2012 surveys in Shetland) and absence from others was due to differing taxonomic resolution in sample analysis and/or the serendipitous retention of small organisms that should not have been retained by the sieve.

a. Multivariate analyses

The multivariate analyses were carried out using PRIMER 6 with the PERMANOVA + extension (Anderson *et al* 2008; Clarke & Gorley 2006). Data was generally square or fourth root transformed depending on the abundance of certain dominant taxa (i.e. *Balanus* spp., capitellid polychaetes, *Corophium* spp. amphipods, ophiuroids and ascidians were sometimes 2 and 3 orders of magnitude more abundant than most other taxa). A Bray-Curtis similarity matrix was created for all the samples from the locations used in the analyses. The matrices were later ordinated using non-metric multidimensional scaling (MDS) and represented by plots to visualise the groupings between the sample stations. Cluster

analysis was undertaken using SIMPROF tests (set at $\alpha = 0.05$) to determine significant groupings in the communities. The data were tested by fitting PERMANOVA mixed models using location as the categorical factor and depth, wave exposure and tidal current as continuous factors, upon subjectively converting the categorical values to the numerical values displayed in Table 2.3 (see McBreen *et al* 2010). The effect of *M. modiolus* abundance on the community associated with the clump and MNCR data were investigated by adding *M. modiolus* abundance as a co-variate in the PRIMER analyses for clumps. The environmental matrices used in the mixed PERMANOVA models were calculated using Euclidean distance as resemblance indexes following normalization of the variables.

Taxanomic abundance in both the DDV and *in situ* MNCR Phase II type dive surveys was semi-quantitative in nature therefore MDS plots and PERMANOVA analysis were undertaken using Bray-Curtis similarity indexes based on a conversion of the SACFOR scale to a ranked numerical category. This method is similar to that used to analyse the communities associated with the *M. modiolus* bed off Pen Llŷn (Keenan *et al* 2010). SIMPER analyses were used in all cases to determine which taxa typified resulting groups and which were responsible for most of the dissimilarities between the groups.

Variable	Categorical value	Assigned numerical value
SACFOR abundance scale	Superabundant (S)	6
	Abundant (A)	5
	Common (C)	4
	Frequent (F)	3
	Occasional (O)	2
	Rare (R)	1
	N (Absent)	0
Exposure	Extremely exposed	7
	Very exposed	6
	Exposed	5
	Moderately exposed	4
	Sheltered	3
	Very sheltered	2
	Extremely sheltered	1
Tidal current	Very strong	3.5
	Moderately strong	2.5
	Strong	1.5
	Weak	0.5
	Very weak	0

Table 2.3 Equivalences between categorical variable classes (SACFOR / Exposure / Tide) and discrete ordinal values used to statically evaluate community indicators. For percentage cover and density equivalences see http://incc.defra.gov.uk/page-2684.

b. Univariate analyses

A suite of diversity indices were calculated to investigate the effect of *M. modiolus* on the associated faunal assemblages and to determine the suitability of *in situ* and remote techniques in obtaining metrics for community indicators. These indices, described in Appendix 3, are total number of taxa (S), total abundance of individuals (N), Margalef's species richness (d), Shannon-Wiener's diversity (H') and Pielou's evenness (J). All univariate indices were calculated using the PRIMER v6 software package (Clarke & Gorley 2006). PERMANOVA was used to investigate the importance of sampling location, depth, exposure and *M. modiolus* abundance in the observed variability in univariate community indices. To account for non-independence of errors as result of spatial and temporal

autocorrelation, quadrats and sampling site (nested within location) were incorporated as random factors in the PERMANOVA models.

Diversity indices estimated from DDV, MNCR dive surveys and clump removal samples were compared if possible (Shetland, Orkney and Pen Llŷn *M. modiolus* beds). The approach involved fitting GLMM models which are suited to unbalanced, non-independent data (Anderson 2005; Bolker *et al* 2009; Venables & Dichmont 2004; Zuur *et al* 2009). Retrospective power analyses were undertaken for all three methods to determine the sampling effort needed to detect 10-40% change in mean diversity at significance alpha 0.05. The power (1-beta) used (ie the probability of rejecting the null hypothesis when it is false) was set at 0.8 and 0.95 (probability of type II errors 0.2 and 0.05). Regression and correlation between *M. modiolus* density and diversity indices were investigated by fitting linear and non-linear regression models to determine best fit to the scatterplots. Finally, coefficients of variance were calculated for each index and method tested to determine the error introduced by each method.

2.5 Results and Discussion

2.5.1 Density indicators

i. Existing data on the density of *M. modiolus* beds

One of the earliest published records of *M. modiolus* bed densities are from guadrats taken in 1976 at Long Sheelah, in the North Basin of Strangford Lough where there were 276 mussels m⁻² (Roberts *et al* 2004). Roberts (1975) estimated that, in Strangford Lough's South Basin, *M. modiolus* occupied an area in excess of 1km² at approximate densities of 170 mussels m⁻². These historical records are of particular importance because they put into context recent monitoring in Strangford Lough by Roberts et al (2011) who found that, following intense impact from mobile fishing gear, remnant beds had experienced a significant decrease in mussel densities (as low as 5 mussels m⁻² at Long Sheelah). Tightly packed semi-infaunal *M. modiolus* forming clumps on undulating structures (Wildish et al 1998; Lindenbaum et al 2008; Sanderson et al 2008) can reach densities of 600 mussels m⁻² in Pen Llŷn a'r Sarnau SAC (Rees et al 2008). In Scotland, densities range from 28 mussels m⁻² in the north basin of Loch Creran to 150 mussel m⁻² over a 2km² area in Sullom Voe. Shetland (see Table 2.4). However, the majority of *M. modiolus* beds are structurally fragmented and consist of clumped aggregations of variable densities of infaunal and semi-infaunal mussels among clear patches of mud and empty shells, particularly towards the edges of the beds, (Mair et al 2000; Moore et al 2006; Moore et al 2013).

Most estimations of density in UK beds have been calculated as percentage cover using the cross-hair method (see Table 2.4). Mair *et al* (2000) suggested that cross-hair counts were a faster, non-destructive alternative to *M. modiolus* abundance estimations using quadrat clearance carried out by divers. They found a significant correlation between the number of mussels in the quadrats and the cross-hair counts (Spearman's correlation index r = 0.542; p<0.05) thus concluding the percentage cover calculated using this method was a suitable proxy for mussel density. Percentage cover estimations carried out in Scotland suggest most beds across the range (Loch Creran, Loch Alsh, Shetland) cover no more than 45% of the seafloor at their densest points (Loch Alsh in 1999; Mair *et al* 2000) while the majority range between 1 and 36%. Cross-hair counts therefore suggest a substantial variation in percentage cover across the distribution range as well as within each bed surveyed (Emu Ltd 2004; Marine Bio-images 2007; Table 2.4).

SACFOR scale estimations have been used by Hirst *et al* (2013) from DDV off Noss Head (north-east Scotland) where *M. modiolus* covered more than 80% of the seabed. Similar estimations are given by Bunker *et al* (1999) for the Pen Llŷn *M. modiolus* structures. Outside the UK, extensive, dense beds have also been described in Iceland where densities of up to 150 mussels m⁻² have been estimated (Ragnarsson & Burgos 2012). Epifaunal aggregations of *M. modiolus* at densities ranging between 140 and 280 mussels m⁻² have been recorded as bioherms in the Bay of Fundy (Kenchington *et al* 2007) and on rocky substrate off the coast of New England by Witman (1985).

Table 2.4 Range in M. modiolus density and percentage cover throughout its known distribution according to existing literature. The majority of Scottish records originated from SNH commissioned surveys using cross-hair quadrat counts.

Location	Site	Density (mussels m ⁻²)	%Cover (mean)	Method	Source
Canada	Bay of Fundy	4-158	40	Dredge	Wildish & Fader (1997); CPAWS report
Shetland	Busta Voe	45	8-16	Clearance/Cross- hairs	Mair <i>et al</i> (2000)
Scotland	Ardyne (Firth of Clyde)	10		Dredges	Comely (1978)
Scotland	Firth of Lorne	37		Dredges	Comely (1978)
Iceland		20->150		Photographs	Ragnargsson & Burgos (2012)
lsle of Man		20-40		Not known	Holt <i>et al</i> (1998)
Scotland	String Rock (Loch Alsh)	106	45 (29)	Clearance/Cross- hairs	Mair <i>et al</i> (2000)
Scotland	String Rock (Loch Alsh)		1-22.5 (11.5)	Cross-hairs	Marine Bio-images (2007)
Scotland	String Rock (Loch Alsh)		5-24.8 (15)	Cross-hairs	Emu Ltd (2004)
Scotland	String Rock (Loch Alsh)		3-18 (6.83)	Cross-hairs	Moore <i>et al</i> (2013)
Scotland	Loch Creran	28	20	Clearance/Cross- hairs	Mair <i>et al</i> (2000)
Scotland	Loch Creran		0.6-34 (6.3)	Cross-hairs	Moore et al (2006)
Scotland	Creagan Narrows (Loch Creran)	4		Dredge	Comely (1978)
Scotland	Port Appin (Loch Linnhe)	10		Dredge	Comely (1978)
USA	Maine	14.4		Direct quadrat counts	Ojeda & Dearborn (1989)
USA	New England	140-280	26-57	Direct quadrat counts	Witman (1985)
Scotland	Noss Head		SACFOR scale C-S (20 to >80%)	DDV estimates	Hirst <i>et al</i> (2012)
Wales	Pen Llŷn (ridges)	600		<i>In situ</i> counts (air lift suction)	Sanderson <i>et al</i> (2008); Rees <i>et al</i> (2008)
Wales	Pen Llŷn (troughs)	6		In situ counts	Sanderson <i>et al</i> (2008)
Wales			SACFOR A (40-79%)	MNCR survey	Bunker <i>et al</i> (1999)
Northern Ireland	Long Sheelah (Strangford Lough)	276		quadrats	1976 data reanalysed by Roberts <i>et al</i> (2004)
Northern Ireland	Black Rock (Strangford	170		Dredge	Roberts (1975)

Location	Site	Density (mussels m ⁻²)	%Cover (mean)	Method	Source	
	Lough)					
Northern Ireland	Black Rock (Strangford Lough)	42		Clearance quadrats	Roberts et al (2004)	
Northern Ireland	Long Sheelah (Strangford Lough)	69		Clearance quadrats	Roberts <i>et al</i> (2004)	
Northern Ireland	Round Island (Strangford Lough)	77		Clearance quadrats	Roberts <i>et al</i> (2004)	
Northern Ireland	Long Sheelah (Strangford Lough)	60		Clearance quadrats	Roberts <i>et al</i> (2011)	
Northern Ireland	Black Rock (Strangford Lough)	32		Clearance quadrats	Roberts <i>et al</i> (2011)	
Northern Ireland	Round Island (Strangford Lough)	80		Clearance quadrats	Roberts <i>et al</i> (2011)	
Shetland	U /	37		Dredge	Comely (1978)	
Shetland	Calback Ness (Sullom Voe)	62-150	3-36	Cross-hairs	Mair <i>et al</i> (2010)	
Shetland	Voxter Voe (Sullom Voe)	1	1-10	Cross-hairs	Mair <i>et al</i> (2010)	
Shetland	Calback Ness (Sullom Voe)		6-30	Cross-hairs	Mair & Sanderson unpublished 2012 data	
Shetland	Voxter Voe (Sullom Voe)		1	Cross-hairs	Mair & Sanderson unpublished 2012 data	

About half the records for *M. modiolus* beds (Table 2.4) had >30% cover and a third had densities >100m⁻². Two thirds of the records contain densities >50m⁻². Undoubtedly there will be methodological biases in these various measures (and it is most likely that dredging will under-record) but, given that many records are derived from protected areas, indicators that can can be defined and validated for *M. modiolus* beds of either >30% cover or >50 mussels m⁻² will likely be the most useful to nature conservation management.

a. Analyses of spatial variability in *M. modiolus* reef density indicators in the UK

Differences in percentage cover estimated using cross-string counts were investigated using data from Loch Alsh, Loch Creran and Shetland (see Section 2.2 for sources). Additionally, unpublished *M. modiolus* percentage cover estimates were obtained from cross-string surveys undertaken by HWU in Loch Creran in 2008 and Orkney (North Cava) during field trials in May 2013 as part of the present project. Median percentage cover values varied between 5% (Loch Creran) and 40% (Loch Alsh, 1999 surveys) while maximum covers of ca. 60% were recorded in Loch Alsh in 1999 and in the beds recently surveyed off North Cava (Figure 2.16). The results of the binomial GLMM indicated variation was significant only between Loch Creran and Orkney *M. modiolus* beds (z=4.09; P<0.001) but not between other beds.



Figure 2.16 *M. modiolus* percentage cover recorded in surveys across Scotland. Cover was estimated from cross-hair quadrat counts. Box plots show interquartile range, median and maximum / minimum observed values as whiskers (1.5 times the interquartile region). Black dots indicate values above and below the 1.5 times the 3rd and 1st quartiles, respectively.

The decline in percentage cover of *M. modiolus* reported in co-located quadrats in Loch Alsh by Emu Ltd (2004); Marine Bio-images (2007) and Moore *et al* (2013) were also investigated as part of the present work. The result of the binomial GLMMs indicated a significant temporal effect in percentage cover ($F_{(3,30)}$ =18.57; P<0.001). Post-hoc tests showed that the decline in percentage cover was only significant between 1999 (mean=32%) and all subsequent years. Cover estimations recorded in 2012 were significantly lower than those recorded in 2004 (mean cover = 8% and 16%, respectively). However, although the trend continued between 2006 and 2012 the declines between these years were not statistically significant. Variation in percentage cover for *M. modiolus* beds surveyed in Loch Creran between 2005 and 2008 (~5% in both) was not statistically significant (binomial GLMMs; t=-0.32; P=0.74).

Despite *M. modiolus* being a slow-growing and long-lived species (Fariñas-Franco 2012; Richardson *et al* 2001; Seed & Brown 1975), *M. modiolus* beds are most likely to be stable features in the absence of physical anthropogenic impacts (Holt *et al* 1998; Lindenbaum *et al* 2008). Previous reports by Marine Bio-images (2007) and Emu Ltd (2006) could not satisfactorily interpret the decline observed in Loch Alsh *M. modiolus* beds: there were no obvious signs such as dredge tracks and benthic fishing was probably excluded by boat traffic. In 2004 divers were seen landing sacks of *Buccinum*, *Modiolus* and various other shellfish at Loch Alsh but it was not clear if this activity was a rare or common event. However, Moore *et al* (2013) described a "cogent pattern of a decline in *Modiolus* density consistent with an increase in *Limaria* density". Competition between the two reef building species could be a plausible explanation for the observed dynamics.

The significant variation in percentage cover between the Loch Creran and Orkney beds probably reflects structural differences between reefs rather than being the result of anthropogenic pressures in the former. The decline in percentage cover in Loch Alsh also suggests that this supposedly stable habitat can sometimes vary. Inaccurate re-location of sample stations, insufficient replication and surveyor bias may have contributed to the variance recorded at Loch Alsh (see Emu Ltd 2006; Marine Bio-images 2009) but these sources of error do not satisfactorily explain the ongoing declines in density. Overall, it is evident from the Loch Alsh case study that changes in density can only be attributed to anthropogenic activity and therefore be relevant to management with additional corroborating activity data.

Existing historical data collected from the String Rock beds in Loch Alsh were used to carry out power analyses using α =0.05 and powers (1- β)=0.95 and 0.80. Bootstrapped mean % cover of *M. modiolus* was estimated as 16% with a corresponding standard deviation of 15%. Unsurprisingly, the number of replicates necessary to detect a 10% change in percentage cover was exceedingly high (n=1144): Changes of 20, 30 and 40% respectively needed 287, 128 and 73 replicate 0.25m² quadrats across the area occupied by *M. modiolus* (see maps in Moore *et al* 2013; Mair *et al* 2000).

Other density metrics such as cell frequency counts or total quadrat counts may provide more accurate estimations with lower sampling effort. These methods were therefore tested during the field trials undertaken in Orkney in May 2013 and the results of these surveys are described in the following section.

- ii. Comparative analyses of *in situ* and remote methods for constructing density indicators
 - a. Evaluation of density indicator monitoring using *in situ* and remote (camera) approaches results of field trials

Cross-hair and cell frequency counts as monitoring measures of M. modiolus density

Bootstrapped regression and correlation results for the comparison between cross-hair counts, cell frequency counts and photo quadrats counts and their corresponding total *in situ* counts of live mussels in each quadrat are presented in Table 2.5. Both correlation and regression analysis assumed total counts as the 'true' value against which the other methods were compared because these figures are the maximum visible counts in the same location. Sampled *M. modiolus* have nevertheless shown lower totals compared to what is actually amongst the clumps (Rees *et al* 2008). Bootstrapped Spearman's correlation index and 95% confidence intervals were highest for estimates obtained using cell frequency and *in situ* counts by divers, indicating a significant positive relationship between both metrics (Table 2.5). The correlation between photo quadrat counts and total *in situ* counts was low and borderline significant (0.04) while cross-hair counts, contrary to previous reports, were not significantly correlated with total quadrat counts (Table 2.5).

Table 2.5 Spearman's correlation indexes and corresponding bootstrapped (10,000 resamples) 95%confidence intervals. Calculations were obtained using R following routines described by Crawley(2013).

Method	Spearman's r	S.E.	95% CI	t	Р	df
Cross-hairs	0.24	0.11	0.027-0.47	1.92	0.06	58
Cell frequency	0.36	0.11	0.16-0.61	2.96	<0.01	58
Photo quadrats	0.26	0.10	0.07-0.46	2.07	0.04	58

The relationship between density values obtained from alternative methods and the corresponding *in situ* total counts was poorly fitted by linear regression equations (Figures 2.17A-C; Table 2.6). Overall the relationships were negatively allometric as slopes were significantly less than one in all cases (t-test, P<0.001). Goodness-of-fit measured by the r^2 value (fraction of the total variation in y explained by variation in x; where $r^2=1$ is a perfect fit) was poor across the methodologies (Table 2.6). Bootstrapped regression indicated that, following 10,000 iterations with replacement, the highest upper and lower bounds for 95% confidence intervals for cell frequency estimates were higher than for any other method.

Table 2.6 Parameters of bootstrapped regression between proxy methods for density indicators and total *in situ* counts of *M. modiolus* from $0.25m^2$ replicate quadrats. Bootstrapping assumed 10,000 iterations without replacement. Values were obtained from estimates from a ~ $2m^2$ bed surveyed by 7 divers in Orkney in May 2013.

Independent Variable	Slope	S.E.	95% CI	Intercept	SE	R ²	SE	95%CI
Cross-hairs	0.12	0.06	-0.04-0.25	0.32	1.62	0.06	0.06	0.00-0.19
Cell frequency	0.22	0.05	0.02-0.33	9.94	1.99	0.13	0.08	0.02-0.34
Photo quadrats	0.37	0.17	0.01-0.60	3.90	4.56	0.07	0.05	0.00-0.19



Figure 2.17 Linear regression plots showing the relationship between total *in situ* counts of live mussels recorded by divers using 0.25m² quadrats (abscissa) and: A) Cross-hair counts; B) Cell frequency counts and; C) Counts of live mussels from digital photographs of the undisturbed seafloor (i.e. photo quadrat counts).

Inter-surveyor variability

Binomial GLMMs with logit link functions indicated significant differences in mussel counts density depending on the method used ($F_{(3, 207)}$ =385.43; P<0.001) and the surveyor who carried out the counts ($F_{(5,207)}$ =10.97; P<0.001). Best fit models also incorporated a significant interaction term suggesting some surveyors performed differently depending on the method used for counting ($F_{(15,207)}$ =8.947; P<0.001). Because the methods used provided different density metrics (percentage cover/counts) they were each investigated separately.

There were significant differences between methods and surveyors, and the interaction between the two ($F_{(1,99)}$ =711.85; $F_{(5,99)}$ =16.67; P<0.001; and $F_{(5,99)}$ =2.84; P<0.05). Figure 2.18 shows percentage cover, estimated using cell frequency counts, ranged from 43 to 85% across all surveyors while cover derived from cross-hair counts ranged from 0 to 55%. The patterns displayed by the boxplots, also in Figure 2.18, indicate the highest variability in cover estimations between the surveyors was obtained when the cross-hair method was used. Post-hoc analyses confirmed between-surveyor differences in percentage cover estimations derived from cell frequency counts were not significant. However, variability in the results obtained from cross-hair counts were significant between surveyor E and surveyors B, C and D; and between surveyors F and B (see Figure 2.18).

Density estimates (in mussels m⁻²) using total counts varied between 75 and 142 across all surveyors showing only slight inter-surveyor variation (Figure 2.19). Boxplots of density estimations derived from counts of *in situ* photographic stills suggested more substantial variation between the surveyors doing the counts. Densities derived from photo stills were also lower than those from direct total counts, ranging from 20 to 80 mussels m⁻². Model results indicated significantly different estimations of density dependent on the method used (camera or *in situ* counts) and surveyor carrying out the method (Method: $F_{(1,99)}$ =269.5; Surveor: $F_{(5,99)}$ =10.22; Interaction: $F_{(5,99)}$ =14.88; P<0.001). A Tukey's *post hoc* test with adjusted p values showed no significant inter-surveyor differences for total *in situ* counts of mussels. However, counts obtained from photo digital stills were significantly different between most surveyors.



Figure 2.18 Variation in *M. modiolus* percentage cover derived from cross-hairs (green boxes) and cell frequency counts (red boxes). Box plots show interquartile range, median and maximum / minimum observed values.



Figure 2.19 Variation in density of *M. modiolus* (m⁻²) estimated from *in situ* counts (purple) and counts from digital stills (green) of 0.25m² quadrats. Box plots show interquartile range, median and maximum / minimum observed values.

Barplots displayed in Figure 2.20 represent differences in inter-surveyor variability calculated as coefficients of variance (CV) for each sampling method (see Lindenbaum *et al* 2002). Overall, the results suggest 'real' spatial variability in *M. modiolus* density (using total counts as the reference value for density) within the surveyed area is low. Of the methods of measuring *M. modiolus* density, cell frequency estimates out-performed other methods because results best matched those obtained from 'total' mussel counts and yielded the lowest CV scores. Photograph estimations and cross-hair counts yielded the highest overall and between-surveyor CV. It has been previously suggested (Marine Bio-images 2007), as well as by personal observations from diving surveyors, that cross-hair counts are probably unsuitable as a proxy for *M. modiolus* density, particularly in situations where beds are fragmented and mussels are aggregated into discrete, small clumps. The method is also more susceptible to surveyor error because the angle at which the diver observes the quadrat or slight changes in the position of the latter, even if fixed to the seafloor using pegs, can dramatically alter the counts.

In situ quadrat photographs were used as a simulation of drop down camera systems, therefore the cover of ophiuroids that characterised the Cava Island bed had not been brushed away to facilitate the counts. The comparatively high variability in mussel density estimated from photographs (Figure 2.20) is, therefore, probably a consequence of the difficulty in clearly establishing presence of live mussels underneath the brittlestar canopy (Figure 2.21). Additionally there were substantial differences in density estimates between some surveyors despite all viewing the same high resolution images (300 dpi TIFFs) and receiving the same instructions and training (e.g. 'only count mussels showing orange mantle/siphons'). Because some observers were more familiar with *M. modiolus* it is also possible that previous experience surveying *M. modiolus* biotopes was an influential factor.



Figure 2.20 Surveyor and method variability represented by a standardised measure of the variance (mean coefficient of variation) based on that used by Lindenbaum *et al* (2002). Each coloured bar represents a different surveyor.



Figure 2.21 Cross-hairs 0.25m² quadrat deployed onto the *M. modiolus* beds northwest of Cava, Orkney. The beds are characterised by abundant (> 60% cover) *M. modiolus* covered by a dense canopy of brittlestar (*Ophiothrix fragilis*).

Spatial variability

Field trials undertaken in Orkney were also analysed to determine if *M. modiolus* density indicators were capable of capturing natural spatial variability in reef density. If horse mussel beds are naturally spatially variable this could make detection of anthropogenic impact problematic. Poisson GLMMs were fitted to the density data obtained from direct counts of live *M. modiolus* by divers using quadrat position within each sampling station as the random factors. The results indicated that adding location as an explanatory, categorical variable significantly improved the null model ($F_{(6.54)}$ =3.248; P<0.01). The boxplots displayed in

Figure 2.22 showed that density was relatively homogenous amongst the seven stations sampled within the core of the *M. modiolus* bed (see Figure 2.13). Station 7, however, was likely to be significantly less dense according to the boxplot chart (non overlapping interquartile boxes). A post-hoc Tukey's test confirmed location was indeed a significant explanatory variable in the model due to lower densities of *M. modiolus* (median=60%) at Station 7. The results suggest Station 7 was at the edge of the *M. modiolus* bed, probably signalling a transition between different biotopes because there were no signs of anthropogenic or natural impacts such as storm damage or predation, either of which might be expected to result in empty or broken up *M. modiolus* shells, as found in impacted *M. modiolus* beds in Strangford Lough or the Isle of Man.



Figure 2.22 Range of densities (mussels m⁻²) of live horse mussels counted within 0.25m² quadrats. Station positions are displayed in Figure 2.13. Box plots show interquartile range, median and maximum / minimum observed values.

All three methods used to test inter-surveyor variability (see previous section) were also tested for their ability to capture spatial variability in density across the Cava Island bed. According to the GLMMs most of the variation of the null model using percentage cover as the dependent variable was explained by methodological differences ($F_{(1,117)}$ =268.7; P<0.001) and, to a lesser extent, by spatial differences ($F_{(6,117)}$ =2.93; P<0.05). Patterns of detection were also different for each method across the surveyed area (interaction: $F_{(6,117)}$ =2.82; P<0.05).

Figure 2.23 and the GLMM results indicate that percentage cover was significantly underestimated if the cross-hair method was used. Cover ranged from 0 to almost 60% when extrapolated using crosshair counts. If the cell frequency method was chosen, percentage cover varied from ~15% to 100% (median values between 50 and 70%). Both methods captured spatial variability in percentage cover across the area surveyed, showing higher variability than those estimations obtained from direct counts of live mussels (Figures 2.18 & 2.19). The significant interaction term was further investigated using Tukey's *post hoc* tests with the adjusted p value indicating spatial change in percentage cover, calculated using the cell frequency method, was not statistically significant (P>0.05). Between-site variation in percentage cover derived from cross-hair counts was significant between most stations sampled.



Figure 2.23 Variation in percentage cover of *M. modiolus* using cross-hair (green) and cell frequency (red). Data from across the area surveyed northwest of Cava Island, Orkney. Box plots show interquartile range, median and maximum / minimum observed values.

Spearman's correlation analysis between cell frequency counts and total counts (taken as the reference value, closest to the real density) indicated a significant, positive correlation (r_s =0.49; 95% ci = (0.29, 0.65); t=4.64; df=68; P<0.001). Cross-hair counts were not significantly correlated with total mussel counts (r_s =0.16; t=1.34; df=68; P=0.18; not significant). The poor correlation between both methods was also highlighted by 95% confidence intervals ranging from negative correlation values (-0.07) to weakly positive ones (0.38). The results are therefore in agreement with those obtained during the inter-surveyor variability trials suggesting cell frequency is the most accurate metric for constructing indicators of *M. modiolus* density.

Overall, the greatest variation in metrics for *M. modiolus* density was found to be with the cross-hair estimations across all sampled stations. For example in Station 2 variability in crosshair counts for all 10 replicate quadrats was close to 60%; for cell frequency counts, also in Station 2, variation was 30% while the least variation was obtained by counting all mussels within each quadrat (18%). The same pattern was repeated in Stations 1 and 3. However total counts yielded higher within-site variation in Stations 4-7 when compared to those obtained using the cell frequency method (Figure 2.24).



Figure 2.24 Mean coefficient of variation for counts of *M. modiolus* using different methods. Each bar represents each station sampled across the *M. modiolus* located northwest of Cava, Orkney. Abscissa numbers represent sampling stations grouped by survey method.

Power analyses

Power analyses results reflected the patterns of variance shown in Figure 2.24. A high level of variance (measured as standard deviation) characterised the estimates of density obtained using cross-hair counts. Consequently, the number of replicates (in this case, quadrats) needed to detect 10-40% changes in mean densities were very high for the area surveyed (~1km²).

Figure 2.25 shows the number of guadrats needed to detect changes in mean *M. modiolus* counts. At the lowest resolution (detection of a 40% change in density) all methods needed less than 30 random guadrats distributed across the surveyed area to detect 40% changes in mean *M. modiolus* density with the same power $(1-\beta)$. Realistically, for the bed studied, the cost of high resolution sampling (i.e. detecting changes of 10-20%) is probably not justifiable. Most habitat monitoring approaches use 30-50% change as the detection thresholds (Crawley 2013; TMAP Blue Mussel Group 2009). Out of the three methods tested, cell frequency counts and total mussel counts were the least sampling intensive, needing just five and six replicates (respectively) to detect 40% change in mean M. modiolus density across the area. Counting all the mussels within each guadrat also required six stations. In comparison, 18 quadrats were needed to detect the same change with 80% certainty using cross-hair counts. By increasing the resolution (i.e. decreasing the percentage of change that needs to be detected) sample size increased to levels which, for cell frequency and total counts, were acceptable (less than 40 replicates across the range to detect 20% change). The progression in sample size requirements to detect the same amount of change using cross-hair counts is almost an order of magnitude higher if compared with the other in situ methods. Diving surveyors reported similar counting times (<30 seconds) for all tested methods, therefore, there are no advantages of using cross-hair counts that could justify the high variance and sample effort needed to measure density.

Marine Strategy Framework Directive Indicators for Biogenic Reefs formed by Modiolus modiolus, Mytilus edulis



Figure 2.25 Estimated sampling effort (ordinate) for *M. modiolus* density (abscissa) in beds sampled off North Cava, Orkney. Three different *in situ* counting methods were compared using $0.25m^2$ quadrats: cross-hairs (green); cell frequency (red); and total mussel counts (blue). The probability level is set at 0.05. Power analysis used minimum power (1 - β) set at 0.80 (A) and 0.95 (B). Notice the difference in scale for the number of replicates displayed on the ordinate axis.

b. Evaluation of epifauna as a proxy for *M. modiolus* density based on video quadrat analysis

High definition video footage collected by divers has been used to monitor temporal and spatial change in subtidal benthic communities (Sanderson *et al* 2008; Van Rein *et al* 2009). Although *in situ* video is used by, for example, NRW to monitor *M. modiolus* abundance, the presence of abundant epifauna (especially brittlestars) can make counting difficult thus introducing error during the analytical phase. However, it is possible that conspicuous *A. digitatum* colonies could be used as a proxy for *M. modiolus* because significant correlations have been recorded between the two (Sanderson *et al* 2008). Historic data were obtained from NRW to investigate the suitability of *in situ* handheld video to monitor the density of *M. modiolus* using such a proxy indicator.

The data consisted of counts for *M. modiolus* and *A. digitatum* colonies obtained from fixed 0.25m² quadrats deployed yearly from 2007 to 2009, on the *M. modiolus* beds located off Point of Ayre in the Isle of Man (PoA) and two locations within the Pen Llŷn a'r Sarnau SAC (PL) *M. modiolus* reefs (nPL1, nPL2; Figure 2.26), *A. digitatum* over 15mm in size were counted because recording every colony down to single polyps had resulted in large intersurveyor variation in abundance estimates (Sanderson, pers. obs.). *M. modiolus* were counted if the mussels were clearly alive (gaping and with the mantle visible).



Figure 2.26 Location of *M. modiolus* beds surveyed by NRW as part of their annual biotope monitoring program using hand held video cameras of fixed quadrats. The Point of Ayre (PoA) was surveyed during separate surveys (see Section 2.5).

Variations in abundance of both taxa between years and locations were investigated using GLMMs fitted with a logit link function and Poisson distribution errors using year, site and the interaction between year and taxon as fixed factors. Quadrats were used as a random factor to account for spatial and temporal pseudo-correlation. Correlation analyses were also undertaken to establish if A. digitatum can be used as an adequate proxy for M. modiolus density across the sites. Boxplots (see Figure 2.27) indicated recorded M. modiolus were less abundant than A. digitatum across all sites. There was also a decline in abundance of A. digitatum in the North Pen Llŷn site 1 (nPL1) with the exception of 2008, when significant increases were recorded. In the North Pen Llŷn site 2 (nPL2) and at PoA there was a decline in abundance, although because the interquartile boxes overlapped, these changes were not significant. For *M. modiolus* the trends were largely similar to those observed for *A. digitatum* in the PoA and nPL2. In nPL2 there was an increase in abundance in 2009 compared to previous years when abundance remained largely stable. In the PoA, abundance of M. modiolus remained largely unchanged with time. In nPL1 there was a steady decrease in abundance of *M. modiolus* from 2004 to 2006. Subsequent surveys indicated abundance remained stable throughout the surveyed period (2006-2009). Poisson GLMMs confirmed the differences were significant between sites ($F_{(2,314)}$ =62.31; P<0.001), year ($F_{(5,314)}$ =16.61; P<0.001; taxon ($F_{(1,314)}=183.76$; P<0.001) and the interaction between year and taxon (F_(5.314)=11.88; P<0.001).

Bootstrapped correlation analyses across all sites found a significant positive correlation between *A. digitatum* counts and those of live *M. modiolus* (r_s =0.37; 95% ci (0.24, 0.49); t=5.21; P<0.001). When the analysis was partitioned between sites the correlation was found to be positively significant at nPL1 (r_s =0.49; P<0.001) and PoA (r_s =0.54; P<0.001) but not at nPL2 (r_s =0.11; P=0.43). The results therefore suggest there are significant, natural changes in abundance or the visibility of either *M. modiolus* or *A. digitatum* between some years. *M. modiolus* abundances were largely unchanged across time which further suggests that, with the exception of the early trends observed in Loch Alsh, *M. modiolus* densities are stable features thus validating density estimates as indicators of GES although targets will most likely have to be site or type-specific. The significant, relatively high positive correlation between the abundances of *M. modiolus* and *A. digitatum* indicates the species could be an appropriate site-specific candidate as an indicator of density of *M. modiolus*, particularly if remote DDV methods are used in determining density of deep, inaccessible beds (see previous sections). However, the lack of significant correlation in the second site in North Pen Llŷn could indicate this relationship may not be used at all sites.



Figure 2.27 Boxplots showing temporal trends in *M. modiolus* and *A. digitatum* abundances from fixed quadrats in *M. modiolus* beds in A) North Pen Llyn site 1 (nPL1); B) North Llyn site 2 (nPL2); and C) Isle of Man (PoA).

Box 2.1a Summary of findings for density indicators using in *situ* methods.

Variation in *M. modiolus* density across the UK range

- *M. modiolus* beds vary in their density, from continuous beds of high density up to 600 mussels m⁻² (Wales) to patchy clumped aggregations of <10 to ~300 mussels m⁻²;
- The indicators tested respond to spatial and temporal changes in *M. modiolus* bed density;
- Repeat surveys from Loch Alsh in Scotland show declines in percentage cover with time but the causes remain unclear;
- Elsewhere, reductions in *M. modiolus* density have been linked to physical impact and fragmentation caused by demersal fishing (Strangford Lough);
- The majority of existing density data is estimated using cross-hair counts from quadrats. Variability between observers and co-location inaccuracy are potential sources of error influencing observed trends of density declines.

In situ methodological approaches for *M. modiolus* density indicators

- Density indicators detected natural spatial variability in *M. modiolus* which could be linked to edge effects in field trials at north-west Cava;
- There were no time advantages in using alternative methods compared to direct total counts of live mussels. However, total counts had higher variance than cell frequency counts;
- Of the two *in situ* methods tested only cell frequency counts were significantly correlated with total number of mussels;
- Cross-hair counts were not correlated with total numbers of mussels and yielded significantly lower cover estimations compared to other methods tested, and therefore this sampling method is not recommended;
- Cell frequency counting is the most efficient method due to its low variance and low, non-significant inter-surveyor variability. It is also the most cost-effective method, requiring the lowest number of replicates to detect the same amount of change in mean density;
- The use of divers is recommended as the most accurate method to capture spatial and temporal variability in density of *M. modiolus* beds;
- Soft corals (e.g. *A. digitatum*) are stable features in some *M. modiolus* beds at particular locations and could be used as proxies for density of *M. modiolus* if using DDV methods in some areas;
- Remote methods could prove useful alternatives if the use of divers is precluded (see **Box 2.2a & b**).

Box 2.1b Recommendations for density indicators using in situ methods

- For *in situ* methods, diving surveyors should use cell frequency counts to estimate the density of *M. modiolus*. Cell frequency counts correlates with the number of *M. modiolus* present and are capable of detecting spatial and temporal variation in *M. modiolus* beds that change in response to anthropogenic pressures;
- *In situ* cell frequency counts are no more time consuming than other methods but have lower inter-surveyor variability and corresponding greater statistical power; they require a lower sample effort to detect a certain amount of change compared to other methods;
- Key recommendations related to towed still images are captured in Box 2.2b

c. Evaluation of density indicator monitoring using remote sampling methods

Past efforts to obtain accurate measures of *M. modiolus* densities using remote imaging systems such as Remote Operated Vehicle (ROV) or Drop down video (DDV) systems have proven unsuccessful (Emu Ltd 2006; Marine Bio-images 2007; Roberts *et al* 2011). Technical problems and image quality factors aside, the main problem is the difficulty of counting live mussels due to the high densities of epifauna, particularly ophiuroids, covering most beds and the uncertainty in determining which mussels are dead or alive (Emu Ltd 2006; Marine Bio-images 2007). Overall, most reports recommend remote imaging systems as a way to ground-truth presence of *M. modiolus* and as a rough method of determining relative abundance (i.e. using SACFOR scales, eg Hirst *et al* 2012; 2013).

The usefulness of remote imaging systems was investigated in the present study using high resolution DDV datasets collected by NRW in Pen Llŷn a'r Sarnau SAC (Keenan *et al* 2010) and high resolution (300 dpi) towed camera stills collated by Hirst *et al* (2012) off Noss Head, north-east Scotland. The latter are analysed for the first time in the present study.

Drop down video systems

The DDV surveys undertaken by NRW in the Pen Llŷn a'r Sarnau SAC aimed to identify the dominant benthic biotopes and to determine the scales of temporal change in community structure from 2007 to 2010. The video analyses conducted by Keenan *et al* (2010) focussed on acquiring information on substrate and semi-quantitative abundance SACFOR scores for the components of the biotic community. Because methodologies employed in 2007 were different to those used in subsequent years the authors decided not to include data from 2007 in the analyses. Overall they found no significant temporal trends in biodiversity (number of taxa (S), number of individuals (N), Shannon-Wiener's (H') and Simpson's (λ) indices). SACFOR transformed abundance data for the most abundant species, including *M. modiolus*, were used to determine the temporal trends. The latter approach, whereby numerical values are equated to SACFOR abundance scales, has also been used to obtain quantitative data amenable to statistical analyses by Roberts *et al* (2011) and Strain *et al* (2012).

The results of Keenan *et al* (2010) indicated that the relative abundance of the most abundant, conspicuous taxa (including *M. modiolus*) was stable across the studied period (i.e. differences in abundance between years not statistically significant). This result is also of relevance to species that could be considered indicators for presence of *M. modiolus*, for example (previous section), because a significant correlation existed between the abundance of both *A. digitatum* and the underlying *M. modiolus*: the former could be used as a proxy for abundance of the latter. If, as found by Keenan *et al* (2010), temporal variability

in the abundance of A. digitatum is low and not significant, the implications for the acquisition of density indicators sensitive to anthropogenic change are clear. The use of A. digitatum as an indicator would be, nonetheless, limited to locations where these two species are associated and significantly correlated in high densities (such as the Isle of Man or North Wales) and where remote surveying techniques such as DDV and camera systems are more suited. High definition DDV footage obtained during the present work from NRW was of sufficient quality to determine presence and, in most cases, relative SACFOR abundance of *M. modiolus.* Nonetheless, the presence of epifauna including ophiuroids and large barnacles precluded the acquisition of accurate estimation of densities of live mussels. A. digitatum is clearly more conspicuous hence easier to identify and count than live M. modiolus. The results obtained by Keenan et al (2010), supported by gualitative analyses of DDV footage examined during the present work, suggest that if DDV is to be used to monitor the density of *M. modiolus*, this method should be restricted to beds where *in situ* approaches (Section 2.4.1. iii) are precluded. Under those conditions the use of proxy species such as A. digitatum could provide satisfactory estimates of density for M. modiolus (see section below).

Evaluation of drop down camera stills: Noss Head case study

In 2009 Triscom Marine (Orkney) and MMT Consultancy surveyed the seabed off Noss Head, in north-east Scotland and found a deep, and potentially extensive, dense *M. modiolus* bed. Imagery obtained from the ROV was poor due to the high currents that dominate the area but, nonetheless, was sufficient to confirm the presence of horse mussels (Moore & Roberts 2011). Similarly to reports from Emu Ltd (2004) in Loch Alsh and Roberts *et al* (2011) in Strangford Lough, Moore and Roberts (2011) found it impossible to differentiate between live and dead *M. modiolus* solely using ROV video footage. This is further confirmation that remote video, although adequate to determine presence and extent, lacks the required resolution to obtain accurate estimations of *M. modiolus* density that could be used to determine GES for Criterion 1.6 (Habitat Condition).

In 2011 Heriot Watt University, in partnership with Marine Scotland and Scottish Natural Heritage (Hirst et al 2012) surveyed the Noss Head area. One of the objectives was 'to compare the quality and size of the Noss Head mussel bed to others within Scotland and the UK'. To that effect, 51 stations were surveyed using a combination of DDV, stills camera and multibeam sonar. Using DDV to ground-truth the extent of the bed calculated from multibeam bathymetric maps, Hirst et al (2012) obtained an estimated bed area of 3.85km². DDV footage was also used to estimate SACFOR scale abundance of the epifaunal component of the community which, for *M. modiolus*, ranged between Rare (1-5% cover); Occasional at the bed edges (5-9% cover); Abundant (40-79% cover); and Superabundant (>80% cover). One of the unused outputs of the survey was an extensive library of high definition digital photographs collected using a towed digital stills camera. That library was accessed as part of this report with the aim of determining if high definition images could circumvent issues found in other remote imaging systems and, therefore, be used to acquire estimates of *M. modiolus* density or percentage cover. Out of the 11 stations with *M.* modiolus present, seven were identified as *M. modiolus* biogenic reefs (ModT biotope; Connor et al 2004) and only these were used in the evaluation process. Four replicate photographs were chosen from each station resulting in 28 images analysed in total.

The correlation between the three sampling methods trialled (cross-hair counts, cell frequency counts and count of total live mussels) for each photograph were tested using Spearman's rank tests. Bootstrapped correlation showed a significant positive correlation between cell frequency counts and total counts of live mussels (r_s =0.72; P<0.001) and low and non significant relationship between cross-hair counts and numbers of live mussels

($r_{s=}0.2$; P=0.08; n.s.). Changes in cross-hair counts (number of intersects with live mussels underneath) were poorly explained by changes in total live mussels (bootstrapped R²=0.04; P=0.3; y=0.09+4.32; Figure 2.28). However, the relationship between cell frequency counts and total live mussels had a good linear fit (bootstrapped R²=051; P<0.001; y= 0.66+8.1x; Figure 2.28B).



Figure 2.28 Linear regression plots showing relationships between total live mussel counts and (A) cross hair counts and (B) cell frequency counts. Dotted lines represent 95% confidence intervals.

А

Although the initial qualitative assessment of both DDV and photographic stills suggested the presence of a continuous and dense mussel bed, the boxplot charts (Figure 2.29) show potentially significant spatial variation in *M. modiolus* density and cover across the surveyed area (see also Figure 2.30). A Poisson GLMM with logit link function using raw counts as the independent variable yielded significant results for the factor station ($F_{(5,22)}$ = 5.6; P<0.01) and, as expected considering the different nature of each metric, between methods ($F_{(2,22)}$ =22.93; P<0.001). There were no significant interactions between counting method and surveyed station ($F_{(10,22)}$ =0.94; P=0.50; not significant).



Figure 2.29 Variation in *M. modiolus* density (top) and percentage cover (bottom) estimated from drop down digital stills (Noss Head horse mussel bed). Estimates for density are derived from counts of live, gaping mussels (top) while percentage cover is based on cell frequency and crosshairs counts. Box plots show interquartile range, median and maximum / minimum observed values as whiskers (1.5 times the interquartile region). Black dots indicate values above and below the 1.5 times the 3rd and 1st quartiles, respectively. Station names follow Hirst *et al* (2013; see also Figure 2.30).



0 0.45 0.9 1.8 Kilometers

Figure 2.30 Multibeam bathymetric map of seafloor off Noss Head, North East Scotland. Rugosity patterns were consistent with *M. modiolus* beds ground-truthed using drop down video and camera by HWU (Hirst *et al* 2012). Circles represent *M. modiolus* density estimated from high definition digital stills from drop down cameras as part of the present work.

Image quality was consistently high throughout the Noss Head dataset, whereby individual live mussels could be easily discerned and counted when the siphons and/or mantle were visible. However, in some stations where *M. modiolus* was abundant or superabundant according to the DDV record (for example NH47 and NH21, the latter not included in Figure 2.29), it was impossible to determine if the mussels were alive or dead. While some mussels had their valves closed others were horizontally positioned, which interfered with the counting process. In some photos live mussels were difficult to detect due to the presence of upright emergent hydroids (*Kirchenpaueria pinnata*, *Halecium* spp.; Figure 2.31).

Nonetheless, other reef areas were naturally characterised by patches without or with low density of mussels. An example of such areas is station 47 where disturbed zones with empty or broken shell were apparent. Whether those fragmented areas are the result of anthropogenic impacts or the natural edge of the Noss Head bed is difficult to know without corroborating pressure data (Figure 2.32).

Mean cover estimated using cell frequency counts across the bed was $47\% \pm 27\%$ SD while grid intersects yielded $15\% \pm 18\%$ SD. Total counts on the other hand produced an estimate of 44.68 ± 39.74 SD mussels m⁻². All methods were characterised by extremely high within site variances. Standardised variances calculated using coefficients of variance were lowest for cell frequency estimations of percentage cover (58%) and highest for intersect counts (an unacceptable 118%).



Figure 2.31 Close-up of benthic community found off Noss Head, north-east Scotland. The assemblage is formed by dense *M. modiolus*, brittlestars *Ophiothrix fragilis*, bryozoans *Parasmittina trispinosa* (pale orange crust) and hydroid turf *Sertularia* spp. Although most of the bed is likely to consist of live mussels, as indicated by the presence of numerous gaping specimens, it was not possible to accurately determine mussel densities using high definition drop camera still.



Figure 2.32 Photograph captured using a high definition digital stills camera (Kongsberg OE14-208[®]) at Station NH47, Noss Head, north-east Scotland. The seafloor was characterised by *M. modiolus* forming a bed with clear patches and common empty *M. modiolus* shells.

High variance was the determinant factor in the number of replicate estimates obtained from power analysis calculations (Figure 2.33). Using a conservative power $(1-\beta)$ of 0.8 at α =0.05, it would be necessary to process more than 1000 photographs to be able to detect a 10% change in mean percentage cover across the ~4km² Noss Head *M. modiolus* bed; 490 using total counts; and 342 using the cell frequency method. Under less ambitious aims (abilty to detect a 20% change) there is a substantial reduction in the number of sampling stations needed for the same amount of power: 268 (crosshairs); 124 (total counts); and 87 (cell frequency).



Figure 2.33 Sampling effort estimations for remote drop down camera methods to detect changes in mean *M. modiolus* densities in reef sampled off Noss Head, Scotland. The probability significance level is set at 0.05. Power analysis used minimum power $(1 - \beta)$ of 0.80.

Although Drop down camera (DDC) imagery has the usual caveats associated with calculating estimations of density using remote imagery, high definition digital stills were useful as they provided quantitative density information that was amenable to enumeration and statistical analyses. High variance for all methods resulted in an exceedingly high requirement of samples to detect small changes in *M. modiolus* densities. Nonetheless, change could still be detected at a coarser resolution (20-40%) without the need for a prohibitively high number of samples (i.e. 87 using cell frequency counts). Percentage cover estimation using cell frequency counts can be regarded as the most efficient reef density metric as it was the fastest method with the lowest co-efficient of variance (CV) of all three tested. Although detection of live mussels from the digital stills was difficult overall it was obvious to the observer that most of these mussels were alive.

DDC imagery can, therefore, be used to measure mussel density and determine human impacts at Noss Head or other deep *M. modiolus* beds (e.g. south of Canna, Moore & Atkinson 2012), where greater water depths preclude the use of more accurate *in situ* methods (i.e. by divers).

Box 2.2a Summary of findings for density indicators using remote methods

- Density estimations using photographs were significantly correlated to total numbers of *M. modiolus*, however, the method was prone to high variance and significant inter-surveyor variability;
- It also took longer for surveyors to obtain estimations of density from photographs than for *in situ* divers using any of the other methods;
- Overall, cell frequency counts by divers is the most effective method to obtain density estimates in *M. modiolus* beds, particularly under conditions of high density of epifauna covering the mussels. Towed cameras could be employed to obtain proxies for density or cover of deep, tidal *M. Modiolus;*
- For remote systems, high resolution imagery is needed to reduce variance in density estimations;
- Drop down video (DDV) can cover larger areas in a given time compared to divers but only provides rough approximations of *M. modiolus* cover using SACFOR scales;
- Towed camera systems provide higher resolution images compared to DDV footage which can be processed to obtain more accurate estimations of density and percentage cover;
- Image processing software is recommended to extract density estimates from digital stills; proxy methods can also be adapted to obtain cover and density estimates from high resolution images (Box 2.1);
- Cell frequency counts provide a more accurate estimation of density compared to line intersects. It also requires the lowest amount of replication to achieve an acceptable power;
- High resolution towed cameras can extract estimates of density and detect anthropogenic impact but they are prone to error particularly in dense horse mussel areas with high percentage cover of epifauna where some individuals may be obscured;
- For density estimation, the use of remote imagery may be enhanced by sampling at a time of year when epifauna is seasonally less. However, this may restrict the value of the imagery for community indicators (see Section 2.5 (2.5.2)).

Box 2.2b Recommendations for density indicators using remote methods

- The use of remote imagery is recommended for density monitoring *M. modiolus* of beds that are too deep, large or otherwise inaccessible for divers. Again, quantification using cell frequency from still images produces the least inter-surveyor variability and the highest statistical power requiring fewer replicates to detect a given amount of change;
- Risk of obtaining inaccurate monitoring results caused by changes in the epifauna (particularly brittlestars) obscuring *M. modiolus* may be mitigated by counting *A. digitatum* colonies as a proxy, but only at some sites.
2.5.2 Community indicators

The faunal assemblage associated with *M. modiolus* reefs are usually described as one of the richest and most diverse in European waters (Erwin 1977; Thorson 1957). The *Modiolus* community has a multi-layered structure (Magorrian *et al* 1995) heavily dependent on *M. modiolus* as the keystone species. This community is composed of a rich array of epifaunal species attached to the mussels and mobile scavengers and predators attracted by the feeding opportunities enhanced by the complexity of the habitat. There is also a highly diverse component of small invertebrates attracted to the habitat and food resources offered by the matrix of live *M. modiolus* bound by their byssus threads to shell fragments and accumulated sediments and pseudofaeces.

Good community indicators should be able to capture and discriminate the sources of spatial and temporal change experienced by benthic communities (Pearson & Rosenberg 1978) and align with the MSFD requirements for indicators to include prevailing conditions for the assessment of GES. At the same time monitoring community indicator metrics should be cost-effective and subject to low levels of error. Community indicators considered for *M. modiolus* bed in this report are total abundance of individuals (N), total number of taxa (S), Margalef's richness (d), Shannon-Wiener's diversity (H') and Pielou's evenness (J). Although many other diversity indices exist (see Magrran 2004 and references therein), the chosen community metrics are the only ones that appear in the published literature and, consequently, the only ones amenable to comparative statistical analyses needed to validate them. These metrics can, therefore, also be used to determine baseline conditions against which results from new surveys can be compared.

i. Variability in typical *M. modiolus* community across the UK

Published records for *M. modiolus* communities suggest species richness depends on the method used to calculate it and the specific community component investigated (epifauna, crevice fauna or infauna). For example, existing records collated by divers for epifaunal assemblages range from 23 species in *M. modiolus* troughs in Pen Llŷn a'r Sarnau SAC, Wales (Bunker 1999; Sanderson *et al* 2008) and 27 in Strangford Lough's South Basin (Roberts *et al* 2011) to 100 taxa recorded by Roberts *et al* (2004) also in Strangford Lough (North Basin). Destructive methods such as dredges and quadrat clearances are able to capture the much richer infaunal and crevice faunal component suggesting species richness can range from as little as 36 taxa recorded in clumps collected by Mair *et al* (2010) in Calback Ness (Shetland) to well over 200 taxa found in dredge, quadrat clearance and suction samples (see Table 2.8; also Mair *et al* 2000; Kenchington *et al* 2007; Rees *et al* 2008).

Magurran (2004) indicated that the value of species diversity measured using Shannon-Wiener's index (H') usually ranges from 1.5 to 3.5 and rarely surpasses 4, with 5 being an extremely rich assemblage. Diversity H' indices reported for *M. modiolus* range from 1.7 in impacted beds in Strangford Lough (Roberts *et al* 2011) to well over 3 for most communities across their UK range (see Table 2.7). Shannon-Wiener's diversity values as high as 5 have been reported from cumulative records from clump samples collected in Loch Alsh, West Scotland (Moore *et al* 2013), confirming the presence of an extremely rich and diverse community associated with *M. modiolus* compared to the muddy habitats that are found in the absence of the mussels. Absolute Pielou's evenness (J) values vary between 0 and 1, values close to 1 being indicative of assemblages where most species are equally abundant in contrast with communities with high dominance where only a few species dominate the community. Evenness can be a more informative measurement to define communities than richness or diversity. Faunal assemblages associated with *M. modiolus* beds are usually very even; with Pielou's J values ranging between 0.6 and 0.9 according to published records (see Table 2.8). The index is probably very sensitive to variations in sampling effort, the degree of zeal of the taxonomic expert processing the samples and also the sieve size used. Datasets which include dominant macro-invertebrates associated with the finer fractions of the sediment such as capitellids, oligochaetes and nematodes would suggest communities with low evenness. Meaningful comparisons of historical data can therefore only be achieved if the sampling methods are consistent.

Community composition, and in particular the infaunal component, is also very variable across its British and Irish range. Overall, *M. modiolus* assemblages are largely dominated by the following conspicuous biota:

- Upright emergent sessile species, including soft corals Alcyonium digitatum and hydroid Abietinaria abietina, e.g. in Pen Llŷn a'r Sarnau SAC and the Isle of Man (Cook et al 2013; Sanderson et al 2008); also hydroids Serturlaria spp. and Halecium spp., e.g. in Loch Alsh (Moore et al 2013); and tunicates, mostly Ascidiella aspersa (e.g. Strangford Lough, Farinas-Franco & Roberts in press).
- Echinoderms, such as Antedon bifida (Roberts et al 2011); ophiuroids, Ophiothrix fragilis and Ophiocomina nigra, which can be super-abundant, e.g. in Creagan, in Loch Creran (Moore et al 2006) and in Strangford Lough (Roberts et al 2011).
- Algal species such as *Laminaria hyperborea* and *Phycodrys rubens* can also be codominant in some *M. modiolus* beds, e.g. in Loch Leven (Moore *et al* 2012).

Some taxa are clearly biotope-defining according to Connor *et al* (2004), for example the association between *M. modiolus* and the scallop *Chlamys varia* characterised *M. modiolus* beds in the north basin of Strangford Lough (Magorrian & Service 1998). Other characterising species for *M. modiolus* assemblages include the holothurians *Thyonidium drumondii* and *Thyone roscovita*, the sponge Lophon hyndmani and the tunicate *Pyura microcosmus* (Erwin 1990; Roberts *et al* 2004).

Location	Site	S (MNCR surveys)	S (infauna)	H'	J	Method	Source
Canada	Bay of Fundy		150-341				Fuller 1998 in Kenchington <i>et al</i> (2006)
Canada	Gulf of Saint Lawrence		39-46			0.1m ² quadrats. Decanting, not sieving	Rowell (1967)
lsle of Man	Point of Ayre		270			Dredge	Holt & Shalla (unpublished)
Northern Ireland	Long Sheelah (Strangford Lough)		84			Dredge	Roberts (1975)
Northern Ireland	South Basin (Strangford Lough)		90			NA	Brown & Seed (1976)
Northern Ireland	North Basin (Strangford Lough)	100	120 (182)	2.7	0.7	Clearance 0.25m ² quadrats	Roberts <i>et al</i> (2004)
Northern Ireland	South Basin (Strangford Lough)	60-70	102 (130)	2.6	0.8	Clearance 0.25m ² quadrats	Roberts <i>et al</i> (2004)
Northern Ireland	North Basin (Strangford Lough)	45	109	1.7	0.5	Clearance 0.25m ² quadrats	Roberts <i>et al</i> (2011)
Northern Ireland	South Basin (Strangford Lough)	23	144	2.9	0.8	Clearance 0.25m ² quadrats	Roberts <i>et al</i> (2011)

Table 2.7 Number of taxa (S), diversity (H') and community evenness (J) associated with *M. modiolus* biotopes in published reports. Range indicates minimum and maximum numbers of taxa in discrete clumps. Numbers in brackets indicate cumulative total species richness.

Location	Site	S (MNCR surveys)	S (infauna)	H'	J	Method	Source
Northern Ireland	Experimental <i>M.</i> <i>modiolus</i> reef (Strangford Lough)		223	2.67 -3.6	0.67- 0.89	Clearance 0.0625m ² quadrats	Farinas-Franco <i>et al</i> (2013)
Scotland	Busta Voe	66	76-85 (129)	3.25 - 4.08	0.57- 0.71	Clumps	Mair <i>et al</i> (2000)
Scotland	Firth of Lorne		62			Dredge, 1mm sieve	Collins (1986)
Scotland	Kyleakin (Loch Alsh)		93-100 (163)			Clumps	
Scotland	String Rock (Loch Alsh)	77	93-122 (203)	3.46 - 5.40	0.57- 0.86	Clumps	Mair <i>et al</i> (2000)
Scotland	String Rock (Loch Alsh)		84-100 (145)	4.88 - 5.27	0.76- 0.81	Clumps	Emu Ltd (2006)
Scotland	String Rock (Loch Alsh)		125-146 (222)			Clumps	Moore <i>et al</i> (2013)
Scotland	Upper Basin (Loch Creran)	62	80-91 (153)	4.88 5.00	0.79- 0.81	Clumps	Mair <i>et al</i> (2000)
Scotland	Loch Creran	58	88-104 (169)			Clumps	Moore <i>et al</i> (2006)
Scotland	Port Appin (Loch Linnhe)	35	133-137 (247)			Clumps	Moore <i>et al</i> (2012)
Scotland	Corpach (Loch Linnhe)	50	89-98 (162)			Clumps	Moore <i>et al</i> (2012)
Scotland	Loch Leven	44	97-128 (199)			Clumps	Moore <i>et al</i> (2012)
Scotland	Calback Ness (Shetland)	71	36			Clumps/MCR	Mair <i>et al</i> (2010)
Scotland	Voxter Voe (Shetland)	46				MNCR	Mair <i>et al</i> (2010)
USA	Maine		60			0.25m ² quadrats	Ojeda & Dearborn (1989)
USA	New England		80			0.25m ² quadrats	Witman (1985)
Wales	Pen Llŷn a'r Sarnau	23-27 (troughs) 61 (ridges)	81-134 (213)	4.77 - 5.40		0.0625m ² quadrats	(2008); Bunker (1999); Sanderson <i>et al</i> (2008)

ii. In situ sampling methods

a. Phase II MNCR dive surveys

Phase II type surveys follow MNCR protocols laid out by Hiscock (1996). The method has often been applied by divers descending a shotline in pairs, each surveying a 2m band either side of a transect line, noting the species they encounter and assigning SACFOR values to them (Table 2.3). MNCR transect data used in this report were collated from a total of 20 discrete *M. modiolus* beds from six broad survey locations: Pen Llŷn a'r Sarnau SAC in Wales and Loch Linnhe; Loch Alsh; Loch Creran; Orkney; and Shetland, all in Scotland. Although the methodology was broadly the same, some transects were delineated for either a fixed distance (i.e. 50 or 100m) or depth range, as in the 2005 surveys carried out in Loch Creran (Moore *et al* 2006). On occasions the *in situ* records were supplemented by video footage which was used to help improve the accuracy of the record. Table 2.8 shows the variations in MNCR Phase II survey data.

The main aim of this section is to assess the usefulness of *in situ* dive survey techniques in acquiring community indicators relevant to MSFD Descriptor 1 as well as to estimate the level of error introduced and their ability to detect spatial and temporal change in community metrics. Variability in diversity and community composition of *M. modiolus* beds was investigated using univariate and multivariate statistical analyses of historical and recent MNCR phase II community data, published or unpublished, gathered across the UK. Recent transect faunal data collated in Strangford Lough by Roberts *et al* (2011) was not incorporated into the analyses because, contrary to the Scottish and Welsh surveys, the quantative photoquadrat method was not consistent with MNCR Phase II type protocols.

Univariate analyses of Phase II surveys

In total 327 conspicuous epibiotic taxa were recorded across all surveyed sites. Total number of taxa per transect ranged from 23 in the Pen Llŷn *M. modiolus* beds (Bunker *et al* 1999) to 77 from beds off String Rock, in Loch Alsh (Mair *et al* 2000). Between and within-location changes in community richness, diversity and evenness indices based on SACFOR abundances were converted to numerical values (see Table 2.3), displayed in

Figure 2.34 and summarised in Table 2.8.

Mixed PERMANOVA models were used to determine if the patterns of spatial variation observed in Figure 2.34 were influenced by location, abundance of *M. modiolus* and environmental co-variates (depth, exposure, tidal current). Table 2.9 summarises the results of the PERMANOVA models which indicated variation in total number of species (S); species richness (d), Shannon-Wiener's diversity (H') and Pielou's eveness (J) was not significant amongst the surveyed beds. Recorded environmental conditions did not have any significant effect on community indices while within and between-site variability (confounded with survey) explained most of the random variance of the models.

Table 2.8 Community diversity indices for Phase II (MNCR) survey data collated from 15 different sites across the UK. Repeat transect data were also available for Loch Creran and String Rock (Loch Alsh). S=Total number of taxa; d=Margalef's richness; H'=Shannon-Wiener's diversity; J=Pielou's evenness.

Transect	Site	S	d	Н'	J'
PLL99-ST10	Pen Llŷn a'r Sarnau SAC	27	6.04	3.20	0.97
PLL99-ST11	Pen Llŷn a'r Sarnau SAC	23	5.62	2.99	0.95
PL99-ST15	Pen Llŷn a'r Sarnau SAC	25	5.61	3.11	0.97
LL11_LL	Loch Leven	44	9.90	3.61	0.95
LL11_CP	Corpach	50	11.15	3.70	0.95
LL11_PA	Port Appin	35	8.24	3.41	0.96
LA99	String Rock 1999	77	14.87	4.15	0.96
LA12_SR	String Rock 2012	73	14.86	4.12	0.96
LA12_KA	Kyleakin 2012	46	10.42	3.62	0.95
LC99	Loch Creran 1999	62	12.38	3.99	0.97
LC05	Loch Creran 2005	58	11.53	3.91	0.96
LC12	Loch Creran 2012	35	8.09	3.36	0.94
OK11-GS	Gutter Sound	43	9.24	3.66	0.97
OK11-K	North Cava	43	9.56	3.59	0.95
SH-BV99	Busta Voe 99	67	12.97	4.09	0.97

Transect	Site	S	d	H'	J'	
SH04-CN	Calback Ness	68	13.25	4.05	0.96	
SH04-VV	Voxter Voe	48	10.32	3.76	0.97	
SH12-BV	Basta Voe	54	11.93	3.88	0.97	
SH12-US	Uyea Sound	33	7.58	3.30	0.94	
SH12-HS	Hascosay Sound	30	6.49	3.18	0.94	



Figure 2.34 Total number of taxa; Margalef's richness; Shannon-Wiener's diversity (S) and Pielou's evenness from *M. modiolus* beds surveyed using Phase II protocols (*c.f.* MNCR, Hiscock 1996).

Table 2.9 Results of mixed PERMANOVA models for diversity indices calculated from Phase II transect surveys conducted on *M. modiolus* biotopes across the UK range. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

A) Total number of Species (S). Random variance: Between sites=2.89; Error=6.88.							
Source	df	SS	MS	Pseudo-F	Р		
M. modiolus abundance	1	434.46	434.46	2.01	0.20		
Exposure	1	117.58	117.58	0.41	0.63		
Depth	1	1726.10	1726.1	5.27	0.08		
Tide	1	72.20	72.20	0.34	0.69		
Location	5	1026.7	205.34	0.88	0.57		
Site	8	1625.3	203.16	4.30	0.19		
Residual	2	94.60	47.30				
Total	19	5097					
B) Margalef's richness (d). Random [·]	variance: Between	sites= 2.3; Error	= 0.92			
Source	df	SS	MS	Pseudo-F	Р		
M. modiolus abundance	1	20.09	20.09	3.24	0.11		
Exposure	1	6.52	6.52	0.66	0.51		
Depth	1	48.37	48.37	5.14	0.09		
Tide	1	4.15	4.15	0.63	0.54		
Location	5	32.60	6.52	0.96	0.52		
Site	8	46.43	5.80	6.90	0.12		
Residual	2	1.68	0.84				
Total	19	159.85					
C) Shannon-Wiener's div	ersity (H'). I	Random variance:	Between sites =	0.23; Error=0.21			
C) Shannon-Wiener's div Source	ersity (H'). I df	Random variance: SS	Between sites = MS	0.23; Error=0.21 Pseudo-F	Р		
C) Shannon-Wiener's div Source M. modiolus abundance	ersity (H'). I df 1	Random variance: SS 0.25	Between sites = MS 0.25	0.23; Error=0.21 Pseudo-F 2.54	P 0.16		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure	ersity (H'). df 1 1	Random variance: SS 0.25 0.99	Between sites = MS 0.25 0.99	0.23; Error=0.21 Pseudo-F 2.54 0.78	P 0.16 0.46		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure Depth	ersity (H'). I df 1 1 1	Random variance: SS 0.25 0.99 0.81	Between sites = MS 0.25 0.99 0.81	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48	P 0.16 0.46 0.07		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure Depth Tide	ersity (H'). I df 1 1 1 1 1	Random variance: SS 0.25 0.99 0.81 0.43	Between sites = <u>MS</u> 0.25 0.99 0.81 0.43	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46	P 0.16 0.46 0.07 0.62		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure Depth Tide Location	ersity (H'). I df 1 1 1 1 5	Random variance: SS 0.25 0.99 0.81 0.43 0.45	Between sites = MS 0.25 0.99 0.81 0.43 0.09	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88	P 0.16 0.46 0.07 0.62 0.55		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure Depth Tide Location Site	ersity (H'). I df 1 1 1 1 5 8	Random variance: SS 0.25 0.99 0.81 0.43 0.45 0.74	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.09	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07	P 0.16 0.46 0.07 0.62 0.55 0.35		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure Depth Tide Location Site Residual	ersity (H'). I df 1 1 1 1 5 8 2	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.09 0.45	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07	P 0.16 0.46 0.07 0.62 0.55 0.35		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total	ersity (H'). I df 1 1 1 5 8 2 2 19	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.09 0.45	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07	P 0.16 0.46 0.07 0.62 0.55 0.35		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J):	ersity (H'). I df 1 1 5 8 2 19 Random va	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s	Between sites = <u>MS</u> 0.25 0.99 0.81 0.43 0.09 0.09 0.09 0.45 sites=-0.006; Error	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 pr=0.01	P 0.16 0.46 0.07 0.62 0.55 0.35		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source	ersity (H'). I df 1 1 1 5 8 2 2 19 Random va df	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS	Between sites = <u>MS</u> 0.25 0.99 0.81 0.43 0.09 0.09 0.45 sites=-0.006; Error <u>MS</u>	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 pr=0.01 Pseudo-F	P 0.16 0.46 0.07 0.62 0.55 0.35 P		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source M. modiolus abundance	ersity (H'). I df 1 1 1 5 8 2 2 19 Random va df 1	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS 2.75*10 ⁴	Between sites = <u>MS</u> 0.25 0.99 0.81 0.43 0.09 0.09 0.45 sites=-0.006; Erro <u>MS</u> 2.75*10 ⁻⁴	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 pr=0.01 Pseudo-F 3.06	P 0.16 0.46 0.07 0.62 0.55 0.35 P 0.12		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source M. modiolus abundance Exposure	ersity (H'). I df 1 1 1 5 8 2 2 19 Random va df 1 1	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS 2.75*10 ⁴ 1.56*10 ⁴	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.45 sites=-0.006; Error MS 2.75*10 ⁻⁴ 1.56*10 ⁻⁴	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 pr=0.01 Pseudo-F 3.06 1.61	P 0.16 0.46 0.07 0.62 0.55 0.35 P 0.12 0.29		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source M. modiolus abundance Exposure Depth	ersity (H'). I df 1 1 1 5 8 2 19 Random va df 1 1 1 1	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS 2.75*10 ⁴ 1.56*10 ⁴ 4.63*10 ⁴	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.45 sites=-0.006; Error MS 2.75*10 ⁴ 1.56*10 ⁴ 4.63*10 ⁴	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 0r=0.01 Pseudo-F 3.06 1.61 0.58	P 0.16 0.46 0.07 0.62 0.55 0.35 P 0.12 0.29 0.53		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source M. modiolus abundance Exposure Depth Tide	ersity (H'). I df 1 1 1 5 8 2 19 Random va df 1 1 1 1 1	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS 2.75*10 ⁴ 1.56*10 ⁴ 4.63*10 ⁴ 2.30*10 ⁴	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.45 Sites=-0.006; Error MS $2.75*10^{-4}$ $1.56*10^{-4}$ $4.63*10^{-4}$ $2.30*10^{-4}$	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 0r=0.01 Pseudo-F 3.06 1.61 0.58 2.47	P 0.16 0.46 0.07 0.62 0.55 0.35 P 0.12 0.29 0.53 0.17		
C) Shannon-Wiener's div Source M. modiolus abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source M. modiolus abundance Exposure Depth Tide Location	ersity (H'). I df 1 1 1 5 8 2 19 Random va df 1 1 1 1 1 5	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS 2.75*10 ⁴ 1.56*10 ⁴ 4.63*10 ⁴ 2.30*10 ⁴ 6.18*10 ⁴	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.45 Sites=-0.006; Error MS $2.75*10^{-4}$ $1.56*10^{-4}$ $4.63*10^{-4}$ $2.30*10^{-4}$ $1.23*10^{-4}$	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 0r=0.01 Pseudo-F 3.06 1.61 0.58 2.47 1.43	P 0.16 0.46 0.07 0.62 0.55 0.35 P 0.12 0.29 0.53 0.17 0.34		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source <i>M. modiolus</i> abundance Exposure Depth Tide Location Site	ersity (H'). I df 1 1 1 5 8 2 19 Random va df 1 1 1 1 1 5 8 8 8	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS 2.75*10 ⁴ 1.56*10 ⁴ 4.63*10 ⁴ 2.30*10 ⁴ 6.92*10 ⁴ 6.92*10 ⁴	Between sites = MS 0.25 0.99 0.81 0.43 0.09 0.09 0.45 Sites=-0.006; Error MS $2.75*10^{-4}$ $1.56*10^{-4}$ $4.63*10^{-4}$ $2.30*10^{-4}$ $1.23*10^{-4}$ $8.62*10^{-5}$	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 0r=0.01 Pseudo-F 3.06 1.61 0.58 2.47 1.43 0.71	P 0.16 0.46 0.07 0.62 0.55 0.35 P 0.12 0.29 0.53 0.17 0.34 0.70		
C) Shannon-Wiener's div Source <i>M. modiolus</i> abundance Exposure Depth Tide Location Site Residual Total D) Pielou's evenness (J): Source <i>M. modiolus</i> abundance Exposure Depth Tide Location Site Residual	ersity (H'). I df 1 1 1 5 8 2 19 Random va df 1 1 1 1 5 8 2 2 2 19 8 2 2 19 8 2 2 19 8 2 2 19 10 10 10 10 10 10 10 10 10 10 10 10 10	Random variance: SS 0.25 0.99 0.81 0.43 0.43 0.45 0.74 0.90 2.49 ariance: Between s SS 2.75*10 ⁴ 1.56*10 ⁴ 4.63*10 ⁴ 2.30*10 ⁴ 6.92*10 ⁴ 2.43*10 ⁴ 2.43*10 ⁴	$\begin{array}{r} \hline \text{Between sites} = \\ \hline \textbf{MS} \\ 0.25 \\ 0.99 \\ 0.81 \\ 0.43 \\ 0.09 \\ 0.09 \\ 0.09 \\ 0.45 \\ \hline \textbf{sites} = -0.006; Error \\ \hline \textbf{MS} \\ \hline 2.75*10^4 \\ 1.56*10^4 \\ 4.63*10^4 \\ 2.30*10^4 \\ 1.23*10^4 \\ 8.62*10^5 \\ 1.22*10^4 \\ \hline \end{array}$	0.23; Error=0.21 Pseudo-F 2.54 0.78 5.48 0.46 0.88 2.07 0r=0.01 Pseudo-F 3.06 1.61 0.58 2.47 1.43 0.71	P 0.16 0.46 0.07 0.62 0.55 0.35 P 0.12 0.29 0.53 0.17 0.34 0.70		

Multivariate analyses of Phase II surveys

Cluster analyses of the multivariate Bray-Curtis similarity matrix show significant (P<0.05) SIMPROF groupings which, in some cases, roughly coincided with surveyed locations (

Figure 2.35). Each of these groupings was assigned a code to facilitate SIMPER analyses (Groups I to VII). Gutter Sound (Orkney) communities were the most dissimilar diverging at 20.54% similarity (Group I) followed by Pen Llŷn a'r Sarnau SAC (24.42%; Group II) and a broad group including all remaining sites (27.17%) with the following subgroups: Group III formed by Loch Alsh (1999), Busta Voe (1999), Loch Creran (1999 and 2005), and Shetland (2004-Calback Ness and Voxter Voe) diverging at 39% similarity; Group IV formed by Loch Linnhe (2011, Corpach), Loch Creran (2012) diverging at 31.42%; Group V (North Cava bed in Orkney, Loch Leven and Port Appin in Loch Linnhe) at 37.41%, Group VI (String Rock

and Kyleakin, Loch Alsh 2012) at 43.33% and Group VII (all Shetland surveys for 2012) at 48.91% (see Table 2.8).

Epibiotic communities were largely aggregated by survey and location although there was an evident temporal shift in multivariate records where repeated surveys had been undertaken (such as Loch Alsh and Loch Creran). A subsequent PERMANOVA analysis (Table 2.10) further confirmed the lack of overall structure found in the dendrogram indicating that none of the environmental covariates or the categorical factor (location) significantly explained the variations in epifaunal assemblage composition across the surveyed range. Between-site variation was responsible for most of the residual, unexplained variation although not at significant levels. The ranked faunal (Bray-Curtis) and environmental (Euclidean distance) similarity matrix was compared using a Spearman correlation coefficient obtained from the RELATE method in PRIMER. The results suggested low correlation between environmental factors (depth, exposure, tide) and biotic composition of the communities (rho=0.26; P=0.16). According to the BIOENV procedure, also in PRIMER, of all environmental variables, tide and depth were the most influential in the faunal distribution patterns although the Spearman correlation value was nevertheless quite low (0.29).

Table. 2.10 PERMANOVA mixed model for biotic (flora and fauna) data obtained from *M. modiolus* beds across the UK based on MNCR Phase II surveys. Significance value established at α =0.05. Random variation (akin to standard deviation: Between-sites=32.05; Within-sites=29.57). Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

Source	df	SS	MS	Pseudo-F	Р
<i>M. modiolus</i> abundance	1	2470.6	2470.6	1.29	0.32
Exposure	1	4479.2	4479.2	1.50	0.33
Tide	1	4978.8	4978.8	1.84	0.22
Depth	1	2469.4	2469.4	1.21	0.35
Location	5	18225	3645.1	1.73	0.20
Site	8	14700	1837.5	2.10	0.06
Residual	2	1748.6	874.31		
Total	19	49072			

SIMPER was used to characterise species composition of the broad significant SIMPROF cluster groupings. Overall *M. modiolus* was the taxa responsible for the highest within-group similarity (5.78 – 10.59%) with the exception of Group VI (Loch Alsh surveys in 2012; see Appendix 4.1 for full SIMPER results) where Limaria hians and Ophiopholis aculeata represented 14% of the total cumulative similarity followed by *M. modiolus* at 6.10%. Most of the dissimilarities between groups were due to the presence of algal species, for example Chorda filum was recorded in Pen Llŷn but was absent from most beds. The same could be said for Laminaria hyperborea which was an integral component of the shallow M. modiolus community found in Corpach (Loch Linnhe) and Loch Creran in 2012 (both forming Group IV). Balanus balanus and Verruca stroemia were abundant in Groups III and VI but were either absent or not recorded in others, the latter probably being the case for the Pen Llŷn beds since they were abundant in records from the same site published by Rees et al (2008). The communities found off Corpach in Loch Linnhe, Loch Alsh or the Pen Llŷn a'r Sarnau SAC bioherms stood apart largely due to the respective presence of either kelp, flame-shell Limaria hians beds and the high abundance of the sponge Amphilectus fucorum. However, it was generally difficult to disentangle the differences due to specific taxa across the broader community spectrum because numerous species were exclusive to one site.



Figure 2.35 Cluster dendrogram based on Bray-Curtis similarity matrices of multivariate epibiotic community data obtained from MNCR dive surveys. Symbols a-k denote significant (P<0.05) groupings while labels indicate broad survey locations.

The presence of site specific taxa, particularly species difficult to identify in the field such as sponges, hydroids and, particularly, bryozoans is most likely the result of inter-surveyor variability. Group III (Figure 2.35) highlights the issue of inter-surveyor variability because it contains all the records of one survey team, regardless of site or year. It also seems likely from the SIMPER analysis that some habitat heterogeneity and therefore key modifying species such as kelp are being captured in the Phase II records, introducing more variance to the analysis.

Overall, significant variation between locations based on available data using just Phase II survey data cannot be explained but is likely to result from a combination of site-specific environmental factors as well as confounding error encountered while recording these factors. Variation between survey teams and seasonality between surveys in different locations can also contribute to the unexplained variance.

Univariate analyses of community indices derived from Phase II MNCR style surveys suggested *M. modiolus* bed diversity and evenness are high and stable across the broad (>1km) geographical range used in this study. Both the boxplots and the PERMANOVA models show high random variability, albeit not significant, between transects in some of the broad locations for all indices. For some locations (Loch Alsh, Loch Creran) this can be interpreted as a degree of temporal variability because the survey team was consistent. In the remaining broad locations the variation was the result of spatial variability between transects located farther apart (>1km, i.e. Shetland, Loch Linnhe). The effects of tide, exposure and depth on epifaunal community indices and composition were not significant again suggesting variability was mostly the result of unexplained spatial and temporal factors. Although this result may seem strange, environmental factors in these surveys were not recorded accurately. Additionally, while some transects were surveyed at an approximately constant depth, others were run along a depth gradient, thereby confounding factors.

Overall, Phase II (MNCR) surveys were able to show some level of spatial and temporal variation in diversity and species composition of *M. modiolus* communities. The variation is likely the result of site specific variation (possibly confounded by different surveyors) as none of the sites are regarded as impacted or experiencing anthropogenic pressures. Overall high diversity indices in all sites provides some confidence in the dataset. Phase II techniques have, nonetheless, detected temporal changes in benthic communities (including *M. modiolus* beds) which was probably from anthropogenic pressures such as trawling (Van Rein *et al* 2011; Strain *et al* 2012). Phase II would appear to generate satisfactory monitoring data but issues of inter-worker variability would need rigorous quality control measures.

b. Community analysis of *M. modiolus* clump samples

Univariate analyses of clump samples

Following data cleaning and standardisation, a total of 989 different taxa belonging to all major phyla were recorded from samples collected using the same clump sampling method (described in Section 2.4.2i.a) across 15 survey sites and, in some cases, different sampling years at the same site. The total number of taxa varied from ~40 to 160 per clump. In horse mussel ridges in Pen Llŷn a'r Sarnau SAC suction samples yielded 100 taxa on average while troughs were less rich, with 84 taxa on average. A total of 572 taxa were recorded following removal of colonial and encrusting species to facilitate quantitative analyses. In Scotland the highest average richness (as opposed to cumulative total species richness in Table 2.7) was recorded from String Rock samples collected in Loch Alsh in 2011 (149) and in 2004 (147). High mean numbers of taxa were also obtained from Port Appin (132); Loch Leven (114); Kyleakin, also in Loch Alsh (104); Loch Creran in 2005 (100); and Basta Voe in Shetland (102). Lowest mean community richness (S) was recorded in Uyea Sound, Shetland (65), followed by Busta Voe (71), North Cava (76), Loch Creran in 1999 (88), Corpach (89) and Hascosay Sound (91). The samples from North Llŷn M. modiolus ridges yielded well over 1,500 individual specimens, however, average numbers (1331) were not much higher than those recorded off String Rock by Emu Ltd. in 2004 (1298) where high abundances (N) were composed largely of polychaetes Pholoe inornata and Sphaerosyllis hystrix. In the Pen Llŷn ridges the taxa responsible for the high N values were crevice fauna and infauna, mostly bivalves such as Hiatella arctica, Kurtiella bidentata and Nucula spp., the detritivorous crab *Pisidia longicornis* and *M. modiolus* itself.

Overall, diversity indices were very high (Figures 2.36-2.40) with the exception of Loch Leven (located within Loch Linnhe) where the abundance of Balanus spp. and Spirobranchus spp. drove the diversity and evenness indices down. If these encrusting species were removed (see Figures 2.37B-2.40B) community diversity and evenness were consistently very high across the range. Average Shannon-Wiener's values (calcareous polychaetes and Balanus spp. included) were higher than 3 in most beds, most notably North Cava (3.77), Port Appin (3.71), Basta Voe (3.63), Loch Creran (3.62) and String Rock in 2011 (3.54) and 2004 (3.52). Mean H' varied from 3.4 to 3.32 in ridges and troughs in Wales while the lowest diversities were recorded in Uyea Sound and Busta Voe (2.56 and 2.45 respectively). Results from clearance quadrats investigated in impacted and relatively intact reefs in Strangford Lough (Farinas-Franco & Roberts, in prep.) were included in the boxplot charts to obtain a visual comparison between spatial (due to natural, site-specific conditions) and pressure-related change. The boxplots suggest that untrawled *M. modiolus* reefs in Strangford Lough were similarly diverse to untrawled reefs in Scotland and Wales (3.07). The impacted sites, however, were substantially less diverse (1.92) indicating that, although spatial and temporal variations might occur under natural conditions, community change due to anthropogenic physical pressures is discernible from background variations in community indicators (diversity, richness, evenness).

Mixed PERMANOVA models for all indices were not significant except the total abundance (N) which showed a highly significant relationship with *M. modiolus* abundance and a significant relationship with tidal speed. The total number of species had a near significant relationship with *M. modiolus* abundance (Table 2.11). As the samples with highest number of *M. modiolus* were collected using suction samplers (Pen Llŷn a'r Sarnau) it is possible that sample effort influenced the results. Diversity indices, on the other hand, were independent of sample size as *M. modiolus* did not significantly explain the variations in those indices. If the data were re-analysed removing barnacles and encrusting polychaetes there were no significant differences in total abundances (N) between sites aside from a significant variation between surveys. This result indicates that it is the provision of additional niches in the form of shell substrata that is a strong driver of the community biodiversity value. The remaining PERMANOVA models found no significant broad-scale spatial variation in any of the chosen community diversity metrics (Tables 2.11A-D). However, survey (either different site within location or different sampling year for the same site) was a significant random effect, explaining most of the random variability across the assemblages. The residual error was interpreted as random differences in the assemblages between each of the four clumps sampled at each surveyed site and year.

Overall it appears that high diversity (Shannon-Wiener's index) and high total abundance of individuals (N) are characteristic metrics for 'natural' *M. modiolus* infaunal communities. In an indicator actual values for diversity may need to be site specific and adjusted for tidal regime and the density of the bed but diversity (Shannon-Wiener's index) will occur when impacted by trawling activity.



Figure 2.36 Total number of taxa recorded in *M. modiolus* clumps across seven broad geographical locations in the UK. Each box corresponds to different *M. modiolus* beds surveyed within the same broad location. Locations with numbers (see legend) represent repeat surveys carried out at those sites, eg String Rock99 or Loch Creran99 are surveys from those sites in 1999. The box represents the interquartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values.

A) Burts Vas 00 1500 Basta Voe Round North Cava Total abundance (N) Stei 1000 String Redid4 String Redit1 Kyleakin Loch Lever 500 Loch Cretan@ Looh Cresan08 Pes Livo Pen Llyn_trough Trawled Untravied Shetland Loch Linnhe Loch Creran Orkney Loch Alsh Pen Llyn Strangford Lough B) Busta Voe99 1200 Basta Voe Hassesay Soun Uyea Sound 1000 North Cava Total abundance (N) Guttersound String Rosk96 800 String RockO4 String Rock11 Kyle akin 600 Looh Leves Carpach Part Appin 400 Looh Creran00 Losh Cresan05 Pen Llyn_ridger 200 Pan Live trough Travled 📫 Untrawied Loch Creran Loch Alsh Loch Linnhe Pen Llyn Strangford Lough Shetland Orkney

Figure 2.37 Total abundance of individuals (N) recorded in *M. modiolus* clumps across seven broad geographical locations in the UK. A) With *Balanus* spp. and *Spirobranchus* spp. present. B) With *Balanus* spp. and *Spirobranchus* spp. removed. Each box corresponds to a different *M. modiolus* bed surveyed within the same broad location. Locations with numbers (see legend) represent repeat surveys carried out at those sites, eg String Rock99 or Loch Creran99 are surveys from those sites in 1999. The box represents the interquartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values.



Figure 2.38 Shannon-Wiener's diversity (H') recorded in *M. modiolus* clumps across seven broad geographical locations in the UK. A) With *Balanus* spp. and *Spirobranchus* spp. present. B) With *Balanus* spp. and *Spirobranchus* spp. removed. Each box corresponds to a different *M. modiolus* bed surveyed within the same broad location. Locations with numbers (see legend) represent repeat surveys carried out at those sites, eg String Rock99 or Loch Creran99 are surveys from those sites in 1999. The box represents the interquartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values.



Figure 2.39 Margalef's species richness (d) recorded in *M. modiolus* clumps across seven broad geographical locations in the UK. A) With *Balanus* spp. and *Spirobranchus* spp. present. B) With *Balanus* spp. and *Spirobranchus* spp. removed. Each box corresponds to a different *M. modiolus* bed surveyed within the same broad location. Locations with numbers (see legend) represent repeat surveys carried out at those sites, eg String Rock99 or Loch Creran99 are surveys from those sites in 1999. The box represents the interquartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values.



Figure 2.40 Boxplots comparing Pielou's evenness (J) recorded in *M. modiolus* clumps across sevem broad geographical locations in the UK distributional range. A) With *Balanus* spp. and *Spirobranchus* spp. present. B) With *Balanus* spp. and *Spirobranchus* spp. removed. Each box corresponds to a different *M. modiolus* bed surveyed within the same broad location. Locations with numbers (see legend) represent repeat temporal surveys carried out at each site (i.e. String Rock, Loch Creran). The box represents the interquartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values.

Table 2.11 Results of mixed PERMANOVA models for diversity indices calculated from replicate *M. modiolus* clumps collected from beds across their UK range. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

A) Total number of specie	es. Survey ra	andom variance=	4.11; Error varia	nce= 3.05	
Source	df	SS	MS	Pseudo-F	Р
M. modiolus abundance	1	115.38	15.38	4.19	0.05
Exposure	1	130.51	130.51	1.66	0.22
Depth	1	217.24	217.24	2.08	0.17
Tide	1	23.15	174.63	0.62	0.39
Location	5	873.14	73.71	2.29	0.13
Survey	8	589.69	73.71	7.89	0.001
Residuals	2	457.97	9.35		
Total	19	2407.1			
B) Total abundance of in	dividuals (N)	. Survey random	variance=7.87; E	rror variance=6.18	
Source	df	SS	MS	Pseudo-F	Р
M. modiolus abundance	1	1518.10	1518.1	14.44	0.002
Exposure	1	865.28	865.28	2.95	0.11
Depth	1	399.69	399.69	1.05	0.38
Tide	1	805.82	805.82	5.75	0.01
Location	5	2743.40	548.68	1.93	0.20
Survey	8	2199.8	274.98	7.20	0.001
Residuals	2	1872.2			
Total	19	10404			
C) Margalef's richness (d	I). Survey rar	ndom variance=4	.09. Error variand	e=2.74	
Source	df	SS	MS	Pseudo-F	Р
M. modiolus abundance	1	23.41	23.41	0.92	0.31
Exposure	1	45.48	45.48	0.60	0.50
Depth	1	145.62	145.62	1.44	0.27
Tide	1	0.59	0.59	0.02	0.92
Location	5	660.97	132.19	1.79	0.23
Survey	8	571.42	71.43	9.51	0.001
Residuals	2	367.89			
Total	19	1815.4			
D) Shannon-Wiener's div	<u>versity (H'). S</u>	urvey random va	riance=5.02; Erro	or variance= 2.25	
Source	df	SS	MS	Pseudo-F	Р
M. modiolus abundance	1	27.16	27.16	0.84	0.35
Exposure	1	7.90	7.90	0.01	0.93
Depth	1	39.97	39.97	0.29	0.88
Tide	1	18.24	18.24	0.39	0.42
Location	5	255.27	51.05	0.49	0.77
Survey	8	811.26	101.41	19.92	0.001
Residuals	2	249.45			
Total	19	1409.3			
E) Pielou's evenness (J).	Survey rand	Iom variance=4.4	5; Error variance	= 2.09	
Source	df	SS	MS	Pseudo-F	<u>P</u>
<i>M. modiolus</i> abundance	1	59.76	59.76	2.32	0.14
Exposure	1	29.78	29.78	0.35	0.63
Depth	1	7.80	7.80	0.08	1
lide	1	28.03	28.03	0.76	0.23
Location	5	131.76	26.35	0.32	0.89
SURVEY	8	641.99	80.25	18.40	0.001
Desiduala	õ	040.07			
Residuals	2	213.67			

Multivariate analyses of clump samples

The stress value for the MDS ordination plot was 0.21 suggesting the two-dimensional plot (Figure 2.41) was of borderline use in representing the relationships between the clump samples. Multivariate data were largely aggregated by location and survey within location (site or year). Distinct year groups suggest a temporal shift in community composition where repeated surveys had been undertaken (such as Loch Alsh and Loch Creran).

SIMPROF analysis identified significant differences between *M. modiolus* clump samples grouped according to location, with all significant (<0.05) clusters diverging at the 40% similarity threshold. The only exception were the horse mussel clumps collected in Loch Alsh and Loch Creran in 1999 by Mair *et al* (2000) which grouped together (Figure 2.42).



Figure 2.41 Two-dimensional MDS plots for biotic data with *M. modiolus* abundance (number of mussels per sample) overlaid as bubble plots. High two-dimensional stress value (0.21) is borderline for good representation of multivariate relationships suggesting the results should be interpreted with caution. Label acronyms: PL=Pen Llyn (R=Ridge; T=trough); LC=Loch Creran; SH=Shetland (BV=Busta Voe (99 only); BV=Basta Voe (2012); US=Uyea Sound; HS=Hascosay Sound); LA=Loch Alsh (SR/MOD=String Rock; KA=Kyle Akin); OY=Orkney (G=Guttersound; K=North Cava); LLn=Loch Linnhe (CP=Corpach; PA=Port Appin; LL=Loch Leven). The number after the first two letters of the label represents sampling year.



Figure 2.42 Cluster diagram based on Bray-Curtis similarity matrices of multivariate biotic community data obtained from *M. modiolus* clumps. Pen Llŷn data were collated using suction samplers whereas all others were collected as clumps (see methods). Symbols a-t denote significant (P<0.05) SIMPROF groupings while labels indicate survey locations. Label acronyms: PL=Pen Llyn (R=Ridge; T=trough); LC=Loch Creran; SH=Shetland (BV=Busta Voe (99 only); BV=Basta Voe (2012); US=Uyea Sound; HS=Hascosay Sound); LA=Loch Alsh (SR/MOD=String Rock; KA=Kyle Akin); OY=Orkney (G=Guttersound; K=North Cava); LLn=Loch Linnhe (CP=Corpach; PA=Port Appin; LL=Loch Leven). The number after the first two letters of the label represents sampling year.

PERMANOVA models (Table 2.12) found that depth and abundance of horse mussels exert a significant effect in the species composition of assemblages associated with *M. modiolus* clumps. The categorical factor (location) significantly explained assemblage composition. Between-site variation was significant across individual survey sites and it was responsible for most of the residual, unexplained variation. The bubble-plot displayed in Figure 2.41 helps visualise the influence of sampling effort in the distribution of multivariate community data because samples with highest numbers of horse mussels were obtained from Loch Linnhe and the ridges in the Pen Llŷn a'r Sarnau SAC bioherms but the trough samples collected in the Pen Llŷn contained low numbers of mussels (<7) but nevertheless grouped with the high density samples. Although not included as a factor in the models, community composition in *M. modiolus* might also be influenced by type of dominant substratum (OSPAR 2009; Holt *et al* 1998). The absence of accurate PSA data for all sites surveyed precluded its evaluation for this project.

Table 2.12 PERMANOVA mixed model for biotic composition obtained from *M. modiolus* clump removal surveys across the UK. Significance value established at α =0.05. Random variation (akin to standard deviation) for survey nested in broad location =32.53; Residual variance=25.86. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

Source	df	SS	MS	Pseudo-F	Р
M. modiolus abundance	1	4765.1	4765.1	2.63	0.001
Exposure	1	7073.3	7073.3	1.42	0.06
Tide	1	7071.7	7071.7	1.44	0.06
Depth	1	13011	13011	1.98	0.001
Location	5	51996	10399	2.12	0.001
Survey(Location)	8	33843	4834.7	7.05	0.001
Residual	49	34273	685.46		
Total	66	1.52*10 ⁵			

Analysis of clump biotic data provided an in-depth view of the community with almost 1,000 different taxa, mostly macro-invertebrates, characterising *M. modiolus* reefs across their UK range. The community consisted of typical crevice fauna (e.g. *Hiatella arctica, Pisidia longicornis, Harmothoe* spp., *Polynoe* spp., *Pholoe* spp.), infauna (e.g. *Nucula nucleus, Abra alba*, cirratulids, *Polycirrus* spp. and other terebelids) and epifauna attached to the mussels (e.g. Anomiidae, *Balanus* spp., *Spirobranchus, Ascidia* spp.) as well as vagile species, particularly brittlestars *Ophiothrix fragilis* and *Ophiopholis aculeata* which were super-abundant in some beds.

SIMPER analysis results (Appendix 4.2) indicated that, apart from *M. modiolus* itself, taxa contributing the most to within group similarities for each survey area were the polychaetes *Nereymira punctata, Pholoe* spp. and *Spirobranchus* spp.; the crevice dwelling bivalve *Hiatella arctica*; saddle oysters *Anomia* spp.; brittlestar *Ophiopholis aculeata* and the crab *Pisidia longicornis*. It was interesting to note that *Ophiothrix fragilis* and *Ophiopholis aculeata*, while typifying some beds, such as those in North Cava and String Rock, were absent from Loch Linnhe. All significant SIMPROF groups were highly dissimilar from each other (>75%) suggesting community composition in *M. modiolus* beds is highly variable, even across these diverse and relatively unimpacted beds, largely as a result of site specific random unexplained variation and the influence of depth and abundance of horse mussels (all significant factors in PERMANOVA, see Table 2.12). The least dissimilar beds were those with similar abundances of horse mussels found within the same broad survey areas: North Cava and Guttersound communities were 51% dissimilar with 53 sediment and crevice dwelling taxa responsible for 50% of the dissimilarities while in Loch Alsh, communities

surveyed in Kyleakin and String Rock in 2011 were just 40% dissimilar. Community composition also varied in those locations where repeated sampling took place (String Rock and Loch Creran). Dissimilarity between the String Rock communities sampled in 2004 and 2011 was 60% with notable decreases in *Ophiothrix fragilis*, for example. Loch Creran communities sampled in 1999 and 2005 were 50% dissimilar.

Overall it was difficult to establish patterns in community differences using high resolution taxonomic data due to the broad range of species found and the large area covered. While many differences between groups were the result of species commonly found in *M. modiolus* communities being recorded in differing densities (*Pholoe* spp. *Polynoe* spp., *Flabelligera affinis*), the majority of community composition differences were site specific. Of these, it is possible that some common taxa such as spirorbids (i.e. *Jugaria granulata*), ostracods, *Balanus* spp. or isopods (i.e. *Janira maculosa*), which were responsible for a high percentage of the dissimilarities between some areas, were simply overlooked in some of the analyses.

According to the PERMANOVA models, depth has a significant effect in community composition in *M. modiolus* reefs. The shallowest beds were found off Corpach in Loch Alsh and Hascosay Sound in Shetland, both very sheltered areas with weak to moderate tidal streams. In the case of Corpach, where the *M. modiolus* bed was just 7 m deep, the area was also subjected to inputs of freshwater run-off. Both communities were dominated by a detritivorous polychaete fauna (terebellids, Capitella capitata, Mediomastus fragilis, spionids and cirratulids including Cirratulus serratus and Chaetozone spp.) likely associated with abundant soft sediments and faeces and pseudofaeces deposited in high quantities amongst the mussel clumps as a result of prevailing hydrodynamic conditions. One of the most relevant outcomes of the multivariate analyses of the clump faunal data is that monitoring the fauna associated with small clump samples can capture spatial and temporal change in horse mussel reef community composition but there is a substantial amount of site-specific variation, i.e. a standard *M. modiolus* community is hard to define using the infaunal component. It would therefore be difficult to detect anthropogenic pressures based on community composition analyses, which suggests that univariate indices (see previous section) are more suited for detecting human impacts on the reef community.

Relationship between community and density indicators

Spearman's correlation indices showed a positive, non-significant correlation between abundance of *M. modiolus* in the clumps and: A) number of species S (r = 0.21; P=0.08); B) total abundance of individuals N (r=0.20; P=0.1); C) Margalef's richness index d (r = 0.22; P =0.08); D) Shannon-Wiener's diversity H' (r=0.22; P=0.08); and E) Pielou's Evenness J (r=0.18; P=0.14). Non-linear regression models indicated an asymptotic exponential relationship between abundance of *M. modiolus* and community evenness (J) and diversity indices measured as Margalef's richness (d) and Shannon-Wieners' diversity (H) (Figure 2.38A & B). However, the relationship between total abundance of individuals and numbers of horse mussels was only weakly linear (Figure 2.43C).

The regression models suggest a rapid increase in community diversity and evenness towards a climax community even at very low mussel densities (<10 mussels m⁻²) which further reinforces the theory that *M. modiolus* has a crucial role as a habitat engineer and ecosystem engineer. Similar dynamics were found for *Serpula vermicularis*, another reef building foundation species, in Loch Creran (Chapman *et al* 2011). One of the main outcomes following these findings is that, above a density of 10 mussels m⁻², a large clump sample size is not crucial to monitor condition of *M. modiolus* beds using univariate

community indicators. A high sampling effort is not required to obtain a meaningful and informative snapshot of the condition of the associated communities because past a threshold of mussel abundance community metrics of biodiversity and evenness reach an asymptote.

The poor correlation between abundance of *M. modiolus* and cumulative abundance of individuals associated with the reef (N; Figure 2.43C) suggests the total abundance of taxa (N) is not a good indicator of the condition of *M. modiolus* communities. High abundance (N) values could actually be indicative of impacted conditions e.g. high numbers of *Pisidia longicornis* and tubificids and capitellids were found in impacted beds in Strangford Lough (Roberts *et al* 2011; Fariñas-Franco & Roberts in prep.). Tubificids and capitellids are known to be super-abundant in stressed benthic habitats (Nilsson & Rosenberg 2000; Rosenberg *et al* 2004).



Figure 2.43 Relationship between the abundance of *M. modiolus* in clump samples and diversity (A, B); total fauna (C); and community evenness (D) recorded *M. modiolus* reefs in Scotland and Wales.

c. Comparison between in situ methods

In situ Phase II MNCR surveys carried out by divers are adequate methods to map biotopes at fine scales (10s of metres), however, their taxonomic resolution is low because they focus on conspicuous epifaunal and mobile species. The method requires skilled divers with a high level of taxonomic expertise, for example, certain sponges, hydroids and bryozoans are closely associated with *M. modiolus* reefs and failure to obtain a positive identification in the field as a result of inter-surveyor variability can be interpreted as absence or decrease and may be a substantial source of error in the data presented here. For the purpose of this study, MNCR records were pooled into broad-scale locations to provide appropriate

replication. However, the results of the PERMANOVA analyses for each diversity index and overall community composition indicated MNCR surveys failed to capture significant spatial or temporal changes in community diversity or species composition. Considering that the diversity and evenness indices were high across all sites it is likely that, in the absence of natural or anthropogenic pressures, the epifaunal assemblages remain temporally and spatially stable. This finding is in accordance with the results of the monitoring surveys undertaken by CCW in the Pen Llŷn a'r Sarnau SAC (discussed by Keenan *et al* 2010).

Although MNCR surveys could detect abrasion and other physical impacts in the same way that other visual methods such as video-quadrats have achieved (Cook et al 2013; Section 2.5) they are probably less objective compared to the use of a quadrat or photographic still. Clump collection, on the other hand, is a method of much higher taxonomic resolution because most components of the reef assemblage are captured. Superficially, as a field method, clump collecting might appear more cost effective because it does not require benthic taxonomists in the field, but in practice, properly trainined scientific divers are still needed to do this sampling. Sample processing and taxonomic identification from clumps in the laboratory is nevertheless costly and requires highly trained staff if using the highest possible taxonomic resolution (species level). Statistical analyses following substantial data cleaning and standardisation suggested clump communities were species rich (N, D, d), diverse (H') and very even (J) across all sites, even though the communities varied considerably. When community data from a known impacted site (Strangford Lough's north basin) were introduced into the analyses the differences in community indices were apparent, suggesting variability across non-impacted sites in Scotland and Wales was down to natural, site-specific differences. Both in situ and clump methods indicated M. modiolus beds host highly species diverse and even communities, however, the mean diversity and evenness indices calculated for the clumps were higher than those obtained using SACFOR scale numerical equivalences (Table 2.13; Figure 2.44A-D) and the coefficient of variation (standard deviation*mean⁻¹) was lower from clump samples compared to MNCR phase II community data (see Chi-square and significance values in Table 2.13). This means that, of the two methods, clump sampling has comparatively lower inherent variability and is thus more likely to detect changes in community metrics in a monitoring programme.

Variable	X^2	df	Р	Coefficient of va	ariation (%)
				Clumps	MNCR
S	7.4	1	<0.001	13.0	28.4
d	7.4	1	<0.01	10.2	24.9
H'	83	1	<0.01	34	79

< 0.01

0.2

2.5

8.4

Table 2.13 Results of non-parametric Kruskal-Wallis ranked analysis for *M. modiolus* community indices comparing *in situ* MNCR diver surveys and clumps of horse mussels. The comparisons are between mean indices calculated for each broad survey area. S=Number of taxa; d=Margaelf's richness (d); H'=Shannon-Wiener's diversity; J=Pielou's evenness.



Figure 2.44 Mean community indices and standard error bars for *M. modiolus* communities surveyed using *in situ* methods (MNCR phase II type surveys, in blue; and *M. modiolus* clump removal, in red). A) Total number of taxa (S); B) Margalef's richness (d); C) Shannon-Wiener's diversity (H'); and D) Pielou's evenness (J).

Box 2.3 Summary of findings for community indicators using in situ methods

- Univariate indices of community richness, diversity and evenness are high and stable across unimpacted *M. modiolus* beds in the UK;
- There is spatial and temporal variability in univariate community metrics but these are site-specific and only significant at medium to fine spatial scale (within survey area and between replicates);
- Univariate metrics for community indicators appear robust to fluctuations in sample size with the exception of total abundance of fauna (N);
- Tidal energy was also a significant factor affecting N; these relationships were driven by encrusting species associated with the mussels;
- Univariate indices respond to anthropogenic physical damage (e.g. bottom trawling in Strangford Lough);
- Variation in community composition (multivariate analysis) is largely site-specific although the infaunal assemblages are also affected by *M. modiolus* density and depth. Community composition analyses did not yield useful information for potential indicator species or a 'type' community;
- Phase II MNCR style surveys can capture high diversity and evenness however they are susceptible to higher methodological variability compared to metrics obtained for community indicators using clump sampling;
- Overall, Shannon-Wiener diversity index and Pielou's evenness are stable across all sampled sites but sensitive to anthropogenic impact (abrasion and removal). These two indices may provide the metric for comparing condition of *M. modiolus* communities. Reference conditions for these habitats could be calibrated incorporating these or other similar indices such as AMBI (Borja *et al* 2009) or IQI;
- The relationship between mussel density and community indicators is exponentially asymptotic suggesting that the condition of the community measured using those indices remains largely unchanged above a cut-off mussel density value (~10 mussels m⁻²);
- Regular monitoring of the epifaunal community coupled with density estimations could prove sufficient to detect sudden shifts in reef health as a result of physical impact or other anthropogenic pressures (Section 2.5 Validation).

iii. Remote community sampling methods

Community data from DDV footage were obtained for eight broad locations where *M. modiolus* beds are found, namely: 2011 DDV surveys by NRW off North Pen Llyn (Wales, nine stations); Noss Head (50 stations); Copinsay and Shapinsay (Orkney, 153 stations); Uyea Sound, Colgrave Sound, Hascosay Sound and Basta Voe (Shetland, 93 stations). Four raw datasets containing SACFOR information from 305 video tows were scanned for presence of *M. modiolus* yielding a total of 32 datasets for communities containing *M. modiolus*. The data were supplemented with existing biotope GIS layers to select *M. modiolus* biotopes. All datasets containing *M. modiolus*, regardless of the community being classified as a reef or not, were used in multivariate analyses to detect the effect of different densities of horse mussels on the benthic community compared to areas without them. The data were interrogated by means of univariate and multivariate analyses of species abundances calculated using an arbitrary numerical scale of equivalences (Keenan *et al* 2010; Table 2.3).

a. Univariate analyses

In total, 200 taxa were identified in all DDV datasets used. Cumulatively, the highest number of taxa were recorded in Shetland beds (106), followed by Orkney (96), Wales (74) and Noss Head (68). Mean community diversity (H') and Margalef's richness (d) were relatively low if compared with those captured for *M. modiolus* reefs using *in situ* methods (2.5-2.7 for H' and 3.5 to 4.4 for Margalef's d). However, mean community evenness (J) was very high across all beds (J=0.95-0.97). The results displayed in Figure 2.45 suggest some degree of spatial variability for all indices across and within each surveyed area. Within-site variation, measured as coefficient of variance (CV), is displayed in Table 2.14 showing high variability for S and d, particularly in Wales. The least variable indices across all sites were H' and J, the latter having the lowest CV of all metrics used. PERMANOVA tests indicated that DDV was not able to capture significant changes in diversity indices across the studied areas (Table 2.16). A preliminary assessment of the environmental covariates using Draftman's plots indicated they were correlated and therefore the full PERMANOVA models were simplified to include only one continuous environmental variable; tidal current. PERMANOVA results showed none of the diversity indices could be statistically to the physical environment (exposure, depth or tidal current).

	-				
Index	Noss Head	Orkney	Shetland	Wales	Total method
S	20%	42%	14%	73%	34%
d	16%	35%	12%	62%	28%
н	8%	22%	6%	24%	14%

1.6%

0.9%

1.4%

1.4%

1.2%

J

Table 2.14 Coefficients of variance (CV) for community diversity metrics obtained from drop down video tows in four representative *M. modiolus* beds. S=Number of taxa; d=Margaelf's richness (d); H'=Shannon-Wiener's diversity; J=Pielou's evenness.



Figure 2.45 Number of taxa (S); Margalef's richness (d); Shannon-Wieners' diversity index (H'); and Pielou's evenness (J) calculated for SACFOR abundance of benthic taxa observed in DDV footage collated from eight *M. modiolus* beds. The box represents the interquartile range, with a line indicating the median and whiskers representing the maximum and minimum values observed in each discrete video tow.

It should be taken into consideration that DDV is normally used as a broad-scale habitat monitoring method to collate coarse community data where conditions are usually too difficult for divers to undertake fine-scale surveys (Holt *et al* 2001b). The footage obtained from the Welsh surveys was qualitatively evaluated in parallel with the statistical analyses showing some variability resulting in poor visibility in some video clips. Poor image quality could likely explain the high variability observed in number of species (S) and species richness (d) in Welsh *M. modiolus* beds (Table 2.15).

Diversity (H') and particularly evenness (J) had the lowest variability across sites, which might suggest those metrics could perform best as candidate community indicators for *M. modiolus* reef condition but equally may be a product of the low resolution of the method and the relatively few visible organisms. It also remains untested if diversity indexes derived from DDV footage using SACFOR abundance data would be responsive to anthropogenic pressures.

Table 2.15 Results of mixed PERMANOVA models for diversity indexes calculated from DDV footage at eight UK *M. modiolus* beds. Significance value established at α =0.05. Sums of squares type was Type I (sequential). A total 999 permutations of the residuals were undertaken under a reduced model. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

A) Total number of species.					
Source	df	SS	MS	Pseudo-F	P
M. modiolus abundance	1	13.01	13.01	2.02	0.3
Tide	1	17.72	17.72	2.38	0.21
Location	3	406.26	135.42	4.18	0.13
Survey	5	154.36	30.87	0.25	0.90
Residual	18	2507.6	125.38		
Total	30	3099			
B) Margalef's d richness.					
Source	df	SS	MS	Pseudo-F	Р
M. modiolus abundance	1	12.29	12.29	1.98	0.31
Tide	1	23.23	23.23	2.46	0.25
Location	3	315.04	105.01	4.05	0.14
Survey	5	118.9	23.78	0.26	0.89
Residual	18	2298.70	91.46		
Total	30	2298.7			
C) Shannon-Wiener's diversit	ty (H').				
Source	df	SS	MS	Pseudo-F	Р
<i>M. modiolus</i> abundance	1	0.35	0.35	2.21	0.28
Tide	1	1.81	1.81	2.75	0.22
Location	3	78.48	26.16	5.11	0.08
Survey	5	24.32	4.86	0.20	0.91
Residual	18	481.7	24.08		
Total	30	586.67			
D) Pielou's evenness (J).					
Source	df	SS	MS	Pseudo-F	Р
<i>M. modiolus</i> abundance	1	3.30*10-4	3.30*10-4	0.26	0.95
Tide	1	0.15	0.15	0.81	0.57
Location	3	1.43	0.48	1.59	0.24
Survey	5	0.70	0.14	1.68	0.17
Residual	18	1.67	0.08		
Total	30	3.95			

b. Multivariate analysis

DDV systems were able to detect significant differences in community composition between broad areas where *M. modiolus* beds were surveyed (PERMANOVA P<0.001; Table 2.17) although there was no detectable variation in the structure of the assemblages among beds (sites in each location). There was a highly significant relationship between the covariates (abundance of *M. modiolus* and tidal stream) and community structure (Table 2.16; Figures 2.46A & B).

M. modiolus communities defined using DDV systems were grouped into nine significant SIMPROF subgroups which could be aggregated into six larger groups after cluster diagram (40% similarities) investigation. SIMPER analysis (Appendix 4.3) indicated that, in most beds, *M. modiolus* was the highest contributor to the within-group similarities. Samples S16B and S72 from Shetland formed a distinct separate community of *M. modiolus* dominated by mobile species *Echinus esculentus*, *Pagurus bernhardus*, *Asterias rubens*, crabs and scallop spp. *Pecten maximus* with no apparent sessile epifauna contributing significantly to the similarities. *M. modiolus* beds from Copinsay (C4 and C14) and Pen Llŷn formed a larger group, beds from Wales having high abundances of *Ophiothrix fragilis*. Beds from Noss Head and Copinsay (C10, C11 and C13) were characterised by superabundant *M. modiolus* and common or abundant *Ophiothrix fragilis*. *M. modiolus* assemblages from Shapinsay were distinct at the 40% level as a result of the higher abundance of calcareous polychaetes, bryozoans *Flustra foliacea* and encrusting coralline algae. The dissimilarities were, however, very apparent in Pen Llŷn where, apart from the very distinctive *A. digitatum* colonies, very few taxa were recorded in comparison to other beds.

DDV has overall succeeded in detecting broad-scale spatial differences in community composition for apparently natural *M. modiolus* beds under different environmental conditions of tide and exposure. The differences in species composition picked-up by SIMPER analysis indicated that, for example, *A. digitatum* was one of the biggest contributors to the dissimilarities between groups mostly as a result of being absent from some beds and being superabundant in others. Although barnacles *Balanus* spp. and tubiculous polychaetes *Spirobranchus* spp. were also important contributors to most of the between-group differences, confidence in these results is lower because these taxa are more susceptible to surveyor bias as they can be either ignored or not spotted. There were also noticeable differences in the relative abundances of mobile opportunistic taxa (e.g. *E. esculentus* and *A. rubens*) and ophiuroids.

Temporal shifts in community composition from sessile species towards opportunistic scavengers have been linked to physical damage in Strangford Lough by Roberts *et al* (2011) and Strain *et al* (2012). However, natural variation in species composition of stable benthic communities is common (Bell & Barnes 2001; Balata *et al* 2006). Without temporal data and proper controls it is difficult to determine if the absence of *A. digitatum* and the abundance of opportunistic species found in some beds are pressure related (Cook *et al* 2013) or linked to site-specific natural conditions. Nonetheless, the results here suggest composition of the epifaunal *M. modiolus* communities could be determined using DDV to establish reference conditions. These surveys should be site specific as the communities are largely influenced by environmental factors as well as the abundance of *M. modiolus*. Once benchmark conditions have been established, multivariate analyses of semi-quantitative community data obtained from DDV footage has the potential to detect shifts in the assemblages that could be linked to impact if the proper experimental controls (e.g. Before-After-Control-Impact (BACI) designs) are established.

Table 2.16 Mixed PERMANOVA model for biotic composition obtained from *M. modiolus* DDV surveys across three UK *M. modiolus* beds. Significance value established at α =0.05. Random variation (akin to standard deviation) for survey nested in broad location =13.00; Residual variance=34.22. Sums of squares type was Type I (sequential). A total 999 permutations of the residuals were undertaken under a reduced model. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

Source	df	SS	MS	Pseudo-F	Р
M. modiolus abundance	1	7056.2	7056.2	3.65	0.002
Tide	1	7290.5	7290.5	3.37	0.001
Location	3	17756	5918.8	2.38	0.004
Site(Location)	5	6199.9	1240	0.97	0.53
Residual	18	25699	1284.9		
Total	30	64002			



Figure 2.46 Two-dimensional MDS plots for biotic data with *M. modiolus* abundance (A) and tidal stream values (B) overlaid as bubble plots. Green ellipses delineate significant SIMPROF groups. High two-dimensional stress value (0.2) is borderline for good representation of multivariate relationships suggesting the results should be interpreted with caution.

2.0

iv. Comparison between *in situ* and remote methods for community indicator monitoring

An ideal monitoring method should be able to detect spatial variability while minimizing the random, within-replicate variance (Bart *et al* 2002; Elphick 2008; Underwood & Chapman 1998). The surveys carried out in Orkney and Shetland by HWU and at Pen Llŷn a'r Sarnau by NRW produced species composition lists for all three different *in situ* and remote methods that allowed a joint analysis of sensitivity to spatial change as well as variance estimations for each method. Figure 2.47 shows the barplots and standard error bars for all diversity indices calculated for all 15 survey data sets acquired and re-analysed for this study. The results of a mixed PERMANOVA model, using location and method as fixed factors and site nested in location as a random factor, indicate significant differences between all three data collection methods across the sites for all community indices tested:

- Number of taxa S (Pseudo-F_(2,47)=110.35; P=0.001);
- Margalef's richness d (Pseudo-F_(2,47)=109.11; P=0.01);
- Shannon-Wiener's diversity H' (Pseudo-F_(2,47)=59.15; P=0.01);
- Pielou's eveness J (Pseudo-F_(2,47)=41.29; P=0.01).

The only non-significant methodological differences were found for evenness (J) obtained from MNCR Phase II dive surveys and DDV at any survey site. Between-site differences in community indices were significant for d (Pseudo-F $_{(2,47)}$ =3.32; P=0.04), H (Pseudo-F $_{(2,47)}$ =3.62; P=0.03) and J (Pseudo-F $_{(2,47)}$ =57.51; P=0.001). A post-hoc analysis found that for all methods, only clump analyses had high enough spatial resolution to identify significant (P=0.001) spatial variation among all sites for each of the community indices considered. Of all the methods considered, the MNCR Phase II dives yielded the lowest co-efficients of variance (CV) for S, d and H' calculations (0-33% depending on the surveyed site). For evenness estimates, analyses of clump replicate samples were the least variable (0.3-0.5%). The highest variance was consistently recorded for DDV surveys. The indices showing the least variance (CV) across all methods were Shannon-Diversity (H, 1-4% for MNCR and clump data and 6-24% for DDV) and Pielou's evenness J (0.1-1.3%).

Pielou's evenness (J) is able to capture spatial variability while experiencing the least variability across all methods. It performs well as it works on the basis of expected diversity, should all the species be equally abundant in the community (as opposed to dominance). Other indices are more heavily influenced by the method of data collection used or sample effort. For example, communities sampled by clearance methods (e.g. clump collection or suction sampler) appear richer (in terms of S and d) than if they were sampled using visual observations by divers or imaging methods (Figure 2.47). Nonetheless, it is possible that the stability in evenness (J) is indicative of this index not being able to detect the impacts of pressures on *M. modiolus* reefs. However Figure 2.47 shows a distinct reduction in evenness (J) between possibly unimpacted beds in Scotland and Wales and those from Strangford Lough where a known impact has occurred. Fariñas-Franco & Roberts (in press) also found significant temporal increases in evenness (J) in translocated *M. modiolus* clumps suggesting the index is also able to capture temporal variability and recovery in impacted M. modiolus communities, from an altered state towards a more stable one. Cook et al (2013) found significant reductions in community evenness (J) in impacted reefs while Figure 2.47 in this section shows a significant reduction in evenness (J) between Welsh and Scottish reefs as a result of including data from *M. modiolus* troughs, which are typically species poor (Rees et al 2008).



Figure 2.47 Mean community indices with standard error bars for *M. modiolus* communities surveyed using in–situ and remote methods A) Total number of taxa (S); B) Margalef's richness (d); C) Shannon-Wiener's diversity (H'); and D) Pielou's evenness (J).

Box 2.4 Summary of findings for community indicators using remote methods.

- Epifanal community composition was site-specific at a broad-scale (>10km) but no differences were detected within each surveyed area.
- Epifaunal community varied with *M. modiolus* density and tidal flow.
- DDV could be used to monitor presence and abundance of characterising sessile taxa for each discrete bed as an indicator of, for example, abrasion but otherwise lacks ability to represent the biodiversity of the site in any detail.
- Shannon-Weiner's diversity (H') and Pielou's evenness (J) had the lowest coefficients of variance of all indices and could be put forward as the most reliable of all metrics for the purpose of reef community condition monitoring.
- The overall poor performance of DDV in capturing community information and metrics (if compared with *in situ* methods), suggests high resolution still cameras (with better resolution) should be limited to monitoring of extensive or deep *M. modiolus* beds where diving is impractical or, comparatively, too expensive.
- H' and J obtained from Phase II survey would, on the whole, be likely to show the effects of anthropogenic pressures because variance in these measures is lower. There appears to be scope for reduction in interworker variability by introducing task-specific training.
- Clump sampling provides greater community detail, greater resolution and lower variance.
- DDV would be the next best option for monitoring these indicators where diving would be too expensive or impractical.

Box 2.5 Recomendations for community indicators.

- In shallow *M. modiolus* beds, Phase II *in situ* diver survey should be used in a spatially restricted (belt transect) way to record community composition. The deployment of this method will need to be carefully quality controlled. The monitoring metrics used will need to be based on H' and J.
- Clump sampling nested within Phase II *in situ* diver survey (above) could be used to provide greater community resolution and more sensitive biodiversity measures but, by virtue of expense and limited spatial coverage, is more likely to be relevant to diffuse pressures such as enhanced sedimentation or contamination.
- In deeper water, remote method towed still cameras are likely to be the best monitoring option using metrics based on H' and J.

2.6 Validation of *Modiolus modiolus* reef indicators

2.6.1 Sensitivity of Modiolus modiolus to anthropogenic pressures

The term 'validation' in the context of the present work is the process by which the indicator is tested to demonstrate that it actually works i.e. it responds to a pressure of interest and it is possible to measure the change. Once an indicator is described and validated it can then become 'operational' when appropriate monitoring, quality standards and a process for disseminating the results has been put in place (Moffat *et al* 2011).

i. Pollution and other chemical changes

As a bio-accumulator, *M. modiolus* stores aromatic hydrocarbon compounds and very high levels of heavy metals in the shell (Richardson *et al* 2001) and soft tissues; well above EU legislative limits (Julshamn *et al* 2008). The impact of these components on the reproductive ability or the survival fitness of the species is unknown. However, pollution from organic carbon, PAHs and PCBs can reduce the richness and diversity of some *Modiolus* biotopes (Stewart & White 2001).

Statutory monitoring programmes started in 1978 on the licensed discharge area off Calback Ness Oil Terminal, in Sullom Voe. Bi-annual surveys were conducted until 1985 when surveys were then carried out only once a year. The surveys, carried out by ERT Ltd, aimed at collecting sediment samples for chemical analysis and measuring total hydrocarbon in M. modiolus specimens (Davies & Matheson 1995). Shetland Oil Terminal Environmental Advisory Group (SOTEAG) surveys continued to focus on *M. modiolus* (unpublished confidential reports). Shellfish tissues were tested for hydrocarbon presence prior to the operation of the oil terminal (1976-1979) and thereafter during the operational phase, starting in October 1979. The presence of alkanes in the pre-operational phase sample results were the result of contamination during collection (Davies & Matheson 1995). During the operational phase, patterns of increased tissue accumulation of petroleum hydrocarbons were consistent with increased outputs from the diffuser and decreased from 1986 onwards. Cadmium and lead levels increased from background during the construction phase then stabilised to pre-operational ambient levels. Zinc and Vanadium on the other hand consistently increased in concentration in *M. modiolus* tissues. Davies and Matheson (1995) concluded that short term accumulation of waterborne heavy metals and hydrocarbons had occurred.

Although the physiological effect of these accumulations on the mussels themselves were unknown, the communities associated with *M. modiolus* in Calback Ness seemed unaffected by the discharge, with consistently high values for diversity and species richness throughout (Pearson and Eleftheriu 1981; May 1995; ERT unpublished data). Historical benthic monitoring survey data at Sullom Voe was accessed as part of the present study and it showed the number of species ranging from 31-50 in clumps and 71 from MNCR Phase II surveys (Mair *et al* 2010). While the dive surveys suggest the community was similarly diverse compared to other *M. modiolus* beds in the UK (Table 2.7), the number of species obtained from the clumps is unusually low. Whether these differences are related to the effluent discharge is not known but May and Pearson (1995) found lower biotic diversity in sediments with high hydrocarbon concentration close to the terminal in Sullom Voe.

ii. Removal of target species

M. modiolus is not regarded as a species of commercial interest in Britain or Ireland but recreational gathering of horse mussels does occur, particularly in Scotland. In the Faeroe Islands and Norway, *M. modiolus* is used for human consumption (Strand & Volstad 1997). In Norway demand for horse mussels is increasing with annual landings ranging from 0.46 to 1.48 tons between 2000 and 2006 (Julshamn *et al* 2008). *M. modiolus* fisheries are likely to be unsustainable due to the life strategies of the species i.e. they are very slow-growing and long-lived with irregular reproductive cycles and sporadic or absent recruitment of spat (Comely 1978; Elsäßer *et al* 2013). Wiborg (1946) reported that once exploited by gatherers, most *Modiolus* beds in Norway declined and some never recovered. Harvesting of *M. modiolus* populations could lead to Allee effects (Courchamp 2008; Elsäßer *et al* 2013; Roberts *et al* 2011) resulting in local extinction and, therefore, should not be encouraged without proper consideration of its probable negative impacts (Hussey 2007; OSPAR 2009).

The effect of predator removal in food web interactions is well known (Leibold 1996; O'Gorman *et al* 2010; Paine 1966). As fishing in and around horse mussel beds usually targets predators (e.g. *Buccinum undatum*, *Necora puber*, *Cancer pagurus*) there could be ongoing effects which might be affecting the functioning of trophic chains associated with *M. modiolus* beds.

iii. Smothering

Although *M. modiolus* can be partially endobenthic, in some cases just barely protruding above the seafloor (Comely 1978; OSPAR 2009), smothering as a result of excessive sedimentation could result in physiological stress, as found with other benthic organisms (Fabricius 2005; Hall-Spencer 2000; Trigg & Moore 2009). High siltation (mean Mass Accumulation Rate= 0.63±0.09 g cm⁻² year⁻¹) following bottom trawling in Strangford Lough was suggested as a driver for potential negative effects on the physiological condition of remnant populations by Strong & Service (2008) and Magorrian et al (1995). Fariñas-Franco et al (under review) found that increased sediment deposition as a result of trawling could be linked to observed phenotypical adaptations in shell shape (Figure 2.48). Mesocosm experiments using Vortex Re-suspension Tanks (pVoRTs) (Last et al 2011) found that *Mytilus edulis* tolerates short term and repeated burial although mortality increased with finer sediments. It is unknown whether M. modiolus responds to excessive siltation in a similar way to those of *M. edulis* but changes in respiration and feeding efficiency are likely. Emerging research suggests *M. modiolus* are far more sensitive to sediment burial than had previously been thought (Zoe Hutchison / Kim Last pers. comm. 2013).



Figure 2.48 Infaunal *M. modiolus* in soft substratum. Strangford Lough, Northern Ireland (Photo: Jose M. Fariñas-Franco).

iv. Physical damage: selective extraction and abrasion

Although in Iceland large *M. modiolus* seem to be the dominant species in some areas subjected to high fishing pressure (Guijarro García *et al* 2006), the stability of a highly diverse and self-regulated community created by a slow growing organism such as *M. modiolus* is likely to be compromised by physical disturbance (i.e. bottom trawling or direct gathering). As most populations have poor and irregular recruitment rates, while others fail to recruit juveniles altogether (Elsäßer *et al* 2013; Mair *et al* 2000), recovery from physical damage is likely to be slow or nonexistent. Repeated trawling can also reduce the availability of suitable settlement and crevice habitat causing a decline in biodiversity (Norse & Watling 1999).

Some of the most detailed accounts of the decline of the *M. modiolus* habitat are from the United Kingdom where scallop dredging and trawling have resulted in a reduction in quality and extent of historical *M. modiolus* dominated habitat (Bradshaw *et al* 2002; Service & Magorrian 1997). The effects of mobile fishing gear on the *M. modiolus* faunal assemblages have also been reported from Canada (Hussey 2007; Kenchington *et al* 2006) where there is a growing concern that these types of unsustainable fishing practices may be causing irreversible damage. Strangford Lough is to date the best case study of the effects of bottom trawling on *M. modiolus* communities.

The *M. modiolus* communities found in Strangford Lough were described as 'luxuriant' by Erwin (1977) who regarded them as the climax subtidal communities in the Lough. The important effect that the presence of *M. modiolus* has on the biodiversity of the mostly mud dominated areas where it can be found, was also acknowledged by Roberts (1975) who identified 25 epifaunal, 18 crevice dwelling fauna and 14 infaunal species from just five specimens collected off Brown Rocks. Considering he also estimated these particular beds occupied an area of up to 1km², with densities of 170 mussels m⁻² the ecological importance
of the beds as hotspots of benthic biodiversity was clear. The sublittoral surveys carried out by the Ulster Museum in the 1980s (Erwin 1990) revealed *M. modiolus* beds were found extensively throughout Strangford Lough. However, Service & Magorrian (1997) and Magorrian & Service (1998) concluded that trawling had significantly affected the *Modiolus* communities, destroying the epifauna and disrupting and flattening the *M. modiolus* clumps confirming previous observations (Brown 1989). The reefs lost their three-dimensional structure, increasing accessibility to predators and reducing recruitment of juveniles to the population.

More than seven years later, Roberts *et al* (2011) recorded significant declines in the density and frequency of *M. modiolus* in impacted areas in the Lough's North Basin along with decreases in the community indices. On the other hand, communities in the South Basin, not targeted by the scallop trawlers, registered increases in the number of species while there were no clear trends for diversity and evenness indexes. Strain *et al* (2012) studied the bottom trawled areas and found the *M. modiolus* epibiotic community had been reduced as had the *M. modiolus* itself and characterising species such as *C. varia* and *A. opercularis*. Predators and scavengers increased in abundance, with the exception of species targeted by an existing pot fishery. Roberts *et al* (2011) concluded that long-term impacts on the structure of the benthic communities in Strangford Lough were consistent with intense past physical disturbance caused by bottom trawling and dredging.

2.6.2 Impact case study

i. Mobile fishing gear disturbance in Isle of Man and Pen Llŷn *M. modiolus* beds

The pressures most often linked to declines in *M. modiolus* beds are those associated with demersal fisheries (Service & Magorrian 1998; Roberts *et al* 2011; Strain *et al* 2012). As part of the present study a case study on the physical impact of fishing gear was developed and published (Cook *et al* 2013). Habitat rarity can prohibit elegant experimental approaches to support sensitive management (Stringell *et al* 2013), but providing the impact evidence - base may also be unethical or illegal if it is necessary to willingly damage a habitat or species in a protected area. In the present study, benthic marks attributed to the single passage of two types of bottom fishing gear were identified during routine monitoring operations on *M. modiolus* reefs. This provided a unique opportunity to investigate, directly, the scale of the epifaunal and infaunal impact.

ii. Methods

Benthic marks attributed to the single passage of two types of bottom fishing gear were identified during routine monitoring operations on *M. modiolus* reefs at Point of Ayre (Isle of Man) and Pen Llŷn a'r Sarnau SAC (Figure 2.49A). Side scan sonar imagery (Figure 2.49C) was used to identify an impact study site in June 2012 on another *M. modiolus* reef 5km north of the Lleyn Peninsula. Scallop dredging vessels had been recorded in the area during the preceding season (November 2011 – April 2012) and the marks had not been recorded in all previous annual side scan sonar surveys. The Point of Ayre (PoA) and north Lleyn Peninsula (nLP) sites both contained raised reef structures (1m+) and high densities of *M. modiolus*.

Divers systematically filmed the 25 cells that made up $0.25m^2$ quadrats at close range (<0.5m) using high-definition handheld colour video-cameras (quadrats were then removed from the site). At the PoA quadrat records were made between August and September in 2007, 2008 and 2009 at 12 positions relocated using fixed plastic pins on top of ridges of *M. modiolus*. In 2008 notification of the survey and a position was given to local shipping and

fishing organisations in a Notice to Mariners a week before the survey. During the subsequent 2008 survey, six of the original quadrat positions were found to be impacted by a pair of parallel furrows and a 'swept' area between that was tangential to the ridges of the natural bedform (Figures 2.49A & B) and consistent in size and orientation with the passage of an otter trawl. Recording was conducted in a similar way at nLP in July 2012, except that quadrats were randomly placed in areas with conspicuous dredge marks and adjacent undredged areas. Frame grabs of each of the quadrat sub-cells were stitched together to create a high-resolution mosaic of the benthic community under each quadrat, from which conspicuous species were enumerated (Figure 2.50). Fauna were recorded to the highest taxonomic resolution possible.

At PoA four random 0.0625m² infaunal samples were taken in 2009 from each of three *M. modiolus* ridge locations: two outside of the marks recorded in 2008 where there was no evidence of trawl damage (Figure 2.49A: "Control") and one where a ridge was found damaged in 2008 (Figure 2.49A: "Impacted"). Divers sampled to 20cm depth and recovered material into 0.5mm drawstring mesh bags. Samples were preserved in 5% buffered formaldehyde and sieved on a 0.5mm mesh. Infaunal samples were sorted separately and all fauna identified to a coarse level of taxonomic resolution (Class level) sufficient to detect impacts (Clarke & Gorley 2006). Porifera, Hydrozoa, Anthozoa and Bryozoa were not used in subsequent analysis because they were better represented in the video analysis.



Figure 2.49 (A) Map of fixed quadrat locations (dotted squares) on raised ridges (grey polygons) at Point of Ayre study site. Dotted ellipses indicate infaunal sample areas for impacted and control treatments. Two trawl door marks in 2008 are indicated by dashed lines. One trawl door mark in (A) is visible in the video-grab image (B) where the more extreme impact (compared to the net) in the path of the trawl door is also illustrated with dashed lines. The numbers "7" and "3" in (A) are quadrat numbers referred to in Figure 2.50. Metal waypoint pins enabled navigation around the site. (C) Side scan sonar image from 2012 at the study site off the north of the Lleyn Peninsula: marks from two gangs of scallop dredges are visible across the surface of the *M. modiolus* ridges.



Figure 2.50 Mosaic quadrat images. Quadrat 7 (indicated in Figure 2.49) from Point of Ayre in 2007 (A) and 2009 (B). (C) Unimpacted quadrat and (D) impacted quadrat from N. Lleyn Peninsula in 2012. Numbers indicate conspicuous epifauna: 1 *Alcyonium digitatum*, 2 *Modiolus modiolus*, 3 *Echinus esculentus*, 4 *Ophiothrix fragilis*, 5 *Antedon bifida*.

iii. Data treatment and statistical analysis

Multivariate analyses were conducted on Bray-Curtis similarity coefficients of square root transformed species abundance data, using PRIMER v6 with PERMANOVA+ (Anderson *et al* 2008). Non-metric multidimensional scaling (MDS) was applied to Bray Curtis similarities using the Kruskal fit scheme and, in the case of epifaunal data from PoA, a dummy variable was used to stabilise dispersion of sparse data (Clarke & Gorley 2006). Variation between impacted and unimpacted quadrats at the two sites were tested as fixed effects in one-way (nLP) and mixed two-way designs with year as random factor (PoA) using PERMANOVA based on 9999 permutations and Type III sums of squares (SS). Type III SS is the most conservative SS method for PERMANOVA, fitting every term simultaneously and ensuring independence of all factors in unbalanced designs (Anderson *et al* 2008). Within-site correlation differences through time in the PoA site were tested using PERMDISP

(permutation of dispersion) (Anderson *et al* 2008). Taxa contributing to dissimilarities between treatments were investigated using a SIMPER.

Number of individuals (N), Shannon-Wiener's diversity (H'), Margalef's richness (d) and Pielou evenness (J) were imported into R and tested for normality and heteroscedasticity. Effects of physical impact on diversity and evenness indices from quadrat records at both sites were tested (α of 0.05) by fitting linear mixed effects models (LMMs: Ime4 package) with individual quadrats (both sites) and sampling year (PoA site) as random factors to account for spatial and temporal pseudoreplication. Impact (impacted vs non-impacted) was the categorical predictor (fixed factor) in the mixed model. Generalized LMMs with Poisson error distribution and logit link function were fitted to the abundance data (N; *M. modiolus* and epifauna) incorporating the same fixed and random factors as the LMMs to cope with non-normal data in unbalanced, mixed-effect experiments (Bolker et al 2009; Venables & Dichmont 2004). Overdispersed Poisson models were refitted using Penalized Quasi Likelihood approximations (glmmPQL: MASS package (Venables & Ripley 2002)). The Akaike Information Criterion (AIC) was used to assess the effect of the physical impact on the null model for PoA and nLP while controlling for the random effects. Model selection was based on the lowest AIC score. Infaunal count data from PoA cores conformed to the parametric assumptions and were therefore tested against impact treatments using standard one-way ANOVAs. All models were tested using residual plots to confirm that the assumptions of normality and sphericity of the residuals were met.

iv. Results

In total, 29 different taxa were recorded in video guadrats at the two study sites. According to PERMANOVA models at both sites there were significant impact effects on community composition (pseudo F=24.37, p=0.0001; pseudo F=2.86, p=0.03 for PoA and nLP respectively). There was also significant variability among years in the structure of the community at PoA (pseudo F=2.52, p=0.005). PERMDISP analysis indicated significant larger dispersion across time in epifaunal community samples following impact (deviations from centroid: $F_{(1,36)} = 2.07$; p<0.01). However individual pairwise tests showed significant difference in dispersion occurred only after the trawling event in 2008 (2007 and 2008: t=4.99; 2007 and 2009: t=5.57; p<0.001) with no significant within-site differences between 2008 and 2009 (t=0.56; p=0.69). The average dissimilarity between impact treatments at PoA site was high (85%) in the SIMPER analysis and driven by reductions in all but one (Paguridae) of the taxa in the impacted guadrat records. More than 90% of the average differences between unimpacted and impacted quadrats were accounted for by reductions in Alcvonium digitatum (L.), Actinaria, Antedon bifida, Hydrozoa and M. modiolus (SIMPER), At nLP the impact was less pronounced with 31.3% average dissimilarity between impacted and unimpacted treatments and reductions in the abundance of Modiolus modiolus, A. digitatum, Ophiothrix fragilis, Ascidiella sp, Flustra foliacea, Pyura sp. and Anomiidae accounting for 57% of the dissimilarity between treatments (SIMPER). Some encrusting and low-lying taxa at nLP were more abundant in records from impacted guadrats because upright emergent epifauna had reduced and revealed them (e.g. increased Crisia eburnea) contributed 5.5% to dissimilarity). Overall, for both *M. modiolus* reefs there was compelling evidence of physical impact on the epifaunal communities (Figures 2.51 & 2.52) and the significant differences in dispersion between 2007 and 2009 at PoA indicated no recovery.



Figure 2.51 MDS plot showing the relationship between impacted and unimpacted epifaunal communities. (A) Point of Ayre. Dummy variable (present everywhere) used to create coherence in low abundance [impacted] data (see (36). (B) North of the Lleyn Peninsula.

In video guadrat data from PoA, significant reductions in number of individuals (N), numbers of upright emergent epifauna and total numbers of visible *M. modiolus* occurred at the impacted areas (Figure 2.52). Species richness (Margalef's d) and Shannon-Wiener's diversity (H') and community evenness (J) were significantly lower in impacted quadrats (Figure 2.52). Overall, mean number of total individuals (N) was significantly reduced by 90.3% in trawled guadrats (2.63 \pm 1.96) compared to untrawled guadrats (27 \pm 12.23) (GLMM: t=-11.41; p< 0.001). Most of the variation in N in impacted and unimpacted quadrats occurred between quadrat locations (σ^2 site=0.39), varying little between years (σ^2 year=0.09). At nLP there was a 59% lower mean abundance of total individuals (N) in video records from the scallop dredged areas $(97.3 \pm 21.7 \text{ compared to } 164 \pm 32.0; \text{ LMM})$ = 3.42; d.f. = 6; p < 0.05). Lower abundances of *M. modiolus* and total upright emergent epifauna (mostly A. digitatum and F. foliacea) in dredged areas were significant only for the latter (LMM; *M. modiolus* t=1.75; p=0.13; upright emergent epifauna t= 3.06; p < 0.05; Figure 5). Shannon-Wiener's diversity (H'), Margalef's d richness and eveness (J) of the associated community were not significantly altered by impact (H': t=-1.74, p=0.13; d: t=-1.55, p=0.17; J: t=-1.14, p=0.29).

Using low taxonomic resolution 19 broad groups were recorded from infaunal samples at PoA. The trawled infaunal community in 2009 varied significantly from the two control sites (PERMANOVA: pseudo F=9.02, p=0.002) a year after the impact was first observed. In the SIMPER analysis, reductions in the abundances of bivalves, malacostracans, ophuroids and polychaetes accounted for 60% of the average differences between impacted and unimpacted samples. Each of these reductions in abundance was significant (Figure 2.52D; ANOVA:Polychaeta, $F_{(2,9)}$ = 9.69, p<0.01; Bivalvia, $F_{(2,9)}$ =24.75, p<0.001; Malacostraca, $F_{(2,9)}$ = 6.52, p<0.05; Ophiuroidea, $F_{(2,9)}$ = 11.44; p<0.01).



Figure 2.52 Reductions in epifauna and infauna following bottom-towed fishing gear. Total number of individuals (A), upright emergent epifauna (B) and numbers of *M. modiolus* (C) recorded on impacted and unimpacted 0.25 x 0.25m video quadrats off Point of Ayre (PoA) and North Lleyn Peninsula (nLP). (D) Abundance of infaunal taxa contributing the most to the dissimilarities between impacted and unimpacted treatments at the PoA site (SIMPER). Box plots represent interquartile range, median, maximum and minimum values. The effect of physical impact was significant at α of 0.05 for all measures except *M. modiolus* abundance at nLP.

v. Discussion

The present study investigated the effects of single passes of bottom-towed fishing gear on rare protected *M. modiolus* reef communities. The null model was rejected because there were substantial declines in the abundance of epifauna in response to both trawl and scallop dredges as well as declines in all major taxonomic groups in the infaunal community at the trawled site. The present study provides the most direct evidence yet of physical impacts on the community associated with this type of complex habitat. Abrasion of epifauna is undoubtedly one mechanism responsible for the changes observed but loss of structure formed by *M. modiolus*, and the role that the species plays in pelagic-benthic coupling, also probably account for reductions in most taxonomic groups (especially at PoA). The post impact increase in Paguridae at PoA is consistent with increased scavenging in other fishing gear impact studies (Kaiser 1998; Kaiser et al 2000). The results are consistent with indirect studies elsewhere where biogenic reefs formed by mussels and associated epifaunal declines have been documented in dredging and trawling grounds in Strangford Lough (Strain et al 2012), New Zealand (Cranfield et al 2003) or Canada (Kenchington et al 2007) and where *M. modiolus* as a species (not forming biogenic structures) has been shown to decline in experimentally trawled areas (Kenchington et al 2006). Similarly, other biogenic reefs formed by oysters Ostrea chilensis and horse mussels Modiolus areolatus in the Faveux Strait (New Zealand) have shown widespread reductions in the associated community and reef habitat following prolonged dredging (Cranfield et al 2004).

The magnitude of changes in the present study are similar to the differences in fauna between ridge and trough structures in naturally occurring beds (62%: Rees *et al* 2008; Sanderson *et al* 2008). In essence, the physical impact from bottom-towed gear removed ridge structure and appeared to reduce the community to a 'trough' habitat (*sensu* Sanderson *et al* 2008) at PoA and, although declines in *M. modiolus* were not significant at nLP, clump structures were visibly flattened as well as showing significant epifaunal declines. The scale of change in epifaunal abundance, in particular and *M. modiolus* density in PoA, suggest that density and community indicators are responsive to anthropogenic disturbance. Large changes in the variance (see PERMDISP results) associated with epifaunal abundances might be expected across a reef experiencing low levels of physical impact that only cover part of it, whereas a substantial significant decline in mean epifaunal abundance (60% or more) would be expected across a reef experiencing fishing throughout; both scenarios indicate a severe impact on the communities that is unlikely to be 'sustainable' or commensurate with GES.

2.6.3 Key findings and research needs

The work of Roberts *et al* (2011) in Strangford Lough demonstrates that the scale of variation for *M. modiolus* density and community indices can be measured and successfully predicted (Roberts *et al* 2011). The impact monitoring data described in Section 2.5.2 further supports the validation of the proposed indicators in terms of the response of the community metrics to a single physical abrasion event (see Cook *et al* 2013). Case studies from Strangford Lough where 100% reductions in density and percentage cover of *M. modiolus* occurred have provided the benchmark against which we can predict the effects of physical abrasion on *M. modiolus* beds described in Table 2.17. The clump data analyses described in Section 3.4.2b indicate trawled *M. modiolus* communities were substantially less diverse, rich and even than un-trawled ones across the UK range. Section 3.5.2 also provides evidence that physical damage reduced *M. modiolus* density and epifaunal abundance.

Quantitative analyses carried out for this study prove there is a strong, significantly negative response of density indicator metrics to physical impacts. Community indices, particularly Shannon-Wiener's diversity and Pielou's evenness are well suited as community indicators using *in situ* and towed still cameras because they are less variable across surveyed sites and respond to anthropogenic pressures such as physical impact and possibly pollution. The density and community indicators can therefore be considered validated for the purpose of establishing GES for Descriptor 1 under Criteria 1.6.1 of the MSFD for *Modiolus modiolus* reef habitats.

It is not known if the indicators can respond in the same way to other impacts or pressures. It is likely that smothering as a result of increased siltation have a physiological effect on *M. modiolus*, increasing stress levels and reducing feeding rates. Dynamics where benthic disturbance results in coupled deleterious effects (e.g. physiological stress as a result of reduced feeding rates) have been suggested for fragmented *M. modiolus* beds in Strangford Lough, for example (Roberts *et al* 2011). At the time of writing, contaminants are not considered a significant threat to *M. modiolus* reefs; however, evidence suggests that such effects would be manifested in declines in the richness and diversity of the associated community (May & Pearson 1995). The use of biotic indices based on soft bottom communities such as the AZTI Marine Biotic Index (AMBI) and the Infaunal Quality Index (IQI) could be trialled to help calibrate conditions for existing, impacted and unimpacted, *M. modiolus* habitats. These reference conditions could be used as basis for collation and comparison of newly acquired community data to detect the influence of a variety of anthropogenic pressures, not only abrasion.

Box 2.6 Key recommendations

- Descriptor 1 indicators for reef density and community diversity associated with *M. modiolus* beds (Recommendations 1 & 2) will respond to major physical abrasion pressures associated with demersal fishing and are therefore recommended for consideration in an MFSD monitoring strategy. Quality standards and procedures should be scoped in order to make the indicator fully operational.
- Diversity indices (e.g. Shannon-Weiner/Pielou) are recommended as appropriately validated metrics of *M. modiolus* community condition because they will respond to physical abrasion. WFD multimetric indicators also use a component of diversity therefore, a trial investigating the performance of IQI indicators (and perhaps others such as AMBI) is recommended for possible alignment between Directives.
- The effects of re-suspended sediments (from demersal fishing plumes *etc*) on *M. modiolus* reefs and *M. modiolus* communities should be investigated as an important pressure that is not yet accounted for in monitoring and management.

Table 2.17 Summary of the known effects and magnitude of change in mussel density and in the biodiversity of communities associated with *Modiolus modiolus* reefs in response to environmental change based on a review of the literature and field observations.

	M. modiolu	<i>is</i> Density	Associated Community		
Impact / Pressure	Anticipated Change	Magnitude of Change	Anticipated Change	Magnitude of Change	
Changes in suspended solids (water clarity): increased turbidity	Physiological stress, low recruitment, smothering leading to increases in mortality.	Unknown	Impacts on the fauna associated with <i>M.</i> <i>modiolus</i> reefs will be species specific with some species having similar tolerance levels to <i>M. modiolus</i> and others less tolerant. A reduction in diversity is therefore likely.	Unknown	
Changes in suspended solids (water clarity): decreased turbidity	Reduced feeding rate. Energies diverted to somatic growth. No reproduction growth.	Unknown	Impacts on <i>M. modiolus</i> may be reflected in the associated fauna. Could cause changes in the balance of deposit and filter feeders (<i>A. bifida</i> , ophiuroids) associated with the reef.	Unknown	
Siltation rate changes including smothering	It could affect physiological condition and reproduction. Little is known about escape behaviour in <i>M. modiolus</i>	Unknown	Likely to cause complete or near complete die-off.	Unknown	
Physical Disturbance damage: selective extraction and abrasion	Causes reductions in density and extent. Recovery unlikely but depends on larval sources and connectivity. Areas of reef will become more patchy with increasing physical impact.	80-100% decrease as a result of direct destruction and ensuing cascading effects Reduction from 100% cover to 0- 20% cover (Fragmentation) Loss of elevation (100%)	Reduces complexity and diversity. Increases dominance by opportunistic species. Increased number of soft- bottom species.	50-90% declines in number of species. Declines in Shannon Weiner diversity to ~1.5 and evenness ~0.5. Increase dominance of opportunistic species.	
Pollution and other chemical changes	<i>M. modiolus</i> is a bioacumulator. Pollution could affect physiological condition and reproduction.	Unknown	A decrease in diversity is anticipated as found in case studies in Sullom Voe and Nova Scotia.	Unknown	

3. The development of Descriptor 1 (Biological Diversity) indicators for *Mytilus edulis* reefs

3.1 Introduction

Mytilus edulis is a mytilid mussel widely distributed as a result of its high tolerance to different environmental gradients including temperature, desiccation, salinity and wave exposure and resilience to anthropogenic pressures such as pollution (Seed & Suchanek 1992; Goslin 1992). Although ubiquitous throughout the world's temperate waters, *M. edulis* is more commonly found in exposed or moderately wave-exposed areas (Seed & Suchanek 1992; Witman & Suchanek 1984). In the UK, *M. edulis* is hybridised in varying degrees with two other mytilids, *M. galloprovincialis* and *M. trossulus*, and authors often refer to a *M. edulis* complex rather than to a single species (Wood *et al* 2003; Beaumont *et al* 2008). This report, however, keeps in line with nomenclature in EUNIS and MNCR biotope classification systems (<u>http://eunis.eea.europa.eu/</u>; Connor *et al* 2004) and will refer to *Mytilus edulis* biotopes which are of relevance to the MSFD.

Mytilus edulis is a gregarious species that forms beds of variable thickness (usually less than 50cm deep, Dare *et al* 2004; Holt *et al* 1998) in the lower intertidal and the shallow subtidal (less than 10m; see Jones *et al* 2000 for a full review of environmental requirements). Mussel beds are usually fragmented, constituting metapopulations with dynamics controlled by a wide range of factors including supply of larvae, environmental conditions (e.g. cold winters, storms, exposure), predation and anthropogenic activities, particularly fishing (Burrows *et al* 2008; Büttger *et al* 2008; Hilgerloh 1997; Kritzer & Sale 2004; Levin & Rasmussen 2011; McGrorty *et al* 1990; Paine 1969; Seed 1969). Site specific mussel growth patterns, disease and emigration to other beds are also important processes that control bed density (Bignell *et al* 2008; Kirk *et al* 2007; McGrorty *et al* 1990; Svane & Ompi 1993).

Mytilus edulis are ecosystem engineers and keystone species (Borthagaray & Carranza 2007; Jones et al 1997; Jones et al 2000; Mills et al 1993; O'Connor et al 2012). Mussels action on the ecosystem is both direct (autogenic), resulting from their physical presence, and indirect (allogenic) as a result of physiological processes (e.g. filtration). Direct and indirect effects include habitat creation, substrate stabilization, de-nitrification, bio-deposition and nutrient sequestration (Commito & Rusignuolo 2000; Meadows et al 1998; Norling & Kautsky 2007; Snover & Commito 1998). The physical presence of the mussels and the bioaccumulation of sediment are two processes crucial in the definition of what constitutes a biogenic reef (see the Manual for Interpretation of EU Habitats (2013 version) or Holt et al 1998). Both processes increase habitat complexity in otherwise species-poor areas of mud or sand, thus enhancing biodiversity by attracting new colonists that can take advantage of newly opened feeding grounds (Dekker & Drent 2013; Norling & Kautsky 2008; Wilding & Nickell 2013). Subtidal and intertidal mussel beds on sand, mud and mixed soft substrata increase the structural complexity of the seafloor and therefore qualify as biogenic reefs (Beck et al 2009; Buschbaum et al 2009; Coen & Grizzle 2007). In common with biogenic structures created by other mussel species (e.g. Modiolus modiolus, see Chapter 2 of this report), *M. edulis* beds can have a multilayer arrangement of living mussels and dead shells, accumulated sediments (silt, faeces and pseudofaeces) and relatively rich floral and faunal communities (Seed & Suchanek 1992; Suchanek 1972).

In comparison to *M. modiolus* beds, *M. edulis* beds are neither particularly diverse nor host rare or endemic species (Dekker & Drent 2013; Holt *et al* 1998; Ragnarsson & Burgos 2012; Saier 2002). The communities are largely dominated by littorinid gastropods, barnacles

amphipods as well as fucoids and polychaetes *Arenicola marina* and *Lanice conchilega* (Connor *et al* 2004; Jones *et al* 2000). However, *M. edulis* beds can be highly productive systems compared to the surrounding non-mussel habitats (Dittmann 1990; Lintas & Seed 1994). Even if the effect on the associated fauna and flora macroinvertebrate communities was not substantial, *M. edulis* reefs are also of high conservation value as they maintain large populations of breeding and wintering waterfowl particularly waders and diving ducks (Hilgerloh 1997; Jessop *et al* 2010; Koivisto & Westerbom 2010; Laursen *et al* 2010; Saier 2002; Stillman *et al* 2010).

3.1.1 Legislation relevant to Mytilus edulis beds/reefs

Mytilus edulis differs fundamentally from the other two biogenic reef forming species dealt with in this report insofar as it is a relatively important UK fishery (Marine Management Organization 2013). Intertidal beds are harvested to supply markets in the UK and continental Europe (Dare *et al* 2004; Jessop & Maxwell 2011) while subtidal beds are dredged to supply seed for the mussel aquaculture industry (Dolmer *et al* 2001; Maguire *et al* 2007).

Different acts and by-laws regulate the exploitation of *M. edulis* across the UK while the European Shellfish Growing Waters Directive (2006/113/EC) ensures water quality in shellfish growing areas, including those where *M. edulis* beds are found, is maintained. Those legislative instruments are also relevant to *M. edulis* beds qualifying as biogenic reef features. From a conservation perspective it should be noted that legislation applies to the habitat created by *M. edulis* and not the species itself. Mussel beds can be features of conservation importance either directly (as biogenic reefs, for example) or indirectly if they are subfeatures within other protected habitats (e.g. estuaries).

Protection of *M. edulis* beds constituting biogenic reefs is theoretically duplicated by fisheries and conservation legislation both at European and UK levels (Table 3.1). Where statutory duties and ministerial commitments exist towards the active management of *Mytilus edulis* habitats, either explicitly or implicitly, some form of monitoring and / or assessment is required. This is particularly relevant to mussel beds forming features within Natura 2000 sites as almost all of them are exploited (www.ukmarinesac.org.uk). The approach for beds within SACs/SPAs is to ensure mussel stocks remain above a threshold that secures the long-term sustainability of the resource and that guarantees wading bird populations are not impacted (Moore 2009; Stillman *et al* 2010).

Table 3.1 Table summarising the legislative instruments used to protect subtidal and intertidal *Mytilus edulis* bed habitats in the UK. Fisheries and water quality legislations are also included.

Legislative Instrument	Mechanism for Protection
European Habitats Directive 1992	Special Areas of Conservation (SACs)
European Birds Directive 1979-updated and replaced in 2009	Special Protection Areas (SPAs)
OSPAR Convention 1992	OSPAR Marine Protected Areas (MPAs)
Nature Conservation (Scotland) Act 2004	Scotland's Biodiversity Strategy
Natural Environment and Rural Communities Act 2006	England's Biodiversity Strategy Environment Strategy for Wales
Marine Strategy Framework Directive 2008 Marine and Coastal Act 2009	"Good Environmental Status" targets Marine Conservation Zones (MCZ)
Marine (Scotland) Act 2010	Nature Conservation Marine Protected Areas (MPAs)
Environment (Northern Ireland) Order 2002	<i>Mytilus edulis</i> beds protected as natural features of Areas of Special Scientific Interest (ASSI)
Ramsar Convention on Wetlands of International Importance Especially as Waterfowl Habitat 1971 through the Wildlife & Countryside Act 1981, Nature Conservation (Scotland) Act 2004 and Nature Conservation and Amenity Lands (Northern Ireland) Order 1985	Sites of Scientific Interest (SSIs)/Areas of Special Scientific Interest(ASSIs) in Northern Ireland
The Seafisheries (Shellfish) Act 1967	By-laws can be enforced to manage the stocks including fisheries closures if they reach minimum thresholds (i.e. below conservation objectives)
Shellfish Growing Waters Directive 1979-updated and replaced in 2006	Shellfish water designation

i. European Habitats Directive

Although not specifically listed as Priority Habitats, subtidal *M. edulis* beds qualify as Habitats of Community Interest (HCI) under several habitat types listed in Annex I of the European Habitats Directive (92/43/EEC), namely 1130 (Estuaries); 1140 (Mudflats and sandflats not covered by seawater at low tide); 1110 (Sandflats which are slightly covered by sea water at all time); 1160 (Large Shallow Inlets and Bays); and 1170 (Reefs). According to the Interpretation Manual of EU Habitats (European Commission 2013), corresponding categories relevant to biogenic concretions formed by *Mytilus* spp. listed under 'Habitat type 1170' include the German Classification habitats "Miesmuschelbank des Eulitorals der Nordsee (050107)", "Miesmuschelbank des Sublitorals der Nordsee (030207)", French ZNIEFF-MER 'Moulière médiolittorale à *Mytilus* sp.' and the Tri-lateral Wadden Sea Classification "Sublittoral (old) blue mussel beds (03.02.07)".

In the UK the presence of *M. edulis* beds is explicitly listed as one of the qualifying features chosen for the designation of three Special Areas of Conservation (SACs):

- 1. Studland to Portland (UK0030382): *Mytilus edulis* beds are found to occur in very high densities on bedrock associated with strong currents to the southeast of Portland Bill.
- 2. Pen Llŷn a'r Sarnau (UK0013117): presence of reefs of *Mytilus edulis* on cobble in different locations on the north coast of Pen Llyn.
- 3. Berwickshire & North Northumberland Coast (UK0017072): large intertidal beds of *Mytilus edulis* within habitat type 1140.

Special Areas of Conservation (SACs) cannot be designated on the basis of intertidal reef areas. The reefs have to connect to a contiguous subtidal reef (Holt *et al* 1998). *Mytilus edulis* beds that qualify as reefs are found in numerous other SACs but they are not the primary reason for their designation: For example, The Wash and North Norfolk; Burry Inlet and Three Rivers (Carmarthen Bay and Estuaries); Morecambe Bay; Dornoch Firth; Tay and Eden Estuaries; Plymouth Sound and Estuaries; and Solway Firth. Subtidal *M. edulis* beds are also found present in the Inner Dowsing, Race Bank and North Ridge and Haisborough, Hammond and Winterton SACs (Natural England SAC Selection Assessment documents 2010).

ii. European Birds Directive (2009/147/EC), 79/409/EEC

Special Protection Areas (SPAs) are designated to provide protection for birds by establishing a network of protected areas for birds including those that feed on intertidal shellfish or shellfish beds such as those formed by mussels. Mussel beds are important supporting habitats for birds in numerous SPAs. As such, and under Article 3 of the Directive, these biotopes should be preserved, maintained and re-established.

iii. Country Biodiversity Strategies

In England and Wales *Mytilus edulis* beds ('blue mussel beds') are listed as 'Habitats of Principal Importance' (HPI) under Section 41 of the Natural Environment and Rural Communities (NERC) Act 2006 (http://www.naturalengland.org.uk). Unlike the OSPAR list habitat, 'blue mussel beds' under NERC include both subtidal and intertidal *M. edulis* beds. In Northern Ireland there is no Habitat Action Plan (HAP) specific for *M. edulis* beds (DOENI 2005). The Scottish Biodiversity List, published to satisfy <u>Section 2(4) of The Nature</u> <u>Conservation (Scotland) Act 2004</u>, includes blue mussel beds under the marine section of the list with the following comments: 1) conservation action is needed; 2) avoid negative impacts; 3) legally protected under Annex I of the EU Habitats Directive; 4) listed on the UK BAP; and 5) important habitat for supporting marine plants and animals. Although *M. edulis* beds are listed as threatened under OSPAR, the Scottish Biodiversity List does not recognise that status. In Northern Ireland there are no provisions for a dedicated HAP for *M. edulis* beds although from 2012 the Environment (Northern Ireland) Order 2002 affords them direct protection as features within Areas of Special Scientific Interest (ASSI). The Order makes an offence to intentionally or recklessly damage the natural features of ASSIs.

iv. Ramsar Convention (Transposed into legislation by the Wildlife and Countryside Act 1981)

Ramsar sites are designated under the Convention on Wetlands of International Importance (Ramsar 1971). Government policy statements grant wetlands listed as RAMSAR sites the same level of protection as that afforded to Sites of Special Scientific Interest (SSSIs / ASSIs) under the Wildlife and Countryside Act 1981 and SACs and SPAs (Natura 2000) sites under the EU Birds and Habitat Directives. Examples of Ramsar sites with mussel beds include Morecambe Bay, Poole Harbour, The Wash, Carlingford Lough, Dundrum Bay, Belfast Upper Solway Flats and Marshes, Severn Estuary; Dornoch Firth and Loch Fleet; and Cromarty Firth, among others.

v. OSPAR Convention

Intertidal *Mytilus edulis* beds, covering at least 30% of mid and lower shores on mixed, sand and mud substrata are listed in the "OSPAR list of threatened and / or declining habitats" (OSPAR 2008). This list is based on the Texel-Faial criteria for identification of species and

habitats in need of protection (OSPAR 2003). The main criteria used for the inclusion of *M. edulis* beds in the OSPAR list were: (1) Sensitivity (particularly to chemicals); (2) Ecological significance, as providers of ecosystem services including food and refuge for other species; (3) Threat due to ongoing exploitation; and (4) Substantial decline and localised rarity. For *M. edulis*, substantial decline has occurred particularly in the Wadden Sea (Germany, the Netherlands and Denmark) following unregulated fishing, mainly spat collection for aquaculture (OSPAR 2008). The OSPAR list is used as one of the criteria to designate MPAs in the U.K. (Cork *et al* 2006).The EUNIS *M. edulis* biotopes included in the OSPAR list are A2.7211 and A2.7212 (see Table 3.2 for equivalences).

vi. Water Framework Directive (2000/60/EC)

Macrozoobenthos is included as a biological quality element for Good Ecological Status (GEcS) of surface water. The biotope 'intertidal blue mussel bed at stable sites' has such an element. In the Netherlands subtidal mussel beds have also been proposed as biological quality element for water (Wolff *et al* 2010).

vii. Marine Strategy Framework Directive (2008/56/EC)

Biogenic reefs formed by *M. edulis* are suitable for the establishment of Good Environmental Status (GES) indicators for Descriptors 1 (Biological diversity) and 6 (Seafloor integrity) under the MSFD (Cochrane *et al* 2010). As *M. edulis* beds are identified under Community (European Habitats Directive) and International (OSPAR) legislation they are considered a 'special' habitat as defined in Table 1 of Annex III of the MSFD.

viii. Marine (Scotland) Act 2010

Blue mussel beds are identified as both a Priority Marine Feature (PMF) and an Marine Protected Area (MPA) search feature, including all four EUNIS biotope classifications for *M. edulis* biotopes (<u>http://www.snh.gov.uk/docs/B1064114.pdf</u>; Moore & James 2011). Provisions are also made within the Marine (Scotland) Act 2010 for the regulation of exploitation of mussel beds. The blue mussel bed MPA search feature and PMF includes the following component biotopes within the broad habitat 'Blue mussel beds':

- *Mytilus edulis* beds on littoral sediments (LS.LBR.LMus.Myt), with examples from Western Scotland and estuaries in the east coast such as the Moray Firth and the Firth of Forth;
- Mytilus edulis and Fabricia sabella in littoral mixed sediments (LS.LSa.St.MytFab). Reported as an unusual example of mussel bed at the strandline with no records outside Scotland. Recorded in Loch Ridden; Loch Bracadale; Dornoch Firth; Moray Firth; the Tay Estuary; and the Houb of Fugla Ness in Shetland. According to Holt *et al* (1998) this biotope is not a biogenic reef;
- *Mytilus edulis* beds on sublittoral sediment (SS.SBR.SMus.MytSS). Examples are given from the Solway Firth; Loch Creran; Loch Ailort; the Firth of Tay and Whiteness Vow in Shetland;
- Mytilus edulis beds on reduced salinity infralittoral rock (IR.LIR.IFaVS.MytRS). Reported as a predominantly Scottish biotope, with records from Shetland, west coast and the Outer Hebrides, Holt *et al* (1998) does not consider it a biogenic reef as the species associated with the mussel beds are able to be survive in the absence of the mussels. Moreover, Holt *et al* (1998) also indicates these are not significantly raised beds. Therefore, because the PMF *M. edulis* beds on reduced salinity rock is not raised and, more importantly, does not host a distinct biotic community it does not qualify as biogenic reef, making it unsuitable for inclusion as a "special' habitat' in the process of developing and monitoring of MSFD indicators for GES.

ix. Marine and Coastal Access Act (2009)

Provisions relevant to *M. edulis* beds are also made in Part 5 of the Marine and Coastal Access Act 2009 for designation and protection through a new type of MPA, called Marine Conservation Zones (MCZs). In England MCZs will exist alongside European Marine Sites (SACs and SPAs), to form a MPA network. *Mytilus edulis beds are* listed in the "Ecological Network Guidance" both as the Broad Scale Habitats 'Intertidal biogenic reefs A.26' and 'Subtidal biogenic reefs A5.6' and the Habitat Feature of Conservation Importance (FOCI) 'Blue mussel beds (including intertidal beds on mixed and sandy sediments)'. The Habitat FOCI 'Blue mussel beds' only covers 'natural' beds on a variety of sediment types and specifically excludes artificial mussel beds and those beds found on rocky substrata.

The Act also aims to make provision in relation to marine functions and activities in England and Wales including bylaws enforced by the Inshore Fisheries and Conservation Authorities (IFCAs) which apply to each regional IFCA. The Act places a duty on the IFCAs to manage the exploitation of sea fisheries including shellfish in a sustainable way and to protect and promote recovery of the marine environment including protecting and furthering the conservation objectives of the MCZs (www.ne-ifca.gov.uk).

x. Shellfish growing Waters Directive 1979

Shellfish waters are designated by the country governments where water quality must be protected or improved to protect the shellfish growth and protect the quality of the shellfish for human consumption. The Directive sets imperative and guideline standards to be achieved for each parameter monitored as well as minimum sampling frequency and the reference methods for the analysis. As such the Directive affects areas where mussels are harvested including those where subtidal and intertidal mussel beds might constitute biogenic reefs. The Directive is transposed into each country through several pieces of legislation (regulations). Each country has a list of designated waters that meet EU standards or should meet them under improvement plans.

xi. Shellfish regulations relevant to *M. edulis* beds

a. Scotland

- Mussels were removed from the public fishery by the Mussel Fisheries (Scotland) Act 1847. Therefore the Crown, who owns the foreshore, grants licenses for mussel exploitation, but few are normally issued. SNH is consulted before licenses are granted;
- In the Dornoch Firth the rights were ceded to the Commons by James I and the Crown acceded to transfer the rights to the Highland Council;
- Regulating Orders are granted by the Scottish Ministers under the terms of the Sea Fisheries (Shellfish) Act 1967 (as amended). A licence from the private owners or Crown Estate may be necessary for harvesting mussels. It also provides protection of shellfish stocks. However, according to McKay & Fowler (1997) those Acts are not known to have been relevant to mussel fisheries in Scotland where the simplest method of control of the mussel fisheries has been through the provisions of the lease from the Crown Estates;
- Amendments to the Shellfisheries Conservation Act 1967 were introduced in the Marine (Scotland) Act 2010. In relation to mussel fisheries it removed Crown Estate consent to several and regulating orders and extended the existing protection of private oyster beds to all privately owned shellfish beds;
- The Aquaculture and Fisheries (Scotland) Act 2013 establishes protection of shellfish

waters. An Act of the Scottish Parliament to make provision about fish farming but also shellfish farming and waters for shellfish. Part 4 of the Act ('Shellfish') relates to protection and improvement of shellfish waters. It amends the Sea Fisheries (Shellfish) Act 1967;

• Inshore Fishing (Scotland) Act 1984: enables the regulation of fishing in inshore waters. It can establish closures and prohibition of fishing methods for certain areas including mussels. The Inshore Fishing (Scotland) order 2004 prohibits dredging for mussels in the Dornoch and Cromarty Firths.

b. England

- The Sea Fisheries (Shellfish) Act 1967 provides for the regulation of the exploitation of shellfisheries including mussels within waters adjacent to England (also Wales and Scotland) through by-laws established by each regional IFCA to help managing their fisheries and protected areas;
- The Sea Fisheries (Shellfish) Act 1967 provides the legal route to establishing Fishery Orders (Regulating or Several Orders) which enable local management of shellfish e.g. The Wash Fishery Order 1992 (WFO 1992) established a mechanism for local management of the cockle and mussel fishery within the Wash estuary. Similarly, the Poole Fishery Order 1985 enables the IFCA to control effort and exploitation in Poole Harbour;
- The IFCAs were set up in April 2011 as a revised approach to fisheries and conservation management replacing the Sea Fisheries Committees with extended responsibilities and new powers under the Marine and Coastal Access Act (2009) including the power to establish closures and determining minimum landing sizes. Additionally some IFCAs such as the north-west IFCA regional branch have codes of conduct for intertidal shell fisheries (www.nw-ifca.gov.uk).

c. Wales

- The Sea Fisheries (Shellfish) Act 1967 applies in Wales but there are specific by-laws and orders that establish further fishing restrictions (i.e. Cockles and Mussels (Specified Area) (Wales) Order 2011);
- Until 2011 by-laws were issued by two Sea Fisheries Committees but these were dissolved following article 3 of the Marine and Coastal Access Act 2009 being brought into force. Responsibilities have been transferred to the Welsh Government Assembly. A new marine and fisheries division has been created and will regulate the fishery in the future (<u>http://wales.gov.uk/</u>). The Welsh Government are carrying out a review of existing By-laws including those for *M. edulis* fisheries.

d. Northern Ireland

- Unregulated harvesting is an issue in Northern Ireland. The National Trust Act 1946 was overruled by Common Law and, at the moment, there are no regulations in place that affect intertidal shellfish gathering (AFBI 2011);
- The Department of Agriculture and Rural Development of Northern Ireland (DARDNI) issues seed mussel dredging licences. Depending on the vessel size VMS and black boxes might be necessary.

3.1.2 Mytilus edulis reef in the context of the MSFD

According to the Interpretation Manual of EU habitats (European Commission 2013), biogenic concretions are reefs if they consist of hard compact substrata on solid or soft sediment arising from the seafloor in either the sublittoral or the littoral zones. The manual also indicates that, when a layer of sediment covers the hard concretions, the community associated with such sediments must be also dependent on the presence of such distinct hard substrata. The working definition of biogenic reefs used by the UK's SAC project report on biogenic reefs (Holt et al 1998) also takes into account elevation of hard substratum above the seafloor as an important defining feature. However, these elevated, solid structures also need to be substantial in size (a somewhat arbitrary extension of 1-2 metre across is given) and the community associated with them needs to be sufficiently distinct from that inhabiting the surrounding substratum (see also Buschbaum et al 2009). The Wadden Sea Tri-lateral Monitoring and Assessment Program (TMAP), also defines a mussel bed in similar terms as both 'a spatially well defined collection of more or less protruding smaller beds' and 'a benthic community structured by mussels' (Nehls et al 2009). Biotopes defined by the presence of *M. edulis* are common throughout the United Kingdom but not all of them meet the criteria to be considered biogenic reefs. The OSPAR case study report for intertidal *M. edulis* beds indicates that the ecosystem engineering effect caused by the mussels is most apparent under high densities, more than 30% cover, when substrate binding and habitat provision for flora and fauna occur. Nonetheless the document does not imply that less dense beds have been excluded from the list of threatened/declining habitats. It can be very difficult to establish the boundaries of a mussel reef due to the fragmented condition of most of them. A working definition used in the Wadden Sea monitoring programmes (Nehls et al 2009) establishes that groups of patches less than 25m apart constitute beds only if they cover more than 5% of the substratum. Nearby groups of patches more than 25m away are considered distinct, separate beds (Figure 3.1).



Figure 3.1 Schematic representation of fragmented beds and definition criteria (extracted from Nehls *et al* 2005).

In the UK, *M. edulis* beds are not particularly elevated if compared to, for example, bioherms of over 1m height formed by *M. modiolus. Mytilus edulis* beds are usually much less than 50cm deep (Holt *et al* 1998). For example, mussel beds in The Wash at the beginning of the 20th century, at the prime of their condition since records began, had maximum depths of

30cm (Dare et al 2004).

The best examples of *M. edulis* aggregations forming biogenic reefs are found when the mussels grow on species-poor mixed substrata of cobble and pebble and on sand or mud. Under these conditions most studies indicate that the effect mussels have as biodiversity facilitators is significantly positive, even at low densities (Buschbaum *et al* 2009; Koivisto & Westerbom 2010; Ragnarsson & Raffaelli 1999). Extensive mussel beds found on rocky substratum, although being functionally similar to other beds on soft or mixed substratum may not be considered as biogenic reefs as the effect of the mussels on the biological community is indistinct from that of the underlying rocky habitat (Holt *et al* 1998) Moreover, mussel beds on rock and coarse gravel substratum might not intrinsically be hot-spots of biodiversity and, on occasions, the communities associated with beds might be less diverse than those communities found in adjacent substrata (Mandy Knott, north-west IFCA, pers. comm.). This effect could be linked to negative synergistic effects whereby mussels in absence of predation can outcompete other fauna when growing on hard substrata (Dürr & Wahl 2004).

Both intertidal and subtidal *M. edulis* biotopes are relevant as 'special' habitats under the MSFD for Descriptor 1 indicator development (Cochrane *et al* 2010; Moffat *et al* 2011). Holt *et al* (1998) consider four *M. edulis* bed biotopes can be truly considered biogenic reefs based on elevation, habitat modification and distinctiveness of the associated assemblage. Table 3.2 lists all *M. edulis* biotopes that are regarded as truly biogenic reefs including those selected as PMFs and MPA search features under the Marine (Scotland) Act 2010 (Baxter *et al* 2011). The biotopes 'LS.LSa.St.MytFab' and 'IR.LIR.IFaVS.MytRS' are also included in this review as they are PMFs although, as previously mentioned, Holt *et al* (1998) do not consider these beds (mostly formed by juveniles) of sufficient scale to qualify as massive, solid biogenic reef structures with distinct biotic communities. Figure 3.2 presents the distribution of those four biotopes as well as the additional two biotopes listed as PMFs in Scotland's Marine Atlas (Baxter *et al* 2011).

Due to the scarcity of targeted survey data for *M. edulis* biogenic beds, in particular data relevant to community indicators, we broadened the literature review to include all methodological approaches and comparative studies relevant to *M. edulis* beds in general. A wealth of information is available from stock assessment surveys of commercial mussel beds in England and Wales and the Wadden Sea (Netherlands, Germany and Denmark). Although some of these beds might not qualify as biogenic reefs, and most are probably impacted, these survey approaches are very relevant to the definition of density indicators for *M. edulis* beds.

There is virtually no information on communities from mussel beds in the UK. Diversity indices have been calculated and compared from datasets from subtidal mussel beds in the Firth of Tay (Bates *et al* 2004). Faunistic studies from protected mussel beds in the Wadden Sea and the Baltic Sea were the only source to help put the biodiversity of mussel beds into context for the development of community indicators in the present study. In the Wadden Sea mussel beds are protected and monitored, not as reefs, but as features within 'Sandbanks' and 'Mudflats and sandflats' Annex I habitat types. Strictly these studies might not refer to reefs but all the studies consulted acknowledged the significant positive effect the mussel aggregations have as habitat modifiers and biodiversity enhancing structures. Studies on populations of relaid mussels were not considered for this report.

Table 3.2 Potential *M. edulis* biogenic reef biotopes listed in the EUNIS classification and their correspondence with National Marine Habitat Classification (NMHC) biotopes (Connor *et al* 2004). Asterisks denote biotopes included as MPA search features and PMFs in Scotland which do not qualify as biogenic reefs according to Holt *et al*'s 1998 review report.

Eunis code	EUNIS Title	Equivalent NMHC code	Brief description
A2.212*	<i>Mytilus edulis</i> and <i>Fabricia sabella</i> in littoral mixed sediment	LS.LSa.St.Myt Fab	Pebbles, gravel, sand and shell debris with mud in sheltered Firths with a strandline of fucoid algae. The fauna is characterised by juvenile mussels <i>M. edulis</i> , often in very high numbers Dense aggregations of <i>M. edulis</i> on the mid and lower
A2.721	<i>Mytilus edulis</i> beds on littoral sediments	LS.LBR.LMus. Myt	shore, on mixed substrata, on sand, or on sheltered muddy shores. In high densities the mussels bind to the substratum and provide a habitat for many infaunal and epifaunal species
A2.7211	[Mytilus edulis beds on littoral mixed substrata	LS.LBR.LMus. Myt.Mx	Mid and lower shore mixed substrata (mainly cobbles and pebbles on fine sediments) in a wide range of exposure conditions and with aggregations of the mussel <i>M. edulis</i> colonizing mainly the sediment between cobbles, though they can extend onto the cobbles themselves. The mussel aggregations can be very dense and support various age classes. Examples in the UK: Berwickshire & North Northumberland Coast; Morecambe Bay This sub-biotope occurs on mid to lower shore sand and
A2.7212	<i>Mytilus edulis</i> beds on littoral sand	LS.LBR.LMus. Myt.Sa	muddy sand. Mussels <i>Mytilus edulis</i> grow attached to shell debris and live cockles <i>Cerastoderma edule</i> , forming patches of mussels on consolidated shell material, and often growing into extensive beds. Examples in the UK: Burry Inlet; Dornoch and Cromarty Firths.
A2.7213	<i>Mytilus edulis</i> beds on littoral mud	LS.LBR.LMus. Myt.Mu	Dense mussel beds found in sheltered conditions on mud. There is a buildup of pseudofaeces that results in a bed that is very soft to walk on, and sediment that is anoxic to the surface. The sediment infauna is very poor due to anoxic conditions. Examples in the UK: Burry Inlet; The biotope accurs in shallow, often tide swept reduced
A3.361*	<i>Mytilus edulis</i> beds on reduced salinity infralittoral rock	IR.LIR.IFaVS. MytRS	salinity conditions. Dense beds of the mussel <i>M. edulis</i> with the occasional barnacle <i>Balanus crenatus</i> . A wide variety of epifaunal colonisers on the mussel valves, including seaweeds, hydroids and bryozoans can be present.
A5.625	<i>Mytilus edulis</i> beds on sublittoral sediment	SS.SBR.SMus .MytSS	Shallow sublittoral mixed sediment, in fully marine coastal habitats or sometimes in variable salinity conditions in the outer regions of estuaries, are characterised by beds of the common mussel <i>M. edulis</i> . Examples in the UK: Solway Firth; Dornoch Firth; probably the Firth of Tay



Figure 3.2 UK distribution of *M. edulis* biotope records that might qualify as biogenic reef habitats. Records were obtained from a July 2013 snapshot of Marine Recorder and overlaid on Google Earth base-layers using the open source QGIS software (version 2.0). Names refer to locations where published quantitative stock data was used or reviewed for this report.

3.2 Definition of *Mytilus edulis* bed indicators

3.2.1 Evidence base

Monitoring of blue mussel (M. edulis) beds in the UK has been driven by the need to guarantee the sustainability of the existing fisheries while ensuring the bird populations that depend on them are not negatively affected (Jessop et al 2012; Stillman et al 2010). However most if not all the data collated from *M. edulis* beds in the United Kingdom has solely focussed on stock management metrics, useful only for the development of Descriptor 1 density indicators. The associated faunal and floral communities have received limited attention and datasets that could be used to define and validate community indicators, in the same way that was done for other biogenic reef forming species, were not available. Video footage and digital photographs collated incidentally from subtidal *M. edulis* reefs were reviewed to determine their suitability as non-destructive methods to capture density and community metrics. The footage originated from surveys not specifically targeted at monitoring blue mussel beds (e.g. Regional Environmental Characterisation (REC) and Natural Resources of Wales (NRW) monitoring surveys) or did not necessarily show beds that could be regarded as biogenic reefs. The footage was reviewed in a gualitative manner because there were insufficient replicated datasets to allow comprehensive, quantitative analyses with an adequate level of statistical confidence.

A review of the existing data held in Marine Recorder (June 2013 snapshot) for *M. edulis* biogenic reef biotopes indicated there is an estimated area of 258km² in the intertidal and subtidal coastal zones occupied by *M. edulis* beds. More than 70 reports and scientific publications were consulted during the preparation of this section: stock assessment reports from agencies and fisheries committees in the UK and abroad (e.g. IFCAs and DEFRA in the UK; IMARES in the Netherlands) were supplemented with existing, relevant peer-reviewed scientific literature. Of particular importance to the development of the community indicators were research papers investigating the communities associated with *M. edulis* in the Wadden and Baltic Seas (i.e. Dekker & Drent 2013; Koivisto *et al* 2011; Norling & Kautsky 2007, 2008, among others).

Methodological approaches for density indicators were obtained from annual stock assessment reports and reviews from England and Wales, chiefly:

- The Wash (eastern IFCA; Dare *et al* 2004; Jessop & Maxwell 2011; Jessop *et al* 2010, 2012);
- Solway Firth (north-west IFCA, Solenvo 2004, 2006);
- The Burry Inlet and Three Rivers, Severn Estuary (Mercer 2002, Moore 2009);
- Fenham Flats (Green & Royle 2011, 2012, Green 2010) and
- Outer Thames area (Wright & Bailey 2009).

Outside the UK, reports from Wadden Sea countries as well as Ireland and Sweden were useful sources of information on monitoring approaches and indicator validation. The 'Waddenzee Blue Mussel Group', part of the Trilateral Monitoring and Assessment Programme (TMAP), has published a chapter in the TMAP Habitat Monitoring Handbook (Common Wadden Sea Secretariat 2008). This handbook is intended to provide common guidelines for monitoring of parameters for habitats in the Wadden Sea, including blue mussel beds. The TMAP Handbook as well as other reports from the Dutch IMARES Institute (Dekker & Drent 2013; Drent & Dekker 2013a, 2013b) were consulted. In Ireland, subtidal mussel beds have been surveyed both by public governmental bodies (e.g. Marine Institute, An Bórd Iascaigh Mhara (BIM)) and private contractors (Brendan O'Connor pers.comm.).

Footage obtained from drop down cameras and towed video systems has been tested in other sections of this report to establish the scale of error associated with extracting density estimations for S. spinulosa and M. modiolus reefs. Because the images and videos were collated as part of surveys specifically targeted at monitoring those other two biogenic reef types, there was sufficient replication to determine the sources of variance and identify spatial and temporal change using statistical analysis. The same approach was not possible for subtidal *M. edulis* beds due to the dearth of images and videos from areas that could be classified as *M. edulis* reefs. The Regional Environmental Charactisation (REC) and Thanet survey spreadsheets and media libraries were scanned for blue mussel beds resulting in just 19 photographs from three sites: Humber (13 images, not a reef), Thanet (two images, raised bed, probably a reef), East Coast REC (four images, insufficient quality to determine if reefs were present). The images obtained during the pre-construction survey at the Lincs windfarm site by EGN Ltd. indicated the presence of dense *M. edulis* juveniles. Although visibility was poor to attempt any quantitative estimation of mussel density, the bed was likely an ephemeral seed bed, as later demonstrated by the eastern IFCA surveys. Video and stills obtained by NRW off the Llyn Peninsula did cover some *M. edulis* beds at one station. That footage was reviewed to determine if the quality was good enough to assess density and community indicators.

Experts on mussel bed monitoring from the UK and abroad were consulted during the preparation of this chapter including: Nicolas Chopin, BIM; Francis X, O'Beirn, Irish Marine Institute; Dr. Brendan O'Connor, marine biologist, Ireland, Ron Jessop, eastern IFCA; Mandy Knott, north-west IFCA; Sarah Clarke, Devon and Severn IFCA and Karin Troost, IMARES. Some of these and other experts were present at a workshop (under the present project) on mussel stock assessment with the aim of discussing best practice for both community and density indicator monitoring.

Stock assessment datasets from historical mussel surveys in the Burry Inlet (Moore 2009; Stillman *et al* 2010) were accessed with the aim of establishing if temporal and spatial trends in density metrics were statistically significant. Bootstrapped power analyses were undertaken to determine the power to detect change obtained with the design undertaken by the surveyors while recommendations on the number of stations needed to detect different variations in density for the system at a standard power were made. Data was also extracted from some IFCA reports (Jessop & Maxwell 2011; Jessop *et al* 2010, 2012, Solenvo 2006) to establish if the trends observed were of statistical significance.

3.2.2 Density indicators

i. Natural range and variability of the proposed density indicator metrics

Historically, most mussel stock monitoring programmes for intertidal *M. edulis* beds in the UK and the Wadden Sea measure three mandatory quantitative parameters to describe their structure (Herlyn 2005; Jessop *et al* 2012):

- 1. Area of the mussel bed in km²; calculated with the aid of portable GPSs and Geographical Information Systems (GIS).
- 2. Surface covered by mussels; percentage cover estimated using different standard methodologies.
- 3. Biomass: wet weight in grams m⁻²; wet weight can be directly calculated by weighing the fresh weight of mussels or indirectly from length-weight regression equations.

These three metrics are subsequently used to obtain extrapolations of total biomass (tonnage) or total abundance of mussels for each individual bed (multiplying either biomass

or abundance m⁻² and % cover by the area of each bed). The total tonnage or numbers of mussels for the area studied are later calculated by adding the values together for each discrete bed. These parameters are then used as guidance for stock management purposes. Additional information can include size-frequency distribution data to help monitor recruitment history or condition indices to determine the reproductive status of the breeding stocks or sustainability for different wading bird species. The latter are, however, not relevant for the purpose of MSFD density indicator development but may be relevant to the OSPAR indicator on shellfish population demographics.

All three metrics (area, percentage cover and biomass) are potential density indicators for *M. edulis* reef condition. Monitoring recovery of impacted mussel beds involves the measurement of biomass and abundance of individuals (particularly adults) that contribute to the reproductive effort in the context of metapopulation dynamics including connectivity (Jennings 2000). Marine Recorder yielded 258km² of *M. edulis* biogenic reef biotopes on sand, mud or mixed substrata. The majority of existing records with density-related metrics for UK mussel beds originate from stock assessment surveys of intertidal mussel beds. With the exception of the ephemeral seed mussel beds, subtidal *M. edulis* aggregations are not subjected to the same level of regular monitoring as intertidal beds. The lack of commercial activity on mature subtidal *M. edulis* due to its relative rarity has resulted in a corresponding lack of survey records. Recent SAC monitoring surveys in Wales (Keenan *et al* 2010) suggested subtidal *M. edulis* densities are relatively stable over time. Nonetheless, there is still a knowledge gap regarding the variability in density and coverage of unexploited subtidal blue mussel beds.

In the present work we reviewed nine stable *M. edulis* bed locations in the UK (Figure 3.2) suggesting that intertidal mussel beds can be relatively stable features at large scales (i.e. in the same area for a number of years). However, they are usually very fragmented and each individual bed can experience substantial spatial and temporal variability in extent, density and total stocks as a result of, among other factors, recruitment failure and overexploitation (Dare *et al* 2004; Dolmer *et al* 1999, 2001). The data, including the nine selected and others not included in the analyses, are summarised in Table 3.3, which also contains estimates of extent, density and coverage from representative examples of *M. edulis* beds outside the UK for comparison.

Table 3.3 shows that discrete *M. edulis* metapopulations in the UK can range from $<1km^2$ to $15km^2$ (i.e. The Wash in the 1920s) and tonnages vary from <100t to more than 30,000t. Biomass or abundance (mussels m⁻²) provide a more intuitive measure of density in the present context. Most beds are less than $\sim15kgm^{-2}$, with recorded peaks of 30kg m⁻² while mussel abundance can vary from less than 100 mussels m⁻² to over 20,000 mussels m⁻² following spatfall events. Mussel beds with a mixed population of adult and young mussels usually range between 500 and 5000 mussels m⁻².

Table 3.3 Range of known extent and density parameters for intertidal (IT) and subtidal (ST) M. edulis	3
beds in the UK and continental Europe reported in the literature.	

Location	Туре	Area (km²)	Biomass (tonnes)	Density (kg m ⁻²)	Density (N m ⁻²)	% Cover	Source
Solway Firth (England)	IT	1.65- 1.98	6,000- 19,000	0.7-9.61	16-12,448	11-81%	North-west IFCA (2004-2006)
Fenham Flats (England)	IT	0.3-0.4	3,000- 6,000	7.5-16.41	536-1,323	64.91- 79.81%	Northumberland IFCA (2006-12)
The Wash (England)	IT	4-15	< 7,000 to 30,000	5-15	-	11-47%	Eastern IFCA annual stock survey reports (2009-2012)
Essex Estuary	IT	0.6-0.8	-	-	400-800	-	McGrorty <i>et al</i> (1990)
Kent coast	IT	4.36	3,000	-	-	-	Kent IFCA (Wright & Bailey 2009)
Lough Foyle (N. Ireland)	ST		968		5.5-19.6 (excluding re-laid mussel beds)		CEFAS (2009)
Lough Foyle (N. Ireland)	IT	0.06			1000- 3,000		Briggs (1982)
Burry Inlet (Wales)	IT	1.24- 1.66	2,160- 4,759		580-2250 (including Whiteford Scar)		Moore <i>et al</i> (2009)
Nigg Bay (Scotland)	IT	-	-	-	10-1,000	-	Trendall <i>et al</i> (2011)
Lincs Windfarm	ST				15,000		EGS Ltd.
Limfjorden (Denmark)	ST	19-83	616,000	0.31-3.3	-		Dolmer <i>et al</i> (1999; 2001)
Norderney (Germany)	IT	0.24- 0.31		1.4-11.9	918- 26,995	11-14%	Herlyn (2005)
Ho Bight (Denmark)	ST	11		0-63.9	486		Munch-Petersen & Kristensen (2001)
Dannish Waden Sea	ST	8.7	15,500- 117000	7.1-19.5	1,500 3,800	5-100	Kristensen (ND)
Sylt (Germany)	ST				1301- 4000		Buschbaum & Saier (2001); Dittman (1990); Asmus, 1987
Western Wadden Sea (Netherlands)	ST	10-60	10,000- 70,000		70-600		Nehls <i>et al</i> (2009); Dekker & Drent (2013)

a. England

The Wash

The Wash is a large embayment located on the east coast of England, between Lincolnshire and Norfolk. It is an area of very high conservation importance because it supports the largest populations of wintering waterfowl in the UK (Atkinson *et al* 2000). The birds mostly depend for food on stocks of common cockle (*Cerastoderma edule*) but also predate on mussels to a lesser extent (Atkinson *et al* 2003; Dare *et al* 2004). Wintering birds are therefore attracted by the presence of some of the most extensive intertidal mussel beds in the UK. The Eastern IFCA (EIFCA) is responsible for the management of the inshore fisheries in The Wash including the intertidal and subtidal mussel beds.

In The Wash, the intertidal beds have been subjected to intensive, large scale fishing pressure in the past and are also prone to substantial natural variability following cold winters, lack of recruitment and disease (Jessop & Maxwell 2011; Jessop et al 2012). In the past there have been at least 71 discrete mussel beds in the site and major fluctuations in stocks since the beginning of the 20th century. Peak total biomass records have ranged from 30,000t in the 1920s to 12,000t in the 1980s (Dare et al 2004). The decline continued to levels well below the 7,000t set by Natural England as a conservation objective for adult stocks resulting in total closures of the fishery in the early 2000s. This was followed by recovery to pre-1990 levels by 2004 (>12,000t) which led Natural England to change the conservation status of the Site of Special Scientific Interest (SSSI) from 'unfavourable declining' to 'unfavourable recovering'. Since then, fluctuations in tonnage have continued, forcing the total closure of the fishery in the 2010/2011 period. The reasons for these increases and declines in the population remain unclear (Jessop et al 2012). Besides fishing and bird predation there are natural factors that can affect the dynamics of the mussel beds in The Wash including ice scouring, storms, poor spatfall, and infestation by the parasite Myticola intestinalis. Incidentally, some beds that have been protected from fishing have continued declining, which might be explained by intraspecific competition (Dürr & Wahl 2004).

The total area covered by mussel beds in The Wash has also been dramatically reduced, from 15km^2 in the 1920s to less than 5km^2 in 2012. In the last three years, however, there has been a moderate increase in overall extent from 3.83km^2 in 2009 to 4.05km^2 in 2012) while mean density of stocks is also on the rise (Figure 3.4). This apparent recovery masks important declines in beds that held substantial stocks in the recent past, such as the Gat beds, likely as a result of poor recruitment and high mortality of older age classes (Jessop *et al* 2012). The map displayed in Figure 3.3 shows the location of The Wash mussel beds which are colour coded to indicate temporal trends in stock biomass for each discrete mussel bed between 2011 and 2012.





Figure 3.3 Location of *M. edulis* beds surveyed in The Wash by the EIFCA in 2012 and trends in stock biomass compared to surveys undertaken in 2011 (extracted from Jessop *et al* 2012).

Figure 3.4 Trends in mean *M. edulis* biomass across The Wash population between 2010-2012. Data manually extracted from EIFCA annual stock survey reports.

Data presented in the EIFCA reports (see references above) were re-analyzed for the present study using generalized mixed models (GLMMs) to determine the resolution of the density metrics used (response to spatial and temporal changes and associated variance). Time was included as a fixed factor in the GLMMs while bed was a random factor. The results showed biomass (kg) per m² remained relatively stable in 2010 and 2011 (t=-0.067; p>0.05). The increase in density (kg m⁻²) in 2012 was statistically significant (t=2.064; p<0.05). Percentage cover across the beds fluctuated between 11% and 47% with a general, non significant, declining trend since 2010 (2011: t=-0.56; 2012: t=-0.20). The boxplots displayed in Figure 3.5 show between and within-bed variability in mussel density was large and increased with time (CV=44%). Variability in mussel coverage across the sampling area was lower compared to density records (CV=35%). With the exception of 2011 (when a noticeable decrease in coverage in some beds occurred) mussel coverage remained spatially and temporally stable. The GLMMs (Table 3.4) indicated differences between discrete beds explained most of the random variability in density and coverage (73% and 51% of the total model residual variance, respectively). The significant spatial variability in the density metrics indicate that, although a positive trend for the beds as a whole exists, mussel bed dynamics are those of a highly fragmented metapopulation (Hanski & Simberloff 1997, Kritzer & Sale 2004).



Figure 3.5 Variability in intertidal mussel stock density and percentage cover in recent years in The Wash, England. Data extracted from annual stock survey reports published by the EIFCA.

Table 3.4 Summary of GLMM fitted to A) density and B) percentage cover of mussels surveyed in The Wash by (Jessop & Maxwell 2011; Jessop *et al* 2010; 2012).

A) Term	Estimate	Standard Error	t value
Intercept	7.45	0.76	9.77
Year 2011	-0.04	0.60	-0.07
Year 2012	1.23	0.60	2.06
Random effects	Variance	Standard Deviation	
Mussel bed	8.06	2.84	
Residuals	3.58	1.89	
Number of observations	60		
Groups (bed)	20		
B) Term	Estimate	Standard Error	t value
Intercept	28.45	2.24	12.71
Year 2011	-1.25	2.21	-0.57
Year 2012	-0.45	2.21	-0.20
Random effects	Variance	Standard Deviation	
Mussel bed	51.49	7.18	
Residuals	48.75	6.98	
Number of observations	60		
Groups (bed)	20		



Figure 3.6 Scatterplot matrix showing the frequency distribution and relationship between parameters relevant for density indicators (mussel bed area and mussel % coverage, density and total stocks) for data extracted from EIFCA mussel stock surveys in The Wash. Red lines represent lowess best fit lines and 95% confidence intervals. Green lines are best fit linear regression lines. Scatterplot constructed in R using the car package (Fox *et al* 2011).

Subtidal mussel beds also exist in The Wash, off the Lincolnshire coast near the Lincs and Inner Dowsing windfarms (Jessop & Maxwell 2011). Pre-construction reports by EGS Ltd. for Centrica Energy recorded 15,000 mussels m⁻² within the Lincs windfarm licensed area, although they were mostly juveniles. The bed was later surveyed by the EIFCA using a combination of AGDS and grab sampling and only empty young mussel shells were found. Another bed off the Norfolk coast (Sea Palling) was also surveyed for which a total 22,000 tonnes of mussel seed were calculated. These are very transient, subtidal beds formed by seed mussel and do not constitute biogenic reefs.



Figure 3.7 Digital image of mussel bed found by ENG Ltd. during the Lincs windfarm pre-construction survey, Lincolnshire. The presence of mussels is confirmed although turbidity and image quality do not allow for the extraction of meaningful metrics for density and community indicators.

Solway Firth

The Cumbrian section of the Upper Solway Firth has been managed and monitored by the north-west IFCA (NWIFCA - formerly the Cumbrian Sea Fisheries Committee) since 1994 (Solenvo 2006). The mussel scars in the Solway Firth are known to be ephemeral, with newly formed beds appearing each year following spat settlement events only to be washed away by storms. Some beds (i.e. Dubmill scar) expand or contract as a result of competition for space with the reef building polychaete *Sabellaria alveolata* (Mandy Knott, NWIFCA, pers.comm.; Solenvo 2006). Similarly to the results reported by the EIFCA in The Wash, stable beds dominated by adult age classes show little or no recruitment and stocks have diminished as a result of natural mortality.

According to the NWIFCA, from 2004 to 2006, the total area occupied by the mussel scars decreased from 1.98 to 1.65km². Scar extent was also very variable from a minimum of 0.05km² to maximum records of 0.7km². Records of total biomass suggest a steady decline from 19,212t in 1999 to 6,284t in 2006 (Figure 3.8). Abundance of mussels was very dependent on spatfall events and, as a result, could vary from just 12 mussels m⁻² to 21,000 mussels m⁻² (including spat).The Cumbrian Flats mussel beds are of moderate density

(mean biomass (kg) m^{-2} =4.68±2.41), with a maximum per bed ranging from 4 to 16kg m^{-2} . Coverage for the whole system varied little from 40-50% but variance was large in 2005. Figure 3.9 shows variations in density and coverage during the 2005-06 period. Boxplots clearly show that temporal variation in both parameters occur although is not very high and not significant (GLMM, t=-1.05, p>0.05). Spatial variability in density and coverage across the surveyed area was relatively low in 2004 (CV = 38 and 34%) and 2006 (CV = 54 and 27%) compared to the records obtained in 2005 (64 and 61%). The GLMMs (Table 3.5) indicated that between-bed variability explained 50% of the residual variation across all surveys. These patterns demonstrate that indicator metrics for mussel bed density in the Cumbrian Flats fluctuate in space and time. According to the NWIFCA reports (Solenvo 2004; 2006) these can be explained by natural processes of spat recruitment and adult mortality. These beds were exploited each year and the combination of increased fragmentation and poor spatfall in 2004/05 probably resulted in the increased variance in 2005. Increases in 2006 followed a large spatfall event that probably compensated for the fishing pressure, which incidentally doubled that year. Therefore, although indicators for density can detect spatial and temporal changes, more information would need to be incorporated into the models if spatial and temporal changes are to be interpreted within the context of natural factors and human pressures.



Figure 3.8 Temporal trends in *M. edulis* mean bed tonnage across beds in the Cumbrian Solway Firth. Data extracted from NWIFCA reports for the 2004-06 survey period.



Figure 3.9 Temporal trends and spread in *M. edulis* density and coverage across beds in the Cumbrian Solway Firth. Data extracted from NWIFCA reports for the 2004-2006 survey period. Boxplots represent the interquartile range. Whiskers represent the maximum and minimum values (1.5 times the box range). Dots are extreme values beyond the range covered by the whiskers.

Table 3.5 Summary of GLMM fitted to A) density and B) percentage cover of mussels surveyed in Solway Firth by Solenvo (2006).

Term	Estimate	Standard Error	t value
Intercept	4.71	0.83	5.68
Year 2005	-0.31	0.93	-0.34
Year 2006	0.32	1.10	0.29
Random effects	Variance	Standard Deviation	
Mussel bed	2.81	1.68	
Residuals	3.56	1.89	
Number of obsservations	23		
Groups (bed)	11		

Term	Estimate	Standard Error	t value
Intercept	48.50	6.37	7.63
Year 2005	-7.57	6.85	-1.11
Year 2006	-4.93	8.16	-0.60
Random effects	Variance	Standard Deviation	
Mussel bed	186.68	13.66	
Residuals	193.09	13.90	
Number of observations	23		
Groups (bed)	11		



Figure 3.10 Scatterplot matrix showing the frequency distribution and relationship between parameters relevant for density indicators for Solway Firth survey data using adapted Defra survey methodologies. Red lines represent lowess best fit lines and 95% confidence intervals. Green lines are best fit linear regression lines. Scatterplot constructed in R using the car package (Fox *et al* 2011).

Other English populations

The sheltered intertidal beds in the Exe Estuary (Devon; Figure 3.11) are found on shingle and mud (McGrorty *et al* 1990). Over a period of seven years the adult population showed relatively small variations in density (400-800 mussels m⁻²) and the area occupied (0.6-0.8km²). In the absence of anthropogenic impact and inclement weather conditions, density and abundance parameters were largely controlled by density-related mortality, immigration and recruitment.

Surveys carried out by the Kent and Essex IFCA in intertidal mussel beds around the Kent coast estimated up to 3,000t of mussels divided into six discrete beds. The smallest bed occupied 4 x 10^{-3} km² and had an estimated biomass of 45t while the largest was 1.8km² in extent but was dominated by juveniles resulting in a total biomass of 603t. The highest biomass was found at the Margate bed with 853t across 1.3km². Finally, stock assessment surveys carried out on a large (0.3-0.4km²) bed on the Fenham Flats (near Holy island) by the Northumberland IFCA (Green & Royle 2011, 2012, Green 2010) estimated a total biomass varying from 3,101t to 6,022t. These annual surveys have detected slight variations in total area as well as annual oscillations in biomass (7 to 16kgm⁻²) and abundance (536-1323 mussels m⁻²).



Figure 3.11 Location of mussel beds in the Exe Estuary where McGrorty *et al* (1990) undertook their surveys. The position and extent of the beds is derived from Marine Recorder polygon data provided by JNCC.

b. Northern Ireland

The most extensive mussel beds are in Lough Foyle where subtidal relaid mussels constitute the bulk of the standing stock biomass (8,304t in 2009) with just 968t of subtidal wild mussels at very low densities (ranging from 5.5 to 19.6 mussels m⁻²). Wild mussel densities are too low for these beds to be considered biogenic reefs. There are no published survey records for the intertidal natural beds with the exception of those taken from a 0.06km² intertidal bed by Briggs (1982) where densities of 1000-3000 mussels m⁻² were recorded. Our search for survey data for mussel beds in Northern Ireland (beside ephemeral mussel seed beds) was unsuccessful.

c. Scotland

The largest beds in Scotland exist in the wider Moray Firth, particularly in the Cromarty and Dornoch Firths. Mussel landings aside (historically ranging from 50 to 2,000t for the Dornoch fishery according to McKay & Fowler 1997) there are few published quantitative records of mussel bed extent and density metrics. For Nigg Bay (Cromarty Firth) densities of wild mussels varying from 100 to 1,000 mussels m⁻² are reported (Trendall *et al* 2011). Extensive sublittoral beds (SS.S.BR.SMus.MytSS) were mapped by Bates *et al* (2004) in the mid and outer Firth of Tay using an Acoustic Ground Discrimination System (AGDS) and drop down camera (DDC). Estimations of density from Van Veen grab and pipe dredge samples ranged from 20 to 830 adult mussels m⁻². Some stations had subtantial spatfall densities of up to 3410 mussels m⁻². No other data were found during the review process for Scottish *M. edulis* beds.



Figure 3.12 A: location of intertidal and subtidal mussel beds in the wider Moray Firth; B: location and extent of mussel beds in the Tay Estuary (Marine Recorder data and Bates *et al* 2004).

d. Wales

Since 2004, regular mussel stock surveys in the Burry Inlet Special Protected Area (SPA) have been carried out on an annual basis in order to help balance the needs of the existing fishery and the conservation of wintering waders (Stillman *et al* 2010). Surveys on behalf of NRW (Moore 2009) found temporal and spatial variability in all beds, particularly when including the large and ephemeral Whiteford Scar. The changes were particular important in 2006 and 2007 with 120% increases in mean mussel densities and corresponding 85% increases in mussel coverage. Both temporal and spatial variability were recorded for all metrics, the latter largely the result of differential recruitment levels between discrete beds. A summary of the values is displayed in Table 3.3. Power and GLMM analyses were undertaken for this report, firstly, to determine the adequacy of the sampling effort chosen by the surveyors and, based on those results, establish the recommended replication to detect change in density metrics of coverage and abundance m⁻². Secondly, change in density indicator metrics for *M. edulis* were examined over time and the level of error (variance) which could be explained by natural spatial variability was considered (Section 3.2.2 ii b)

e. Non UK M. edulis beds

Historically, some of the most extensive mussel beds in Europe were found in the Wadden Sea along the Dutch, German and Danish coasts (Figure 3.13). However, a combination of intense fishing pressure helped by poor spatfall resulted in the collapse of these populations. Collaboration between all three countries was necessary to ensure sustainability of the beds, leading to the implementation of tri-lateral policy agreements and management plans. At the moment there is a total fishing ban in place for intertidal mussel beds (Wolff *et al* 2010). The Wadden Sea Tri-lateral Monitoring and Assessment Program (TMAP) has therefore set out a conservation target for the intertidal mussel beds of increased area and more natural development and distribution of natural mussel beds. The parameters (indicators) chosen for monitoring purposes are total mussel bed area and biomass and percentage coverage of mussels in each discrete mussel bed.



Figure 3.13 Protected *M. edulis* fishery in the Wadden Sea (from Wolff *et al* 2010, report available from <u>http://www.waddensea-secretariat.org</u>).

Monitoring surveys in the Wadden Sea have consistently shown a declining trend in extent and biomass in spite of partial recovery following stringent protection measures (Figure 3.14; also Büttger *et al* 2008; Dankers & Brinkman 2001; Dankers *et al* 1999). In The Netherlands, mussel bed area has historically fluctuated between 10 and 60km² in the intertidal and shallow subtidal. Following intense pressure from the fishing industry and low spatfall the Dutch beds completely collapsed, fully disappearing in the late 1980s (Brinkman *et al* 2002). Unlike in the UK, the Dutch fishery employed vessel dredging on intertidal beds whereas UK intertidal fisheries are mainly hand gathered on a small scale. The beds partially recovered as a result of good spatfall years and the strict protection measures put in place (Nehls *et al* 2009). The current fishery is limited to the subtidal seed mussel beds where dredgers are licensed to collect seed for relaying in licensed plots. Mussel dredging is most intense in The Netherlands and Germany and non-existent in Denmark where there are no licenses for culturing. Export of seed is also banned (Nehls *et al* 2009). Recent (2008) stratified surveys have estimated a total 55,000t of subtidal mussels in the whole Dutch Wadden Sea (Goudswaard *et al* 2008).

In Germany, following protection, spatfall has been the main factor regulating the dynamics of the intertidal populations in Niedersachsen (50 to 10km²; 1,000 to 100,000t) and Schleswig-Holstein. The latter are mussel beds that appeared following intense spatfall events in 1987 following a series of cold winters. At that time they occupied an area of 15km². Tonnage has decreased from an initial 60,000t to 8,000t in 2005. Mussel coverage started at 43% but has steadily decreased to values between 27 and 19%. Historical beds in the Ameland region are reported as extinct (Dankers 1990). The Danish beds also collapsed in the 1980s due to large scale unregulated fishing, severe winters and oxygen depletion (Dahl et al 1994; Dolmer et al 1999; Laursen et al 2010) but have recovered to 2,000 mussels m⁻² and biomass densities of 5-50kg m⁻² (Dahl et al 1994; Dolmer et al 1999; Kristensen 1995). Following partial closures of the Danish fishery in 1994, Laursen et al (2010) found mussel beds declined in extent after the closure, from 6.41km² to 3.35km². Biomass, however, did not experience a significant variation before and after the closure. Coincidentally, similar trends have been reported in the Gat beds in The Wash (Ron Jessop EIFCA, pers. comm.). Nevertheless, it is not uncommon for recovery metrics such as species abundance to continue to decrease immediately after a closure before a stable state is reached (Gerber et al 2003; 2005).

In the Republic of Ireland, monitoring surveys in intertidal and subtidal beds have solely focussed on tonnage estimations with a view to commercial exploitation. Tonnage is the only metric reported in stock assessment reports from commercial beds. All the survey report consulted indicated temporal and spatial variation in tonnage among beds in Lough Foyle, Lough Swilly (120–200t in intertidal beds) and estuaries in the Irish Coast including the Boyne River (1,500t), Dundalk Bay (70-230t) and Wexford Bay (724-1199t) (source BIM stock reports: Meany & Edwards 1968; Meany 1970; Edwards 1969 and ensuing reports). Mussel beds in the Irish Sea coast of Ireland are monitored and managed by Bord Iascaigh Mhara (BIM) on an annual basis using Side-scan Sonar ground-truthed with Day grabs and dredges (www.bim.ie/aquaculture, Nicolas Chopin, pers. comm.). These beds contain high densities of mussel spat carpeting the seafloor and although most are thought to be very transient as a result of heavy predation and storm events, they might constitute more permanent features than originally thought (Francis O'Beirn, Marine Institute (Ireland), pers. comm.).


Figure 3.14 Temporal and spatial variation in area (left) and percentage cover (right) of *M. edulis* beds in the Wadden Sea from 1999 to 2007 (from Wolff *et al* 2010, report available from <u>http://www.waddensea-secretariat.org</u>)

ii. Evaluation of existing surveying techniques

Most of the standard quantitative assessment methods for mussel beds originated in the Wadden Sea, where a need to monitor their conservation status arose following the catastrophic stock collapse in the 1980s and 1990s. These methodologies usually rely on a combination of remote sensing and *in situ*, walk surveys to obtain accurate and reliable information including location, area and structure of the population (Herlyn 2005). Although advances are being made whereby predictive models are being used to extract density indicator measures from remote sensing imagery, to date, the recommended approach involves using ground-truthed remote imagery and *in situ* field surveys. Remote imaging is used to locate the beds while estimations of percentage cover and mussel density, among other parameters, are extracted from walk surveys. Those estimates can later be extrapolated to obtain whole-bed biomass or standing stock numbers (Kristensen 1995; TMAP Monitoring Handbook). Overall, the best technique should capture the spatial and temporal variations in the stocks with the lowest possible error. The metrics have to be easily measured and have low levels of quantifiable error (Elphick 2008).

For the purpose of Descriptor 1 indicators relevant to *M. edulis* beds (density of the reef forming species and community abundance) the main aim would be to identify decreases in the indicator metrics following anthropogenic impact and recovery following protective measures (Jennings 2000; Muniz *et al* 2005). At the same time indicators need to be cost-effective and easy to use (Gerber *et al* 2005). The techniques presented below are all resource management approaches aiming at establishing a reference stock threshold below which corrective measures are implemented (Gerber *et al* 2005). For the purpose of establishing GES for *M. edulis* reefs the proposed indicator is '*density of the reef forming species*', however, it remains doubtful that such total standing stock threshold values (i.e. tonnage) are meaningful, because they include ephemeral spatfall.

This section provides a critical review of industry methods for monitoring mussel bed density and includes a descriptive exercise to determine the power and sample replication needed to detect meaningful changes in the indicators using data collected from the Burry Inlet surveys carried out by Jon Moore on behalf of CCW (now NRW) from 2005 to 2009.

a. Remote sensing methods

Aerial photographs

This method is still widely used in the Wadden Sea monitoring programmes to locate the position and determine the area occupied by intertidal mussel beds (Karin Troost pers. comm. See also Dolmer et al 1999; Kristensen 1995; Nehls & Thiel 1993). Weather conditions are extremely important for the success of an aerial survey as they influence the quality of the photographs produced. Aerial surveys need to be conducted under sunny, clear skies, with no wind and at low water when mussel beds are fully exposed (Kristensen 1995). The methodology, described by Herlyn (2005) and adopted by the TMAP Blue Mussel Group, involves delineating the boundaries of the beds with the aid of a stereoscope and georeferencing using GIS. The area of each bed in km² is easily estimated with the aid of GIS. Two main parameters can be extracted from aerial photography: total area of the mussel bed and percentage cover of mussels (expressed as area occupied by mussels in relation to total bed area). The resolution recommended for bed delineation and area calculation is 1:10.000. The main source of error is human related as the method relies on a surveyor to visually estimate the boundaries of the bed (Hendrick & Foster-Smith 2006). Estimates of percentage coverage need much higher resolution (1:2,500) in order to reduce the error. It is possible, however, to obtain estimations of coverage using image processing

software (see Kristensen & Borgstrom 2006, in Wolff *et al* 2009) and the recommended approach is to obtain *in situ* estimations as part of the stock assessment and ground-truthing transect surveys. Laursen *et al* (2010) used aerial photography to detect change linked to bird predation.

Aerial photography could also be used to map shallow sublittoral mussel beds however the scale of error is likely to be higher compared to intertidal beds as a result of light attenuation, sea surface reflection and turbidity, among other processes (Kenny *et al* 2003).



Figure 3.15 Scanned orthorectified aerial photograph of an intertidal mussel bed in the Wadden Sea (Denmark). The digitised image can be processed using software to obtain an estimate of the total area (right). Image from Wolff *et al* (2009) and Kristensen & Borgstrom (2006).

Low altitude aerial photography (~50m) using balloons or kites is a cheaper, less weather dependant alternative to plane-based aerial photography which has successfully been used to map intertidal habitats (Bryson *et al* 2013) and oyster beds (Troost 2010; <u>http://www.blimppics.com/</u>).



Figure 3.16 Low altitude photograph using a blimp balloon in an oyster bed in the Oosterschelde (The Netherlands). Photograph from Troost *et al* 2010 (credited to Johan van de Koppel).

Acoustic Ground Discrimination Systems (AGDS)

Single beam AGDS such as RoxAnn[©] have been used to locate and map sublittoral mussel beds for a number of years (e.g. surveys by the EIFCA, Jessop 2012). This method requires constant calibration against known substrate types and, although it might be suitable as a broad-scale mapping method, the resolution is usually not high enough to detect subtle differences between mussel beds and other seafloor habitats (Limpenny *et al* 2010; Mair *et al* 2010a) or to discriminate between live mussel beds or empty shells (Jessop & Maxwell 2011; Magorrian *et al* 1995). Although cheaper than aerial photography and relatively simple to operate, its low reliability means that AGDS is being replaced by more accurate, high resolution side-scan and multibeam sonar systems as the preferred method to map subtidal mussel beds (Vorberg *et al* 2009). According to Maguire *et al* (2007; Appendix VI), a North-South, zig zag survey approach using several vessels is preferred to ensure maximum coverage while undertaking acoustic seed mussel surveys.



Figure 3.17 AGDS tracks on seed mussel beds (red pattern) surveyed by Bord lascaigh Mhara (BIM) in the Irish Sea, off co. Louth, Ireland. Superimposed labels indicate size and density of seed from dredge tows used to ground-truth the acoustic signal. (Source: Nicolas Chopin, BIM).

Side-scan sonar

Side-scan sonar has recently been used in the Wadden Sea to map intertidal (surveying at high water) and shallow subtidal mussel beds (van Overmeeren *et al* 2009). The results indicated that, once the maps were calibrated using species-specific acoustic signals, a high

level of accuracy can be obtained (van Overmeeren *et al* 2009). Although raw acoustic maps are not suitable to derive estimations of mussel coverage or density, van Overmeeren *et al* (2009) found that backscatter filtering and calibration using *in situ* counts from walking transects allowed quantitative measurements of density for intertidal mussel beds with a high level of accuracy. However, statistical confidence is an issue for subtidal mussel beds, because grab sampling and other remote methods used in the ground-truthing phase usually introduce larger density estimation errors (Dolmer *et al* 1999; McIntyre & Eleftheriou 2005). Side-scan sonar is, therefore, a very useful method to locate and map mussel beds at very high resolution allowing more targeted and precise subsequent *in situ* surveys.

Hydroacoustic methods are more widely used to locate and map subtidal seed mussel beds in the East coast of Ireland. In the Netherlands, side-scan sonar has been successfully used to map shallow and very shallow beds (1.5 – 10m). Alongside Multibeam sonar, Side-scan sonar is the most accurate and cost-effective system to discriminate different seafloor texture patters and relate them to habitat types using ground-truthing. Side-scan postprocessing software can be used to analyse backscatter and automate the identification of mussel beds. Sonarwiz ® software (<u>www.chesapeaketech.com</u>) employs image analysis techniques from the medical imaging sector to produce seabed habitat classification including that of mussel beds (Nicolas Chopin, BIM, pers.comm.).



Figure 3.18 Top: Google Earth satellite image (left) compared with high resolution side-scan sonar of the same area (right) showing a mussel bed in the shallow subtidal. Wadden Sea (Overmeeren *et al* 2009). Bottom: side-scane sonar output map of an area surveyed in the Irish Sea by BIM. Rugosity patterns were consistent with seed mussel bed. The data is ground-truthed with grab samples and biomass estimates are derived for each bed (Nicolas Chopin, BIM).

LiDAR surveys

LiDAR stands for 'light radar' (or 'light detection and radar' depending on the sources) and it is an established technology widely used in precise surface terrain mapping. LiDAR systems produce fine scale digital terrain maps that can be trained to identify intertidal biotic communities (Chust *et al* 2008; 2010). For mussel beds, Schmidt *et al* (2013) used classified random field categorization models to discriminate between water, mudflats and mussel beds in the German Wadden Sea. Although there was some level of error for mussel beds (particularly around the edges between mudflats and mussel beds), misclassification was relatively low (39.9%). Aside from bed classification, LiDAR could be used to obtain density metrics in the same manner as it has been done for terrestrial habitats, such as forests (Clawges *et al* 2008) and volume calculations of intertidal *S. alveolata* reefs (Almeida Noernberg *et al* 2010).

For subtidal marine habitat mapping, water-penetration bathymetric LiDAR has been used to quantify seabed topographical complexity, particularly of biogenic reef structures such as corals. Standard LiDAR is not useful for submerged habitats as the light does not penetrate the water column but advanced, bathymetric LiDAR technologies such as those used by the National Aeronautics and Space Administration (NASA) (low-beam divergence, 5KHz pulse repetition Experimental Advanced Airborne Research LiDAR (EAARL)) can avoid such issues to produce seafloor elevation models of 1m resolution (Zawada & Brock 2009). Although LiDAR can successfully resolve seabed topography, the range of operating depths is shallower than for acoustic methods such as side-scan sonar or multibeam (Costa *et al* 2009) while environmental conditions (i.e. waves) can still affect the outputs (Wang & Philpot 2007).



Figure 3.19 Digital terrain image and overlaid predictions for mussel beds (red) and water (blue) using **LiDAR** (Reproduced from Schmidt *et al* 2013).

Remote and in situ video and photography

Camera stills and video footage can be an alternative to destructive sampling methods, which is a preferable approach in the context of monitoring sensitive habitats such as biogenic reefs. One such method is fixed viewpoint photography, whereby photographs of a fixed area in the intertidal are taken using a camera mounted on a tripod of constant height to monitor temporal variation. A similar approach can be taken, as a non-destructive alternative to standard destructive survey methods (see next section, in situ walking surveys). Following protocols adapted from those used in stock assessment surveys (see *in situ* survey methods, Section 3.2.2 iib), a frame mounted camera can be used to take

quadrat photographs across *M. edulis* beds $(0.25m^2)$. The images can be later processed using image processing software (e.g. Image J[®], Schindelin *et al* 2012) to obtain density indicator metrics such as mussel percentage cover and density as well as recording the associated epifaunal assemblage. This method has been used before in fractal geometry and density studies for mussel beds (Snover & Commito 1998) and could provide a useful, non-destructive alternative to standard quadrat clearance approaches used by fisheries managers. The South Wales Sea Fisheries Committee has used fixed point photography to investigate the dynamics of the large ephemeral mussel beds at Whiteford Point. This study demonstrated the effect of weather on the distribution of seed mussel and the formation and subsequent erosion of mussel herms (Andy Woolmer, pers. obs.).

For subtidal mussel beds, drop down cameras (DDC) and towed video are routinely used to ground-truth acoustically derived maps of the seafloor (see representative footage in Figure 3.20). The most common configuration includes a towed video sledge to determine presence, bed boundaries and percentage coverage to supplement the side-scan sonar outputs (Agri-Food and Biosciences Institute (AFBI) in Northern Ireland, Bord Iascaigh Mhara (BIM) in the Republic of Ireland and the English Inshore Fishery Conservation Agenices (IFCAs) use or are planning to use this method (Mandy Knott, NWIFCA, pers comm. 2013).

Remote imaging systems are never used on their own as a means to obtain stock density estimations as they depend on environmental conditions, particularly good visibility, to produce clear images. Subtidal mussel beds are common in estuaries where turbidity is a major issue and most accounts indicate that image rejection rates are very high (CEFAS 2007; Wright & Bailey 2009).

Notwithstanding the low number of video and images accessed during the present study that contained good quality footage of *M. edulis* beds, a preliminary analysis indicates that the method is prone to the same sources of error listed for *Sabellaria spinulosa* reefs (Chapter 4): turbidity, poor focus, siltation and epifauna obscuring the reef building organism. However, if turbidity is low, high resolution DDCs deployed perpendicularly to the seafloor can be treated as replicate quadrats from which mussel density (as number of mussels per m²) and coverage following the same approach described in Chapters 2 and 4 for *Modiolus modiolus* and *S. spinulosa*. The effect turbidity has on image quality, particularly if operating in estuarine conditions, can be substantially reduced using freshwater lens camera systems (described in Chapter 4). Drop down imagery can be useful to extract density indicators because *M. edulis* beds normally have low presence of epifauna and mobile fauna than could obscure the counts (which is the main caveat when using the system for *M. modiolus* beds).

Remotely Operated Vehicles (ROVs) are difficult to manoeuvre under high currents and produce footage of low quality at the expense of rapid deployment and wider survey range. They are useful ground-truthing systems but unsuitable as means to extract abundance estimates. We have tested (Hirst *et al* 2012) and evaluated (earlier chapter) the use of ROVs on *M. modiolus* beds and found them to be insufficient for the present indicator development and monitoring objectives. There is therefore no reason to believe that they will perform any better on smaller mussel species in probably lower visibility conditions.

An alternative to standard imaging methods is the Habitat Mapping Camera System (HABCAM), developed by the Woods Hole Oceanographic Institution in the USA (<u>http://habcam.whoi.edu/</u>). HABCAM is an integrated, self-contained ROV used in benthic habitat mapping and environmental monitoring surveys. It uses a combination of high definition optical cameras (~1mm pixel size) and side-scan sonar as well as Conductivity,

Temperature, Depth (CTD) and multiparameter probes. The system could be very useful in bivalve stock assessment surveys and it has already been successfully used in partnership with the commercial scallop industry in the east coast of the USA. The HABCAM system produces fine scale maps of bed extent, scallop biomass estimations, dredge efficiency estimations and population structure analyses (height frequency distributions). Although there are obvious applications of this system in density and community indicator monitoring the cost is likely to be excessive. Other imaging alternatives include, for example, the Pyramid© frame camera system (Harris & Stokesbury 2010), which is being trialled for scallop stock assessment by the Devon and Severn IFCA with a view to its future application in surveying subtidal *M. edulis* beds (Sarah Clark, Devon & Severn IFCA, pers. comm. 2013).

In situ videoing/photography of quadrats is commonly used to monitor *M. modiolus* bed coverage and density (as seen in Chapter 2) and it is preferred over DDCs because of the lower level of error associated with *in situ* video data. The use of divers to monitor density indicators for *M. edulis*, however, increases the costs and limits the desired level of replication that could be achieved with DDCs. Drop down systems are preferable to *in situ* videoing because subtidal mussel beds are can be extensive and the mussels are more visible so they usually lack epifauna that can obscure density estimates as reported for *M. modiolus* reefs. However, the use of divers would be a more adequate approach to obtain abundance and cover estimates of very shallow subtidal beds, for example, in combination with video transect surveys to map bed extent and fragmentation (Moore *et al* 2003).



Figure 3.20 A) Freshwater lens drop down camera, Thanet windfarm, Kent; B) Drop down camera, North Llyn, Wales; C) Drop down camera, Lincs Windfarm, Linconshire; D) Towed video, North Llyn, Wales; E) Handheld camera, Irish Sea; F) ROV, Irish Sea seed mussel bed. Image sources: A, Vattenfall; C, ENG Ltd.; B&D, NRW; E&F Nicolas Chopin, BIM.

Remote destructive sampling gear

Remote sampling using destructive equipment including box corers, dredges and grab samplers is the standard method used to ground-truth imagery of subtidal mussel beds. Beam and other trawls are used for qualitative sampling of the epifauna over large areas. On occasions, they can provide samples of the mussels themselves when used over dense beds but they are highly destructive and not very efficient (McIntyre & Eleftheriou 2005; Figure 3.21).

Existing literature indicates that, to extract quantitative estimations of mussel stocks, the most cost-effective strategy involves the use of grabs and dredges to locate the edge of the beds and obtain density estimations once the location and extent of the target mussel bed is known (i.e. following acoustic mapping) (Nicolas Chopin & Ron Jessop, pers. comm.).

Estimates of biomass from dredges can be obtained by multiplying the catch by the efficiency of the dredge and dividing the result by the total area covered (Laursen et al 2010). The area sampled can be calculated by multiplying the tow speed by the tow time and the width of the dredge (Munch-Petersen & Kristensen 2001). The efficiency of a dredge is usually higher in high density mussel beds compared to low density, fragmented ones (CEFAS 2007; Dolmer et al 1999). Grabs used to sample mussel beds are usually Van Veen grabs and Day grabs. Grabs can be prone to misfiring or jamming particularly if the substrate is pebbly and their deployment is more weather dependent than dredges. As the surface sampled by a grab replicate is relatively small (usually 0.1m²) there is a need for high replication to reduce the variance associated with heterogeneous, patchy beds. However, from a statistical point of view, it is advantageous to collect numerous, small samples particularly as if the habitat is very heterogeneous (McIntyre & Eleftheriou 2005). A commercial dredge will only provide a one-off, averaged sample across the track thus losing important information that could be useful from a habitat conservation perspective (i.e. fragmentation). Smaller sampling dredges address these issues but still return only a qualitative sample.

The survey design is also important and should take into account the variation in bed structure by following a stratified design. In the case of commercial dredges it is recommended to follow replicated north-south, east-west transects across the mussel bed (Maguire *et al* 2007). The level of replication should be established based on mean density and standard deviation values obtained from exploratory pilot surveys.



Figure 3.21 Top: Small dredge with coarse mesh bag and catch obtained during epifaunal and mussel stock surveys surveys on mussel beds Ireland (Photograph supplied by Dr. Brendan O'Connor, reproduced with permission); Bottom: Catch showing young mussels and associated epifauna from beds off the Norfolk Coast. A beam trawl, among other gears, was used for large scale ground-truthing acoustic data collated during the geophysical surveys carried out during the East Coast REC (www.marinealsf.org)

b. In situ walking surveys (intertidal beds)

For intertidal beds, once the position and extent has been defined using remote imaging methods, the best approach to obtain an accurate and reliable representation of the mussel standing stocks and the condition of the population is to use *in situ* walking transect surveys. Fisheries and conservation agencies have been using different *in situ* approaches to obtain standing stock values to derive total allowed catches (TACs) or to compare them against set conservation objectives (such as those introduced by Natural England in The Wash, see Jessop et al 2012). Density indicator metrics that could be used to determine GES for mussel beds include percentage cover, biomass or abundance m⁻². Length-frequency or age frequency distributions can also be obtained to follow reproductive output and recruitment rates (www.ospar.org). Most methods have been derived from Wadden Sea monitoring programmes and are recommended by The Wadden Sea Tri-lateral Monitoring and Assessment Program TMAP (Common Wadden Sea Secretariat 2008). In the UK stock assessment surveys are usually carried out in August/ September, two months after the end of the main spawning season for *M. edulis* (May-July, according to Seed 1969; and Maguire et al 2007) and just before the mussel fishing season commences and the arrival of overwintering birds. In the Netherlands the TMAP recommends two annual surveys, in spring and autumn.

Methods based on West et al (2004)

Used in the Burry Inlet cockle and mussel stock surveys on behalf of the Countryside Council for Wales (CCW), now Natural Resources Wales (NRW) (Moore 2009); surveyors are provided with maps featuring the mussel beds known from previous surveys. Before undertaking the survey the surveyor overlays a grid over each bed ensuring that a minimum of 30 equally spaced sampling stations are distributed across the area. Upon choosing the number and location of the sampling stations, the surveyor walks the perimeter drawing a sketch for each bed and entering representative waypoints in a handheld GPS. These positions and hand-written sketches of each bed are then used to derive polygon layers on a GIS map. Working two hours each side of the tide, the sampling procedure involves laying a 0.1m² quadrat right beside the front of the foot to avoid bias. For each quadrat, percentage cover is calculated. All mussels within the quadrat are removed. However, if cover is 100% a proportion of the quadrat is cleared instead (25%-50%). The samples are later counted and measured. A sub-sample is retained for weight-length and Ash-Free Dry Weight (AFDW) analyses. The parameters derived include percentage cover, abundance per m², total abundance, biomass per bed and total biomass (tonnage).

Walker and Nicholson (1986) 'foot on' method

This methodology was adapted from the *stiefelmethode* ('boot method') used in the German and Dutch Wadden Sea. In England it was adopted by Defra and it is currently being used by officers from several IFCAs (NWIFCA, Northumberland IFCA). The bed perimeters of each bed are defined by walking around them taking co-ordinates as waypoints on a handheld Geo-Positioning System (GPS) terminal. The surveyor walks each bed following straight tracks of up to 300 steps following a zigzag pattern to ensure the bed area is randomly covered. Percentage cover for each track is calculated as the co-efficient between the number of steps that land on live mussels and the total number of steps for each track. At the end of each track a 0.1m² quadrat is laid right in front of the surveyor's foot. The percentage cover of mussels is calculated by eye prior to removing all the mussels within the quadrat which are subsequently counted, weighed and the total fresh weight measured. Length and age measurements and net biomass are calculated for every third quadrat sampled. This method relies on the surveyor to determine the best strategy to randomise the layout of the tracks and there is always the danger of human bias at several levels (positioning of the quadrat, presence or absence of mussels under the foot in very muddy areas, Ron Jessop pers. comm.). Moreover, compared to other methods (e.g. the Dutch 'wand' method, see below) the 'foot on' method relies on a lower number of samples distributed across each individual bed, sacrificing sample replication in favour of increased individual sample area.



Figure 3.22 Metallic box quadrat used to obtain mussel samples following the foot on zigzag survey methodology (Solenvo 2004)

Dutch 'MarinX' wand method

Developed in the Netherlands (van Stralen & Boit 2004) as an alternative to even grid stratified approaches, this method aimed at obtaining a more reliable estimate of densities for individual patches in fragmented beds. The rationale of this method is similar to that used by Walker & Nicholson's (1986) method, whereby the beds are first located and their boundaries mapped with a GPS at low water to calculate the area of each bed. Once the bed is mapped, a random zigzag pattern is then delineated by the surveyor to cover the entire bed. The most important difference with the 'boot on' method lies in the use of a 1m long pole ('wand') with a plastic ring (11cm diameter) attached to its end. Every 3 or 4 steps the pole is placed on the ground, without looking, and the presence or absence of mussels are recorded as hits or misses (Figure 3.23). The number of hits is later used to calculate percentage cover of mussels for each track. Samples are collated every 4th or 5th hit by means of a 10cm deep plastic corer with the same diameter as the wand ring (i.e. 11cm). Each transect or track consists of a fixed number of paces (~150).

At the end of each track all samples are pooled together into a 5L bucket and later weighed to obtained density (biomass m⁻²) and total biomass estimations derived from the area of the bed, mussel density and mussel percentage cover.

The main advantage of the MarinX method is the substantially higher replication achieved compared to Walker & Nicholson's (1986) method, and this results in lower variance. The MarinX method has a more efficient randomization approach and a much clearer protocol which is less prone to surveyor bias. The surface area is, nonetheless, smaller to that of a foot and there is a possibility that the method underestimates coverage compared to the Defra method, particularly in beds with very low densities of mussels. The method is less prone to surveyor error because it avoids biased estimates of what is a representative sample or what constitutes a hit (Van Stralen & Bol 2004; Ron Jessop, EIFCA, pers. comm.). Although the methodology relies on the surveyor to establish randomness it is possible that issues of spatial autocorrelation or non-independent errors might arise. These issues could be compensated by using GLMMs (Bolker *et al* 2009; Cook *et al* 2013).



Figure 3.23 Surveyor using the Dutch wand method to survey intertidal *M. edulis* beds in The Wash. Core used to collect mussel biomass samples (photo: Ron Jessop, EIFCA).

c. Intertidal survey optimization: Power and sample effort using Burry Inlet datasets as case study

A power analysis was undertaken using raw percentage coverage and numbers of mussels m⁻² (abundance) data to use as potential variables for density indicator metrics. Total bed abundance or tonnage are not statistically appropriate because they are correlated (i.e. the values are derived from percentage cover and abundance measurements). The main aim of the exercise was to establish:

- 1. The adequacy of the sampling effort used in each annual survey by calculating the achieved power to detect different changes in the mean coverage and abundance of mussels;
- 2. Based on the mean and standard deviation of the mussel abundance and coverage for each, determine the sampling effort needed in future surveys;
- 3. Develop GLMMs to establish if the method used can capture spatial and temporal change in density indicator metrics.

Figure 3.24 shows the evolution of the Burry Inlet mussel beds mapped by Moore *et al* (2009) from 2005 to 2008. The position of the beds does not change much from year to year suggesting these beds are relatively stable features in the Burry Inlet.

The total area covered by the mussel beds each year varied from a minimum of 0.48km² in 2004 (not shown in Figure 3.24) and 1.92km² in 2007. Following an increase in 2005, total area remained between 1.2km² and 1.92km² until 2009 (

Figure 3.25). The boxplots in Figure 3.26 show the interquartile ranges clearly overlapped throughout the period, indicating very little temporal and spatial variability in mussel density and percentage cover. The records from Whiteford Scar, a substantially larger mussel bed dominated by ephemeral juveniles are the outliers (black dots in Figure 3.24) located above each boxplot. A generalized linear mixed effect model (GLMM) using area as the dependent variable, year as fixed factor and bed as a random factor indicated between-year variability was not significant with the possible exception of 2006 (t=2.026; p=0.497). Between- and within-bed variability explained 92% of the residual variability in area.







2007

2008

Figure 3.24 Spatial and temporal variability in location and extent of intertidal *M. edulis* beds (in purple) in the Burry Inlet, Wales (including Whiteford Scar). GIS layers derived from data by Moore *et al* (2009). Maps were developed on Google maps base layers using QGIS v2.0.



Figure 3.25 Top: Temporal variability in area of mussel beds across the Burry Inlet. Bottom: Cumulative average area bar chart for the same period. The box represents the interquartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values. Areas are measured in hectares (ha.).



Figure 3.26 Change in density (top) and mussel coverage (bottom) across the Burry Inlet *M. edulis* beds from 2005 to 2009. Within- and between-bed variability is substantial but the time parameter is temporally stable. The box represents the interquartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values.

GLMMs indicated density was significantly variable with time but solely as a result of the increase recorded in 2007 (possibly due to a successful spatfall). Mussel coverage was much more stable over time with no significant effect of year (p>0.05). Most of the random variability was the result of spatial variability between the beds. Both parameters were highly correlated at a significant level (r=0.95; p<0.001; Figure 3.27) suggesting that estimation of percentage coverage might suffice as an indicator of density. It is likely that this result reflects a steady-state of exploitation maintaining densities on these habitually fished beds (Andy Woolmer, pers. comm.). However, without knowing if anthropogenic impacts are occurring in the area it is not yet possible to validate this relationship.



Figure 3.27 Scatterplot matrix showing frequency distributions and relationships between mussel cover (%) and corresponding mussel density (mussels m^{-2}).

The power analysis of the existing surveys was based on a two-sided t test (Crawley 2013) using the known sample sizes used each year over known *M. edulis* bed areas (displayed in Figure 3.28). The results are displayed as a line-chart displaying detected percentage change in the ordinate axis and mean statistical power $(1-\beta)$ with standard error bars. The chart indicates that the level of sampling resulting from adopting the chosen methodology (based on West *et al* 2004; see Section 3.2.2 iib) is more effective in detecting change in cover than density (a result of the higher error in obtaining density estimates compared to percentage cover estimations).

Overall the replication successfully captures 30-40% changes in mean mussel coverage with an 80% probability of detecting a Type II error (accepting the null hypothesis when it is false, i.e. no change). The power to detect change at much higher resolutions (10-20%) with that level of replication is well below the 80% (0.8) threshold. The results clearly indicate that 40% change could be detected with 80% certainty at α =0.05 for the number of samples used in the previous surveys (n=229-340). However the confidence in detecting changes in mean density of less than 40% was well below the 0.8 threshold (Figure 3.28).



Figure 3.28 Power to detect change in mean density and coverage for historical mussel stock surveys in the Burry Inlet using protocols adapted from West *et al* (2004). Significance α is set at 0.05. Ordinate axis represents percentage change in the mean.

The level of replication needed across the mussel beds mapped in the Burry Inlet was calculated based on power=0.8 and α =0.05 with bootstrapped mean and standard deviations of density and coverage (10,000 iterations without replacement). The results are more easily interpreted by representing detected change in the ordinate axis and the required number of samples needed to detect it using a 0.1m² quadrat over the survey areas displayed in Figure 3.29. The results confirm that the level of replication derived from the protocols used in the Burry Inlet is indeed adequate to detect changes of more than 40% in mean density. Moreover, less replication would have achieved the desired power (0.8) for any desired change, particularly if the variable used was percentage coverage which is subject to lower variances (i.e. 40% density=225 samples; coverage=108 samples). Changes of 40-50% in the mean are the usual cut-off points in monitoring survey design (Crawley 2013; TMAP guidelines 2009). It is recommended that, to avoid unnecessary replication and reduce costs, sampling effort is revised during the planning phase using the results from the preceding survey as baseline data.



Figure 3.29 Sampling effort estimated for mussel cover and density using standard walk and quadrat survey methodology using survey data from historical surveys in the Burry Inlet, Wales. Ordinate axis represents percentage change in the mean.

Box 3.1 Summary of findings for *M. edulis* bed density indicators.

- Studies show inherent spatial variation in density indicator metrics which can be detected using existing methodologies and metrics. At the broad-scale, density indicators are, nonetheless, quite stable through time if recruitment occurs. However, temporal stability can mask declines in individual beds.
- Density indicators are responsive to anthropogenic impacts. Substantial decreases in extent and biomass of intertidal mussel beds have been linked to vessel overfishing in the Wadden Sea and mass mortalities can occur as a result of episodes of hypoxia.
- However, in most cases mussel bed dynamics depends on the balance between recruitment and mortality due to predation, disease, and natural events and not only to overfishing and other anthropogenic impacts (e.g. coastal defences, bait collection).
- There is a significant positive correlation between percentage coverage and mussel abundance.
- Standard industry approaches to monitoring (e.g. Wadden Sea TMAP) are recommended for the purpose of density indicator monitoring. Collaboration and data sharing between home country agencies in the UK is also desirable.

Box 3.2 Recommendations for *M. edulis* bed density indicators.

General Recommendations

- Monitoring of density indicators for GES of *M. edulis* reefs should focus on temporally stable beds (minimum persistence of 2-3 years).
- *Mytilus* biotopes that do not qualify as biogenic reefs (e.g. PMFs) are not suitable for MSFD GES monitoring.
- Mussel spat (<10mm) should be ignored for the purpose of density indicator monitoring to help with the interpreting of bed condition dynamics. However, population structure data could be relevant to community indicators (see **Box 3.3**).
- Spatfall from the autumn is still going to affect the results and specifiying spring is going to be at variance with current fishery / bird prey surveys. Sampling effort should be estimated using density and standard deviation values from previous surveys to calculate 40% changes in density indicators.
- Statistical analyses: generalized linear mixed models can deal with temporal and spatial pseudoreplication, non normal distributions and unbalanced designs. They are suitable for the kind of data generated by these surveys.

Recommendations for intertidal beds

- Aerial photography is needed to identify new mussel beds and determine inter-annual variation in bed area and mussel coverage. The use of side-scan sonar should also be contemplated.
- Area of each discrete bed, mussel abundance, size-class distribution and % cover are industry-tested indicators useful as density indicators for GES.
- The significant correlation between mussel coverage and density suggests monitoring mussel coverage could be sufficient for the purpose of assessing mussel bed condition. Lower numbers of replicates are needed to detect the same change in % cover compared to mussel abundance indicating monitoring of % coverage is the most costeffective method.
- Field trials are needed to generate additional data that could validate the use of % as a stand-alone indicator for mussel density.
- The MarinX ('Dutch wand') survey method is recommended as a straight forward and statistically robust monitoring method.
- Parallel monitoring of potential impacts and environmental parameters is necessary to determine the cause of the observed dynamics including fishing pressure, nutrient and contaminant levels, temperature, oxygen, salinity, weather and storm events.

Recommendations for subtidal beds

- Subtidal beds can be mapped using side-scan sonar, ground-truthed with grab samples and high definition cameras to calculate biomass/abundance.
- Non destructive density estimations could be obtained from high definition photographs captured using freshwater lens drop down cameras. The methodological approach is similar to that tested for *S. spinulosa* and *M. modiolus* within this report.
- If ground-truthing reveals a significant positive correlation between mussel coverage and density, percentage cover can be used as the sole indicator of density (similar to intertidal beds).

3.2.3 Community indicators

i. Diversity and typical fauna and flora associated with *M. edulis* beds

Abundance of the associated mussel community has been selected as a potential indicator of habitat condition within the broader indicator 'condition of the typical species and communities' for biogenic reef habitats (Moffat et al 2011). One of the pre-requisites for biogenic aggregations to be considered as biogenic reefs is the development of an associated biotic assemblage which is sufficiently distinct and, usually, more diverse than those communities found in the adjacent substrata (Holt et al 1998; Seed & Suchanek 1992; Service & Magorrian 1997). Analytical approaches to monitor and define GES in biogenic reefs using community indicators include multivariate and univariate techniques aimed at characterising those communities. If these metrics and analyses have the precision to differentiate between mussel bed and adjacent non-mussel habitats it is likely that they will also be responsive to declines in diversity induced by anthropogenic pressures. For aggregations of Modiolus modiolus and, to some extent, Sabellaria spinulosa, habitat modification through the physical presence of the mussels or worm tubes results in distinctly different and rich assemblages (George & Warwick 1985; Ragnarsson & Burgos 2012; Rees et al 2008). The significant difference between bed and non-bed communities has been useful to detect and quantify the degradation caused by, for example, mobile fishing gear (Cook et al 2013; Roberts et al 2011).

a. Diversity metrics associated with *M. edulis* beds

Table 3.6 lists the range of different abundance and biodiversity metrics relevant to developing community indicators of GES for *M. edulis* beds. Overall there was a broad range of sampling techniques in the literature that could influence the variability observed in total number of species (S) and total abundance of individuals (N). Cumulative numbers of species across broad spatial scales ranged from 108 for the whole of the Wadden Sea (Dekker & Drent 2013, 2013a, 2013b) to 10 species in mussel beds in the Limfjorden (Denmark) and 13 in Wexford Harbour, Ireland (unpublished data). Diversity indices such as Shannon-Wiener's diversity (H'), Margalef's richness (d) and Pielou's evenness (J) allow for more meaningful comparisons as they are standardised to take into account sampling effort (see Appendix 3 for definitions; also Magurran 2004 and McIntyre & Eleftheriou 2005). The values of H' were relatively low, ranging from 0.67 in subtidal mussel beds on mixed sediment to 2.76 in mussel patches on hard substratum in the lower intertidal. Measurements of d were also low (1.28-6.58) but results were not common in the literature and could only be extracted from the few datasets accessed during this study. The same could be said for the estimations of community evenness, which suggest these assemblages, although more diverse than those in non-mussel habitats are still dominated by a few characterising species. Compared to other biogenic reef types (e.g. those formed by *M. modiolus* or Serpula vermicularis) *M. edulis* bed communities are substantially less rich and less diverse (Chapman et al 2011; Fariñas-Franco et al 2013; Rees et al 2008). Interestingly, they compare positively to the communities associated with S. spinulosa as the range in all metrics is similar for both species (see Chapter 4). It is possible that these communities are transient stages towards a more stable community, as the datasets for both species originated from beds or reefs that are probably being impacted or recovering from past impacts.

Table 3.6 Range of macrofaunal diversity associated with *Mytilus edulis* beds reported in the literature review.

Location	Sampling method	Bed type	Ν	S _{total}	S m⁻²	H'	d	J	Source
Wales		Intertidal (across depth shore gradient)	4169	56		1.56 (upper)- 2.76 (lower)	2.53-4.51		Seed 1996
Wadden Sea	Transects; Boxcorer 100 cm ² , 10cm, 0.5mm sieve	Intertidal	125/100 cm ²	96		1.98 (centre)- 2.07 (edge)	6.35 (edge)- 6.58(centre)	1.56 (centre) - 1.59 (edge)	Dittmann 1990
(Germany)	Monthly cores (x5), 100cm ^{2,} 20cm	Intertidal		40					Asmus 1987
Lough Foyle (Ireland)	0.1m ² clearance quadrat	Intertidal		34					Briggs 1982
Baltic Sea (Sweden)	20 x 20cm clearance quadrats	Shallow subtidal	500-2500 0.4m ⁻²	22		1.37-1.65			Koivisto & Westerbom 2010
Cloggerhead (Ireland)			468	26		1.24		0.49	
Rush (Ireland)	100cm ² clearances	Intertidal	155	36		1.74(1.58-1.90 depending on mussel sizes)		0.67	O'Connor & Crowe 2007
Wadden Sea (Germany)		Intertidal	2052	19	6	0.7		0.26	Saier 2002
Ythan Estuary	Cores, 7.5cm diam, 10cm	Subtidal Intertidal (natural)	1184 40-80 (per core)	22 27	12	2.0		0.74	Ragnarsson & Raffaelli 1999
(Scotland) Wadden Sea	deep, 0.5mm sieve 0.06m ² box corer, 15cm	Intertidal (transplanted)	100-150(per core)	25					Dront & Dokkor 2013a
(Netherlands)	deep, 0.5mm sieve	Shallow Subtida		100 (24 exclusive)					Dient & Derkei 2013a
(Denmark)	0.1m ² Van Veen grab	Shallow subtidal	$4000 (\text{por } \text{m}^2)$	10-15					Dolmer <i>et a</i> 2001
Wadden Sea (Germany)	10.5cm corer (85 cm ²), 20cm deep	Shallow subtidal	reported in Buschbaum and Saier, 2001)	32		1.84		0.53	Buschbaum <i>et al</i> 2009
Wadden Sea (Netherlands)	0.06m ² box corer	Shallow subtidal		102	20.4				Drent & Dekker 2013a;
Wadden Sea (Netherlands)	0.06m ² box corer	Shallow subtidal		108					Drent & Dekker 2013b
Wadden Sea (Netherlands)	20 2dm ² samples at equal intervals along a 1km transect. Total area 0.95m ² per site	Intertidal		28-38	13-18				Beukema & Dekker 2010
Baltic Sea (Sweden)	20 x 20cm quadrats, clearance. Cores for infauna.		108,000m ⁻²	24	6-17 (18.2)				Norling & Kautsky 2008
		Shallow subtidal	14356	45 macrofauna (34 maximum at patch); 23 macroalgae					Norling & Kautsky 2007
Anglesey (Wales)	5 x 5cm clearance quadrats	Intertidal	4169	59					Lintas & Seed 1994
Firth of Tay (Scotland)	0.1m ² Van Veen grab	Subtidal	109-688 (340.8)	37	7-19	0.69-1.7 (1.19)	1.28-3.2 (2.01)	0.27-0.75 (0.49)	Extracted from Baker <i>et al</i> (2004)

b. Comparison between reef and non-reef areas

Notwithstanding the comparatively lower number of taxa, beds of *M. edulis* are also 'islands' of biodiversity' (Norling & Kautsky 2008) and the vast majority of existing research indicates the communities associated with mussel beds, particularly those on sandflats, mudflats and areas of mixed substrata, are clearly distinct and usually more diverse than those found in areas without mussels (Buschbaum et al 2009; Koivisto et al 2011; Norling & Kautsky 2008; Ragnarsson & Raffaelli 1999). Buschbaum et al (2009) found that the communities associated with epibenthic species of mytilid mussels growing on soft sediment in tidal flats around the world, including *M. edulis* beds in the German North Sea, were significantly different to those associated with the sandy areas without mussels. For M. edulis in particular the difference between mussel and non-mussel areas was highly significant (ANOSIM, R=0.997; p<0.001). The mussel beds of *M. edulis* also had significantly richer communities, 32 taxa compared to non mussel communities which only had 17 taxa. Other indices, such as Shannon-Wiener's H' and Pielou's evenness J, were much higher than those recorded in the surrounding mud, where dominance of a few opportunistic species was high. However, those observations were not replicated in endobenthic (infaunal) mussel beds; the key enhancing factor is the physical presence of the mussel, protruding from the seafloor and forming an elevated hard substratum where the larvae of sessile species can settle.

Similar studies by Dittman (1990) indicated that, as the mussels are the only hard substratum present in the extensive sandflats of the Wadden Sea, it is the epifaunal component that differentiates the assemblages developing on the mussel beds from the infaunal communities of polychaetes and burrowing bivalves found in the sandflats. Moreover, the communities developing in the sediment under the mussels were also distinct compared to the sandflats but were not necessarily richer. The mussels might develop on top of an anoxic layer of mud where only a few opportunistic species can thrive in significantly high numbers (i.e. capitellid polychaetes such as C. capitata or Mediomastus fragilis and oligochaetes Tubifex spp. and Tubificoides spp.). Compared to the relatively diverse polychaete fauna in the bioturbated and more oxygenated nearby sand, the infaunal community in the mussel bed can be relatively impoverished but clearance and mussel transplantation experiments have demonstrated the abundance of a few opportunistic infaunal species is much higher in the mussel beds than in bare sand and mud (Commito & Boncavage 1989; Ragnarsson & Raffaelli 1999). In spite of differences in the trophic group's response to the presence of mussels, abundance and richness in the biotic communities associated with mussel beds is significantly higher than those found in areas without mussels (Drent & Dekker 2013a; Norling & Kautsky 2007, 2008, Ragnarsson & Raffaelli 1999).

The positive effect mussels have on the biodiversity of benthic communities is not only limited to sandflats and mudflats, as it was also found for mussel beds on hard substratum in the Swedish Baltic Sea by Koivisto and Westerbom (2010). There, the increased habitat complexity resulting from the presence of mussels resulted in higher community diversity compared to bare rock. The cumulative effect resulting from the presence of the mussels and the accumulation of faeces and pseudofaeces facilitated the development of a community of suspension and filter feeders as well as infaunal detritivorous species. The distinction between bed and non-bed communities is, therefore, an important indicator of whether an aggregation of mussels is of conservation importance, or not.

c. Factors affecting variability in community indices for *M. edulis* beds

Koivisto & Westerbom (2010) found that, as well as a distinct separation between mussel and non-mussel communities, a significant relationship exists between the number of taxa (S), number of individuals (abundance N) and mussel biomass (respectively r=0.66; r=0.88; p<0.001). The relationship between mussel biomass and the number of taxa and number of individuals found by Koivisto and Westerborn (2010) was similar to that found by Chapman et al (2011) for Serula vermicularis and for Modiolus modiolus (Fariñas-Franco & Roberts in prep.; also this report) whereby species richness increases rapidly with the increasing biomass of the reef building organism but reaches an asymptote of ca. 20 species at 4-6g (flesh dry weight). For number of individuals, an asymptote of 2500 individuals $(0.04m^2)$ was reached at 10g of mussels. Similarly, highest values of Shannon-Wiener's H' diversity (2 -2.13) were recorded at medium density (no equivalence to mussel m^{-2} given) mussel patches. Norling and Kautsky (2007) found that the significantly positive relationship between mussel patch size and number of macrofaunal species reached an asymptote of ca. 30 species at ~250cm² of mussels. These results are easily explained by the reduction in microhabitats as mussel density increases, competition and amensalism (Dittmann 1990). It could be argued that the results are specific to hard-bottom mussel biotopes, however, Norling & Kautsky (2008) carried out a similar experiment on mussel beds on sandflats and mudlflats (generally the type of mussel bed that fits the definition of biogenic reef) where they also found a significantly positive relationship between patch size and species richness. On this occasion, the asymptote was at 15 species, which was reached at mussel coverages of 350cm².



Figure 3.30 Relationships between number of species associated with mussel beds and mussel biomass and patch size. A) Mussels on hard substratum, Sweden (Norling & Kautsky 2007); B) Mussels on hard substratum, Finland (Koivisto & Westerbom 2010); C) Mussels on mud, Sweden (Norling & Kautsky 2008); D) Relationship between abundance of macrofauna (N) and mussel biomass. Mussels on hard substratum, Finland (Koivisto & Westerbom 2010).

The presence of an asymptotic relationship between density and community indicators for *M. edulis* is important for several reasons:

- 1. Density indicators (e.g. biomass and mussel abundance) could be used on their own to establish mussel bed condition;
- 2. Community indicators (e.g. diversity and evenness indices) are responsive to physical anthropogenic impacts whereby extraction or destruction of the beds result in a reduction of the bed density or fragmentation;
- 3. It affects survey design: there is no need to collect large samples to acquire a representative snapshot of the community.

For the purpose of monitoring condition in mussel beds, it is important that spatial and temporal (seasonal) variation in the metrics in question is low so that natural changes do not confound those induced by anthropogenic impact. Within each bed or mussel patch there is usually some degree of variation, particularly along the depth gradient and from the centre of a bed towards the edges. Significant edge effects are known to occur in intertidal mussel beds (Briggs 1982; Dittmann 1990), with the number of species and diversity increasing towards the buffer zone between mussel beds and bare sandflats (see Table 3.6; also Dittman 1990). Briggs (1982) also found spatial and temporal variation within intertidal *M. edulis* beds in Ireland where, in spite of the study site having the same tidal cover, the number of taxa was significantly higher in the seaward section of the mussel bed.

The existing body of research indicates that the communities associated with mussel beds are relatively stable over time. Dittman (1990) found seasonal variability in characterising taxa in intertidal *M. edulis* beds on sandflats was more pronounced in bare substratum than in mussel beds. Similarly, Saier (2002) found tidal level (i.e. subtidal vs. intertidal) was the only significant factor explaining variability in number of species, abundance of individuals, Shannon-Wiener diversity (H') and Pielou's evenness (J). Wave exposure, tidal regime or years were not significant factors affecting the diversity of the associated communities. There were, however, annual changes in the composition of the macroinvertebrate community for both intertidal and subtidal mussel beds. Dekker and Drent (2013) identified significant decreases in mussels and native species (*Macoma balthica, Peringia ulvae* and *Heteromastus filiformis*) and increases in introduced species *Mya arenaria* and *Ensis directus*. Functionally, though, the ecosystem seemed to remain stable as groups and phyla had similar biomass in 2013 compared to 1981.

The work of Dolmer *et al* (2001), who compared experimentally dredged mussel beds with intact ones, also confirmed the significant effect mussels have on the diversity and composition of benthic communities. Species composition differed significantly between dredged and undredged controls, largely as a result of the significant decreases in species diversity and biomass in the former. Moreover, the indices (diversity and biomass) in the control communities were stable with time and did not experience significant within-site variability, unlike the assemblages in the dredged tracks. This study is particularly relevant as the results confirm both the distinctiveness of the mussel bed community and the validity of the community indicator metrics (diversity, biomass) in responding to a known anthropogenic impact while remaining stable within time and space in the absence of pressures.

d. Differences in diversity and community composition between intertidal and subtidal *M. edulis* beds

Conservation efforts in the Wadden Sea and The Wash have been traditionally geared towards intertidal mussel beds in response to dramatic declines following overexploitation and consequent effects on bird species that feed on mussels such as oystercatchers and eider ducks (Dare *et al* 2004; Laursen *et al* 2010; Nehls *et al* 2009; Wolff *et al* 2010). Subtidal beds, where they occur, are mainly ephemeral seed mussel beds and, in some areas, may be subjected to intense fishing pressure. More persistent, long-lived subtidal beds are rarer and their survival may be as a function of a combination of environmental conditions and reduced predation pressure. However, compared to intertidal beds, the conservation importance of subtidal mussels not only as larval sources but as diverse biogenic structures has been acknowledged much more recently, mainly because they could be qualifying Nature 2000 features (Drent and Dekker 2013a; Saier 2002; Wolff *et al* 2010).

Because of the effects of emersion (heat stress, desiccation, hypoxia), intertidal mussel beds are characterised by less diverse communities compared to subtidal beds (see Table 3.6 for diversity values) where more stable conditions allow for the colonization of a more diverse array of taxa across different major phyla (Drent and Dekker 2013a; Saier 2002). Subtidal beds are, however, subjected to more intense predation pressure from crabs and, more importantly, starfish (Gaymer & Himmelman 2002; Saier 2001), which control community structure (Enderlein and Wahl 2004). Even subtidal seed beds can be important for biodiversity (although they should not be considered in the context of monitoring GES due to the ephemerality). In the Irish Sea for example, Davies (2003, in Maguire 2007) found 33 taxa and 29 families including 11 species of commercial importance including a large array of predators (starfish, crabs, gastropods, lobsters and flatfish).

The epifaunal component of intertidal mussel communities is dominated by barnacles *Semibalanus balanoides* and *Elminius modestus*, periwinkles *Littorina* spp. and, occasionally, anemones *Actinia equina*, *Sagartiogeton undatus* and *Metridium senile* (Briggs 1982; Lintas & Seed 1994; Saier 2002). The green crab *Carcinus maenas* can be common (Connor *et al* 2004). The bio-accumulated and underlying sediment is colonised by a community of oligochaetes, nematodes, nemerteans and detritivorous polychaetes *Capitella capitata* and *Heteromastus filiformis* among others (Büttger *et al* 2008; Dittmann 1990; Lintas & Seed 1994; O'Connor & Crowe 2007; Ragnarsson & Raffaelli 1999). Other macroinvertebrates commonly found across the geographical distribution in Europe include polychaete *Lepidonotus clava*, chiton *Lepidochitona cinereus* and amphipods of the genus *Jaera*, the latter being ubiquitous across all datasets consulted (Briggs 1982; Büttger *et al* 2008; Dittmann 1990; Koivisto & Westerbom 2010; Mercer 2002; O'Connor & Crowe 2007; Saier 2002).

Macroinvertebrate taxa found in subtidal mussel beds that were absent from intertidal ones included hydroids *Sertularia* spp., *Hydrallmania* falcata, *Tubularia* spp.; tunicates *Dendrodoa grossularia*, *Corella paralelograma* and scavengers and predators *Buccinum undatum*, *Asterias rubens* and *Echinus esculentus* (Drent and Dekker 2013a, 2013b; Connor *et al* 2004; Saier 2002). Subtidal beds have a higher abundance and diversity of sessile epifauna (anemones *Sagartiogeton laceratum*, *Metridium senile*) as well as polychaetes *Polydora* spp., and *Nephtys* spp.; different species of Nereididae were are all more abundant in subtidal beds, probably as a result of lower predation from crabs as well as the already mentioned more stable conditions (Saier 2002). In spite of the disparity in their abundances, species found in intertidal beds are also consistently found in subtidal beds including *Jaera albifrons*, which consistently appears in all the sources reviewed in this study regardless of their location. Moreover, the few raw datasets that could be obtained (Firth of Tay

(Scotland); Wexford Harbour and Cromane Harbour in Ireland, the latter provided by Francis O'Beirn, Marine Institute) also had *Jaera albifrons* present in relatively high abundances. In the absence of sufficiently replicated quantative data it is impossible to determine if there is a positive correlation between abundances *M. edulis* and *J. albifrons* that could be used as indication of mussel bed condition. However, data reviewed as part of the present study has confirmed a significant positive relationship between mussel density and community indicators. Although it is possible that *J. albifrons* could be an indicator species for *M. edulis* beds across their distributional range, there are no advantages of using an indicator species when *M. edilus* can be directly and easily sampled instead.

Although the studies reviewed in this report suggest community indicator metrics are sensitive to anthropogenic pressures (based on the results from mussel clearance experiments), stratified sampling approaches are essential to account for their natural variability across tidal gradients and within discrete patches (i.e. edge to centre).

ii. Sampling approaches

The techniques described in Section 3.2.2 ii for capturing density indicator metrics are largely applicable in principle to community indicators, perhaps with the exception of acoustic methods. Direct extraction of indices of biodiversity using remote sensing techniques is currently difficult and the only approach is to measure parameters that are indirectly related to biodiversity such as primary productivity (Kachelriess *et al* 2014) or topographic complexity (Zawada & Brock 2009). In fact the link between topographical complexity and diversity indices is been explored to develop predictive models from bathymetric LiDAR using machine learning and other approaches (see for example Clawges *et al* 2008; Collin *et al* 2011; Pittman & Brown 2011).

Due to the limitations in accessing datasets from specifically targeted mussel beds it was not possible to determine in a quantitative, statistically meaningful way whether remote or *in situ* imagery (video and photography) can capture community indicators for *M. edulis* beds in the same way it most likely does for *M. Modiolus*, for example. Nonetheless, a qualitative review of circumstantial footage and existing datasets from destructive sampling surveys (reviewed in previous sections) suggest it is unlikely that non-destructive approaches are adequate. The reason lies mainly in the comparatively species-poor epifaunal community that characterises *M. edulis* beds (see example photographs and video captures in Figure 3.20).

The studies reviewed in previous sections indicate the infauna and crevice fauna are the main contributors towards the significant increase in biodiversity found in mussel beds when they are compared to sedimentary substrata without mussels (Drent & Dekker 2013a). The number and abundance of taxa found in the samples will largely depend on the sampling gear type and the mesh size used to sieve the samples therefore some standardization is vital for comparative analyses. For subtidal mussel beds the use of dredges with coarse meshes is suitable only if mobile fauna and large sessile epifauna are targeted for gualitative analyses (Davies et al 2001). Bottom trawls (e.g. beam, Agassiz) have been used in the Regional Environmental Characterisation (REC) surveys (CEFAS 2009) with the aim of ground-truthing acoustic maps for broad-scale biotope characterisation. Trawls provide cumulative samples collected over transects, which is difficult to analyse statistically. Moreover, they are very destructive and not recommended for monitoring biogenic reef habitats. However, with the exception of studies where meiofauna were targeted (e.g. Dittman 1990), the most common approach to sample intertidal and subtidal M. edulis beds involves the collection of replicates using destructive methods. For intertidal beds this usually involves the use of cores driven into the mussel bed or the removal of the mussels

and underlying sediment from quadrats of standardised dimension. The same approach can also be adapted to sample subtidal beds using divers. The use of divers is normally recommended as part of small scale experiments or to survey small mussel beds. The technique that provides the highest replication covering the largest areas are grabs and boxcorers deployed from survey vessels (Dekker & Drent 2013; Dolmer *et al* 1999; Drent & Dekker 2013a 2013b).

Overall the monitoring approach for community indicator metrics should follow the same principles as that for density indicators and, in fact, a joint density/community indicator survey design is recommended. Because these surveys are targeted at a specific biotope (*M. edulis* beds in this case) a random stratified design is preferable. Therefore, the first step involves undertaking a preliminary survey to define the location and extent of the beds following the methodological approaches recommended in Section 3.2.2 ii. For intertidal beds this can involve aerial photography (see Turnbull & Davies 2001) or side-scan sonar at high tide while ground-truthed side-scan acoustic mapping is the most accurate tested method for subtidal mussel beds. Once the beds are defined it is possible to combine the monitoring of density and community indicators into one survey by retaining the samples obtained during a walk survey (e.g. using the recommended MarinX approach) and undertake macroinvertebrate faunal analyses during the sample processing phase. The CORE methods described in Dalkin & Barnett 2001) should be taken into account to ensure standardisation of the procedure.

For subtidal mussel beds, once the beds are defined, a gridded, stratified sampling design covering the whole extent of the bed is recommended. At each station 3 replicates using a Day grab / long-armed Van Veen grab (Andy Woolmer, pers. comm.; see also http://www.museumwales.ac.uk/en/1573/)/boxcorer should be taken. If the mussel bed is small or deemed too shallow, alternative sampling could involve collecting removal quadrats or cores by divers. The level of replication should be determined following a baseline survey to extract estimates of the mean and standard deviations for community indices. In shallow subtidal beds where the use of boats might be precluded, collection of core samples by divers is recommended. If the mussel bed is found in hard substratum, clearance quadrats are an adequate alternative. However, for the purpose of monitoring community indicators of GES, monitoring should be limited to *M. edulis* beds that qualify as biogenic reefs, i.e. they are host to a rich biotic community structured by the presence of the mussel which is distinct to that found in the adjancent substrata. The references consulted suggest those criteria are met only mussel biotopes found on sediment (e.g. LS.LBR.LMus.Myt.Mx; SS.SBR.SMus.MytSS). Nonetheless, additional data from UK mussel beds on rock are needed to help decide where the focus for Descriptor 1 monitoring should be.

Mussel beds with high size-class heterogeneity are more diverse than those dominated by similar size classes (O'Connor & Crowe 2007). It is, therefore, possible that population structure (i.e. size frequency distribution data) could be a good indicator of community condition for subtidal and intertidal *M. edulis* reefs should a relationship between diversity indices and size-class heterogeneity be established.

Box 3.3. Summary of findings for *M. edulis* bed community indicators.

- All studies reviewed confirm the presence of significant differences in diversity and community composition between mussel beds found on sediment and the adjacent mussel-free substrata;
- Community indices in *M. edulis* can be naturally low, even lower than surrounding communities if these are associated with hard or coarse substrata. It might be difficult to detect anthropogenic impacts on these assemblages based on the proposed community indicators;
- There is a significant positive relationship between sample size and community diversity indices across the distribution range; the relationship is asymptotic;
- Diversity indices are stable across time and between locations. Variability is significant across shore gradients and edge effects are important;
- Subtidal and intertidal communities are significantly different in composition and diversity. Subtidal mussel beds are more diverse and have a richer epifaunal and polychaete community; intertidal beds are dominated by littorinids and barnacles.
- The amphipod *Jaera albifrons* is ubiquitous in intertidal and subtidal beds across the distribution range; further consideration should be given to its use as a proxy for community indices.

Box 3.4. Summary of recommendations for *M. edulis* bed community indicators.

- The recommended survey method for community indicator monitoring involves collating aerial photographs for intertidal beds or side-scan sonar acoustic maps for subtidal beds to map the location and extent of each bed. The side-scan sonar maps need to be ground-truthed using grab samples;
- The MarinX walk survey method should be adapted to incorporate taxonomic analyses of pooled core samples. The cores will provide both mussel density and community metrics. Standard sampling protocols for intertidal sediments (CORE) can also be followed to ensure consistency across the different nature conservation agencies (see Dalkin & Barnett 2001).
- For subtidal beds, Day, long-armed Van Veen or small (0.1m²) Hamon grabs are recommended to obtain quantitative samples. In shallow subtidal beds, cores operated by divers are preferred. The biotope '*Mytilus edulis* beds on reduced salinity infralittoral rock' should not be included for the purpose of MSFD GES indicator monitoring;
- Power analysis is necessary to determine if the level of replication is adequate to detect 40% change in each community index. The results should inform the design of subsequent surveys;
- Indices calculated should, as a minimum, include Shannon-Wiener diversity (H'), total abundance of individuals (N) and Pielou's evenness (J). The relationship between community and density indicators should be investigated when data becomes available;
- Multivariate analyses using Bray-Curtis similarity matrices will provide information on species dominance and temporal shifts in the community that could identify impacts;
- Univariate analyses using either PERMANOVA or generalized linear mixed models (GLMMs) are recommended to determine spatial and temporal variability in community indicator metrics.

Box 3.5. Final recommendations for *M. edulis* bed density and community indicators.

- A combined density and community monitoring methodology based on the MarinX Dutch approach is recommended. Aerial photographs should be used to locate and map intertidal beds, although side scan sonar is preferable if possible.
- Field trials are needed at a suitable scale (e.g. discrete mussel beds) to examine variation in community composition and biodiversity metrics in UK *M. edulis* beds. These trials will need to evaluate whether these indices are responsive to pressures and how they might be measured within an existing survey protocol.
- There is a substantial gap in information for density and community indicators for subtidal mussel beds. Further field trials are needed to determine the relationship between both indicators and to assess the suitability of remote imaging methodologies to detect, map and capture potential density and community indicators. Size frequency monitoring should also be further investigated to determine its potential as an indicator of mussel community condition.

3.3 Validation of *Mytilus edulis* bed indicators

Although limited analysis of available raw data was possible, this section aimed to be a systematic literature review of existing methodologies for stock and biodiversity assessments of *M. edulis* reefs in the UK and elsewhere. The rationale was that by describing existing survey methodologies, the natural range of potential indicator metrics and impact case studies, a decision might be made about which of these are more likely to provide the information necessary to monitor UK *M. edulis* bed indicators in order to determine GES.

The studies reviewed in this report indicate that, for *M. edulis* beds, metrics of density and community indicators are responsive to change. Community indicators could be validated based on experimental exercises with appropriate controls which indicate:

- 1. A clear distinction between communities and biodiversity of reef and non-reef communities;
- 2. A clear relationship between community and density indicators;
- 3. A significant response of community indicators to impacts that can be separated from natural background variability.

Density indicators, which included mussel biomass m⁻², mussel abundance m⁻² and percentage cover of mussels, are more difficult to validate. They consistently show response to temporal and spatial change but with a high degree of random variability for the latter. Although percentage cover was the most stable of all three indicators, it is not possible to determine if these fluctuations are the result of natural variability, a direct effect of the ongoing fishery in The Wash and Solway Firth populations, or a combination of both factors. However, the results from most case studies reviewed as part of this project indicate mussel density and community indicator metrics are responsive to anthropogenic pressures.

3.3.1 Sensitivity of Mytilus edulis to anthropogenic disturbance

i. Removal of target species: overfishing

Some degree of fishing does not necessarily have negative effects on mussel stocks and, in fact, can result in increases in biomass and expansion of the beds resulting from the opening of settlement spaces and a reduction in inter-specific competition (Laursen *et al* 2010;

Solenvo 2004). Nevertheless, most studies suggest that overfishing is the driving force in the declines recorded in intertidal mussel beds in the Wadden Sea and The Wash (for example Dolmer *et al* 1999 reports a significant correlation between size of standing stocks and landings of mussels in Denmark). In the Wadden Sea significant reductions in number of species along a 10-year time series were linked to intense fishing events (Beukema & Dekker 2010). While fishermen suggested the virtual disappearance of the intertidal mussel beds in the Wadden Sea during the 1990s was the result of annually intense storm events, Ens (2006) provided strong evidence that the fishery was the major factor influencing the decline based on: 1) the existence of substantial stocks immediately after the storms; and 2) the recovery of the beds once the fishing pressure was removed. It should be noted that the Wadden Sea intertidal fishery was a vessel fishery employing vessels with a large fishing capacity where UK fisheries are, in the main, small-scale hand-gathered fisheries and, where dredging does occur, it is usually targeting ephemeral beds. Nevertheless, hand-gathered fisheries can exert significant pressures on intertidal beds and in some cases reduce both density and spatial extent (Andy Woolmer, pers. obs.).

There is, however, an indication that density and extent of fished beds can increase while that of unimpacted beds decline (Jessop *et al* 2012; Laursen *et al* 2010; Wolff *et al* 2010). Density and percentage cover depend on the balance between mortality and recruitment which are the result of weather events, hydrodynamic conditions, predation, and intraspecific competition, among other factors. Overall, more transient populations are found on mobile sediments and in dynamic environments such as estuaries. In sheltered areas, mussel beds can persist for a long time and fragmentation of areal coverage is rapidly recolonised (McKay & Fowler 1997) under the right conditions. The existing body of research clearly indicates that, in soft and hard substrata, the presence of mussels enhances the biodiversity of the benthic communities; mussel beds are significantly more diverse and richer (in terms of abundance of individuals) than nearby areas without mussels (Ragnarsson & Raffaelli 1999 and references therein).

Moreover, experimental removal of *M. edulis* either to simulate depletion or dredging consistently results in an impoverished community compared to unimpacted controls (Dolmer et al 1999; Mercer 2002; Ragnarsson & Raffaelli 1999). For example, the results of the Latin squares design experiments that Mercer (2002) used in the Burry Inlet indicate that dredging to remove smothering mussel crumble from important cockle beds significantly alters the mussel crumble communities, reducing the numerical abundance and taxonomic diversity in the short term. This reduction in diversity was statistically significant one month after the impact. There was also an increase in coarseness of the sediment following mussel removal probably due to winnowing of mussel mud. The metrics used responded to physical impact showing significant decreases in abundance of individuals and taxonomic richness. The index also shows temporal variations in controls indicating a degree of natural temporal change. Although some seasonal declines were significant for some groups such as nematodes these decreases lead to an overall more abundant community if compared to the dredged plots. Multivariate SIMPER analyses showed a clear effect of dredging which was most pronounced one month after the impact. Limited but significant differences between post impact and one month after impact were also found. A long-term study would have provided information on recovery and recruitment at this site.

Due to their high filtration capacity mussels can contribute to enhanced water quality. One of the consequences of overfishing could be an increase in pollutants and eutrophication, particularly in shallow estuaries with poor water exchange (Beukema & Dekker 2010; Coen & Grizzle 2007; Dolmer *et al* 1999; Maguire *et al* 2007).

ii. Pollution and other chemical changes

Mytilus edulis are bio-accumulators due to their filter-feeding habits but the species is relatively resilient to pollution (Roberts 1976). However, displacement of mussel communities or decreases in mussel density as a result of sewage pollution have been reported (Vallarino & Elias 1997; Vallarino *et al* 2002). Community structure may be affected by sewerage pollution rather than the mussels themselves; higher diversity has been recorded in mussel communities close to sewage effluents compared to those further away (Vallarino *et al* 2002). Organic enrichment resulting from sewage or agriculture input can result in phytoplankton blooms which in turn can lead to oxygen depletion events resulting in mass mussel mortalities (Domer 1999; de Jong 1999, see Section 3.3.1 iii). Nonetheless, as Holt *et al* (1998) notes, it is not recommended to make generalisations about the sensitivity of mussels to the wide range of potential pollutants.

iii. Deoxygenation

Although the removal of mussels can augment its negative effects, eutrophication of coastal waters and estuaries largely occurs as a result of diffuse and sewage pollution and agricultural run-off increasing levels of nitrates and phosphates. These nutrients promote algal blooms and the growth of macroalgae. The sudden breakdown of algae and seaweeds has resulted in episodes of hypoxia over mussel beds, which in turn can lead to episodes of hypoxia. Mass mortalities of *M. edulis* resulting from such episodes have been recorded in Limfjorden in the past (Dolmer *et al* 1999).

iv. Smothering (siltation)

Excessive accumulation of sediment can be detrimental to *M. edulis* (Seed & Suchanek 1992). Experiments by Last *et al* (2011) involving Vortex Re-suspension Tanks (pVoRTS) found that *M. edulis* is relatively tolerant to short-term and repeated burial with mortality increasing with increases in percentage of re-suspended finer sediments. However, qualitative photographic surveys carried out by divers in Irish mussel beds close to dredged navigational channels found no apparent smothering or stress caused to nearby mussels following channel dredging operations (Brendan O'Connor, pers. comm.). Estuaries with dynamic river channels may experience changes in hydrodynamic and depositional regime which in turn may result in erosion or smothering of mussel beds. This occurs in the Burry Inlet and Three Rivers Estuary (Andy Woolmer, pers. obs.).

v. Physical damage: abrasion

Abrasion can be caused by anthropogenic pressure by the use of trawls, dredges or pots or by natural events such as ice scouring. In fact, ice coverage is the most important regulating factor for the development of mussel beds in the Danish Wadden Sea according to the Trilateral Monitoring Program (TMAP) report (Wadden Sea Ecosystem 1997). Abrasion caused from bottom trawls and dredges may impact sublittoral beds resulting in impoverished epibenthic communities (Cook *et al* 2013; Sanchez *et al* 2000; Thrush & Dayton 2002). For mussel beds, the studies of Mercer (2002) and Dolmer *et al* (1999) both concluded that physical abrasion had negative effects on the structure of the faunal assemblage associated with the mussels. Dredging affected the sediment structure, reduced diversity and biomass, changed the structure of the community and attracted predators (Dolmer *et al* 1999).

vi. Introduction or spread of non-indigenous species

The introduction of *Crassotrea gigas* in the Wadden Sea is thought to have resulted in a displacement of the mussel beds through competition (Reise 1998; Troost 2010). However, recent reports suggest that newly developed oyster reefs might be enhancing natural recruitment of *M. edulis* and therefore contributing to the re-colonisation of new areas (Karin Troost, pers. comm.) The slipper limpet *Crepidula fornicata* is present on intertidal and subtidal beds around the south coast of the UK. In sheltered areas where they thrive there is a risk of smothering and competition for space and food.

vii. Introduction of microbial pathogens

The Eastern IFCA has consistently recorded high levels of infestation by the parasitic copepod *Mytilicola intestinalis* in The Wash mussel population (Jessop & Maxwell 2011; Jessop *et al* 2010, 2012). The influence of *M. intestinalis* on the mortality of young (< 3 years) mussels in The Wash, observed by Jessop (pers. comm.), merits further investigation because there are reports of mass mussel mortalities elsewhere which could be potentally linked to this parasite (Holt *et al* 1998). Other parasites associated with *M. edulis* include trematodes and the shell boring polychaete *Polydora ciliata*.

Table 3.7 Summary of the known effects and magnitude of change in mussel density and in the biodiversity of communities associated with *Mytilus edulis* reefs in response to environmental change based on a review of the literature and field observations.

Impact /	M. edulis Density		Associated Reef Community			
Pressure	Anticipated Change	Magnitude of Change	Anticipated Change	Magnitude of Change		
Changes in suspended solids (water clarity): increased turbidity	Physiological stress, low recruitment, smothering leading might lead to increased mortality.	Unknown	Impacts on the fauna associated with <i>M. edulis</i> reefs will be species specific with some species having similar tolerance levels to <i>M. edulis</i> and others less tolerant. A reduction in diversity is therefore likely.	Unknown		
Changes in suspended solids (water clarity): decreased turbidity	Reduced feeding rate. Energies diverted to somatic growth. No reproductive growth.	Unknown	Impacts on <i>M. edulis</i> may be reflected in the associated fauna. Could cause changes in the balance of deposit and suspension feeders (hydroids, ophiuroids) associated with the reef.	Unknown		
Siltation rate changes including smothering	It could affect physiological condition and reproduction at high siltation rates.	Unknown	Likely to cause complete or near complete die-off	Unknown		
Physical Disturbance damage: selective extraction and abrasion	Causes reductions in density and extent. Recovery likely but depends on larval sources and connectivity. Areas of reef will become more patchy with increasing physical impact. Increase eutrophication can lead to de-oxygenation and die-offs. Allee effects, genetic depression.	80-100% decrease as a result of direct destruction and ensuing cascading effects Reduction from 100% cover to 0-20% cover (Fragmentation) Loss of elevation (100%) Moderate extraction can lead to increased settlement in cleared areas leading to biomass increases and bed extent.	Reduces complexity and diversity. Increases dominance by opportunistic species and predators. Increased number of soft-bottom species.	50-90% declines in number of species and abundance. Declines in Shannon diversity to ~1.0. Increase dominance of opportunistic species. Evenness might increase as a result of reduction in dominant infauna.		
Pollution and other chemical changes	<i>M. edulis</i> is a bioacumulator. Pollution could affect physiological condition and reproduction.	Unknown	Potential increases in already abundant capitellids, nematodes and oligochaetes. Expected decreases in biodiversity but the opposite effect could also be observed.	Unknown		
4. The development of Descriptor 1 (Biological Diversity) indicators for *Sabellaria spinulosa* reefs

4.1 Introduction

4.1.1 Legislation relevant to Sabellaria spinulosa reefs

Sabellaria spinulosa reefs have been identified as a priority habitat for conservation in European and national legislation as summarised in Table 4.1. It should be noted that all such legislation applies to the habitat created by *S. spinulosa* and not the species itself. Where statutory duties and ministerial commitments exist towards the active management of *S. spinulosa* habitats, either explicitly or implicitly, some form of monitoring and / or assessment is undertaken.

Table 4.1 Table summarising the legislative instruments used to protect Sabellaria spinulosa reefs in the UK.

Legislative Instrument	Mechanism for Protection
European Habitats Directive 1992	Special Areas of Conservation (SACs)
OSPAR Convention 1992	OSPAR Marine Protected Areas (MPAs)
Nature Conservation (Scotland) Act 2004	Scotland's Biodiversity Strategy
Natural Environment and Rural Communities Act 2006	England's Biodiversity Strategy Environment Strategy for Wales
Wildlife and Natural Environment Act (Northern Ireland) 2011	Northern Ireland's Biodiversity Strategy
Marine Strategy Framework Directive 2008	"Good Environmental Status" targets
Marine and Coastal Act 2009	Marine Conservation Zones (MCZ)
Marine (Scotland) Act 2010	Nature Conservation Marine Protected Areas (MPAs)

i. European Habitats Directive (92/43/EEC)

Sabellaria spinulosa reefs qualify under Annex I of the Habitats Directive where they are a type of "reef" protected by a network of Special Areas of Conservation (SACs). The Interpretation Manual of European Union Habitats (Anon 2007) specifically lists *Sabellaria spinulosa* reefs of the sublittoral North Sea "*Sabellaria*-Riff des Sublittorals der Nordsee", though they may also be protected by virtue of their occurrence in broader physiographic habitats listed under the Directive such as "Estuaries" and "Large Shallow Inlets and Bays". In the UK the presence of well-developed and stable *S. spinulosa* reefs was one of the primary reasons considered for the designation of 'The Wash and North Norfolk Coast' SAC (UK0017075). Here the reefs are both an Annex I habitat in their own right and part of the broader 'Large Shallow Inlets and Bays' habitat. More recently, an additional three UK sites were put forward to the EU Commission for the protection of *S. spinulosa* reefs (Tranche 38, August 2010): Inner Dowsing, Race Bank and North Ridge (UK0030370); North Norfolk Sandbanks and Saturn Reef (UK0030358); and Haisborough, Hammond and Winterton (UK0030369). All have been approved by the European Commission as Sites of Community Importance (SCI) (http://jncc.defra.gov.uk/page-1488).

ii. OSPAR Convention

Sabellaria spinulosa reefs were added to the "OSPAR list of threatened and / or declining habitats" based on the Texel-Faial criteria for identification of species and habitats in need of protection. Sensitivity, rarity, ecological significance and decline were cited as reasons for its inclusion with information also provided on threat (OSPAR 2008). The list is used as one of the criteria to designate MPAs in the UK (Cork *et al* 2006).

iii. Country Biodiversity Strategies

In England and Wales *S. spinulosa* reefs are listed as Habitats of Principal Importance (HPI) under Section 41 of the Natural Environment and Rural Communities (NERC) Act 2006. In Northern Ireland a *S. spinulosa* Habitat Action Plan (HAP) is currently under implementation (DOENI 2005). The Scottish Biodiversity List was published to satisfy Section 2(4) of The Nature Conservation (Scotland) Act 2004, however, the list does not include *S. spinulosa* habitats.

iv. Marine Strategy Framework Directive (2008/56/EC)

Biogenic reefs formed by *S. spinulosa* are suitable Good Environmental Status (GES) targets for Descriptors 1 (Biological diversity) and 6 (Seafloor integrity) under the MSFD (Cochrane *et al* 2010). As *S. spinulosa* reefs are identified under Community (European Habitats Directive) and International (OSPAR) legislation they are considered a 'special' habitat as defined in Table 1 of Annex III of the MSFD.

v. Marine and Coastal Access Act (2009)

The Marine and Coastal Access Act 2009 received royal assent on 12 November 2009 and introduced a new framework for managing the many demands placed on the sea, improving marine conservation and opening up access for the public to the English coast. Provisions are made in Part 5 of the Act for designation and protection through a new type of MPA, called Marine Conservation Zones (MCZs). MCZs will exist alongside European Marine Sites (SACs and SPAs) to form a MPA network. *S. spinulosa* reef is identified as a priority habitat for protection in the "Ecological Network Guidance" both as the Broad Scale Habitat, Subtidal biogenic reefs A5.6 and as the Habitat Feature of Conservation Importance (FOCI) Ross worm (*Sabellaria spinulosa*) reefs.

vi. Marine (Scotland) Act 2010

S. spinulosa reefs are not specifically listed as an MPA Search Feature or a Priority Marine Feature (PMF) (Tyler-Walters *et al* 2012; <u>http://www.snh.gov.uk/docs/B1140045.pdf</u>).

4.1.2 Sabellaria spinulosa reef in the context of the MSFD

Sabellaria spinulosa is a gregarious polychaete which is found in numerous different growth forms ranging from solitary individuals and small clumps to low level veneers and reef. The boundary between the different growth types, their ecological significance and the conditions which lead to the development of one growth type to another remains largely unknown. *S. spinulosa* reefs fall under the broad "reef" definition provided by the Habitats Directive as well as the more specific *S. spinulosa* reef definition provided by the OSPAR list of threatened and declining species as detailed in Box 4.1. These habitat definitions are somewhat ambiguous and can be difficult to apply to standard survey data. In recent years there have been a number of attempts to further refine the parameters by which a *S.*

spinulosa reef can be distinguished from other growth forms, most notably Gubbay (2007) and Hendrick & Foster-Smith (2006).

Both of the aforementioned reef definitions rely primarily on physical reef attributes such as elevation from the seabed, extent, consolidation and patchiness, however, there has been no research to date to conclusively link these attributes with reef functioning and health. Pearce *et al* (2011) investigated the influence that reef elevation, extent and patchiness had on macrobenthic communities found in association with the *S. spinulosa* in the North Sea, but found no link between any of these reef attributes and the associated fauna. The results of this study indicate that there is no clear link between the function of an *S. spinulosa* habitat and its extent or shape.

A small number of studies have shown that the diversity of fauna associated with newly developed areas of Sabellariid reefs can support a higher diversity of macrofauna than older, more consolidated reefs as illustrated in Figure 4.1 (see also Dubois *et al* 2002 and Pearce *et al* 2007). Indeed as Sabellariid reefs develop they can become dominated by species such as the porcelain crab *Pisidia longicornis* and the consolidation of the reef can exclude species that might otherwise inhabit sediment accumulations forming in the spaces between aggregations of worms, therefore reducing overall macrofaunal biodiversity.

Box 4.1 Definitions of or encompassing S. spinulosa reef

Definition of 'reefs' from the revised Habitats Directive Interpretation Manual (CEC 2007)

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

Definition of S. spinulosa reef from the OSPAR list of Threatened and Declining Species (OSPAR Commission 2008)

"The tube-building polychaete Sabellaria spinulosa can form dense aggregations on mixed substrata and on rocky habitats. In mixed substrata habitats, comprised variously of sand, gravel, pebble and cobble, the Sabellaria covers 30% or more of the substrata and needs to be sufficiently thick and persistent to support an associated epibiota community which is distinct from surrounding habitats. On rocky habitats of bedrock, boulder and cobble, the Sabellaria covers 50% or more of the rock and may form a crust or be thicker in structure. In some areas, these two variations of reef type may grade into each other. Sabellaria reefs have been recorded in depths between 10-50m Below Chart Datum (BCD) or more. The reef infauna typically comprises polychaete species such as Protodorvillea kefersteini, Scoloplos armiger, Harmothoe spp., Mediomastus fragilis, Lanice conchilega and cirratulids together with the bivalves Abra alba and Nucula spp. and tube-building amphipods such as Ampelisca spp. Epifauna comprise calcareous tubeworms, pycnogonids, hermit crabs, amphipods, hydroids, bryozoans, sponges and ascidians. S. spinulosa reefs are often found in areas with quite high levels of natural sediment disturbance; in some areas of reef, individual clumps of Sabellaria may periodically breakdown and rebuild following storm events. S. spinulosa reefs have been recorded from all European coasts except the Baltic Sea, Skagerrak and Kattegat. Areas of dead Sabellaria reef indicate the site supported reef habitat in the past and should be reported as this habitat type."

Ohanastanistis		'Reefiness'					
Characteristic	NOT a reef	Low	Medium	High			
Elevation (cm) (average tube height)	<2	2-5	5-10	>10			
Extent (m ²)	<25	25-10,000	10,000-1,000,000	>1,000,000			
Patchiness (% cover)	<10	10-20	20-30	>30			

Threshold ranges of reef characteristics proposed by workshop participants (Gubbay 2007).



Figure 4.1 Schematic showing the development of *Sabellaria spinulosa* reefs at Hastings Shingle Bank (reproduced from Pearce *et al* 2007).

Since most reefs are likely to contain a number of different developmental stages and the different stages are likely to have some intrinsic value to reef function, all stages of reef development have been included in the present study. The extent of individual *S. spinulosa* reefs has been delineated using high resolution remote sensing techniques (side-scan sonar and multibeam bathymetry) in a small number of the studies used in this analysis. Unfortunately though this information was not available for the majority of the datasets used and so all of our subsequent analysis has been carried out using the presence of *S. spinulosa* as a proxy for reef presence as well as different density categories and areas identified as reef using high resolution acoustic data, where this information was available.

4.1.3 Sabellaria spinulosa biotopes

Within the Marine Habitat Classification for Britain and Ireland Version 04.05 (Connor *et al* 2004) and the European Nature Information System (EUNIS) classification schemes there are four biotopes in which *S. spinulosa* is noted as being abundant or common (according to the SACFOR scale (Connor & Hiscock 1996) as summarised below in Table 4.2. Two of these biotopes, "*Sabellaria spinulosa* on stable circalittoral mixed sediment" and "*Sabellaria spinulosa* encrusted circalittoral rock" are considered to be equivalent to the OSPAR threatened and declining habitat "*Sabellaria spinulosa* reef" whereas the other two are not. However, it should be noted that there is no measure of extent, elevation or any indication of longevity associated with these classifications. Both the reef and non-reef *S. spinulosa* biotopes are frequently applied to isolated point sample data where the community is dominated by *S. spinulosa*. However, these conclusions should be treated with some caution because they may all have been taken from a spatio-temporal mosaic which, overall, would qualify as a reef.

Because *S. spinulosa* biotopes have not been applied to sample data consistently, a comparison between them was not possible within the scope of this study. These biotope classifications were used however to narrow down the search for seabed images containing *S. spinulosa* aggregations as explained in more detail in Section 4.2.

Table 4.2 Summary of the four main biotopes (Connor *et al* 2004; European Environment Agency 2010) in which *Sabellaria spinulosa* are noted as being common or abundant (using the SACFOR scale (Connor & Hiscock 1996). Whether or not these biotopes are considered to be equivalent to the OSPAR threatened and declining habitat "*Sabellaria spinulosa* reef" is also noted.

Biotope Name	Biotope Code	S <i>. spinulosa</i> Reef	Biotope Description
Sabellaria spinulosa on stable circalittoral mixed sediment	SS.SBR.PoR. SspiMx EUNIS: A5.611	Yes	Sabellaria spinulosa at high abundances on mixed sediment. Sabellaria typically forms loose agglomerations of tubes forming a low lying matrix of sand, gravel, mud and tubes on the seabed. The infauna comprises typical sublittoral polychaete species such as <i>Protodorvillea kefersteini</i> , <i>Pholoe synophthalmica, Harmothoe</i> spp, <i>Scoloplos armiger, Mediomastus fragilis, Lanice conchilega</i> and cirratulids, together with the bivalve <i>Abra alba</i> , and tube building amphipods such as <i>Ampelisca</i> spp. The epifauna comprise a variety of bryozoans including <i>Flustra foliacea, Alcyonidium diaphanum</i> and <i>Cellepora pumicosa</i> , in addition to calcareous tubeworms, pycnogonids, hermit
Sabellaria spinulosa encrusted circalittoral rock	CR.MCR. CSab.Sspi EUNIS: A4.221	Yes	Biotope with an almost entire crust of <i>Sabellaria spinulosa</i> tubes typically found encrusting the upper faces of wave-exposed and moderately wave exposed circalittoral bedrock, boulders and cobbles subject to strong and moderately strong tidal streams in areas with high turbidity. A diverse fauna may be found attached to the crust. There are two variants: The first (Sspi.ByB) contains turfs of bryozoans (including <i>F.foliacea, A. diaphanum</i> and <i>Bugula plumosa</i>); other scour tolerant species such as <i>Urticina felina, Tubularia indivisa</i> and <i>Nemertesia antennina</i> may also be present. The second variant (Sspi.As) has a dense turf of didemnid ascidians and scour-tolerant bryozoans including <i>F. foliacea</i> and <i>Cellaria</i> species. Sparse sponges and patchy occurrences of small ascidians such as <i>Polycarpa</i> spp. may also be observed. The fauna attached to the <i>Sabellaria</i> crust in many cases seem to reflect the biotopes on nearby rock.
Sabellaria spinulosa with kelp and red seaweeds on sand- influenced infralittoral rock	IR.MIR.KR. Lhyp.Sab EUNIS: A3.2145	No	Laminaria hyperborea kelp forest on shallow infralittoral bedrock and boulders characterised by encrustations of <i>S. spinulosa</i> tubes which cover much of the rock, together with sand-tolerant red seaweeds. Some of the richer examples of this biotope also have a rich fauna of ascidians, sponges, hydroids and bryozoans. A similar biotope is also found in the circalittoral zone, where it lacks the algal component (CR.MCR.CSab.Sspi).
<i>Laminaria digitata</i> and piddocks on sublittoral fringe soft rock	IR.MIR.KR. Ldig.Pid EUNIS: A3.2113	No	Soft rock, such as chalk, in the sublittoral fringe characterised by <i>Laminaria digitata</i> and rock-boring animals such as piddocks <i>Barnea candida</i> and <i>Pholas dactylus</i> , the bivalve <i>Hiatella arctica</i> and worms <i>Polydora</i> spp. <i>S. spinulosa</i> often colonises empty piddock burrows. Beneath the kelp forest, a wide variety of foliose and filamentous red seaweeds occurs together with bryozoans and hydroids. The undersides of small chalk boulders are colonised by encrusting bryozoans, colonial ascidians and the tube-building polychaete <i>Pomatoceros lamarcki</i> .

4.2 Evidence base

Data collected from Sabellaria spinulosa reefs for research and Environmental Impact Assessment (EIA) purposes were collated from a variety of private and public sector sources as outlined in Appendix 1.2. All data sources known to the authors of this work were considered for use in this study and the MarLIN and Marine Recorder databases were also queried to ensure that no datasets were missed. Unfortunately not all S. spinulosa datasets were made available to the project and not all that were made available were suitable for inclusion. For example some macrobenthic data analysis did not use standard enumeration methods or could not be made available in electronic format. Similarly, some seabed imagery that was made available for use in this study was only available at a very low resolution (e.g. embedded in a report) or was of insufficient quality to provide accurate species identifications and tube counts. Issues pertaining to seabed image quality are explored in more detail in Section 4.3.2 and a full list of S. spinulosa datasets that were identified is provided in Appendix 1.2 along with any limitations encountered. It is likely that with additional time and resources the evidence base used for the development S. spinulosa biodiversity indicators could be improved significantly and this is discussed further in Section 4.8.

Datasets containing both quantitative grab sample data and good quality seabed images were prioritised in data collection efforts as were datasets where *S. spinulosa* reef habitats had been the specific target for some or all of the sampling. Studies in which attempts had been made to map the extent of *S. spinulosa* habitats using acoustic mapping techniques, in addition to undertaking direct sampling, were also targeted for use in this study. There is a wealth of data available from the private sector collected as part of the licensing process, however, *S. spinulosa* habitat records of this nature tended to be sporadic and incidental. For many years, Statutory Nature Conservation Bodies (SNCBs) in the UK have advised EIA consultants not to collect quantitative samples from these sensitive habitats, due to the perceived negative impact caused by destructive sampling. The consequences of this advice are that there are only a small number of cases where *S. spinulosa* reefs have been quantitatively sampled. The evidence base available for Descriptor 1 biodiversity indicator development may therefore be lacking in some areas.

The datasets that ultimately made up the evidence base for the Descriptor 1 biodiversity indicators for *S. spinulosa* reefs are summarised below in Table 4.3. Where these data are available in the public domain they have been provided in full in Appendix 2.2. Those datasets that have been provided under licence for use in this study only are not provided as raw data but have been used in the analysis. The distribution of the data used in this study is illustrated in Figure 4.2Table 4.3 Summary of the datasets used to inform the development of Descriptor 1 biodiversity indicators for *Sabellaria spinulosa* reefs.

Site	Year	Biological Data	Environmental Data	Remote Sensing Data	Purpose of Data Collection	Source
Hastings Shingle Bank [Aggregate Licence Areas 366- 370]	2006	0.1m ² Hamon grab samples	Particle Size Distribution analysis	High resolution side scan sonar data interpreted to provide charts illustrating the distribution and extent of <i>S. spinulosa</i> aggregations	Applied research programme investigating the recoverability of <i>S. spinulosa</i> aggregations following aggregate extraction	Marine Aggregate Levy Sustainability Fund (MALSF) research report available from: <u>http://www.cefas.defra.gov.uk/</u> <u>media/462262/mal%200027%</u> <u>20final%20report.pdf</u>
Cutline [Aggregate Licence Area 447]	2008 - 2010	0.1m ² Hamon grab samples Seabed Images (fresh- water lens camera)	Particle Size Distribution analysis	Side scan sonar data interpreted to provide charts illustrating the distribution of seabed sediments	Applied research programme investigating the impact of adjacent aggregate extraction activities on <i>Sabellaria spinulosa</i> aggregations	Marine Aggregate Levy Sustainability Fund (MALSF) research report available from: http://www.cefas.defra.gov.uk/ media/462374/mepf%2008%2 0p39%20final%20report.pdf
East Coast REC	2009	0.1m ² Hamon grab samples 2m scientific beam trawl samples Seabed Images	Particle Size Distribution analysis	High resolution multi-beam data interpreted to provide charts illustrating the distribution and extent of <i>S. spinulosa</i> aggregations	Broad-scale seabed mapping programme including more detailed surveys of <i>S. spinulosa</i> habitats where they were identified	Marine Aggregate Levy Sustainability Fund (MALSF) research report available from: <u>http://www.cefas.defra.gov.uk/</u> <u>media/469471/ec%20rec%20fil</u> nal%20report_low%20res.pdf
Humber REC	2009	0.1m ² Hamon grab samples 2m scientific beam trawl samples Seabed Images	Particle Size Distribution analysis	Side scan sonar and multi- beam data interpreted to provide charts illustrating the distribution of seabed sediments	Broad-scale seabed mapping programme	Marine Aggregate Levy Sustainability Fund (MALSF) research report available from: <u>http://www.cefas.defra.gov.uk/</u> <u>media/477115/humber_rec_fin</u> <u>al_report_lowres.pdf</u>
Thames REC	2007	0.1m ² Hamon grab samples 2m scientific beam trawl samples Seabed Images	Particle Size Distribution analysis	Side scan sonar and multi- beam data interpreted to provide charts illustrating the distribution of seabed sediments	Broad-scale seabed mapping programme	Marine Aggregate Levy Sustainability Fund (MALSF) research report available from: <u>http://www.cefas.defra.gov.uk/</u> <u>media/462472/outer%20thame</u> <u>s%20estuary%20rec%20final</u> <u>%20report.pdf</u>

Site	Year	Biological Data	Environmental Data	Remote Sensing Data	Purpose of Data Collection	Source
South Coast REC	2007	0.1m ² Hamon grab samples 2m scientific beam trawl samples Seabed Images	Particle Size Distribution analysis	Side scan sonar and multi- beam data interpreted to provide charts illustrating the distribution of seabed sediments	Broad-scale seabed mapping programme	Marine Aggregate Levy Sustainability Fund (MALSF) research report available from: http://www.cefas.defra.gov.uk/ media/462500/southcoastrec_f inal%20report%20july%2010_I ow%20res.pdf
Thanet Offshore Wind Farm	2005, 2007 and 2012	0.1m ² Hamon grab samples Seabed Images (fresh- water lens camera)	Particle Size Distribution analysis	Side scan sonar data interpreted to provide charts illustrating the distribution and extent of <i>S.</i> <i>spinulosa</i> aggregations	Baseline and pre- constructions surveys for the Thanet Offshore Wind Farm	Vattenfall – data provided under licence for use in this project. More details of the development can be found here: http://www.vattenfall.co.uk/en/t hanet-offshore-wind-farm.htm
Lincs Offshore Wind Farm	2010 2011	0.1m ² Hamon grab samples (data provided but not used) Seabed Images (fresh- water lens camera). Note images taken in 2011 did not show any evidence of <i>S</i> . <i>spinulosa</i> habitats and weren't included in the analysis		Side scan sonar data interpreted to provide charts illustrating the distribution and extent of <i>S.</i> <i>spinulosa</i> aggregations	Cable route survey for the Lincs Offshore Wind Farm	Centrica – data provided under licence for use in this project



Figure 4.2 Chart illustrating the geographical distribution of data (grabs and remote imagery) used to inform the development of Descriptor 1 biodiversity indicators (see also Table 4.3). Boxes A-D refer to maps displayed in Figure 4.3 and Figures 4.5-4.8, respectively. Map constructed using QGIS version 2.0.1-Dufour <u>www.qgis.org</u>.

4.2.1 Study sites

i. Hastings Shingle Bank Aggregate Extraction Licence Areas 366-370

The Hastings Shingle Bank dataset comes from an applied research programme investigating the recoverability of *Sabellaria spinulosa* aggregations following aggregate extraction in the eastern English Channel. The aggregations were mapped in 2005 using high resolution side-scan sonar, ground-truthed with seabed imagery and sediment grab samples. A total of 145 grab samples and co-located seabed images were collected from this site with approximately equal numbers of samples collected from within and outside acoustically defined areas of *S. spinulosa* habitat as shown overleaf in Figure 4.3. All samples were taken from an area of mixed gravel, which is the target of extractive activities in this area. High resolution side-scan sonar data was collected from this area and this was used to map the extent of the *S. spinulosa* aggregations allowing for comparisons to be made between *S. spinulosa* habitats and the adjacent sedimentary habitats.



Figure 4.3 Chart showing the distribution of co-located 0.1m² Hamon grab samples and seabed images collected across the Hastings Shingle Bank site in 2005 (black circles). Note that although seabed images were taken at the same sampling stations as the grab samples they were taken at different times during the survey and will not be exactly co-located. Precision estimated to be <10m. *S. spinulosa* aggregations (dot areas) are delineated based on high resolution side-scan sonar maps. Light brown areas denote coarse sediments. Map constructed using QGIS version 2.0.1-Dufour www.qgis.org.

ii. Regional Environmental Characterisations

The Regional Environmental Characterisation (REC) surveys were funded through the Marine Aggregate Levy Sustainability Fund (MALSF) administered by Defra, as a means of providing some regional context to local Environmental Impact Assessments (EIAs) routinely carried out for the aggregate extraction industry in the UK. Four areas were chosen due to their strategic importance to the industry: the Humber, the east and south coasts of England and the Thames (Figure 4.4). The four areas were surveyed with the aim of developing comprehensive, regional level, geophysical and environmental maps to inform sustainable resource management. *Sabellaria spinulosa* aggregations were identified in all four REC areas and as these datasets are publically available they provide an important component of the evidence base used in this study, providing a good spread of data from the southeast of the UK.

Although the REC projects had common aims, they were undertaken by different organisations and had different budgetary constraints as well as differing regional emphasis. For example, the emphasis for the South Coast REC was on mapping rock habitats unique to this region whereas more emphasis was placed on mapping *S. spinulosa* aggregations in the East Coast REC since this habitat was identified in areas where it had not previously been recorded. Acoustic data have been collected at all four sites but were only collected in corridors with the exception of the East Coast REC. Here, full coverage acoustic data were collected from areas where *S. spinulosa* aggregations were identified with the specific aim of mapping the extent of these aggregations. Additional ground truthing was also targeted in these areas.



Figure 4.4 Chart showing the location of the four Regional Environmental Characterisation (REC) Projects

Broad-scale biotope maps were created for each of the REC areas, using slightly different habitat modelling techniques. In the Thames REC area, biotopes were assigned to point locations and overlaid on a map of the physically defined habitats with no attempt to extrapolate the biological communities between points. In the East Coast REC a composite biotope model was constructed using the combined output of numerous different modelling methods, where those methods were found to be in agreement with one another. In this instance *S. spinulosa* habitats were mapped as a separate exercise using full coverage, high resolution acoustic data. In the Humber and South Coast RECs the physically defined habitats were used to identify suitable habitats for the broad community types identified in the two regions. In the latter two studies the distinction between the predicted occurrence of a community and the predicted occurrence of a habitat suitable for occupancy by a community must be made. For example a very large area of the Humber REC site is predicted to confused with the predicted distribution of the community itself.

iii. Thanet Offshore Windfarm

The Thanet Offshore Windfarm datasets were provided by the site developer, Vattenfall, for use in this study only. A baseline characterisation survey was undertaken at this site in 2005, followed by a pre-construction survey in 2007 and most recently, the first post-construction monitoring survey in 2012. *S. spinulosa* aggregations were identified at this site during the baseline characterisation surveys and these were subsequently re-surveyed and mapped using high resolution side-scan sonar and seabed imagery. Permission was granted for the development of this site on the proviso that turbines were micro-sited to avoid the best parts of the reef, and the pre-construction survey was used for this purpose. This habitat will now be monitored as part of the licence conditions attached to this development using seabed imagery. The use of extractive sampling using a sediment grab to monitor this habitat was limited by the Statutory Nature Conservation Bodies (SNCBs) and prohibited completely in the most recent survey. There is therefore only a limited amount of quantitative data from these aggregations collected before the windfarm was constructed.



Figure 4.5 Charts showing the distribution of 0.1m² Hamon grab samples and drop camera stations across the Thanet Offshore Windfarm site (see also Figure 4.2) in 2005, 2007 and 2012. Precision of co-location estimated to be <10m. *S. spinulosa* aggregations are delineated based on high resolution side-scan sonar maps (2007 and 2012 surveys). Map constructed using QGIS version 2.0.1-Dufour www.ggis.org.

iv. Cutline – Aggregate Extraction Licence Area 447

The Cutline dataset comes from an applied research programme investigating the impact of aggregate extraction on adjacent benthic communities including *Sabellaria spinulosa* aggregations. Grab samples and co-located seabed images were collected at a distance of 250m, 650m and 1000m from a new aggregate extraction site in the outer Thames estuary, in the direction of anticipated net sediment movement. A further six sampling stations were sampled in comparable sedimentary deposits outside the area of anticipated secondary impacts (Figure 4.6). All stations were sampled before aggregate extraction activities commenced in April 2008 and at seven regular intervals after aggregate extraction began between July 2008 and April 2010.

The aim of the study was to measure changes in the benthic communities and in particular the *S. spinulosa* aggregations as a new anthropogenic disturbance was introduced into an adjacent area, testing the hypothesis that increased turbidity associated with aggregate extraction could enhance *S. spinulosa* aggregations. Unfortunately the study site was not closed to other activities and the *S. spinulosa* aggregations that were recorded at this site were apparently subject to trawl damage in early stages of the study, meaning that the secondary impacts of aggregate extraction on this habitat could not be assessed. The Cutline dataset nevertheless provides useful co-located grab and seabed imagery data from *S. spinulosa* aggregations in the Thames sea area.

Although acoustic data were collected from this site as part of the licence application process, these data were not ever used to map *S. spinulosa* aggregations and it is therefore only possible to make comparisons between discrete grab samples and seabed images collected from this area.



Figure 4.6 Chart showing the distribution of co-located $0.1m^2$ Hamon grab samples and seabed images collected across the Cutline site between 2008 and 2010. Note that although seabed images were taken at the same sampling stations as the grab samples they were taken at different times during the survey and will not be exactly co-located. Precision of co-location estimated to be <10m.

v. Lincs Offshore Windfarm

The Lincs Offshore Windfarm datasets were provided by the site developer, Centrica Renewable Energy Ltd, for use in this study only. Pre-construction baseline survey data were provided to the project and these included grab sample data and seabed imagery. Unfortunately the grab sample data could only be provided as text embedded in the final project report and there were insufficient resources in this study to manually re-type this dataset, hence only the seabed images and *S. spinulosa* abundances from corresponding grab sample stations were used in this study to strengthen the density indicator analyses.



Figure 4.7 Distribution of sample locations (red spots) on part of the Lincs pre-construction baseline / baseline survey. Dotted and yellow polygons in A respectively represent medium and low *S. spinulosa* reefs derived from ground-truthed Acoustic Ground Discrimination System (AGDS) maps (data including GIS layers from EGS International Ltd 2011).

4.2.2 Remote sampling data

Photographic data available from sources listed in Table 4.3 (see also Figures 4.2 - 4.8) included data collected using grab mounted cameras (e.g. 'HamCam'), low visibility or 'water curtain' cameras (e.g. Weasel II system) and standard frame mounted drop cameras such as those employed in the East Coast and Humber REC surveys (Figure 4.9 and Table 4.4).



Figure 4.8 Map illustrating case study areas used in the assessment of tube counts as proxies for *S. spinulosa* density. Red dots represent grab sampling stations with positive records for *S. spinulosa* with co-located remote images (from drop down digital cameras).



Figure 4.9 Remote camera systems assessed to determine *S. spinulosa* density: A) 'HamCam' deployed in the East Coast REC © Cefas; B) Freshwater Curtain Camera (FCC) and C) Weasel II FCC deployed at the Cutline 447 aggregate extraction site and Thanet Offshore Wind Farm site © Marine Ecological Surveys Ltd; and D) Drop down camera (DDC) employed in the East Coast REC survey © Cefas.

Table 4.4 Technical specifications and settings of remote camera systems.

Survey	Drop Camera System	Camera	Resolution (dpi)	Aperture	Focal Distance	Area covered in frame (m ²)
East Coast REC	Freshwater curtain	Canon Powershot G5	180	f5.6-f2.2	17.6- 12.7mm	0.08
Humber REC	Drop camera - standard frame	Canon Powershot G5	180	f2.5-f2.0	7.2mm	0.06
Cutline	Freshwater curtain - Weasel II	Olympus SP350	72	f2.9-f2.8	9.3mm	0.27
Thames REC	Freshwater curtain - Weasel II	Olympus SP351	72	n/a	n/a	0.2
Thanet (2005 & 2007)	Freshwater curtain - Weasel II	Olympus SP352	72	n/a	n/a	0.27
Thanet (2012)	Freshwater curtain - Weasel II	Olympus SP352	250	f2.9	9mm	0.27
LINCS	Freshwater curtain	NIKON E990	180	f7.0-f3.4	8.2mm	0.2

All photographic data detailed in Table 4.4 were compiled into a digital library of more than 7,000 images (~16GB). Because of the volume of images collated, and the time available for this study, it was necessary to discriminate between images obtained from established or potential *Sabellaria spinulosa* reefs and those collected from areas were *S. spinulosa* was absent or only present as rubble or isolated crusts and tubes. To that effect, data was collated into a GIS to select areas previously classified as *S. spinulosa* reef biotopes (EUNIS

codes A4.22 and A5.611, see Table 4.2) using either direct ground-truthing observations, habitat suitability models, high resolution remote acoustic methods or a combination of these techniques. All GIS analyses were conducted using ESRI® ARCMAP 10.0 and QGIS version 2.10.

Those sampling stations containing both a photographic record and a quantitative sample, within *S. spinulosa* habitats (see Figure 4.8) were selected for further statistical analyses. Nonetheless, all imagery available was scanned for the presence of *S. spinulosa* aggregations and qualitative presence/absence and density maps were constructed and compared with comparative records from direct sampling using GIS (Figure 4.13 & 4.20).

i. Image analysis

Due to their poor quality, towed sledge video camera stills were not incorporated into the analyses. Out of the 3,257 digital still images obtained, a total of 2,117 were of sufficient quality for evaluation of S. spinulosa density indicators (see Section 4.3.2 ii for quality criteria and error sources). Reefiness scores (Gubbay 2007; Hendrick & Foster-Smith 2006) were also derived during the image analyses and included: A) Elevation; B) Sediment consolidation; C) Area; D) Patchiness; E) Density; F) Biodiversity; and G) Biotope match (see Table 4.5). Only elevation, consolidation and biotope were used as selection criteria to determine whether or not the aggregations of S. spinulosa observed in the photographs qualified as biogenic reefs. As the number of replicate photographs greatly varied for each station (from 1 to 21 images) one to four replicates (where possible) were randomly selected from those of better quality for inclusion in the subsequent analyses and the S. spinulosa tube counts were then averaged for each station. In total 287 photographs of S. spinulosa reefs were used in the analyses. Those photographs corresponded to 149 drop down camera (DDC) stations with co-located grab infaunal data (see Figures 4.8 & 4.19). Of these only 64 stations were positive for S. spinulosa reefs in both grabs and photographs. As part of this study, all epifaunal taxa were identified from the seabed images to species level where possible and were enumerated. Percentage cover by colonial taxa (hydroids, sponges and bryozoans) was calculated and SACFOR scores assigned. Naming followed the World Register of Marine Species (WoRMs; http://www.marinespecies.org/) to ensure naming consistency across datasets. Where possible (i.e. data publicly available) these data are provided in full in Appendix 2.2.

Table 4.5 Summary of reefiness scoring criteria (adapted from Gubbay 2007 and Hendrick & Foster-Smith 2006). Only elevation (based on maximum tube height), consolidation (percentage cover of consolidated tubes in relation to total area covered by the drop down camera) and biotope (based on Connor *et al* 2004 descriptions) were evaluated and used as the selection criteria for *S. spinulosa* reef presence in the seabed images.

	Definition	Not a reef	Low	Medium	High
A. Elevation	Maximum tube height	<2	2-5	5-10	>10
B. Consolidation	Percentage cover of substratum by consolidated <i>S.</i> <i>spinulosa</i> tubes Degree of consolidation	<10 rubble	10-20 veneers, crusts	20-30 upright tubes, some concretion	>30 Matrix of well- developed tubes
C. Area (km²)	Extent of total area	<0.025	0.025-0.1	0.1-1	> 1
D. Patchiness	Percentage cover of consolidated <i>S.</i> <i>spinulosa</i> within overall spatial extent of the reef		10-20	20-30	>30
E. Density	Average density of <i>S. spinulosa</i> m ⁻²		~800	~1500	~3000
F. Biodiversity	Margalef's species richness Shannon diversity index Simpson's diversity index		~5.0 ~2.5 ~0.85	~6.5 ~2.7 ~0.87	~8.0 ~3.0 ~0.90
G. Biotope	MNCR biotope code	Other	CR.MCR.Csab. Sspi	S.SBR.PoR.	SspiMX

The open source program Image J^{\otimes} (Schindelin *et al* 2012) was used to calculate total and consolidated *S. spinulosa* cover (reefiness scores B, C and D) and tube density (score E) and, tentatively, reef elevation (score A) (Figures 4.10). The number of tube openings was used as a proxy for *S. spinulosa* density because animals were completely retracted into their tubes in the majority of photographs obtained. Digital image processing and automating tube counting methods were also undertaken and their feasibility evaluated.

Quantitative abundance data (including the presence / absence of colonial species as 1 / 0 respectively) were standardised to relative abundance (counts m⁻²) and log-transformed prior to carrying out statistical analyses using the open source program R (R Development Core Team 2013). Cumulative *S. spinulosa* density values varied by as much as three orders of magnitude, from less than 100 to well over 10,000 tubes or worms m⁻². Therefore, log transformations of density estimates were necessary prior to statistical analyses to comply with parametric model assumptions of normality and heteroscedasticity. All graphs were constructed using the R package ggplot2 (Wickham 2009).



Figure 4.10 Representative *S. spinulosa* reef images and corresponding worm tube visual counts determined using the image processing software Image J®. (A, B) Weasel II FCC system, 72 dpi, Cutline 447; (C, D) Cefas drop camera, 180DPI, Humber REC; (E-F) Weasel II FCC system, 250 dpi, Thanet Wind Farm 2012 surveys. Yellow marks represent tubes counted during the image analysis. A grid is overlaid to help during the enumeration (F). Analyses carried out using Image J[®].

4.2.3 In situ sampling data

Data used in this study were collected over a number of years by different organisations and for different purposes and hence a certain degree of data cleaning and standardisation was necessary before the data could be re-analysed. Quantitative species abundance data were standardised against the World Register of Marine Species (WoRMs; <u>http://www.marinespecies.org/</u>) to ensure naming consistency across datasets. These data were also truncated to remove non-specific identifications (e.g. *Syllis* Type A) and juveniles,

which are often but not always recorded separately. Colonial species which cannot be enumerated were included in the data as one individual where they were present and zero

where they were not. It was also necessary to assign a project specific label to each sample to avoid duplicated labels, in all instances where this was necessary the original sample label was retained as a factor for cross-referencing purposes.

4.2.4 Geographical grouping

There is some geographical overlap in the study areas chosen for inclusion in the evidence base for Descriptor 1 indicators for *S. spinulosa* reef. The Thanet offshore wind farm site for example, falls within the Thames REC study area and Cutline aggregate extraction site is also very close. Similarly the Lincs wind farm site overlaps with the Humber REC study area and the Hastings Shingle Bank site is adjacent to the South Coast REC site. As one of the objectives of this work is to determine useful indicators of *S. spinulosa* reef condition within the context of natural variability, the data were grouped geographically using the shipping forecast areas displayed in Figure 4.11 in order to carry out post-hoc analysis of variance.



Figure 4.11 Chart showing the UK sea areas as defined by the shipping Forecast. These areas were used to group overlapping or closely adjacent study sites in order to investigate broad geographical trends in density and diversity (Source (GNU license): http://en.wikipedia.org/wiki/File:UK shipping forecast zones.png).

i. Summary of sample replication

The limitations of the data that were made available for this study are such that not all data could be used for all of the different analyses undertaken. For clarity the number of samples available from each of the surveys, under each sample class analysed are summarised overleaf in Table 4.6.

Table 4.6 Summary of the number of sample replicates for each data type included in the analyses used in the development of Descriptor 1 indicators for *S. spinulosa* reef habitats from each of the eight study sites.

	Seabed Images						Grab Sample Data						Grab /
	Presence /Absence	Tube Densities	Epifauna	Presenc	e/Absence		Dens	ity Categ	ories		Acoust Define	tically d Reef	Comparisons*
				Ρ	Α	0	1-19	20- 99	100- 999	1000 +	Reef	Sediment Habitat	
Hastings Shingle Bank	N/A	N/A	N/A	120	20	20	85	24	11	0	82	58	N/A
Cutline Area 447	223	29	29	86	107	107	80	4	2	0	~	~	29
Humber REC	404	40	40	57	78	78	39	9	7	2	~	~	29
East Coast REC	521	73	73	79	76	76	55	12	9	3	56	99	30
Thames REC	244	31	31	25	45	45	17	5	3	0	~	~	19
South Coast REC	5	5	5	36	31	31	25	10	1	0	~	~	2
Thanet Offshore Windfarm	123	125	125	34	118	118	22	4	5	3	20	132	1
Lincs Offshore Windfarm	131	47	47	~	~	~	~	~	~	~	~	~	29
Total No. Samples	1650	350	350	437	475	475	323	68	39	8	158	289	149

*Replicate images averaged per station within defined reef areas; only matching stations simultaneously positive for S. *spinulosa* were considered for the analyses

** East Coast REC samples only

4.3 Methods of detecting and sampling Sabellaria spinulosa reef

Direct sampling is thought to be the most accurate means of determining *S. spinulosa* density and the composition and diversity of associated fauna (Foster-Smith and Hendrick 2003; Limpenny *et al* 2010) however direct sampling is destructive, time consuming and expensive and therefore one of the objectives of this study was to investigate remote methods currently being employed to sample these habitats and to assess their suitability for MSFD indicator monitoring. It was hypothesised that remote sampling methods such as video and still cameras and acoustic remote sensing techniques might provide useful ways of measuring indicators of reef density and faunal associations as well as other reefiness parameters as described by Hendrick and Foster-Smith (2006).

A review of the data available for use in this project and the associated reports was undertaken as a first step in assessing the sampling methods. Methods that were found to be too destructive or of too coarse a resolution for MSFD monitoring were excluded at an early stage to ensure that resources were focussed on the sampling tools which are most likely to yield appropriate information for monitoring purposes. A quantitative assessment of the variability observed in data quality obtained using the short-listed sampling techniques was then made to establish which tools were most appropriate for monitoring *Sabellaria spinulosa* reef habitats in the context of the MSFD.

4.3.1 Method efficiency and suitability for MSFD monitoring

A review of the data and reports that make up the evidence base for this study revealed that the following technologies are used as standard for the detection and monitoring of *S. spinulosa* reefs in the UK, although it should be noted that monitoring *S. spinulosa* reefs is rarely the primary aim of the surveys themselves.

i. Direct sampling methods

a. Scientific beam trawls

Although a small number of surveys have sampled S. *spinulosa* reef habitat using a 2m scientific beam trawl this usually only occurs where the surveys are exploratory (for example in the RECs) and this habitat is not previously known to occur. The level of damage caused by this sampling method is not thought to justify the semi-quantitative data that it yields. Generally a trawl sample is taken over a distance of 250-500m and as *S. spinulosa* reef features may be smaller than this, the positional accuracy of data collected in this way is not considered appropriate for monitoring. Trawling of any kind has not been considered further in this study.

b. Sediment grabs

The sediment grab is the most widely used sampling tool for EIAs in the UK and the $0.1m^2$ Hamon grab is most commonly used in the sublittoral sedimentary habitats where *S. spinulosa* reefs are found, although there are many instances where $0.1m^2$ Van Veen and Day grabs have been used to successfully sample this habitat (see Appendix 2.2 for examples). The sediment grab has proven very useful for detecting and monitoring reefs because it allows direct observations of both the density of living worms and the macrofaunal communities associated with this habitat. Although this sampling method is destructive, the footprint of a mini Hamon grab is only $0.1m^2$ and given the known capacity of *S. spinulosa* to recover from physical disturbances (Pearce *et al* 2007), a moderate sampling regime using this tool is very unlikely to cause lasting damage to *S. spinulosa* reef habitats.

ii. Remote sampling methods

a. Acoustic sampling

High resolution acoustic data have been used as a tool for identifying and mapping the extent of S. spinulosa reefs for a number of years (Marine Ecological Surveys Ltd 2005; 2007, 2012, Pearce et al 2007; 2011) Although it is possible to identify biogenic structures using acoustic techniques, it is not yet possible to accurately differentiate between reefs formed by S. spinulosa and those formed by Mytilus edulis or aggregations of sponges and dense epifauna. Subsequent ground-truthing is therefore required, following which, it is possible to use acoustic data to map reef quality in broad terms (e.g. patchy reef and dense reef). This methodology has been used successfully in the Thanet Offshore Wind Farm site surveys and subsequent monitoring (Marine Ecological Surveys Ltd 2005, 2007, 2012). However, no link has been established between the acoustic signature of S. spinulosa reef and the density of living worms, or the associated reef fauna (Foster-Smith & Hendrick 2003; Pearce et al 2011). The resolution of acoustic data that are currently being used (maximum resolution 23cm, typical resolution 0.5–2m) is very unlikely to yield a strong correlation with reef measures such as S. spinulosa density because the habitat is very variable at this scale (see Figure 4.12). As technology evolves it is possible that the small-scale variation of reefs may be resolved using extrapolation from ground-truthed high definition backscatter (as for *M. edulis* beds, see Section 3.2.2 ii) but at the present time, the remote sampling tools that are most likely to yield useful S. spinulosa density and diversity proxies are underwater video and still imaging. Acoustic methods were not investigated further in the present project.



Figure 4.12 Photograph of a *Sabellaria spinulosa* reef habitat associated with the Silver Pit (© Marine Ecological Surveys Ltd) demonstrating the patchiness that is typically observed in this habitat (colour bands on the scale bar represent 1cm intervals).

b. Towed video

Towed video footage was collected during a number of the studies included in the evidence base for the present work and therefore evaluated as a sampling method within an MSFD monitoring context. However, this sampling method was quickly eliminated because the visibility in this footage was exceptionally poor. Most towed video systems are also

configured to give a landscape perspective of the seabed whereas a 'birds-eye' perspective is required when tube counts or epifaunal enumeration are required. Where turbidity levels were not excessive, and 'birds eye' images were available, image grabs taken from this footage were examined but the resolution was also found to be too poor to resolve individual tubes and most epifaunal species could not be identified with any degree of certainty. The use of towed video to monitor change in reef metrics was therefore not pursued further in the present project.

c. Drop cameras

Standard drop down camera (DDC) and freshwater lens camera systems provided images of sufficient quality to attempt an accurate assessment of tube density, associated epifaunal species and other reefiness measures such as consolidation and height. Grab mounted camera systems were identified as the best source for comparisons with grab data, because the images are obtained from the same location at the same time, allowing for a more meaningful analysis of correlation between direct and remote surveying systems. Unfortunately, grab mounted systems are not commonly used and data from this type of sampling array was only available for eight sampling stations on one survey (East Coast REC) before the image quality was deemed too poor to be used in any further sampling (Limpenny *et al* 2011). It should be noted that image quality issues reported from the East Coast REC are a result of technical problems that preclude further consideration here, this system should not therefore be ruled out of future monitoring programmes.

iii. Efficiency of remote and direct sampling methods for reef detection

Overall, there was a good correspondence between stations where *S. spinulosa* habitats were detected using seabed images and where they were detected using grab samples, as illustrated in

Figure 4.13 and the matrix displayed in Table 4.7. The degree of correspondence quantified in Table 4.7 shows that 22% of Weasel II images obtained from a variety of reefs (Cutline, Thanet, and Thames REC) did not identify *S. spinulosa* where this species was recorded in the co-located grab sample, the highest percentage discrepancy of all methods. In total, the greatest level of agreement in results were obtained from the standard frame DDC used in the Humber REC with only 4% of mismatched images and the fresh water lens camera systems used in the East Coast REC and LINCS surveys (6% mismatch). Therefore, and contrary to Limpenny *et al* (2010), camera stills were able to detect *S. spinulosa* even in conditions of low agglomeration. Remote imaging methods were, however, able to identify the presence of *S. spinulosa* reefs in areas where grab samples had failed to return any records indicating that the two sampling gears were not always aligned and sampling in the same location.

Table 4.7 Matrix summarising percentage correspondence between visual assessments of still imagery and co-located 0.1m² Hamon grab records of *Sabellaria spinulosa* presence and absence. Whilst co-located samples were taken from the same target coordinates they were not taken at the same point in time or at precisely the same location. The accuracy of co-location is estimated to be <10m.

Survey	Drop camera system	Number of photographs	Useable	Sabellaria present	Negatives with grab positives	Positives with grab negatives
EC REC	Freshwater curtain	856	521	404	6%	36%
HB REC	Standard drop down camera	1129	875	455	4%	7%
Cutline	Freshwater curtain - Weasel II	303	223	75		
TH REC	Freshwater curtain - Weasel II	348	244	114	22%	31%
Thanet	Freshwater curtain - Weasel II	173	123	80		
LINCS	Freshwater curtain	448	131	131	0%	10%
Total		3257	2117	1259		

Modelled *S. spinulosa* density distributions based on Inverse Distance Weighting (IDW) interpolations of density estimates obtained from grab counts (Figure 4.13A) and worm tube counts from photographs (Figure 4.13B) suggest that in the Humber REC, cameras underestimated densities in some areas (north-west and south of the study area: Figure 4.13). As previously indicated in Table 4.5, only areas with *S. spinulosa* tubes arising more than 2cm above the seafloor were used in the visual assessments for density indicators. Therefore, the lack of agreement is due to *S. spinulosa* forming dense but low lying crusts rather than a proper reef throughout most of the surveyed area off the Humber estuary. The position of core reef areas detected by stills and grabs roughly matched although high densities of *S. spinulosa* estimated from stills collected from some stations in the Inner Wash (Lincs) and Thanet areas were not matched by grab sample records.

The slight disagreement between remote and direct methods is probably the result of differences in reef elevation and patchiness and environmental conditions that make it difficult to obtain accurate density estimations using remote methods, as well as sample gear positioning. Lower resolution images (72 dpi) and poor focussing and lighting also seemed to have reduced the success of optical sampling methods in some cases (see Figure 2.14).



Figure 4.13 Presence-absence maps for *S. spinulosa* derived from 0.1m² Hamon grab samples (top) and co-located seabed images (bottom). Modelled *S. spinulosa* densities maps are derived from Inverse Distance Weighting (IDW) interpolations (Analysts Tools package; ArcGIS® v.10) using log10 (counts) m⁻² estimated from grabs and photographs (for a theoretical description of IDW see for example Bartier & Keller 1996). Data included in these charts are derived from the Humber REC; East Coast REC and Outer Thames REC, Cutline 447 and Thanet Offshore Windfarm surveys (Table 4.3). Whilst co-located samples were taken from the same target coordinates they were not taken at the same point in time or at precisely the same location. The accuracy of co-location is estimated to be <10m.

a. Reefiness evaluation using seabed images

An added advantage of using seabed images to detect and monitor *Sabellaria spinulosa* reef habitats is that they can yield other useful information regarding the physical nature of the reef. It is possible for example to determine the height and patchiness of the reef, with a reasonable degree of accuracy. Physical reef attributes were recorded for all of the images that were analysed for *S. spinulosa* density and associated fauna, where this was possible. It was possible to record reefiness attributes from between 70 and 100% of the images that were of a suitable quality for density estimates (Table 4.8) showing that this sampling method has significant potential to monitor numerous aspects of *S. spinulosa* reef habitats using a single set of data.

Table 4.8 Summary of remote camera imagery collected and analysed during the present study. Values under reefiness scores (Hendrick & Foster-Smith 2006) represent the percentage of photographs from which it was possible to extract reefiness scores.

		Reefiness scores									
Survey	Camera system	A. Elevation	B. Consolidation	C. Area	D. Patchiness	E. Density	F. Biodiversity	G. Biotope			
EC REC	Freshwater curtain	98%	89%	89%	89%	41%	89%	89%			
HB REC	Standard drop camera	94%	91%	91%	91%	83%	91%	91%			
Cutline	Freshwater curtain - Weasel II	99%	99%	99%	99%	51%	99%	99%			
TH REC	Freshwater curtain - Weasel II	74%	79%	79%	79%	48%	79%	79%			
Thanet	Freshwater curtain - Weasel II	100%	100%	100%	100%	64%	100%	100%			
LINCS	Freshwater curtain	100%	100%	100%	100%	95%	100%	100%			

b. Potential sources of error / variance in seabed image analysis

The quality of DDC images varied considerably between and within the different surveys used in this study (Table 4.7). The main source of variation in image quality was found to be the environmental conditions, although the presence of epifauna and the structure of the reef itself also had some influence on whether images were useful for subsequent analyses.

Environmental Conditions

The differences between the camera systems evaluated were not simply the result of their technical specifications but strongly influenced by poor environmental conditions at the time when the surveys were undertaken. The percentage of 'useful' images was calculated for each survey suggesting the standard DDC produced the highest quality images (78%), the most recurrent source of error being blurred images caused by camera movement (58%). The freshwater curtain camera (FCC) systems were widely used in areas where poor visibility is a recurrent problem. Calculated percentages of usable imagery were similar: 61% (EC REC FCC); 74% (Cutline 447, Weasel II FCC), 71% (Thanet area, Weasel II FCC) and Thames REC survey (70%). The camera deployed in the EC REC surveys produced higher resolution images which could be used to establish most reefiness scores. The most recurrent issues with the system included poor illumination and the presence of large air bubbles that hampered the accurate count of worm tubes in otherwise good quality images

(Figure 4.14A). Air bubbles (22%), poor focus (17%), encrusting epifauna (8%) and horizontally growing tubes (6%) were other factors causing rejection of some of the available imagery obtained from freshwater cameras used in the East Coast REC surveys. Although the system performed moderately well in the predominantly turbid East Coast REC area (39% of images rejected), its performance in The Wash surveys (Lincs) was disappointing. In The Wash, extremely poor visibility resulted in 71% of the images being rejected.

The images produced by the Weasel II system had the lowest resolution (72 dpi; Figure 4.15) and 28% were too dark to be of any analytical use, probably the result of extremely poor visibility conditions. These images were overall poorly lit and lacking in quality to accurately count tubes in low relief *S. spinulosa* reefs or seabed crusts and clumps (Figure 4.14B). A combination of poor lighting, high turbidity and low image resolution was the main cause of rejection in 89% of these images. Nonetheless the system is capable of capturing images of elevated reefs that are sufficient to allow for tube abundance estimations (Figures 4.14C & D). Reef area and patchiness scores were difficult to calculate using the Weasel II FCC systems due to a combination of poor background focus and low visibility (Figure 4.15).



Figure 4.14 Sources of variance identified in digital images obtained from freshwater curtain camera (FCC) systems. A) Air bubble on the Freshwater Lens Camera used in the East Coast REC B) Poor lighting on the Weasel II freshwater lens camera used on Cutline 447; C & D) Poor background exposure and unequal focussing in images taken with the Weasel II freshwater lens camera used on Thanet Wind Farm and the Thames REC.



Figure 4.15 Images captured by the Weasel II freshwater lens camera system in the Outer Thames area (Thanet Wind Farm development) showing low resolution when zooming is performed to facilitate tube counting.

Sabellaria spinulosa reef structure

Density in elevated and intact reefs can be visually estimated with accuracy provided that the images are of sufficient resolution and the tubes grow vertically. Inaccurate density estimations occur if tubes grow horizontally or have been damaged by physical impact. Worm density in reefs consisting of numerous, thin and densely packed tubes can prove very hard to quantify. Less than 10% of images were rejected due to the structural and density characteristics of the reefs.

Siltation/presence of colonial epifauna

Most false negatives obtained from image analyses in the Humber and East Coast RECs were the result of heavy siltation (56%), the presence of smothering or dense colonial epifauna (i.e. *Molgula* spp., hydroid and bryozoans turf found in 5% of the images) or a combination of the two. Under these scenarios tube openings are usually not visible and therefore density estimates are not accurate. Nonetheless, the presence of *S. spinulosa* and other reefiness scores such as reef extension, consolidation, elevation and biodiversity could be estimated.

c. Potential sources of error / variance from grab data

S. spinulosa reef data collected using sediment grabs in the UK is thought to be of a very high quality standard. All of the data used in this study were collected and analysed by taxonomists working in laboratories that actively participate in the National Marine Biological Analytical Quality Control (NMBAQC) scheme. As such the laboratories should all be using the same taxonomic keys and the data should be comparable between labs. Taxonomic nomenclature does however change over time and there will be some minor differences in nomenclature applied between different labs. The data in this study were standardised to the WorMS accepted species nomenclature to ensure that these differences were not significant.

Box 4.2. Summary of methods of detecting / sampling Sabellaria spinulosa reefs.

- High resolution acoustic data are currently being used to detect and map *S. spinulosa* reefs but the resolution is thought to be too coarse at the present time to be useful for assessment of density indicators. However, backscatter analyses could prove a useful proxy for density in the future as it has with *M. edulis* beds (Section 3). The use of side-scan sonar is nonetheless of value to determine changes in reef extent and percentage coverage.
- Towed video has been used to sample *S. spinulosa* reefs in the UK with varying degrees of success. The footage examined as part of this study was not of a sufficiently high resolution to facilitate tube counts or species identifications.
- ROV and AUV have not yet been tested extensively on *S. spinulosa* reefs but these may also become viable deployment strategies in the future.
- Divers have not been used to sample *S. spinulosa* reefs in the UK. Depths and high turbid make dive surveys inappropriate for monitoring.
- Beam trawls have been used to sample *S. spinulosa* reefs in a limited number of cases in the UK. This method of sampling reefs is too destructive to be considered suitable for monitoring and data are only semi-quantitative. Trawls are also likely to have sampled several habitats during a single tow (typically 250-500m long).

Box 4.3. Summary of sources of error in *S. spinulosa* sampling procedures and related recommendations.

- Of the sampling methods currently being employed on *S. spinulosa* reef habitats in the UK, drop down cameras and sediment grab samples are thought to be the most likely to yield information required to assess the environmental status of this habitat.
- The resolution and clarity of seabed images is central to our ability to undertake the tube and species counts that would be required for a Descriptor 1 biodiversity indicator. Both methods are likely to be affected by local environmental conditions, most notably turbidity levels, *S. spinulosa* reef structure (i.e. the direction of tube growth) and siltation / presence of colonial epifauna which may obscure both *S. spinulosa* and the associated fauna. High definition imagery (>250 dpi) and freshwater lens camera systems with adequate lighting are required to minimise these sources of error.
- Sources of error associated with *S. spinulosa* reef grab samples can be appropriately controlled so long as laboratory analysis is undertaken by trained taxonomist working in laboratories that participate in the NBMAQC or equivalent schemes.

4.4 Relationship between the density of *Sabellaria spinulosa* and the associated macrofaunal diversity

The present study set out to develop indicators of *Sabellaria spinulosa* density (where it forms reef habitats) as well as the abundance of other species associated with the reef. It is therefore logical to attempt to understand whether or not there is any relationship between these two aspects of reef ecology. A strong correlation could indicate that monitoring costs could be reduced by focussing sampling on density, for example. Similarly, investigations into the relationship between density and diversity may reveal that the most diverse communities are associated with either an intermediate density of worms or the highest density of worms and therefore better inform sustainable management.

4.4.1 Relationship between the density of *Sabellaria spinulosa* and the diversity of species associated with the reef

i. Measurements of *S. spinulosa* density and associated macrofauna from grabs

The relationship between *S. spinulosa* density and the diversity of associated macrofauna was investigated by undertaking a correlation and regression analysis of *S. spinulosa* worm density against a number of standard diversity indices (as detailed in Section 4.5.2) calculated for the macrofauna identified in the same grab samples (see Section 4.2 and Table 4.6). *S. spinulosa* itself was removed from the faunal data before diversity indices were calculated. Spearman's rank correlation analyses indicated a statistically significant positive correlation between the abundance of live worms in the grab samples and all of the associated diversity indices with just two exceptions; Pielou's Evenness was found to be negatively correlated with *S. spinulosa* density and Simpson's Diversity index showed no significant correlation with the density of living worms (Table 4.9).

Table 4.9. Spearman's rank correlation (r_s) between the densities of *S. spinulosa* recorded in 0.1m² Hamon grab samples and a selection of diversity indices calculated for the associated macrofaunal community. Colonial species that could not be enumerated were recorded as present (1) or absent (0). *S. spinulosa* was removed from the data before diversity indices were calculated. Spearman's rank correlations (r_s) range from -1 (perfect negative correlation) to +1 (perfect positive correlation) with a zero value representing no correlation in the data at all. Statistically significant correlations (P<0.05) are marked with an asterisk.

Variable	r _s	р
Number of Individuals (N)	0.62	<0.001*
Number of Species (S)	0.66	<0.001*
Shanonn's Diversity (H')	0.42	<0.001*
Margalef's Richness (d')	0.64	<0.001*
Pielou's Evenness (J')	-0.27	<0.001*
Simpson's Diveristy (1-λ)	0.06	0.09

The strongest correlations identified were those between *S. spinulosa* density and the number of individuals (N), number of species (S) and Margalef's Richness (d) (r_s =0.62-0.66, P<0.001). The correlation between *S. spinulosa* density and Shannon's diversity (H') was slightly lower (r_s =0.42, P<0.001) which reflects the fact that this index includes a measure of evenness which was found to be negatively correlated with the density of *S. spinulosa* (r_s =0.27, P<0.001). This shows good agreement with earlier work by Pearce *et al* (2007) who found that the density of some associated fauna, most notably the long clawed porcelain crab *Pisidia longicornis* became very dominant as the reefs developed whilst other epilithic and infaunal species began to be excluded. A similar successional pattern has been reported for communities of the Olympia oyster, *Ostrea lurida* (Kimbro & Grosholz 2006). In

this study an increase in the richness of associated species and a decrease in evenness were observed with increasing oyster densities. The authors noted that habitat creation and species exclusion through competition for space were acting as conflicting forces on the assemblages associated with the oyster beds and it is likely that a similar set of forces are acting on the *S. spinulosa* reefs included in this study.

Figures 4.16 A-F represent the corresponding linear regression plots between the live worm abundances and the community abundance, diversity and evenness indices. Overall fit of the regression models was very poor suggesting change in the community and diversity indices cannot be explained solely by standard linear models using density as the explanatory variable. It is likely that multiple regression models, i.e. generalized additive models (GAMs) or generalized linear (mixed or fixed) models (GLMs), could have better explanatory power. However, additional parameterization and construction of such models was beyond the scope and time frame of this study.

The lowess curves in Figure 4.16 as well as results in similar studies (Pearce *et al* 2007) indicate that the relationship between live *S. spinulosa* abundance and community abundance is a hyperbolic one (as found for *M. modiolus* and *M. edulis*, see Sections 2 & 3 of this report). It would appear that the abundance of species associated with a reef increases with increasing densities of *S. spinulosa* until densities reach a point (asymptote) where the reef starts to exclude some species. The increase in community abundance associated with even very low densities of *S. spinulosa* indicates that this habitat is beneficial to benthic communities, presumably through the provision of habitat and food. At higher densities *S. spinulosa* reefs appear to exert a contrasting competitive force on the benthos by excluding some species altogether. This could indicate a high level of habitat modification whereby the habitat no longer contains significant fine sedimentary deposits within their structure or may simply be a result of competition for space.



Figure 4.16 Linear regression charts illustrating the relationship between *Sabellaria spinulosa* density and the associated macrobenthic fauna recorded from 0.1m⁻² Hamon Grab samples (n=741). Red lines represent non-parametric lowess regression curves added to help visualise the trends.

ii. Measurements of *S. spinulosa* density and associated macrofauna from cameras

The relationship between *Sabellaria spinulosa* density and the diversity of associated macrofauna was investigated by undertaking a correlation and regression analysis of *S. spinulosa* tube counts against a number of standard diversity indices (as detailed in Appendix 3) calculated for the macrofauna identified in the seabed images (see Section 4.2. and Table 4.6.). *S. spinulosa* itself was removed from the faunal data before diversity indices were calculated. Spearman's rank correlation analyses indicated a statistically significant positive correlation between the number of *S. spinulosa* tubes in the seabed images and all of the associated diversity indices with just one exception; Pielou's Evenness was found not to be significantly correlated with *S. spinulosa* tube counts (Table 4.10). It is worth noting that although the correlations observed here between *S. spinulosa* tube counts and remote observations of the diversity of associated species show a similar pattern to the correlations observed between in-situ measurements, the relationship is much weaker.

Table 4.10 Spearman's rank correlation (r_s) indexes between the densities of *S. spinulosa* tubes recorded in seabed images and a selection of diversity indices calculated for the associated epifauna recorded from the same images. Colonial species that could not be enumerated were recorded as present (1) or absent (0). *S. spinulosa* was removed from the data before diversity indices were calculated. Spearman's rank correlations (r_s) range from -1 (perfect negative correlation) to +1 (perfect positive correlation) with a zero value representing no correlation in the data at all. Statistically significant correlations (P<0.05) are marked with an asterisk.

Variable	r _s	р
Number of Individuals (N)	0.25	<0.001*
Number of Species (S)	0.29	<0.001*
Shannon's Diversity (H')	0.32	<0.001*
Margalef's Richness (d')	0.22	<0.01*
Pielou's Evenness (J')	0.05	0.55
Simpson's Diveristy (1-λ)	0.17	<0.01*

Figures 4.17 A-F represent the corresponding linear regression plots between remote tube counts and community abundance, diversity and evenness indices calculated from remote species counts. Overall fit of the regression models was very poor suggesting change in the community and diversity indices cannot be explained solely by standard linear models using tube counts as the explanatory variable. It is likely that multiple regression models, i.e. generalized additive models (GAMs) or generalized linear (mixed or fixed) models (GLMs), could have better explanatory power. However, additional parameterization and construction of such models was beyond the scope and time frame of this study. Lowess curves suggested that, similarly to the results described in Section 4.4.1 i, an asymptotic relationship exists between tube density and diversity indices calculated from DDC imagery.


Figure 4.17 Linear regression charts illustrating the relationship between *Sabellaria spinulosa* density (measured as tubes m⁻²) and the associated macrobenthic epifauna recorded from drop down camera stills (n=350). Red lines represent non-parametric lowess regression curves added to help visualise the trends between the parameters measured.

Box 4.4. Summary of findings: relationship between *S. spinulosa* density and the diversity of species associated with the reef.

Relationship between *S. spinulosa* density and measures of the diversity from grab samples

- There is a statistically significant positive correlation of moderate strength between the density of *S. spinulosa* and most diversity and richness measures (r_s = 0.42-0.66, P<0.001).
- There is a weak but statistically significant negative correlation between the density of *S. spinulosa* and the equitability or evenness of species associated with the reef (r_s = -0.27, P<0.001). This indicates that as the density of *S. spinulosa* increases one or more of the associated species also becomes dominant whilst others are excluded.
- There was no correlation between the density of *S. spinulosa* and Simpson's diversity (1-λ) calculated from grab samples (r_s=0.06, P= 0.09). This is because components of species richness and evenness work against each other.

Relationship between *S. spinulosa* density (tube counts) and remote measures of diversity from cameras

- There is a statistically significant positive correlation between *S. spinulosa* tube counts and most diversity and richness measures. However, this is much weaker than the relationship between measurements of density and diversity from grab sampling (r_s=0.17-0.32, P<0.01-0.001).
- There was no correlation between S. spinulosa tube counts and the equitability or evenness of species associated with the reef (r_s=0.05, P=0.55).

Box 4.5. Recommendations for S. spinulosa density and diversity indicators.

- A consistently significant positive relationship was observed between the density of *S. spinulosa* (or tube counts as a proxy for density) and the diversity of species associated with the reef, whether using grab sampling or seabed images. *S. spinulosa* density could therefore be considered as an alternative to measuring density as well as the diversity of associated fauna, but data from grab sampling measurements would be more powerful than seabed imagery.
- Diversity measures that are based on or include a component of species evenness or equitability should be used with caution because changes in richness may be offset by changes in evenness.
- Despite the density-diversity relationship above some form of direct assessment of species associated with *S. spinulosa* reefs is nevertheless important because compositional changes could be early warning signs of stress.

4.5 Sabellaria spinulosa reef density indicators

4.5.1 The range of S. spinulosa densities found within reefs

i. Published records of S. spinulosa density

Published records of *S. spinulosa* density (Table 4.11) are highly variable, ranging from one to a few hundred worms m⁻² in reef habitats surveyed in Selsey Bill, Belfast Lough and offshore of the Humber (Hiscock 2005; Limpenny *et al* 2010) to maximum abundances exceeding 10,000 worms m⁻² in reef habitats surveyed in the southern North Sea (EGS International Ltd 2011; Emu Ltd 2007). All published reports of *S. spinulosa* density originate from sediment grab samples, which sample a relatively small area of the seabed ($0.07m^2 - 0.2m^2$). Since *S. spinulosa* density has been observed to be very patchy at a local scale (see Figure 4.13) differences between locations noted here (Table 4.12) may in part be an artefact of sampling effort.

Table 4.11 Range of *Sabellaria spinulosa* densities and percentage cover reported for samples identified as containing *S. spinulosa* aggregations, biotopes and / or reefs. Numbers in brackets represent mean density.

Location	Density (m ⁻²)	% Cover (m ⁻²)	Method	Source
Borrow head, Luce Bay	n/r	Up to 80	Diver records	Covey (1992)
Amlwych, Anglesey	Up to 9,999	5-79	Diver records (converted from SACFOR)	Hoare & Hiscock (1974)
Bristol Channel	3,028-6,323	n/r	0.07m ² Day grabs	George & Warwick (1985)
Hastings Shingle Bank	Up to 6,400 (400)	n/r	0.1m ² Hamon grab	Pearce <i>et al</i> (2007)
Offshore of the Humber	Up to 299	n/r	0.1m ² Day grabs	Hiscock (2005)
Selsey Bill	Up to 316	n/r	0.1m ² Day grabs	Hiscock (2005)
Thanet Offshore Windfarm	Up to 6,650 (560)	n/r	0.1m ² Hamon grab	Marine Ecological Surveys Ltd, 2005)
Area 447 Cutline	Up to 5,380 (543)	n/r	0.1m ² Hamon grab	(Marine Ecological Surveys Ltd (2008)
Area 107 (Box 1)	870 - 4,840	n/r	0.1m ² Van Veen grab	Hendrick (2007)
Outer Wash (Box 4)	80-7,000	n/r	0.1m ² Van Veen grab	Hendrick (2007)
Area 401	n/r	4-95	Drop down video	(Emu Limited, 2008)
Area 454 Lowestoft	695-8,095 (3,343)	n/r	0.2m ² Hamon grab	Marine Ecological Surveys Ltd (1996)
Benacre	95-2,110 (1,190)	n/r	0.2m ² Hamon grab	Marine Ecological Surveys Ltd (2002)
Lincs (LID6) Development Site	Up to 10,130 (1,320)	n/r	0.1m ² Hamon grab	EGS International Ltd (2011)
Lincs (LID6) Adjacent reef	40-12,730 (4,020)	n/r	0.1m ² Hamon grab	EGS International Ltd (2011)
Race Bank and Docking Shoal Cable Route	570-13,920	n/r	0.1m ² Hamon grab	EMU Environmental Ltd (2007)
Belfast Lough	120	n/r	n/a	Limpenny et al (2010)

Percentage cover of *S. spinulosa* has been reported in a small number of publications where habitat records originate from diver observations (Covey 1992; Hoare & Hiscock 1974) or drop down video (DDV) (Emu Limited 2008). Again, a high level of variation is observed in records reported to have originated from *S. spinulosa* reefs with between 4 and 95% coverage being reported over an area of 1m². Additional records of percentage cover are likely to be available from the early MNCR surveys listed in Appendix 2.2. However, these datasets were not accessed as part of the current study becasue they do not contain quantitative data.

ii. Natural variation in Sabellaria spinulosa density

As a means of investigating the variance in *S. spinulosa* densities in more detail and with a view to determining the typical ranges that could be expected from *S. spinulosa* habitats, the densities recorded within acoustically defined areas of reef were plotted in a box plot (Figure **4.18**). The variability of *S. spinulosa* densities both within and outside acoustically defined areas of reef were found to be incredibly high, illustrating the difficulties associated with developing an indicator for this aspect of reef condition.



Figure 4.18 *S. spinulosa* densities $(0.1m^{-2})$ recorded from grab samples taken within (n=151) and outside acoustically defined areas of *S. spinulosa* habitat (n=266) within the Hastings Shingle Bank, Thanet Offshore Windfarm and East Coast REC study areas. Box plots show interquartile range, median and maximum / minimum observed values as whiskers (1.5 times the interquartile region). Black dots indicate values above and below the 1.5 times the 3rd and 1st quartiles, respectively.

4.5.2 Seabed imagery vs grab sampling for measuring S. spinulosa density

Remote imagery yielded similar density maps to those obtained from Hamon grab samples (

Figure 4.19). In both maps, the highest densities were recorded in areas previously classified as *Sabellaria* reefs either by ground-truthing or through acoustic surveys (see also Figure 4.14, Limpenny *et al* 2011; and Emu Ltd 2008).

Boxplots comparing *S. spinulosa* densities recorded using the remote and grab sampling methods are shown in

Figure 4.20. Boxplot interquartile range and standard error whiskers indicate a higher level of variance in density recorded from seabed imagery for most areas compared to records from grab samples taken in the same area. This result is interpreted as a reflection of the higher numbers of replicate images over a larger reef area obtained from the DDC compared to the number of grab samples collected from the same area. It is also possible that some level of error is introduced as a result of poor illumination and artifacts such as air bubbles, which probably caused underestimation in the counts from some of the survey areas, for example, the East Coast REC (Section. 4.3.1). A more in-depth investigation than was possible in the current study would be required to determine the relative contribution of natural patchiness and different sampling techniques to the variance observed in *S. spinulosa* densities.

The relationship between the mean, log-transformed live worm counts from the grab sample data and log-transformed visual tube counts from images taken at the same station, can be described by the linear equation y = 0.52x+1.28; (F_(1, 50) =13.31; p<0.001; R²=0.21) (Figure 4.21). The results of an ANCOVA analysis (using visual tube counts as the covariate and camera resolution as the categorical, independent variable) indicated the simplest model (equal slope and intersect) was adequate therefore suggesting no significant differences existed between the two density estimates (live worms and tube counts) for each camera system trialled (F_(2.48) =0.95; p=0.39). However, densities were significantly different between locations sampled (F_(3, 189)=9.69; P<0.001) while values recorded from still images were significantly higher than those from grab samples ($F_{(1,189)}$ =4.31; P<0.05) indicating that a large number of the tubes visible in still images were not occupied by living worms. Nonetheless, and as suggested by the overlapping interguartile boxes displayed in Figure 4.20, post-hoc analyses indicated that at each discrete sampling area the differences between densities obtained from grabs or photographs were not significant (P>0.05). Spearman's rank correlation index (r_s) analyses also indicated a significant, positive correlation between mean densities recorded from grab samples and those calculated using visual worm tube counts (global $r_s = 0.55$; df=281; p<0.001). All the results suggest tube counts from remote imagery were an appropriate measure of Sabellaria density. The correlation, nonetheless, varied depending on the camera method used and the image resolution settings. Densities estimated from images collected using the drop camera deployed in the Humber reefs had the highest correlation with the corresponding grab sample abundances ($r_s = 0.76$; df = 63; p<0.001). Freshwater curtain camera (FCC) systems were deployed under low visibility conditions and of the two systems, the lowest correlation value was obtained for the Weasel II cameras set at lower image resolution (72 dpi) ($r_s 0.53$: df = 98; p<0.001) while a higher, positive and significant correlation was obtained for the images collected at 180 dpi in the East Coast REC and LINCS areas ($r_s = 0.6$; df=81; p<0.001).



Figure 4.19 Point-density maps for *S. spinulosa* estimated using (A) a $0.1m^2$ Hamon grab and (B) colocated drop digital cameras. Survey areas (clockwise from the top): Humber REC, East Coast REC and Outer Thames (REC, cutline 447 and Thanet sites; n = 149).



Figure 4.20 Mean *S. spinulosa* tube counts m⁻² obtained from remote imagery and live worms identified from grab samples for all case studies across the English East Coast. Humber Sea Areas: Humber REC (n=54); LINCS (n=44); Thames Sea Area: East Coast REC (n=16); Cutline (n=35); Thames REC (n=33); and Thanet (n=12). Values represent averaged counts for grab sample replicates and replicate photographs from co-located sites. Box plots show interquartile range, median and maximum / minimum observed values as whiskers (1.5 times the interquartile region). Black dots indicate values above and below the 1.5 times the 3rd and 1st quartiles, respectively.



Figure 4.21 Log-log regression between averaged grab sample abundances and corresponding counts from still images captured from three different camera systems and settings. Dotted lines show 95% confidence and prediction bands (red and black lines, respectively).

Variability in tube density across surveyed areas was calculated as coefficients of variance (CV) for each sampling method (see Lindenbaum *et al* 2002). The results indicated that tube density estimates for *S. spinulosa* reefs in the Humber REC area using standard DDCs set at 180 dpi were the least variable (CV=10%) whereas the densities estimated from photographs taken using freshwater lens cameras at 180 and 72 dpi yielded CVs of 19% and 33% respectively. In some areas, the high variance in density estimated using remote data collection methods was probably influenced by image quality and environmental conditions rather than the camera system itself. However, grab samples detected similar trends in the CVs in each survey area indicating natural variability and reef fragmentation were the most likely cause. For example, 34% variation was recorded in the East Coast REC, 42% in the Cutline, Thames and Thanet areas and just 24% in the Humber REC.

4.5.3 Sampling required to detect changes in the densities of S. Spinulosa

A power analysis (Sheppard 1999) on log-transformed mean density data for all samples with S. spinulosa present was used to determine the number of replicate stations (N) needed to detect a given change in the mean using grab samples and drop down cameras (DDC). The digital images generated by the DDCs varied in resolution: those from the CEFAS DDC and from the freshwater curtain cameras (FCC), respectively used in the Humber REC and the East Coast REC/LINCS areas, were set at 180 dpi; on the other hand, the Weasel II freshwater cameras used in the Thames surveys (Thames REC, Cutline 447 and Thanet windfarm in 2005 and 2007) yielded images of much lower resolution (72 dpi). It should also be noted that each camera system covered different areas (0.27m² by the Weasel II system and 0.06-0.08m² by the cameras deployed in the Humber and East Coast RECs). All datasets used in the analyses originated from potential S. spinulosa habitat covering the following areas (see also Figures 4.2-4.8): (1) Humber REC, 14,000km² of habitat suitable for the occurrence of S. spinulosa biotope; (2) East Coast REC and LINCS windfarm, 55km² of ground-truthed acoustic reefs; and (3) Thames area, 184km² (Outer Thames REC, Cutline 447 and Thanet windfarm). N was calculated for the detection of 10, 20, 30, 40 and 50% changes in mean reef density at the 5% significance level. The power of the test is defined as 1- β under the standard significance assumptions (α =0.05; the probability of making a Type II error (β) was 0.05).

Results displayed in Figure 4.22 indicate that 15 replicate stations are needed to detect a 10% change in mean *S. spinulosa* tube density within an acoustically defined area of reef using DDC at 180dpi resolution in the Humber REC reefs compared to 79 grab samples to detect the same percentage of change in the density of live worms. The difference in sampling effort required is a result of the differences in variances associated with the two different sampling techniques, as noted in Figure 4.22.

For more patchy reefs surveyed using freshwater camera systems, 142 and 54 DDC samples were predicted to detect 10% change in tube density using 72 and 180 dpi respectively. The higher number of samples required is highly influenced by high inherent variance caused by broad-scale reef fragmentation or patchiness across the survey areas as well as worse visibility conditions compared to the Humber study. Water curtain systems are usually employed in low visibility conditions thus an increase in sample effort might be expected. Nonetheless, remote imaging offers an advantage over direct, destructive sampling methods because 152 and 240 grab sampling stations would be required to detect the same level of change for the EC REC and Thames areas (respectively). Power analysis indicated that a 20% change in density can be detected using a considerably lower number of samples, even in fragmented reefs under high turbidity conditions (e.g. East Coast and Thames). For example, consolidation was on average 48% according to the digital images obtained from *S. spinulosa* reefs covering an approximate area of 18 km² in the Outer Thames REC. Therefore, a reduction in cover from 48% to 38% could be detected using 37

camera stations or 62 grabs, one order of magnitude lower than required if attempting to detect a 10% change.



Figure 4.22 Sampling effort estimations for camera and grab sampling methods to detect changes in mean *S. spinulosa* densities in reefs sampled as part of marine aggregate and windfarm development environmental surveys. The East Coast and LINCS areas were sampled using freshwater curtain cameras (FCC) set at high resolution (180 and 300 dpi respectively) while the Thames complex was investigated using Weasel II FCCs at 72 dpi. The Humber REC surveys were undertaken by means of a standard drop camera producing 180dpi images. The significance level α is set at 0.05. Power analysis used minimum power (1 - β) set at 0.95.

Box 4.6. Summary of findings for natural range and level of sampling required for *Sabellaria spinulosa* reef density indicators.

- The published literature and data examined in this study indicate that the density of *S. spinulosa* within a reef is highly variable and can range from 0 to 13,000 individuals m⁻².
- This range may encompass densities of *S. spinulosa* recorded from growth forms other than 'reef' such as solitary tubes or small clumps. However, there is an absence of an operational reef definition that can be consistently applied in a monitoring context.
- A much higher level of sampling would be required to detect changes of 10-20% using grabs rather than using seabed imagery.
- The level of sampling required to detect a given level of change in different areas varied. Making indicators operational would therefore require site-specific consideration of sampling effort. However, fifteen replicate samples (using either grab sampling or seabed imagery) should detect a 30-50% change in *S. spinulosa* density in most cases.
- Where a greater level of change detection is required (e.g. 10% change or less) seabed imagery is proposed as the most appropriate monitoring method. Density estimates from seabed imagery are correlated with density estimates from grab sampling anyway and have the added advantage of not being as damaging.

4.6 Sabellaria spinulosa reef community indicators

4.6.1 Fauna associated with *S. spinulosa* reefs in different locations and on different sediment types

i. Published records of S. spinulosa reef communities

There are assertions throughout the published literature that *S. spinulosa* reef support an enhanced community of infauna and epifauna and indeed a distinct faunal complement is listed as one of the characteristics of an *S. spinulosa* reef as defined by OSPAR (OSPAR 2008). Despite this, there are relatively few studies that have investigated the fauna supported by *S. spinulosa* reefs. Those studies that report the diversity of macrofauna supported by *S. spinulosa* reefs in the UK are summarised below in Table 4.12.

Table 4.12 Range of macrofaunal diversity associated with *Sabellaria spinulosa* aggregations/reefs reported in the literature. Macrofaunal diversity is expressed as S (number of taxa), N (number of individuals), d' (Margalefs richness), H' (Shannon's diversity), J' (Pielou's evenness), and $1-\lambda$ (Simpson's diversity).

Location	S	d'	H'	J'	1-λ	Method	Source
East Coast REC	99-214 (312)			0.5-0.9 (0.8)	0.7-0.9 (0.9)	0.1m ² Hamon grab	Pearce <i>et al</i> (2011)
Benacre	30-49 (38)	6.5-8 (6.8)	1.9-2.9 (2.4)	0.5-0.8 (0.7)		0.2m² Hamon grab	Marine Ecological Surveys Ltd (2002)
Area 481	22	5.1	2.3		0.90	Grab	Entek UK
Wash	48	7.0	2.6		0.84	0.1m ² Day Grab	NRA Bailey Unicomarine
Area 107	60	9.2	2.9		0.87	0.1m ² Van Veen grab	Hendrick (2007)
Long Sands	47	8.3	3.0		0.92	0.1m ² Van Veen grab	Hendrick (2007)
Inner Dowsing	41	7.3	2.7		0.87	0.1m ² Van Veen grab	Southeran (2004; 2005c)

A study of *Sabellaria spinulosa* reefs in the Wash found that the reefs supported twice as many species and three times as many individuals as the surrounding sediments (excluding the worms themselves), suggesting that in this area sabellariid reefs are exerting a significant structuring influence on benthic communities (NRA 1994). Unfortunately though, this study has not been published in the peer-reviewed literature and the original report is out of print, and unavailable to use in the present study. George and Warwick (1985) in the Bristol Channel also identified a significant increase in the biodiversity associated with *S. spinulosa* reefs. In this study the number of species associated with the reef was found to be 80% higher than in surrounding sediments. The Bristol Channel, however, presents a case of *S. spinulosa* aggregations having developed in a predominantly sandy environment. The aggregations formed by this worm are also commonly found in association with coarser, mixed gravel deposits, which are known to support a more diverse suite of fauna. Hence the dramatic increase in biodiversity observed in the Bristol Channel is unlikely to be repeated where the reefs have formed on more heterogeneous deposits.

Despite the gaps that exist in our understanding of the fauna associated with sabellariid reefs, some associations are well documented. Crustaceans, for example have been widely reported as showing a preference for sabellariid reefs. Lechapt & Gruet (1993) noted that the

deep water species *Bathysabellaria neocaledoniensis* was associated with pagurids and cirripeds although these associations were based on a small number of observations. In south-east Florida, crustaceans are reported to make up the largest component of reef fauna with at least two species being restricted to the sabellariid reef (Gore *et al* 1978). Crustaceans were also observed in association with *Neosabellaria vitiensis* in Fiji although no analysis of the associated fauna was undertaken (Pohler 2004). In the UK sabellariid reefs have been reported in association with the Ostracod *Hemicythere villosa* (Horne 1982) as well as the pink shrimp *Pandalus montagui* (Warren & Sheldon 1967), the edible crab *Cancer pagurus*, the lobster *Homarus gammarus*, swimming crabs of the genus *Liocarcinus*, squat lobsters *Galathea intermedia* and the porcelain crab *Pisidia longicornis* (Figure 4.23). The widely documented association between Crustacea and sabellariid reefs is perhaps another indication of their stabilising influence since this component of the benthos is usually considered as an indicator of reduced environmental stress (Pearson & Rosenberg 1978).



Figure 4.23 Photographs showing crustaceans associated with *Sabellaria spinulosa* habitats in the UK. A) The swimming crab *Liocarcinus* sp., lobster, *H. gammarus* and numerous squat lobsters, *G. intermedia* B) the edible crab, *C. pagurus* C) the pink shrimp *P. montagui* and D) the long clawed porcelain crab *P. longicornis* associated with *S. spinulosa* aggregations in the southern North Sea © Marine Ecological Surveys Ltd.

ii. Fauna associated with Sabellaria spinulosa reefs used in this study

All of the data used in this study relate to fully subtidal reefs that were not in protected areas at the time of sampling, and therefore it is assumed that all have been subjected to some degree of disturbance, either from current or past commercial fishing, offshore wind farm developments or aggregate extraction. It is acknowledged that mitigation measures are currently being put in place by industry, however, none of the study sites can be considered to be in an unimpacted state.

An earlier study by Pearce *et al* (2011) using data included in the present work, investigated the influence of commercial fishing and aggregate extraction on the communities associated with *S. spinulosa* aggregations identified in the East Coast REC site. This study was unable to find detectable differences between areas subject to different levels of fishing (between 0 and 0.225 Sightings Per Unit Effort (SPUE) or different levels of aggregate extraction (between 0 and >1hr 15 minutes per year). This result is probably because Vessel Monitoring System (VMS) data are only available at a resolution of 0.05 decimal degrees: roughly 5.5 x 3.5km (latitude x longitude) therefore it is impossible to tell whether a $0.1m^2$ grab sample has been taken from an area that was impacted. Furthermore, it is likely that any reef that was directly impacted by these activities, or by aggregate extraction, would have been removed completely.

It is vital that any indicators that are developed for MSFD purposes are able to differentiate between a healthy and a severely impacted reef habitat, and ideally they should also be able to detect smaller changes between these two extremes. For the purpose of this study it is assumed that all of the S. spinulosa aggregations from which data have been collected have been subjected to the same low-level of anthropogenic disturbance; community indicators therefore have been identified through comparisons between reef and non-reef habitats. Non-reef or sedimentary habitats are thought to represent the most extreme form of reef impact - complete physical removal. It is recognised that not all sedimentary habitats included in this analysis will be suitable for S. spinulosa reef development, and therefore may not accurately represent the case of physical removal. However there is insufficient evidence in the literature at this time to determine the precise environmental niche that these reefs occupy. A further extension of this work could usefully investigate this aspect further by making comparisons between reef habitats and different sub-sets of sedimentary samples. In the absence of a robust reef definition that could be applied to the evidence base collated here, comparisons were made between samples in which S. spinulosa was present and absent, between samples with different densities of S. spinulosa and where data were available between areas of acoustically defined S. spinulosa aggregations and the surrounding sedimentary habitats. The sea area and sedimentary habitat in which each of the S. spinulosa habitats occur was also considered because these factors are known to have a significant influence on benthic communities and it is important that these are considered in any stratified monitoring programme design.

iii. The Influence of Sabellaria spinulosa presence on benthic communities

In its most simple form an indicator of associated reef species abundance would need to be able to accurately detect the difference between a *S. spinulosa* reef and a sedimentary habitat. In the following analyses, samples in which *S. spinulosa* were present are compared with samples in which *S. spinulosa* were absent. This approach compares reef with non-reef habitats based on the following two assumptions:

- Heavily impacted S. spinulosa habitats are equivalent to the sedimentary habitats, and
- *S. spinulosa* reef systems will include a range of growth forms and densities from solitary tubes through to densely packed aggregations.

Although none of the *S. spinulosa* habitats included in this study can be considered pristine, it is expected that a good indicator would be able to differentiate between a reef in good condition, a partially impacted reef and a sedimentary habitat. The analyses that follow have been designed with the aim of determining what aspects of these communities are most easily distinguished.

The influence of *S. spinulosa* presence on benthic community composition recorded from grab samples is visualised in Figure 4.24. The Multi-dimensional scaling (MDS) plot suggests some separation in benthic communities in which *S. spinulosa* is present compared to those in which it is absent. The stress level is quite high indicating that this representation may not be completely accurate. Although there is some separation, there is also considerable overlap, indicating that the two communities have many species in common. A PERMANOVA test was carried out on the fourth root transformed abundance data from which *Sabellaria spinulosa* itself had been removed. The broad sea areas (Humber, Thames, Dover and Wight, as delineated by the shipping forecast; Figure 4.11) and sediment classes (Coarse, Mixed, Sand and Mud - UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data) were included in the analysis as a means of testing the influence of *S. spinulosa* presence on macrofaunal communities in the context of other potential community drivers (Table 4.13).



Figure 4.24 Multi-dimensional scaling (MDS) ordination based on Bray-Curtis similarity between fourth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). *S. spinulosa* abundance was removed from the data and colonial epifaunal species which cannot be enumerated are included in the data as present "1" (n= 741) or absent "0" (n= 828).

Table 4.13 Summary of PERMANOVA test carried out on Bray-Curtis similarity between fourth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data but presence of it was used as a factor nested in Sediment (Coarse, Mixed, Sand and Mud - UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data), nested in Sea Area (Humber, Thames, Dover and Wight, as delineated by the shipping forecast; Figure 4.12). Significant test results (at the 5% level) are highlighted with an asterisk. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

Source	df	SS	MS	Pseudo- F	P(Perm)	Unique Permutations
Sea Area	3	46549	15516	2.7561	0.01*	999
Sediment (Sea Area)	16	2.1073E5	13171	1.708	0.007*	996
Sabellaria presence (Sediment (Sea Area))	16	1.6662E5	10414	3.2374	0.001*	995
Residuals	876	2.8179E6	3216.8			
Total	911	3.5707E6	911			

The composition of macrofaunal communities was found to be influenced by sea area, sediment type and by the presence of S. spinulosa (Table 4.13). As a means of investigating this further a SIMPER test was carried out on the data to investigate differences between macrofaunal communities in which S. spinulosa was present and those in which this species was absent. Separate SIMPER tests have been carried out on different sea areas (Tables 4.15 A-D) and in different sedimentary habitats (Tables 4.16 A-D) with a common result as that presented below in Table 4.14. Communities in which S. spinulosa are present have been observed here as being characterised by a higher abundance of macrofaunal species that are otherwise found in low abundances elsewhere. This corroborates the findings of earlier studies of this nature (Pearce et al 2011; 2007). Many of the species that were found in higher abundances where S. spinulosa are present are typical of mixed to coarse sediment where there are surfaces for attachment e.g. the epilithic tubiculous polychaete, Spirobranchus lamarki, and the barnacle. Balanus crenatus, or crevices in which the porcelain crab Pisidia longincornis can inhabit. Other species that were more abundant where S. spinulosa was present are more typical of sedimentary deposits e.g. the polychaete Lumbrineris gracilis which is probably exploiting sediment deposited in gaps within the reef or other micro-habitats created by the S. spinulosa.

Table 4.14 Summary of species contributing to 20% of the dissimilarity between samples containing *Sabellaria spinulosa* (n=475) and those which do not (n=437; from all sea areas and sediment types), derived from a SIMPER test carried out on forth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources as detailed in Table 4.3. Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data and the SIMPER test was carried out using *S. spinulosa* presence / absence as a factor.

Total Dissimilarity 89.55%

Species	S. spinulosa	S. spinulosa	Av.Diss	Diss/SD	Contrib%
	(av. abundance)	(av. abundance)			
	n=475	n=437			
Spirobranchus lamarcki	0.52	1.12	1.85	0.84	2.07
Lumbrineris gracilis	0.41	1.16	1.48	1.15	1.65
Pisidia longicornis	0.14	1.13	1.45	0.92	1.62
Nemertea	0.5	0.94	1.28	0.95	1.43
Notomastus latericeus	0.45	0.7	1.16	0.84	1.3
Ophelia borealis	0.49	0.19	1.09	0.63	1.21
Caulleriella alata	0.34	0.72	1.08	0.89	1.2
Polycirrus spp.	0.33	0.68	1.06	0.83	1.18
Balanus crenatus	0.14	0.74	1.02	0.59	1.14
Actiniaria	0.15	0.66	0.99	0.77	1.1
Spiophanes bombyx	0.42	0.42	0.96	0.74	1.07
Spirobranchus spp.	0.29	0.49	0.95	0.66	1.06
Scalibregma inflatum	0.17	0.62	0.94	0.72	1.05
Glycera lapidum	0.33	0.51	0.93	0.78	1.04
Mediomastus fragilis	0.21	0.66	0.92	0.82	1.02
Ophiuroidea	0.19	0.57	0.89	0.71	1

Table 4.15 Summary of species contributing to 20% of the dissimilarity between samples containing *Sabellaria spinulosa* and those which do not from A) the Humber, B) the Thames, C) Dover and D) Wight sea areas (across all sediment types present), derived from a SIMPER test carried out on forth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources as detailed in Table 4.3. Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data and the SIMPER test was carried out using *S. spinulosa* presence / absence as a factor.

A. Humber – Total Dissimilarity 89.19%

Species	<i>S. spinulosa</i> absent (av. abundance)	<i>S. spinulosa</i> present (av. abundance)	Av.Diss	Diss/SD	Contrib%
	n=78	n=57			
Balanus crenatus	0.37	1.58	1.35	0.91	1.51
Ascidiacea	0.16	1.24	1.03	1.15	1.16
Nematoda	0.53	1.34	1.01	1.26	1.13
Abra alba	0.52	0.98	0.97	0.93	1.09
Lumbrineris gracilis	0.13	1.07	0.96	1.06	1.08
Cirripedia	0.37	1.05	0.94	0.77	1.06
Polycirrus spp.	0.72	1.24	0.92	1.05	1.03
Mediomastus fragilis	0.4	1.05	0.89	1.08	1
Dendrodoa grossularia	0.1	1.05	0.89	0.84	1
Ophelia borealis	0.79	0.21	0.87	0.81	0.98
Urothoe elegans	0.22	0.87	0.83	0.76	0.93
Pholoe baltica	0.36	0.96	0.79	1.12	0.89
Mytilidae	0.08	0.88	0.79	0.72	0.88
Golfingiidae	0.14	0.96	0.79	1.06	0.88

Species	S. spinulosa absent (av. abundance)	<i>S. spinulosa</i> present (av. abundance)	Av.Diss	Diss/SD	Contrib%
Nemertea	0.65	1.12	0.79	0.96	0.88
Ophiuroidea	0.23	0.93	0.77	1.01	0.86
Polynoidae	0.29	0.91	0.73	1.08	0.81
Hiatella arctica	0.13	0.89	0.66	1.02	0.75
Alcyonidium diaphanum	0.22	0.77	0.66	1.01	0.74
Protodorvillea kefersteini	0.16	0.72	0.66	0.86	0.74
Spiophanes bombyx	0.5	0.44	0.64	0.81	0.72
Dipolydora caulleryi	0.09	0.77	0.64	0.9	0.72

B. Thames - Total Dissimilarity 87.83%

Species	<i>S. spinulosa</i> absent (av. abundance)	<i>S. spinulosa</i> present (av. abundance)	Av.Diss	Diss/SD	Contrib%
	n=346	n=224			
Spirobranchus lamarcki	0.64	1.21	2.54	0.9	2.89
Lumbrineris gracilis	0.46	0.99	1.64	1.08	1.87
Nemertea	0.47	0.94	1.63	0.99	1.85
Actiniaria	0.16	0.91	1.51	0.98	1.72
Ophelia borealis	0.47	0.24	1.43	0.66	1.63
Caulleriella alata	0.41	0.73	1.39	0.94	1.58
Notomastus latericeus	0.44	0.53	1.36	0.79	1.55
Spirobranchus spp.	0.36	0.56	1.32	0.73	1.5
Polycirrus spp.	0.24	0.65	1.31	0.82	1.49
Pisidia longicornis	0.11	0.78	1.23	0.79	1.4
Spiophanes bombyx	0.39	0.48	1.23	0.79	1.4
Glycera lapidum	0.34	0.59	1.23	0.86	1.4

C. Dover – Total Dissimilarity 81.59%

Species	S <i>. spinulosa</i> absent (av. abundance)	<i>S. spinulosa</i> present (av. abundance)	Av.Diss	Diss/SD	Contrib%
	n=20	n=120			
Pisidia longicornis	0.68	2.01	2.17	1.37	2.66
Balanus crenatus	1.06	1.39	2.03	1.06	2.49
Spirobranchus lamarcki	0.28	1.07	1.36	1.17	1.67
Lumbrineris gracilis	0.67	1.41	1.32	1.24	1.62
Scalibregma inflatum	0.71	1.24	1.28	1.13	1.57
Galathea intermedia	0.65	0.81	1.2	0.99	1.47
Nemertea	0.35	0.85	1.11	1.04	1.36
Notomastus latericeus	0.36	0.93	1.11	1.13	1.36
Lagis koreni	0.32	0.86	1.11	1.16	1.36
Ampelisca spp.	0.49	0.93	1.09	1.09	1.33
Goniada maculata	0.23	0.84	1.06	1.12	1.3
Harmothoe spp.	0.16	0.88	1.06	1.14	1.3
Poecilochaetus serpens	0.36	0.74	1.02	1.02	1.25

Species	S. spinulosa	S. spinulosa	Av.Diss	Diss/SD	Contrib%
	(av. abundance)	(av. abundance)			
	n=31	n=36			
Balanus crenatus	0.38	1.44	1.16	0.96	1.35
Crepidula fornicata	0.24	1.38	1.05	1.08	1.23
Spirobranchus lamarcki	0.42	1.43	1.03	1.29	1.2
Lumbrineris gracilis	0.4	1.49	1.02	1.47	1.2
Lanice conchilega	0.54	1.07	0.84	1.12	0.99
Maldanidae	0.37	1.15	0.82	1.28	0.96
Notomastus latericeus	0.64	1.34	0.8	1.2	0.93
Dendrodoa grossularia	0.12	1.12	0.78	0.85	0.91
Mediomastus fragilis	0.3	1.09	0.77	1.21	0.91
Amphipholis squamata	0.52	1.16	0.74	1.28	0.86
Pisidia longicornis	0.28	0.99	0.72	1.17	0.84
Caulleriella alata	0.29	0.98	0.7	1.23	0.81
Polycirrus spp.	0.42	1	0.68	1.21	0.8
Galathea intermedia	0.29	0.83	0.67	0.94	0.79
Echinocyamus pusillus	0.51	0.55	0.66	0.8	0.77
Harmothoe spp.	0.24	0.94	0.64	1.29	0.75
Nemertea	0.52	0.94	0.64	1.1	0.74
Nematoda	0.19	0.82	0.63	1.09	0.74
Spiophanes bombyx	0.64	0.43	0.62	0.93	0.72
Aonides	0.52	0.74	0.61	0.98	0.72
paucibranchiata					
Glycera spp.	0.56	0.68	0.6	1	0.7
Asclerocheilus	0.15	0.74	0.59	1.03	0.69
Laonice bahusiensis	0.34	0.67	0.56	0.93	0.66

D. Wight – Total Dissimilarity 85.38%

Table 4.16 Summary of species contributing to 20% of the dissimilarity between samples containing *Sabellaria spinulosa* and those which do not from A) Coarse, B) Mixed, C) Sand and D) Mud sediment (across all sea areas), derived from a SIMPER test carried out on forth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources as detailed in Table 4.3. Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data and the SIMPER test was carried out using *S. spinulosa* presence / absence as a factor.

A. Coarse – Total Dissimilarity 86.32%

Species	<i>S. spinulosa</i> absent (av. abundance)	S. spinulosa present (av. abundance)	Av.Diss	Diss/SD	Contrib%
	n=205	n=248			
Spirobranchus lamarcki	0.75	1.11	1.84	0.88	2.13
Lumbrineris gracilis	0.58	1.1	1.27	1.05	1.47
Nemertea	0.65	0.96	1.15	0.93	1.34
Polycirrus	0.57	0.78	1.15	0.92	1.34
Balanus crenatus	0.28	0.79	1.15	0.66	1.33
Pisidia longicornis	0.21	0.85	1.14	0.85	1.32
Caulleriella alata	0.5	0.72	1.06	0.9	1.23
Spirobranchus	0.45	0.49	1.03	0.73	1.2
Notomastus latericeus	0.45	0.66	1.02	0.88	1.19
Glycera lapidum	0.52	0.57	1.01	0.85	1.17
Aonides paucibranchiata	0.49	0.55	0.98	0.86	1.13
Ophelia borealis	0.47	0.2	0.97	0.63	1.13

Species	<i>S. spinulosa</i> absent (av. abundance)	<i>S. spinulosa</i> present (av. abundance)	Av.Diss	Diss/SD	Contrib%
Spiophanes bombyx	0.44	0.5	0.95	0.8	1.1
Actiniaria	0.21	0.55	0.86	0.74	1
Nematoda	0.34	0.54	0.84	0.77	0.97
Mediomastus fragilis	0.26	0.59	0.82	0.83	0.94
Glycera	0.26	0.42	0.78	0.69	0.9

B. Mixed – Total Dissimilarity 85.70%

Species	<i>S. spinulosa</i> absent (av. abundance)	S. spinulosa present (av. abundance)	Av.Diss	Diss/SD	Contrib%
	n=41	n=89			
Pisidia longicornis	0.19	1.99	1.98	1.2	2.31
Spirobranchus lamarcki	0.51	1.55	1.71	1	1.99
Lumbrineris gracilis	0.75	1.56	1.25	1.15	1.46
Abra alba	0.74	0.85	1.14	0.96	1.33
Scalibregma inflatum	0.43	0.89	1.04	0.93	1.22
Mediomastus fragilis	0.46	0.91	1	1	1.16
Nemertea	0.68	1.01	1	0.98	1.16
Actiniaria	0.19	0.85	0.99	0.93	1.16
Notomastus latericeus	0.76	0.89	0.92	0.97	1.08
Spiophanes bombyx	0.73	0.27	0.91	0.86	1.06
Caulleriella alata	0.49	0.83	0.91	0.99	1.06
Spirobranchus	0.15	0.76	0.9	0.73	1.05
Lagis koreni	0.45	0.6	0.88	0.82	1.03
Balanus crenatus	0	0.87	0.85	0.7	0.99
Harmothoe	0.07	0.79	0.85	0.95	0.99
Goniada maculata	0.36	0.74	0.83	1.01	0.97
Ampelisca spinipes	0.39	0.57	0.8	0.8	0.93

C. Sand – Total Dissimilarity 91.07%

Species	<i>S. spinulosa</i> absent (av. abundance)	<i>S. spinulosa</i> present (av. abundance)	Av.Diss	Diss/SD	Contrib%
	n=159	n=51			
Ophelia borealis	0.74	0.52	2.27	0.79	2.49
Nephtys cirrosa	0.41	0.52	1.97	0.74	2.17
Ophiuroidea	0.16	0.99	1.86	0.9	2.05
Nephtys	0.42	0.52	1.67	0.73	1.84
Nemertea	0.23	0.89	1.61	0.84	1.77
Notomastus latericeus	0.32	0.68	1.44	0.71	1.58
Lumbrineris gracilis	0.08	0.87	1.43	0.89	1.57
Spiophanes bombyx	0.38	0.41	1.29	0.7	1.42
Abra alba	0.23	0.76	1.26	0.69	1.38
Gastrosaccus spinifer	0.17	0.24	1.25	0.49	1.38
Amphipholis squamata	0.03	0.71	1.14	0.68	1.25
Pisidia longicornis	0.01	0.88	1.12	0.65	1.23

Species	S. spinulosa S. spinulosa absent present (av. abundance) (av. abundance)		Av.Diss	Diss/SD	Contrib%
	n=27	n=8			
Scalibregma inflatum	0.5	2.28	3.37	1.25	3.94
Ophiuroidea	0.29	2.02	3.05	1.71	3.57
Ophiura albida	0.19	1.45	2.48	1.41	2.91
Mediomastus fragilis	0.27	1.73	2.4	1.81	2.81
Actiniaria	0.14	1.59	2.35	1.88	2.75
Lagis koreni	0.43	1.41	2.32	1.29	2.71
Abra alba	0.67	1.74	2.28	1.31	2.67

D. Mud – Total Dissimilarity 85.39%

iv. The influence of Sabellaria spinulosa presence on benthic diversity indices

Whilst it is possible to detect differences in community composition associated with the presence of *S. spinulosa*, these may be difficult to separate from the natural variation in the system driven by factors such as sediment type and geographical location. It is therefore useful to understand what (if any) influence the presence of this species has on different diversity metrics as these may ultimately prove to be more consistent monitoring tools. A set of standard diversity indices was calculated for each of the grab samples used in this study, excluding *S. spinulosa* itself. Mean values for each diversity index calculated for samples where *S. spinulosa* were present (n=741) and absent (n=828), are presented in Table 4.17 with a summary of PERMANOVA tests carried out using *S. spinulosa* presence / absence as a factor nested in Sediment (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data), nested in Sea Area (Humber, Thames, Dover and Wight; Figure 4.11).

Table 4.17 shows all indices that differ significantly between samples containing *S. spinulosa* and those that did not. All indices were found to be higher in samples where *S. spinulosa* was present than in samples where *S. spinulosa* was absent with the exception of Pielou's evenness which was found to be lower in the presence of *S. spinulosa*. Several of the diversity indices were also significantly different in different sea areas but were not significantly influenced by sediment type. To explore the relationship between *S. spinulosa* presence, sea area and the different diversity indices, box plots were created as presented in Figure 4.25.

Table 4.17 Summary of PERMANOVA test carried out on Bray-Curtis similarity between the diversity indices S (number of taxa), N (number of individuals), d' (Margalefs richness), H' (Shannon's diversity), J' (Pielou's evenness), and 1- λ (Simpson's diversity) calculated from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present "1" or absent "0". *Sabellaria spinulosa* abundance has been removed from the data and the PERMANOVA test was carried out using *Sabellaria spinulosa* presence as a factor nested in Sediment (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data), nested in Sea Area (as delineated by the shipping forecast). Significant test results (at the 5% level) are depicted by an asterisk. Also shown are the mean values for each index across each of the *S. spinulosa* groups (present and absent).

		S	Ν	ď	H'	J'	1-λ
Mean	Sabellaria absent	17.43	77.84	4.14	2.05	0.81	0.82
	Sabellaria present	45.74	277.45	8.35	2.67	0.75	0.84
P(Perm)	Sea Area	0.021*	0.047*	0.008*	0.234*	0.538	0.889
. ,	Sediment (Sea Area)	0.785	0.735	0.824	0.796	0.473	0.756
	Sabellaria presence	0.001*	0.001*	0.001*	0.001*	0.002*	0.026*
	(Sediment (Sea Area))						

The box plots presented in Figure 4.25 show that there is a clear increase in the total abundance (N), the number of taxa (S), Margalef's richness (d') and Shannon Weiner's diversity (H') in samples which contain *S. spinulosa*. Conversely there is a slight decrease in Pielou's evenness in samples where *S. spinulosa* is present although the difference in this case is not as marked as the preceding diversity indices. Only a very slight difference is observed in Simpson's diversity $(1-\lambda)$ between samples that contain *S. spinulosa* and those which did not.

Of the indices explored, total abundance (N), number of taxa (S) and Margalef's richness (d') showed the greatest degree of difference between samples which contained *S. spinulosa* and those which did not. Furthermore, the observed difference was relatively consistent across all of the sea areas included in the analysis. The one exception to this was the Thames area where the difference between diversity indices calculated for samples in which *S. spinulosa* was present and those in which it was not, was less marked. This could be a reflection of a higher level of anthropogenic and natural disturbance associated with the Thames Estuary, one of the largest in the UK. Nevertheless, this difference does highlight the need to consider the local environment when choosing and applying an environmental indicator.



Figure 4.25 Diversity indices S (number of taxa), N (number of individuals), d' (Margalefs richness), J' (Pielou's evenness), H' (Shannon's diversity) and 1- λ (Simpson's diversity) calculated for benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources as detailed in Table 4.3. *Sabellaria spinulosa* has been removed from the data to allow for comparisons between samples in which this species was present or absent. Box plots show interquartile range, median and maximum / minimum observed values as whiskers (1.5 times the interquartile region). Black dots indicate values above and below the 1.5 times the 3rd and 1st quartiles, respectively.

v. Influence of Sabellaria spinulosa density on the associated benthic community

The MDS plot in Figure 4.26 shows a gradation, which runs approximately left to right, between samples that contained no *Sabellaria spinulosa* and those that contained the highest densities (>1000 0.1m⁻²). The level of dispersion between samples within each density category appears to decrease as the density of *S. spinulosa* increases, indicating that reef communities may become increasingly uniform as they develop. It should be noted though that this pattern may in part be a result of the difference in the number of samples belonging to each category, with the largest number of samples having no *S. spinulosa*

present and the smallest number of samples having >1000 *S. spinulosa* 0.1m⁻² present. To ascertain the statistical significance of the observed differences between communities in which *S. spinulosa* is present in different densities, a PERMANOVA test was carried out on the fourth root transformed abundance data from which *Sabellaria spinulosa* itself had been removed. Sea area (as defined by the shipping forecast) and sediment class (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution data) were included in the test as a means of placing the influence of *S. spinulosa* density on macrofaunal communities in the context of other known community drivers. The results of this test are summarised in Table 4.18.



Figure 4.26 Multi-dimensional scaling (MDS) ordination based on Bray-Curtis similarity between fourth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *Sabellaria spinulosa* abundance has been removed from the data and the MDS plot has been labelled according to the abundance of this species in each of the samples.

Table 4.18 Summary of PERMANOVA test carried out on Bray-Curtis similarity between fourth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data and the PERMANOVA test was carried out using *S. spinulosa* density as a factor nested in sediment (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data), nested in sea area (as delineated by the shipping forecast). Significant test results (at the 5% level) are depicted by an asterisk. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

Source	df	SS	MS	Pseudo- F	P(Perm)	Unique Permutations
Sea Area	3	46515	15505	3.3935	0.001*	998
Sediment (Sea Area)	16	89293	5580.8	1.3337	0.062	995
Sabellaria Density (Sediment (Sea Area))	43	2.8293E5	6579.9	2.0678	0.001*	990
Residual	849	2.7016E6	3182.1			
Total	911	3.5707E6				

The composition of macrofaunal communities was influenced by sea area and by the density of *S. spinulosa* (Table 4.18). A series of SIMPER tests were carried out on the data to investigate differences between macrofaunal communities in which *S. spinulosa* is present at different densities. It should be noted that there is significant difference in macrofaunal communities occurring in different sea areas and hence the patterns observed may vary from site to site.

The results of the SIMPER tests (Table 4.19) showed a general gradation with most of the characteristic species increasing in abundance in parallel with the increase in S. spinulosa density. Most notably, the porcelain crab Pisidia longicornis, which increased from an average abundance of 0.57 individuals per grab where there was no S. spinulosa to an average of 274.88 individuals per grab in samples that contain over 1,000 S. spinulosa. Both macrofaunal diversity and the abundance of *P. longicornis* are likely to be linked to the level of habitat heterogeneity created by the reef forming species rather than the number of living worms. The abundance of this species may therefore be a good indicator of reef condition. Some encrusting species such as the barnacle B. crenatus, the tubiculous polychaete S. lamarki and the ascidian Dendrodoa grossularia were present in their highest abundances at intermediate S. spinulosa densities, indicating that there is less scope for epifaunal colonisation as the reef becomes more consolidated. This is perhaps unsurprising because there is significant niche overlap between these species. What is perhaps more surprising is the sharp increase in some species more typical of sedimentary habitats such as the bivalve mollusc Abra alba and the terebellid polychaete Polycirrus spp. This suggests sediment accumulation within the reef as it develops, which may in part be composed of faecal material produced by S. spinulosa.

Table 4.19 Summary of the average abundance of key macrobenthic species which characterise the dissimilarity between the five *Sabellaria spinulosa* density categories, derived from a SIMPER test carried out on forth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present "1" or absent "0". *S. spinulosa* abundance has been removed from the data and the SIMPER test was carried out using *S. spinulosa* density as a factor.

Key macrobenthic species	0 Av.Abund	Sabellaria 1-19 Av.Abund	density 20-99 Av.Abund	categories 100-999 Av.Abund	1000+ Av.Abund
Pisidia longicornis	0.14	0.78	1.57	2.76	3.58
Spirobranchus lamarcki	0.52	1.14	1.21	0.93	0.43
Lumbrineris gracilis	0.41	1.02	1.42	1.71	2.01
Nematoda	0.23	0.44	0.72	0.81	1.82
Abra alba	0.26	0.39	0.65	1.17	1.94
Balanus crenatus	0.14	0.68	1.14	0.67	0.16
Ophiuroidea	0.19	0.43	0.81	1.12	1.93
Nemertea	0.5	0.84	1.05	1.35	1.89
Ampharete finmarchica	0.09	0.31	0.45	0.57	1.33
Actiniaria	0.15	0.54	0.83	1.1	1.77
Mytilidae	0.11	0.24	0.39	0.88	1.45
Notomastus latericeus	0.45	0.62	0.94	0.89	1.21
Harmothoe spp.	0.03	0.29	0.76	0.95	0.99
Amphipholis squamata	0.16	0.46	0.79	1.05	1.73
Polycirrus spp.	0.33	0.61	0.75	0.96	1.62

vi. Influence of different densities of *Sabellaria spinulosa* on macrobenthic diversity

As in the previous set of analyses, comparisons were made between diversity indices calculated for samples in which *S. spinulosa* was present in different densities across the five different sea areas (as delineated by the shipping forecast) and within different sedimentary habitats (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data). The PERMANOVA test results presented overleaf in Table 4.20 show that all of the indices were found to be significantly different between samples containing different densities of *S. spinulosa* (at the 0.05% significance level). A number of the indices (S, N and d') were also found to be significantly different in different sea areas. None of the indices were influenced by sediment type. Pairwise tests were carried out to investigate the differences observed between sea areas in S, N and d' (Table 4.21) the pairwise tests show that there were no significant differences in these diversity indices between the Thames and Dover, Dover and Humber and Humber and Wight. All three species diversity indices were found to be significantly different between the Thames and Wight sea areas, indicating that these areas are most different from one another.

Table 4.20 Summary of PERMANOVA test carried out on Bray-Curtis similarity between the diversity indices S (number of taxa), N (number of individuals), d' (Margalefs richness), H' (Shannon's diversity), J' (Pielou's evenness) and 1- λ (Simpson's diversity) calculated from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data and the PERMANOVA test was carried out using *S. spinulosa* density categories as a factor nested in sediment (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data), nested in sea area (as delineated by the shipping forecast). Significant test results (at the 5% level) are depicted by an asterisk. Also shown are the mean values for each index across each of the *Sabellaria* density categories.

		S	Ν	ď	H'	J'	1-λ
Mean	0	17.43	77.84	4.14	2.05	0.81	0.82
	1-19	39.38	200.92	7.60	2.62	0.77	0.84
	20-99	67.89	628.74	10.65	2.70	0.65	0.80
	100-999	60.43	372.41	10.39	2.85	0.72	0.84
	1000+	72.25	891.88	10.75	2.99	0.70	0.87
P(Perm)	Sea Area	0.013*	0.026*	0.046*	0.804	0.089	0.252
	Sediment (Sea Area)	0.7	0.826	0.519	0.623	0.539	0.63
	<i>Sabellaria</i> density (Sediment (Sea Area))	0.001*	0.001*	0.001*	0.001*	0.001*	0.019*

Table 4.21 Summary of sea area Pairwise tests carried out on Bray-Curtis similarity between the diversity indices S (number of taxa), N (number of individuals) and d' (Margalefs richness), calculated from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data. Significant test results (at the 5% level) are depicted by an asterisk.

Sea Area Comparisons	5	6	1	N	c	'	
	t	P(perm)	t	P(Perm	t	P(Perm)	
Thames, Dover	0.94819	0.384	0.66865	0.631	1.0184	0.32	
Thames, Humber	2.0543	0.047*	1.5802	0.096	2.0269	0.064	
Thames, Wight	2.9873	0.009*	2.8628	0.008*	2.5462	0.032*	
Dover, Humber	1.4694	0.134	1.4022	0.142	1.3081	0.209	
Dover, Wight	2.3118	0.019*	2.6755	0.006*	1.7341	0.1	
Humber, Wight	0.89786	0.409	0.71557	0.617	0.63899	0.619	

As a means of exploring the relationship between diversity indices, *S. spinulosa* densities and sea areas further, the data were plotted in a series of boxplots as presented in Figure 4.27. The box plots reveal that the Wight sea area has a higher species diversity than the Thames area in terms of S, N and d'. The box plots (Figure 4.27) further demonstrate the general trend for diversity indices to increase in parallel with the increasing densities. Pielou's evenness shows the opposite trend, and generally decreases in parallel to the increasing *S. spinulosa* density, whilst Simpson's diversity does not show a very strong trend at all. The total macrofaunal abundance (N) was again very variable although it also shows the greatest degree of difference between *S. spinulosa* densities.



Figure 4.27 Diversity indices S (number of taxa), N (number of individuals), d (Margalef's richness), J (Pielou's evenness), H' (Shannon's diversity) and 1- λ (Simpson's diversity) calculated for benthic abundance data from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). *Sabellaria spinulosa* has been removed from the data to allow for comparisons between samples containing different densities of this species. The box represents the interguartile range, with a line indicating the median and whiskers extending to the maximum and minimum observed values.

vii. Differences in benthic communities sampled within and outside acoustically defined *Sabellaria spinulosa* reefs

Arguably the best way to examine the differences between the communities associated with *S. spinulosa* reefs and those that are not is to look at samples taken from within and outside defined *S. spinulosa* reefs. A small number of studies have used very high resolution acoustic data to identify the boundaries of *S. spinulosa* aggregations that are likely to be considered as reefs in terms of both the Habitats Directive and the OSPAR threatened and declining habitat definitions, since they represent relatively extensive, topographically distinct seabed features formed by *S. spinulosa*.

Where these habitats have been defined it is possible to make comparisons with the surrounding sedimentary habitats and this allows us to get a much more accurate measure of any differences that exist between reef and non-reef habitats as well as the variability that exists within one particular reef system. The latter is of particular importance to the development of indicators because it is imperative that impacts can be detected against the background of natural variability.

Polygons for *S. spinulosa* aggregation were available for the Hastings Shingle Bank study site (Pearce *et al* 2007), the Thanet offshore wind farm site (Marine Ecological Surveys Ltd 2005, 2007, 2013) and the East Coast REC (Pearce *et al* 2011). In all three of these studies high resolution acoustic data (side-scan sonar and / or multibeam bathymetry data) were used to delineate the extent of the aggregations based on the presence of an irregular texturing. The aggregations had been ground-truthed with a combination of seabed imagery and grab sampling.

A series of MDS plots (Figure 4.28) show some differences between grab samples taken within and outside acoustically defined reef areas, although there is significant overlap indicating that there are many species in common. There is a greater level of dispersion associated with the samples taken outside of the reef (PERMDISP: mean deviation from centroid = 64) than was observed in the samples which were taken within the reef (PERMDISP: mean deviation from centroid = 58) (Figure 4.28). This pattern can be explained in part by the fact that the sedimentary habitats sampled outside of the reef are in some cases quite variable and include both sandy habitats that naturally support a depauperate infaunal community and mixed gravelly sediments which naturally support a higher diversity of infauna in these areas.

A PERMANOVA test was carried out on the fourth root transformed abundance data from which *S. spinulosa* itself had been removed (Table 4.22). Sea area (as defined by the shipping forecast) and sediment class (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution data) were included in the test as a means of placing the influence of *S. spinulosa* reef on macrofaunal communities in the context of other known community drivers. The composition of macrofaunal communities sampled from acoustically defined areas of *S. spinulosa* reef was significantly different from communities sampled from adjacent sedimentary habitats (Tables 4.22 & 4.23). As a means of investigating this further a SIMPER test was carried out (Table 4.24).



Figure 4.28 Multi-dimensional scaling (MDS) ordinations based on Bray-Curtis similarity between fourth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from Hastings Shingle Bank, the Thanet Offshore Wind Farm site and the East Coast REC area where reefs were delineated using high resolution acoustic data (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *Sabellaria spinulosa* abundance has been removed from the data and the MDS plot has been labelled according to the whether the sample was taken from within (blue) or outside (grey) acoustically defined *S. spinulosa* reefs.

Table 4.22 Summary of PERMANOVA test carried out on Bray-Curtis similarity between fourth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from Hastings Shingle Bank, the Thanet Offshore Wind Farm site and the East Coast REC area where reefs were delineated using high resolution acoustic data (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *Sabellaria spinulosa* abundance has been removed from the data and the PERMANOVA test was carried out using acoustic reef classification as a factor nested in Sediment (UKSeaMap EUNIS Level 3 sediment classes determined from Particle Size Distribution (PSD) data), nested in Sea Area (as delineated by the shipping forecast). Significant test results (at the 5% level) are depicted by an asterisk. Df=degrees of freedom; SS= sums of squares; MS=mean squares; Pseudo-F=Fisher's ratio; P=probability associated with pseudo-F value (Anderson 2005).

Source	df	SS	MS	Pseudo-	P(Perm)	Unique
				F		Permutations
Sea Area	2	45608	22804	4.8118	0.001*	998
Sediment (Sea Area)	11	70689	6426.3	0.99134	0.522	995
Acoustic Class (Sediment (Sea Area))	11	91078	8279.8	2.529	0.001*	994
Residual	422	1.3816E6	3273.9			
Total	446	1.764E6				

Table 4.23 Summary of Pairwise tests carried out on Bray-Curtis similarity between the forth root transformed species abundance data from 0.1m² Hamon grab samples collected from a variety of sources (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data. Significant test results (at the 5% level) are depicted by an asterisk. Pairwise tests were carried out on the random factor acoustic class, nested in sediment type, nested in sea area. Note that pairwise tests are not recommended for random factors and hence these results should be treated with some caution.

Sea Area	Sediment type	t	P(perm)
Thames	Coarse	1.3889	0.005*
	Mixed	1.3228	0.028*
	Sand	3.1216	0.001*
	Mud	2.101	0.001*
Dover	Coarse	1.6804	0.001*
	Mixed	1.515	0.004*
	Sand	0.9004	0.676

The results of the SIMPER test between acoustically defined reef areas and adjacent sedimentary habitats, across all sea areas (Table 4.24) shows that the reef habitats are characterised by a higher abundance of macrofaunal species that are otherwise found in low abundances in the adjacent sediments. As there was also a significant difference in sea areas (Table 4.24), separate SIMPER tests have been carried out to investigate the differences between the macrofaunal communities sampled within and outside acoustically defined areas of reef in the Thames and Dover (Table 4.25 A & B respectively).

Many of the species which are found in higher abundances on the *S. spinulosa* reefs are typical of mixed to coarse sediment where there are surfaces for attachment, e.g. the tubiculous polychaete, *S. lamarki*, and the barnacle, *B. crenatus*, or crevices, e.g. *P. longicornis*. Other species that were more abundant where *S. spinulosa* was present are more typical of sand deposits such as the interstitial polychaetes *Scalibregma inflatum* and *L. gracilis*. These species may be taking advantage of the sediment deposits that can accumulate in gaps within the reef. However, they may also be associated with sedimentary deposits in which *S. spinulosa* are only present as solitary individuals or small clumps since acoustically defined reef areas undoubtedly contain all of the different stages in reef development.

It should be noted that there is significant difference in macrofaunal communities occurring in different sea areas and hence the SIMPER test has been repeated using data from the Thames and Dover sea areas separately (Table 4.25 A & B). There are some regional differences in the fauna associated with acoustically defined *S. spinulosa* reef habitats (Tables 4.25 A & B) and more specifically in the fauna that characterise the differences between reef and non-reef habitats. *S. spinulosa* reefs in the Thames sea area are characterised by a high abundance of anemones (Actiniaria), the porcelain crab, *P. longicornis*, and the bivalve *A. alba. S. spinulosa* reefs in the Dover sea area are characterised by a much higher abundance of *P. longicornis*, than the reefs sampled from the Thames area (139 and 15 respectively) and the reef abundance of porcelain crab in the Thames area is in fact very similar to non-reef abundances of this species in the Dover area emphasising the importance of understanding the geographical variation in these communities. In contrast, anemones do not seem to be an important component of reef communities in Dover sea area.

Table 4.24 Summary of species contributing to 20% of the dissimilarity between samples collected from within and outside acoustically defined areas of *Sabellaria spinulosa* reef. The data are derived from a SIMPER test carried out on forth root transformed benthic abundance data from 0.1m² Hamon grab samples collected from Hastings Shingle Bank, the Thanet Offshore Wind Farm site and the East Coast REC area where reefs were delineated using high resolution acoustic data, as detailed in Table 4.3. Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data and the SIMPER test was carried out using acoustic reef classification as a factor.

Таха	Outside	Inside reef	Av.Diss	Diss/S	Contrib%	Cum.%
	reef areas	areas		D		
	Av. abund.	AV. abund.				
	n=289	n=158				
Pisidia longicornis	0.36	1.56	1.84	1.05	2.07	2.07
Lumbrineris gracilis	0.47	1.27	1.63	1.09	1.84	3.9
Nemertea	0.39	1.04	1.51	0.91	1.7	5.6
Notomastus Iatericeus	0.49	0.85	1.37	0.79	1.54	7.14
Ophelia borealis	0.56	0.23	1.36	0.62	1.52	8.66
Scalibregma inflatum	0.38	0.92	1.31	0.94	1.47	10.14
Balanus crenatus	0.33	0.66	1.21	0.58	1.36	11.5
Caulleriella alata	0.26	0.77	1.18	0.79	1.33	12.83
Ophiuroidea	0.27	0.75	1.14	0.79	1.28	14.1
Spirobranchus Iamarcki	0.31	0.73	1.12	0.81	1.25	15.36
Mediomastus fragilis	0.17	0.75	1.09	0.84	1.22	16.58
Lagis koreni	0.28	0.73	1.08	0.88	1.21	17.79
Actiniaria	0.16	0.78	1.06	0.77	1.2	18.99
Nephtys spp.	0.29	0.54	1	0.68	1.13	20.11

Table 4.25 Summary of species contributing to 20% of the dissimilarity between samples collected from within and outside acoustically defined areas of *Sabellaria spinulosa* reef. Based on a SIMPER test carried out on untransformed benthic abundance data from 0.1m² Hamon grab samples collected from A) the Thames (East Coast REC and Thanet Offshore Wind Farm) and B) Dover (Hastings Shingle Bank) sea areas where reefs were delineated using high resolution acoustic data (Table 4.3). Colonial epifaunal species which cannot be enumerated are included in the data as present '1' or absent '0'. *S. spinulosa* abundance has been removed from the data and the SIMPER test was carried out using acoustic reef classification as a factor.

A) Thames

Species	Outside reef areas Av.Abund	Inside reef areas Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
	n=231	n=76				
Nemertea	0.33	1.19	2.04	0.97	2.26	2.26
Ophelia borealis	0.67	0.37	1.93	0.7	2.14	4.4
Lumbrineris gracilis	0.31	1.08	1.8	0.95	2	6.4
Actiniaria	0.15	1.22	1.73	1.03	1.92	8.32
Ophiuroidea	0.27	1.11	1.72	0.97	1.91	10.23
Notomastus Iatericeus	0.45	0.72	1.65	0.72	1.83	12.06
Caulleriella alata	0.22	0.8	1.52	0.75	1.68	13.74
Mediomastus fragilis	0.15	0.89	1.51	0.93	1.68	15.42
Abra alba	0.24	0.91	1.42	0.76	1.58	17
Polycirrus spp.	0.15	0.86	1.41	0.83	1.56	18.56
Pholoe baltica	0.09	0.9	1.32	0.96	1.47	20.03

Species	Outside reef areas Av.Abund	Inside reef areas Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
	n=58	n=82				
Pisidia longicornis	1.29	2.19	1.75	1.3	2.37	2.37
Balanus crenatus	1.6	1.16	1.59	1.02	2.15	4.52
Spirobranchus Iamarcki	0.74	1.11	1.01	1.09	1.36	5.88
Scalibregma inflatum	1.09	1.21	0.94	0.97	1.27	7.15
Galathea intermedia	0.84	0.75	0.93	0.97	1.26	8.41
Notomastus Iatericeus	0.68	0.96	0.9	1.07	1.21	9.62
Nemertea	0.63	0.89	0.89	1	1.21	10.82
Harmothoe spp.	0.5	0.97	0.89	1.15	1.2	12.03
Lumbrineris gracilis	1.1	1.46	0.88	0.98	1.18	13.21
Goniada maculata	0.55	0.9	0.84	1.07	1.14	14.35
Ampelisca spp.	0.77	0.94	0.84	1	1.14	15.49
Poecilochaetus serpens	0.67	0.7	0.83	1	1.12	16.6
Ampharete finmarchica	0.7	0.49	0.82	0.92	1.11	17.71
Echinocyamus pusillus	0.71	0.47	0.81	0.91	1.09	18.81
Crepidula fornicata	0.37	0.78	0.81	1.01	1.09	19.9
Lagis koreni	0.62	0.9	0.81	1.01	1.09	21

B) Dover

4.6.2 Suitability of seabed imagery for acquiring diversity indices of the fauna associated with *S. spinulosa* reefs

Earlier sections of this Chapter determined that *S. spinulosa* density counted from grabs and tube counts counted from images, is positively correlated with the diversity of fauna associated with these habitats (grabs: $r_s = 0.42-0.66$, P<0.001; imagery: $r_s = 0.17-0.32$, P<0.01-0.001). The density of *S. spinulosa* could therefore be used as a proxy for the diversity of associated fauna, especially if measured from grab samples.

i. *Pisidia longicornis* as a proxy for the diversity of fauna associated with *S. spinulosa* reef habitats.

All of the analyses detailed above have highlighted the strong affinity between *P. longicornis* and *S. spinulosa* reefs investigated in this study. An affinity between *S. spinulosa* reefs and the pink shrimp, *Pandalus montagui*, has also been reported in the literature (Warren & Sheldon 1967) although the relationships are unlikely to be mutually exclusive since high abundances of *P. longicornis* have been recorded in association with reefs that have also been reported to support high abundances of pink shrimp. Since pink shrimp are highly mobile and are only effectively sampled by trawling, they are unlikely to make a good indicator for use in MSFD monitoring. Conversely, *P. longicornis* is most effectively sampled with sediment grabs and could therefore have significant potential as an indicator of *S. spinulosa* reef condition.

There is evidence that the relationship between *S. spinulosa* habitats and *P. longicornis* may differ geographically (Tables 4.25 A & B) and it is possible that this species is absent from areas not included in this study. However, there is a relatively consistent relationship between the different aspects measured to identify *S. spinulosa* reef (presence / absence,

diversity classes and acoustically defined reefs) explored in this study and high abundances of *P. longicornis* and this warrants further investigation. It also seems likely that this species could be an effective proxy for the macrofaunal diversity associated with reefs because its affinity with the reef structure stems from its use of crevices. Therefore, unlike the density of living *S. spinulosa*, we might expect there to be a strong correlation between abundance of *P. longicornis* and the other fauna utilising the complex reef structure.

To test this hypothesis Spearman's rank correlation values were calculated for *P. longicornis* abundance and a range of diversity indices derived from the macrofaunal grab data (with *S. spinulosa* and *P. longicornis* removed), and these are summarised below in Table 4.26. This analysis was limited to samples in which *S. spinulosa* were present (n= 388). Regression plots were also constructed for *P. longicornis* abundance and the total abundance (N) and number of taxa (S) recorded in each grab sample (Figure 4.29). All of the diversity indices show a significant positive correlation with *P. longicornis* abundance, with the exception of Pielou's evenness which is perhaps unsurprising as *P. longicornis* itself was contributing significantly to the patterns observed in evenness under different densities of the reef forming polychaete *S. spinulosa*. The strength of the correlation between *P. longicornis* abundance and macrofaunal diversity indices is higher than that observed between measures of density (tube counts) and diversity ($r_s = 0.17-0.32$) from images but slightly lower than the correlation observed between measures of *S. spinulosa* density and macrofaunal diversity from grabs ($r_s = 0.42-0.66$).

Although the variation in diversity and abundance of fauna associated with the *S. spinulosa* reefs is poorly explained by the variation in *P. longicornis* abundance (Figure 4.29), the effort required to obtain abundance estimates of *P. longicornis* is substantially less than that for obtaining full faunal abundance data and hence this may well be a useful rapid assessment tool for monitoring the community condition of *S. spinulosa* reefs.

Table 4.26 Summary of the Spearman's correlation values (df=910) calculated for *Pisidia longicornis* abudance and diversity indices calculated for the macrofauna identified in the same $0.1m^2$ Hamon grab samples. *S. spinulosa* and *P. longicornis* were removed from the data before diversity indices were calculated. Spearman's rank correlations (r_s) range from -1 (perfect negative correlation) to +1 (perfect positive correlation) with a zero value representing no correlation in the data at all. Statistically significant correlations (P<0.05) are marked with an asterisk.

Diversity Index	r _s	Р
Number of individuals (N)	0.39	<0.001*
Number of taxa (S)	0.47	<0.001*
Margalef's richness (d)	0.46	<0.001*
Shannon-Wiener's diversity (H')	0.36	<0.001*
Pielou's evenness (J)	-0.08	0.07
Simpson's lambda (1-λ)	0.13	0.005*



Figure 4.29 Regression plots showing the relationship between *Pisidia longicornis* density and A) Total abundance of individuals (N), B) Total number of taxa (S), C) Margalef's richness (d), D) Shannon-Wiener's diversity (H'), E) Pielou's Evenness (J) and F) Simpson's index (1- λ) recorded from same 0.1m² Hamon grab samples in which *S. spinulosa* were present. Red lines represent non-parametric lowess regression curves added to help visualise the trends.

4.6.3 Sampling required to detect changes in the diversity of *S. spinulosa* biogenic reefs

A power analysis (Sheppard 1999) of diversity index data was used to determine the number of replicate samples needed to detect a given change in the mean using $0.1m^2$ grab samples. Simpson's diversity $(1-\lambda)$ was excluded from this analysis because this index was not found to be significantly different between reef and non-reef habitats. All datasets used in the analyses originated from potential *S. spinulosa* habitat covering the following areas (see also Figures 4.2 & 4.4): (1) Humber REC, 14,000km² of predicted *S. spinulosa* biotope; (2) East Coast REC and LINCS windfarm, 55km² of ground-truthed acoustic reefs; and (3) Thames area, 184km² (Outer Thames REC, Cutline 447 and Thanet windfarm). The number of samples was calculated for the detection of 10, 20, 30, 40 and 50% changes in mean species diversity at the 5% significance level. The power of the test is defined as 1- β under the standard significance assumptions (α =0.05; the probability of making a Type II error (β) was 0.05).

Results displayed in Figure 4.30 indicate that 70 to 105 samples would be needed to detect a 10% change in the mean diversity of associated communities (H) using grab samples depending on the density of the reef. This figure fell considerably to between 20 and 30

samples when 20% change detection was required. If changes in total abundance of organisms was required (N) then the number of samples required for a 10% change detection would need to increase to more than 700 samples depending on the type of reef targeted. This figure fell considerably to less than 600 samples for all reef types when 20% change detection was required. All other diversity indices fell between the extremes of sampling requirements indicated for diversity (H) and total abundance (N).

The quantity of sampling required is within the realms expected for a monitoring programme over moderate spatial scales. It is likely that more stratified sampling to target specific areas or reef sub-types (i.e. patchy elevated aggregations or extensive, low relief crusts) will also improve power by reducing the intrinsicly high variance resulting from reef fragmentation.



Figure 4.30 Sampling effort estimations for grab sampling to detect changes in mean *Sabellaria spinulosa* diversity indices in reefs sampled as part of marine aggregate and windfarm development environmental surveys. The probability level is set at 0.05. Power analysis used minimum power $(1-\beta)$ set at 0.95. Arbitrary categories: Sparse (< 300 worms m²); Dense (>300 worms m²).

Box 4.7. Summary of findings for Sabellaria spinulosa reef diversity indicators.

- In the absence of an operational *S. spinulosa* reef definition that could be applied consistently to the available data it was necessary to use the following three approaches to identifying/categorising reef:
 - 1. Presence of *S. spinulosa*
 - 2. Categorised densities of *S. spinulosa* (0, 1-19, 20-99, 100-999, 1000+m⁻²)
 - 3. Acoustically distinguishable S. spinulosa reef areas.

Diversity and composition of fauna typically associated with S. spinulosa reefs

- The limited published literature on *S. spinulosa* reef diversity indicates that the diversity of fauna associated with these reefs is very variable;
- Using the three approaches to defining reef (above) there was an increase in the number of species (S), total abundance (N), species richness (d') and Shannon's diversity (H') attributed to the presence of this habitat. Pielou's evenness, a measure of the equitability of species abundance, showed a significant decline in the presence of reef, whilst in some cases no differences were detected;
- The composition of the species associated with *S. spinulosa* reefs varied geographically and in terms of the sediments upon which the reefs had developed. Reef communities were found to be characterised by an increased abundance of fauna that were typically more sporadic in adjacent sedimentary habitats, including epifaunal and crevice dwelling species as well as species that are more typically associated with areas of fine sediment.

Suitability of seabed imagery and grab sampling for acquiring diversity measures

- Earlier sections of this Chapter determined that *S. spinulosa* density, derived from grabs or from tube counts from imagery, is positively correlated with the diversity of fauna associated with these habitats. However, the strength of these relationships was markedly different. Density measurements from grabs were moderately correlated with diversity those from seabed imagery were only weakly correlated;
- A significant positive correlation was also identified between the abundance of *Pisidia longicornis* and the diversity and richness of fauna associated with *S. spinulosa* reefs;

Level of sampling required to detect changes in S. spinulosa reef diversity

- The level of grab sampling required to detect a 10-30% change in *S. spinulosa* reefs is likely to be prohibitively expensive and damage the reef (n=30-200+ grab sampled per reef);
- Shannon's diversity (H') and Pielou's Evenness (J') are the most effective community metrics [diversity indices] to monitor because they are subjected to the lowest variance.
Box 4.8. Recommendations for Sabellaria spinulosa reef diversity indicators.

- Reef areas defined by high resolution acoustic data are thought to be the best way to identify the presence of a reef for sampling. However, such data were only available from three sites. Further work to classify *S. spinulosa* reef habitats using acoustic methods has been identified here as a key research priority for the advancement of MSFD indicator development;
- Incorporating a measure of species equitability into a monitoring programme is recommended, because it will reveal important information about the developmental stage of the reef. Certain species become very dominant (reducing species equitability) only in the most developed growth stages;
- S. spinulosa density measures and *P. longicornis* abundance both represent potential proxy measures for the rapid assessment of reef diversity. However, it is recommended that these rapid assessment measures are used to complement full species counts from grab samples rather than to replace them because compositional changes in the community may constitute an early sign of stress could otherwise be missed. However, evidence that community composition changes in response to human pressures is still lacking;
- The variance in species diversity indices presented here is likely to have been inflated through imperfect identification of *S. spinulosa* reef (e.g. presence / absence of *S. spinulosa*). A key priority moving forward should therefore be to undertake more stratified sampling using remote sensing data and more extensive seabed imagery to investigate reef structure and in particular reef patchiness and how this influences species diversity indices.

4.7 Validation of Sabellaria spinulosa reef indicators

The preceding sections of this Chapter investigated the variation in *S. spinulosa* density and the macrofaunal communities associated with *S. spinulosa* reef habitats and potential indices that could be used to monitor these habitats. These indicators have been highlighted because:

- 1. They have been shown to respond to differences between reef and non-reef habitats.
- 2. They reflect the biodiversity qualities of the reef.
- 3. The variance in these indicators is low enough for us to have some confidence in detecting change with an acceptable level of sampling (as suggested by power analyses).

However, all of evidence used in this study has been collected from reefs that are already subject to some anthropogenic disturbance and we do not yet have a good grasp on what an unimpacted reef might look like. There is therefore some way to go before we can be certain the proposed indicators will be able to detect anthropogenic impact and therefore link closely to any particular management measure. Additional field studies on *S. spinulosa* were beyond the scope of the present study and in the absence of any good "baseline" reef data, or any good quality impact studies, the project team have reviewed the literature relating not only to *S. spinulosa* reefs but also reefs created by conspecifics elsewhere in the world. Through this review the evidence-base was evaluated for the types and levels of changes that might be expected to occur in response to different types of anthropogenic activities. This review was supported by a short study using extant data to test the hypothesis that the quality of *S. spinulosa* reef habitats in terms of the density of worms and diversity of

associated fauna would be correlated with the level of anthropogenic disturbance that the reef was exposed to. In the absence of any 'before and after' data from *S. spinulosa* reefs this was the best way of determining the relationship between definite impact and the response of the potential indicators.

4.7.1 Sensitivity of S. spinulosa reefs to anthropogenic disturbance

By their very nature biogenic reefs formed by polychaete worms are thought to be sensitive to physical loss and damage in the marine environment (Holt *et al* 1998). Their sessile nature also makes them more vulnerable to changes in the environment than mobile animals which have the capacity to move away from unfavourable conditions. The occurrence of sabellariid reefs in areas that are also utilised by man means that there is significant potential for adverse anthropogenic impacts, and yet there have been very few studies to determine what these impacts might be.

Significant losses of *S. spinulosa* reefs have been reported in the Wadden Sea (Reise & Schubert 1987; Riesen & Reise 1982) and more locally with the disappearance of Saturn Reef (Hendrick 2007) suggesting that *S. spinulosa* reefs may be particularly sensitive to anthropogenic disturbances such as commercial fishing and eutrophication. However, the lack of targeted sampling means that it is impossible to attribute the cause of these declines, much less quantify the response of *S. spinulosa* reef communities to different levels of disturbance.

In the context of the Marine Strategy Framework Directive and more specifically to the development of Descriptor 1 habitat condition indicators it is essential to understand how *Sabellaria spinulosa* reefs respond to anthropogenic pressures. The following section reviews the limited literature available on this topic and discusses the potential threats and disturbances likely to affect *S. spinulosa* with a view to providing some validation and to highlight areas requiring further research.

i. Changes in suspended solids (water clarity)

A significant physical impact associated with most marine developments is the release of fine sediment into the water column, increasing turbidity. For example, aggregates extracted by dredging are often screened, a process which adjusts the composition of the sediment load to meet consumer requirements and therefore usually release finer (less valuable) sediments back into the sea. This can create a significant sediment plume (Marine Ecological Surveys Limited 2004). Sediment is also released as a result of substrate disturbance during the process of cable laying and other offshore construction activities. It is not thought likely, however, that there would be significant impacts on *S. spinulosa* associated with increased sediment loads. This is due to the worm's apparent preference for turbulent waters, and because of their ability to utilise sediment in their tube building and any associated organic matter as a source of food. With this in mind it is possible that an increased sediment load could even have a positive impact on the development of *S. spinulosa* aggregations.

This is certainly an area requiring further investigation because tolerance to turbidity may vary depending on the composition of the suspended sediment. Davies *et al* (2009) have developed Vortex Resuspension Tanks (VoRT) which allow for the long term study of the response of benthic species to suspended matter in a controlled way. Preliminary studies using the VoRTs have shown that the growth rates of *S. spinulosa* were significantly lower under zero sediment conditions than in intermediate and high sediment regimes (Davies *et al* 2009). This further supports the view that *S. spinulosa* requires at least some suspended sediment and is likely to be tolerant of elevated levels. Further work is underway using

VoRTs which will look in more detail at this species tolerance to differing sedimentation levels and compositions (Kim Last, pers. comm. 2012; Last *et al* 2011).

The exception to *S. spinulosa's* apparent tolerance to change in turbidity is likely to arise where sediment loadings are reduced and indeed Davies *et al* (2009) have found that net erosion of tube structures occurs in sediment starved conditions. Reduced turbidity might occur where water movements are altered, perhaps as a result of marine constructions. This, however, is likely to be a rare occurrence in subtidal environments, with the exception perhaps of tidal barrages. *S. spinulosa* are therefore likely to be more susceptible to these impacts when they (rarely) occur intertidally. Shore defences or harbour extensions could, for example, interrupt sediment transport. The offshore wind farm industry is in its infancy and the degree to which these structures alter the flow of water and sediments remains largely unknown. Large arrays are likely to alter water movement, but the discontinuous nature of these structures makes increased turbidity more likely than a complete interruption to flow. More research would certainly be beneficial in this regard particularly given the scale of Round 3 wind farm developments (The Crown Estate 2009).

Although it is unlikely that the reef building organism itself will be adversely impacted by the levels of turbidity associated with offshore developments and may even thrive under these conditions the impacts on the fauna associated with the reefs has not as yet been determined.

ii. Siltation rate changes including smothering

Pohler (2004) observed intermittent episodes of sedimentation which lead to smothering and mortality of *Neosabellaria vitiensis* reefs. Resettlement occurred within a few weeks of the events suggesting a high turn-over of reefs as well as a high availability of larvae. Miller (2001) also noted regular smothering of intertidal sabellariid reefs where a near complete kill occurred every winter. Nevertheless, recruitment was reported each spring indicating that these intertidal reefs were essentially being maintained by more stable subtidal reefs. Similarly, Pohler (2004) concluded that new recruitments had originated from more stable colonies elsewhere and that larvae were distributed through long shore drift. Pohler (2004) also noted that large amounts of seaweed and litter washed up after tropical storms had a devastating effect on sabellariids by smothering and killing large areas of the reef.

Smothering is likely to represent a very real threat to Sabellaria alveolata which occurs in intertidal and shallow sublittoral environments where new constructions and beach nourishment programmes occur. Sabellaria spinulosa reefs, however, are less likely to experience smothering through anthropogenic activities given the localities within which they exist. However, increased sediments released during marine construction, or through spoils dumping could present a potential threat of smothering. Smothering is perhaps most likely to occur through natural processes, particularly given the habitat preferences of this species. S. spinulosa reefs are often reported on the boundaries between mixed gravel deposits and mobile sands (Hendrick 2007; Pearce et al 2007). It is not unlikely then that smothering could occur through natural storm events that redistribute mobile sand deposits. A recent survey off the coast of East Anglia revealed evidence of exactly this phenomenon with dead S. spinulosa tubes found below layers of sand or new S. spinulosa growth (Figure 4.31; Limpenny et al 2011). The East Anglia area is characterised by very sandy mobile sediments and it would seem that the topographically distinct reefs that were identified here have built up over time in a cycle of inundation and new growth (personal observations). Whilst sabellariid reefs are clearly vulnerable to damage from smothering events, their capacity to recover is such that, providing larval availability is not interrupted, this impact is likely to be temporary, with new reefs developing over old buried ones.

Unlike the impacts of turbidity no studies have been undertaken to examine what levels of smothering are tolerated by *S. spinulosa* and the fauna associated with the reef and although the reefs themselves are capable of growing back in environments where an adequate larval supply is maintained it is likely that many of the reef inhabitants will take much longer to recover especially slower growing epifaunal species such as anemones. Given that many of the reefs incorporated in this study are likely to undergo regular exposure to high turbidity and smothering events from both natural events (e.g. storms) and anthropogenic sources (e.g. adjacent aggregate extraction or wind farm development activities), it is possible that the associated fauna we have observed is not representative of the climax community.



Figure 4.31 Photograph showing the new *Sabellaria spinulosa* growth on top of a relict reef with a thin layer of sand in-between. The relict reef was found to be full of sand and showing signs of anoxia suggesting some sort of smothering event (Limpenny *et al* 2011).

iii. Pollution and other chemical changes

Studies relating to water quality and in particular contamination of water caused by effluent discharges (Hoare and Hiscock 1974, Hoare & Peattie 1979, Walker and Rees 1980) have found that *S. spinulosa* is more tolerant to these conditions than other marine organisms. Hoare and Hiscock (1974) found that species richness and diversity showed a significant reduction within 150m of the outfall of a bromide extraction plant in North Wales. The effluent had a pH of 4 and contained contaminants including free halogens. *S. spinulosa* was found closer to the outfall than any other marine species and was found at higher densities at an intermediate distance. Therefore whilst *S. spinulosa* may show some sensitivity to very marked reductions in water quality, it appears to be more tolerant than other species, providing it with a competitive advantage in intermediate levels of contamination. This is further supported by work carried out by Walker and Rees (1980) who found that sludge dumping in Dublin Bay appeared to encourage the growth of *S. spinulosa*.

Although *S. spinulosa* itself is likely to be tolerant of poor water quality this is unlikely to be true of all of the reef inhabitants. There have not been any studies that look directly at the impact of poor water quality on *S. spinulosa* reef communities so it is not possible to know what the full biodiversity impact might look like.

iv. Physical damage: selective extraction and abrasion

The removal of substrate and physical destruction associated with marine activities is arguably the largest anthropogenic threat that exists for *S. spinulosa* aggregations (Jennings & Kaiser 1997; Reise & Schubert 1987). However, the significance of this threat has yet to be assessed in terms of longevity. It has been suggested in the Wadden Sea Red List that regeneration of this habitat could take between 15 and 150 years (UKBAP 2007) although others have asserted that the recoverability of this species is high (Jackson & Hiscock 2008). There have been several instances in the UK where *S. spinulosa* aggregations have been reported to appear where aggregate extraction activities have ceased. Foster-Smith (2001) reported that the best reefs in an area of the Wash were associated with ground clearly scarred by dredging activities. It was suggested that this was most likely due to a reduction in the overburden of sand resulting in a substrate more suitable for *S. spinulosa*. The recent discovery of significant *S. spinulosa* aggregations within and adjacent to the active aggregate licence area at Hastings Shingle bank (Pearce *et al* 2007) and other areas in the North Sea (Emu Limited 2008) provides further evidence that the physical impacts of dredging activities on this species are short-lived and restricted in extent.

Techniques for surveying biogenic reefs have advanced considerably in the last five years with seabed imagery and high-resolution side-scan sonar becoming standard tools for this purpose (Marine Ecological Surveys Limited 2005, 2006, 2009). Figure 4.32 shows a high-resolution side-scan sonar image taken at the boundary of a dredged area in the English Channel. *S. spinulosa* can be identified in this image as an irregular texturing (labelled speckled sonar response) and the physical damage caused by beam trawls and aggregate dredging can clearly be seen. In fact visible trawl scars are a good indicator of the presence of *S. spinulosa* reef since the scars appear more pronounced when they have taken out topographically distinct structures (personal observations).



Figure 4.32. High resolution side-scan sonar image taken at an aggregate extraction site in the English Channel © CEMEX UK Marine Limited, Hanson Aggregates Marine Limited and United Marine Dredging Limited.

Trawling, dredging, potting and net fishing are all thought to cause damage to *S. spinulosa* reefs (Hendrick 2007; Holt *et al* 1998). Where parts of the reef are broken off or damaged the resulting hole may be enlarged further by wave action (Cunningham *et al* 1994). Towed fishing gear is thought to represent the largest global anthropogenic disturbance to the seabed (Jennings and Kaiser 1998; Kaiser *et al* 2006; Queiros *et al* 2006). In most cases these gears are used in direct contact with the seabed to ensure adequate capture rates of target species that live on or within the seabed (Jennings and Kaiser 1997). The physical damage of bottom trawling on *S. spinulosa* reefs is clearly evident in Figure 4.32. It has long been accepted practice amongst commercial shrimp fishermen to search for *S. spinulosa* using small hand held dredges (Warren and Sheldon 1967). The strong association between these habitats and demersal fish has also made them a target for beam trawlers (personal communications with fishermen) which is in keeping with the observed enhancement to infaunal abundance and presumably, benthic productivity. Fishing is, therefore, thought to be the single biggest threat to *S. spinulosa* reefs.

Shrimp fishing was implicated in the decline of *S. spinulosa* reefs in the Wadden Sea between 1924 and the 1980's (Reise and Schubert 1987; Riesen and Reise 1982). Local fishermen were reported to have deliberately ground the reefs with heavy gear because it ripped apart the nets when fishing for shrimp (Riesen and Reise 1982). Fishermen at Ramsgate do the same to reefs in the Thames sea area (pers. comms.). There was no specific evidence of fishing having caused the Wadden Sea demise and others have speculated that coastal eutrophication, favouring *Mytilus*, contributed to the collapse (Reise & Schubert 1987). *S. spinulosa* reefs of the Thames still exist (Marine Ecological Surveys Limited 2005; this report) despite fishermen's claims of destruction. Similarly, *S. spinulosa* reefs at Hastings Shingle Bank remain extensive despite clear damage from bottom trawling (Marine Ecological Surveys Limited 2006; Pearce *et al* 2007). The fast growth rates of sabellariids reported in the literature (Chen and Dai 2009; George and Warwick 1985; Gruet 1982; Wilson 1971) perhaps goes some way towards explaining this. Bottom fishing is unlikely to remove complete reef systems in a single fishing event hence if fast re-growth is possible then the impact may be short lived.

Evidence in the literature and from observations in the field seem to indicate current fishing levels are not sufficiently high to completely remove *S. spinulosa* reef habitats from UK waters, although it is likely that these activities have reduced their extent. Given that many of the reefs included in this study are likely to have been subjected to repeated trawling it is also possible that the reefs and, in particular the fauna associated with the reefs are being kept at in intermediate stage of development. A study on a newly developed reef in the Hastings Shingle Bank area (Pearce *et al* 2007) found there to be no detectable differences in the multivariate community structure of fauna inhabiting a reef known to have developed in just six months, and the fauna associated with a nearby reef for which there were records going back five years. This could indicate that the fauna associated with *S. spinulosa* reefs are able to recover quickly but, because the five year old reef was subject to on-going commercial fishing it is more likely that the reefs in this area are not in their climax state. Additional research and more specifically, sampling of a *S. spinulosa* reef habitat that is not subjected to on-going anthropogenic impacts is required before we can have confidence in what a 'healthy' reef looks like.

Although there are indications of the types of impact we might anticipate in the face of different anthropogenic disturbances acting on *S. spinulosa* reefs there is no documentation of the magnitude of change that we could expect (Table 4.27), and hence the indicators that have been suggested for *S. spinulosa* monitoring cannot be validated with certainty at this stage of development.

Table 4.27 Summary of the nature and magnitude of change in worm density and in the biodiversity of communities associated with *Sabellaria spinulosa* reefs in response to environmental change based on a review of the literature and field observations.

Impact /	S. spinulosa Density		Associated Community	
Pressure	Anticipated Change	Magnitude of Change	Anticipated Change	Magnitude of Change
Changes in suspended solids (water clarity): increased turbidity	Negligible or positive.	Unknown	Impacts on the fauna associated with <i>S. spinulosa</i> reefs will be species specific with some species having similar tolerance levels to <i>S.</i> <i>spinulosa</i> and others less tolerant. A reduction in diversity is therefore likely.	Unknown
Changes in suspended solids (water clarity): decreased turbidity	Could cause complete or near complete die-off in extreme cases but may also give rise to intermediate impacts.	Unknown	Impacts on <i>S. spinulosa</i> may be reflected in the associated fauna. Could cause changes in the balance of deposit and filter feeders associated with the reef.	Unknown
Siltation rate changes including smothering	Likely to cause complete or near complete die-off (but is likely to recover quickly if an adequate supply of larvae is maintained).	Unknown	Likely to cause complete or near complete die-off.	Unknown
Physical Disturbance damage: selective extraction and abrasion	Likely to cause complete or near complete die-off (but is likely to recover quickly if an adequate supply of larvae is maintained). Areas of reef will become patchier with increasing physical impact.	Unknown	Likely to cause complete or near complete die-off.	Unknown
Pollution and other chemical changes	Negligible or positive as tolerance gives competitive advantage.	Unknown	Impacts on associated fauna are likely to be worse than for <i>S. spinulosa</i> which is known to be robust to poor water quality. A decrease in diversity is therefore anticipated.	Unknown

4.7.2 Case study to validate the response of potential Descriptor 1 indicators to anthropogenic disturbance: East Coast REC

The East Coast REC area is subjected to significant levels of anthropogenic disturbance including aggregate extraction (Figure 4.33) and commercial fishing (Figure 4.34). Commercial fishing activities using mobile gear are widespread across this area and increase in intensity with distance offshore. Conversely, static fishing activities are concentrated in the inshore region with the highest intensity being observed in the far southwest corner. Aggregate extraction occurs over a much smaller area in the centre of the study site with some activity also observed in the far south. The level of anthropogenic disturbance at this site and the presence of significant *S. spinulosa* reefs make the East Coast REC a useful case study to attempt to validate the indicators identified earlier.

Intensity data for each of the three main forms of anthropogenic disturbance, to which the reefs in this area are subjected (mobile fishing, static fishing and aggregate extraction), were extracted for the grab sampling stations and these data were used, on their own and in combination, to assess whether or not the density of *S. spinulosa* and the diversity of the

associated fauna correlates in any way with differing levels of disturbance. We would anticipate that any changes in the density of *S. spinulosa* and the diversity of associated fauna would correlate with the respective gradients in the level of disturbance at any one place, so that an area that is subjected to a greater level of disturbance is more impacted. RELATE tests were therefore carried out on similarity matrices based on ranked levels of disturbance at each station and corresponding matrices based on the density of *S. spinulosa* in each sample, and the number of species recorded in each sample, in order to test for changes in these indicators.



Figure 4.33. Chart showing the extent and intensity of aggregate extraction between 2000 and 2010. These data are derived from Electronic Monitoring System (EMS) data collected from all dredgers operating in the area (British Marine Aggregate Producers Association-BMAPA 2010).



Figure 4.34. Charts illustrating the intensity of A) mobile fishing and B) static fishing activities in the East Coast REC study area (Limpenny *et al* 2011). These data are derived from a combination of inshore fishery observations made by fisheries committees and Vessel Monitoring System (VMS) data collected from vessels over 15m and are displayed as Sightings Per Unit Effort (SPUE). (Vanstaen *et al* 2010). Pale green polygons represent areas of *S. spinulosa* reef identified from high resolution acoustic data (Limpenney *et al* 2011; Pearce *et al* 2011). Dots show the locations of grab samples and seabed images taken across these areas, with those coloured black representing an absence of *S. spinulosa* and those coloured representing the presence of *S. spinulosa*.

i. Sabellaria spinulosa density

The selected anthropogenic disturbances were categorised and ranked as summarised in Table 4.28. The ranks were then used to create model matrices to test for correlations between the density of *S. spinulosa* and the different disturbance gradients using the RELATE test (Somerfield *et al* 2002).

Table 4.28 Table summarising the categorisation and ranking of anthropogenic disturbance data. The aggregate extraction data are derived from Electronic Monitoring System (EMS) data and are displayed as total time dredged in 2009. The fishing data are derived from a combination of inshore fishery observations made by fisheries committees and vessel monitoring system (VMS) data collected from vessels over 15m and are displayed as Sightings Per Unit Effort (SPUE). (Vanstaen *et al* 2010).

Disturbance	Rank			
	0	1	2	3
Aggregate Extraction	0	<15 mins	15 mins – 1hr 15 mins	>1hr 15 mins
Mobile Fishing	0	0.001 – 0.025	0.026 - 0.075	0.076 – 0.225
Static Fishing	0	0.001 – 0.025	0.026 - 0.075	0.076 - 0.225

The results of the RELATE tests are summarised in Table 4.29 which shows that there is no statistically significant correlation between the density of *S. spinulosa* and the level of anthropogenic disturbance. This result could be misleading however as it would be unthinkable that extracting a reef through aggregate extraction or through trawling would not change the density of *S. spinulosa* occurring in that area. This result is most likely to be an artefact of the coarse spatial resolution of the human pressures data. This result may also reflect the fact that all of the samples included in this test are subject to a low-moderate level of impact i.e. there are no samples that are not subject to disturbance, nor are there any that we know are subjected to a very high level of impact, making comparisons of this nature very difficult.

Table 4.29 Table summarising the results of a series of RELATE tests carried out between *Sabellaria spinulosa* density (untransformed) and model matrices based on ranked categorised anthropogenic disturbance gradients (Table 4.28).

Disturbance	Sample Statistic	Significance Level
	(Rho)	(%)
Dredging	0.044	21.1
Static Fishing	0.010	34.9
Mobile Fishing	0.033	15.3
All Disturbance	0.038	15.4

ii. Faunal Diversity

In order to test for correlations between the diversity of fauna associated with the reefs and anthropogenic disturbances, the number of species recorded in each grab sample taken from within the reefs identified in the East Coast REC area, was extracted. A series of RELATE tests were then performed as described for *S. spinulosa* density. The results of the RELATE tests (summarised in Table 4.30) show that there was no statistically significant correlation between the number of species associated with *S. spinulosa* reefs and the level of anthropogenic disturbance. As with the tests on *S. spinulosa* density, it is noting that the data used here were not collected for the purpose of testing for the impacts of fishing and aggregate extraction and a more robust test could easily be implemented by targeted sampling in areas with known levels of disturbance. Furthermore, as the anthropogenic

impact data were only available at a very coarse resolution the statistical power of this analysis is reduced.

Table 4.30 Table summarising the results of a series of RELATE tests carried out between the number of species per grab sample (untransformed) and model matrices based on ranked categorised anthropogenic disturbance gradients within *Sabellaria spinulosa* reefs (Table 4.28).

Disturbance	Sample Statistic (Rho)	Significance Level (%)
Dredging	0.105	11.9
Static Fishing	-0.052	85.2
Mobile Fishing	0.075	15.5
All Disturbance	0.071	13.7

Box 4.9. Summary of findings relevant to validation of Sabellaria spinulosa reef indicators.

- There are insufficient evidence of the magnitude of anthropogenic impact on *S. spinulosa* reefs to adequately validate indicators proposed in this study.
- A case study investigating the impact of aggregate extraction and fishing (mobile and static) on *S. spinulosa* reefs within the East Roast REC site could not detect any changes in *S. spinulosa* density or the diversity of associated fauna. This is probably due to the coarse nature of the impact data and insufficient sampling within the most disturbed areas.
- Research into the response of *S. spinulosa* reefs to anthropogenic disturbance (especially demersal fisheries) has been identified as a key research requirement for the advancement of MSFD indicator development.

4.8 Key findings and research needs

Significant limitations were identified in the evidence base available for the development of S. spinulosa reef habitat condition indicators, most notably the lack of data from unimpacted reefs. Recent sampling (photographic only) of the S. spinulosa reefs associated with the Thanet offshore wind farm indicate that these reefs have been afforded some protection from the larger and therefore more damaging commercial beam trawlers. Whilst there are no statutory fishing exclusions in place at this site the presence and spacing of the wind turbines make fishing with towed gear much less desirable and even dangerous (pers. comms. from local fishermen). Between the pre-construction baseline survey and the first post-construction monitoring survey, some improvement in epifaunal reef condition is evident from the seabed images (Figure 4.35; Pearce et al in press). Construction of the wind farm took place in 2009 and was not completed until 2010 meaning that fishing activities have been limited in this area for a maximum period of two years. The reefs in this area may not have yet recovered to a full climax community but as the reefs are showing signs of developing an epifaunal community dominated by sea anemones, which has not as yet been reported from any other reef systems in the UK, quantitative grab sampling is strongly advised. On-going quantitative monitoring at this site and in the Wash where the reefs are protected from fishing through local byelaws would greatly improve the validation of Descriptor 1 condition indicators for this habitat, as would experimental impact assessments.



Figure 4.35 Images obtained from the Thanet Offshore Wind Farm licensed area in A) 2007, preconstruction; and B) 2012, two years following completion of the construction phase. Digital photographs taken using freshwater lens drop down cameras. Note these images have not been taken at precisely the same location but are typical of the images collected in each year within acoustically defined areas of *S. spinulosa* reef.

Box 4.9. Key research requirements and recomendations for *Sabellaria spinulosa* reef indicators.

- The evidence currently available to assess *S. spinulosa* density and community diversity responses to anthropogenic pressures is insufficient and therefore it is not possible at this time to fully validate MSFD indicators of *S. spinulosa* reef condition. An indicator validation case study is recommended using Thanet offshore wind farm as an un-impacted / low physical impact site. With appropriate pressure data from demersal fisheries there is good reason to believe a study of this type will be successful.
- There is a significant positive correlation between the density of *S. spinulosa* worms (derived from grabs and tube counts from seabed imagery) and community diversity (S, N, d and H') as well as a significant negative correlation with species evenness (J). There is also a weak to moderate significant positive correlation between the density of the long clawed porcelain crab, *P. longicornis* and associated diversity (S, N, d and H'). An indicator using one, or a combination of these rapid-assessment metrics is recommended.
- Both the density of *S. spinulosa* and the diversity of associated species was found to be highly variable within and between reef systems and it will therefore be difficult to determine the environmental status of a reef based on any single threshold value of a diversity indicator. Variability is likely to have been inflated in this study through the use of imperfect identification of 'reef' in sampling, therefore better remote sensing for stratified sampling is necessary.
- Given the significant gaps in the evidence base and our understanding of the complex interactions between *S. spinulosa* reefs and the environment in which they occur we would recommend that initially, a combination of diversity measures are used as metrics to monitor *S. spinulosa* reef condition; these should include Shannon's diversity (H') and Pielou's Evenness as well as *S. spinulosa* density.
- Assessment of the multivariate community structure is also advocated as this is likely to detect smaller compositional changes which could be early indicators of environmental stress. Investigations into other potential habitat condition indicators such as trait based indicators and condition indexes (e.g. AZTI Marine Biotic Index (AMBI)) are also recommended. Such indicators may provide further insight into the ecological functioning of these reef habitats and may allow for a more holistic approach to monitoring.
- It is likely that our ability to monitor *S. spinulosa* reef habitats remotely will improve in the future as sampling technologies, such as underwater video and camera systems, Remotely operated Vehicles (ROVs), Automatic Unmanned Vehicles (AUVs) and acoustic systems continue to advance. Monitoring programmes should be adjusted to make the most of new technology as it becomes available.

Box 4.9. Key research requirements and recomendations for *Sabellaria spinulosa* reef indicators (continued).

- Overall, there are significant gaps in both the evidence base and our understanding of the ecology of *S. spinulosa* reefs and until these research gaps are addressed, a precautionary approach to monitoring is recommended:
 - High resolution acoustic data to ensure sampling is properly stratified in target areas;
 - Seabed imagery is then recommended for widespread, cost-effective monitoring of density and community diversity;
 - Quantitative grab sampling is then recommended to verify the assessment based on imagery (above);
 - The verification from grab sampling could itself be staggered with more rapid, cost-effective *S. spinulosa* counts and *P. longicornis* counts. More time consuming analysis of the fauna could be reported at a later date or if there was any concerns raised.
- SNCB advice on the restriction of grab sampling in S. spinulosa reefs should be revisited in light of evidence here of their likely rapid recovery and the exceedingly small scale of damage from monitoring.

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6. List of Appendices

Appendix 1. Procedural Guidance

Appendix 1.1. Procedural guidance for Modiolus modiolus reef indicators

Appendix 1.2. Procedural guidelines for Mytilus edulis reef indicators

Appendix 1.3. Procedural guidance for Sabellaria spinulosa reef indicators

Appendix 2. References and Datasets

Appendix 2.1. Summary of references and datasets (Modiolus modiolus)

Appendix 2.2. Summary of references and datasets (Sabellaria spinulosa)

Appendix 3. Statistical Methods

Appendix 3.1. Univariate Statistical Methods

Appendix 3.2. Multivariate Statistical Methods

Appendix 4. Statistical Analyses

Appendix 4.1. Summary of taxa contributing to 90% of the within-group similarity and 60% between-group dissimilarities for SIMPROF Modiolus modiolus groupings

Appendix 4.2. Summary of taxa contributing to 30% of the within-group similarity and between-group dissimilarities for Modiolus modiolus reef communities

Appendix 4.3. Summary of species contributing to 90% of the within-group similarity and 60% of the dissimilarity between group similarities for SIMPROF Modiolus modiolus groupings

Contents

Appendix 1. Procedural Guidance1
Appendix 1.1. Procedural guidance for <i>Modiolus modiolus</i> reef indicators1
i. Use of non-destructive <i>in situ</i> methods to determine <i>Modiolus modiolus</i> reef density and community indicators
ii. Use of remote methods to determine <i>Modiolus modiolus</i> reef density and community indicators
Appendix 1.2. Procedural guidelines for <i>Mytilus edulis</i> reef indicators6
ii. Use of combined acoustic mapping and grab sampling to determine <i>Mytilus edulis</i> reef density and community indicators
Appendix 1.3. Procedural guidance for Sabellaria spinulosa reef indicators 10
ii. Use of grab sampling to determine <i>Sabellaria spinulosa</i> reef density and community indicators
Appendix 2. References and Datasets15
Appendix 2.1. Summary of references and datasets (Modiolus modiolus)15
Appendix 2.2. Summary of references and datasets (Sabellaria spinulosa)
Appendix 3. Statistical Methods 29
Appendix 3.1. Univariate Statistical Methods29
Appendix 3.2. Multivariate Statistical Methods
Appendix 4. Statistical Analyses 32
Appendix 4.1. Summary of taxa contributing to 90% of the within-group similarity and 60% between-group dissimilarities for SIMPROF Modiolus modiolus groupings
Appendix 4.2. Summary of taxa contributing to 30% of the within-group similarity and between-group dissimilarities for Modiolus modiolus reef communities
Appendix 4.3. Summary of species contributing to 90% of the within-group similarity and 60% of the dissimilarity between group similarities for SIMPROF Modiolus modiolus groupings75

Appendix 1. Procedural Guidance

Appendix 1.1. Procedural guidance for *Modiolus modiolus* reef indicators

In 2001 JNCC published *Procedural Guidelines* (PGs) on marine monitoring methods as part of the UK national guidance on monitoring for marine nature conservation management and reporting (Davies *et al* 2001). The intention of the work by Davies *et al* (2001) was that PGs should remain 'live' documents and be supplemented and updated.

In the present study, available data from a variety of methods have been reviewed for monitoring the density and community composition of *M. modiolus* beds. Data have primarily been sourced from published reports and field trials carried out to refine existing methodologies. Most sites used in this report are from low impact *M. modiolus* beds of high diversity and evenness which could be potentially used as metrics for future monitoring programmes. Impact case studies from protected sites in Wales, the Isle of Man and Strangford Lough are the strongest evidence found that the proposed epifaunal and infaunal indicators respond to physical anthropogenic impacts.

Overall, for MSFD or MPAs, the most cost-effective *M. modiolus* bed monitoring programme would involve rapid assessments using cell frequency counts from seabed images as a proxy for density. *A. digitatum* counts can be used as a proxy for density in beds where abundant or superabundant *A. digitatum* obstruct accurate estimations of live *M. modiolus*. If ophiuroids regularly hinder the accuracy of remote methods, *in situ* density estimates by divers using cell frequency counts of 0.25m² quadrats is preferred. This method is also recommended as a more accurate alternative to remote cameras for smaller beds in shallower (<30m) waters.

High resolution camera stills are also the most cost-effective monitoring method for community indicators where depths preclude divers from operating safely. However, the use of towed cameras to survey small *M. modiolus* beds in coastal areas with complex topography can be difficult. Under those conditions, Phase II semi-quantitative surveys carried out by divers are an alternative method and are also less susceptible to variability caused by environmental conditions. Erect epifauna are the most sensitive to abrasion caused by mobile fishing gear (Cook *et al* 2013; Galand 2007; Hall-Spencer 1999). Therefore, by predominantly targeting epifaunal communities, both remote cameras and *in situ* diver methods represent advantageous, non-destructive methods that can easily detect community change from physical impact. As most of the community associated with *M. modiolus* reefs are infauna or crevice fauna, a less frequent full infaunal sampling campaign whereby replicate mussel clumps are collected by divers and all the infaunal component extracted and indentified would be appropriate to support the conclusions of rapid epifaunal assessments. Shifts in the community from epifaunal species to opportunistic and soft bottom taxa could be indicative of anthropogenic impact.

Of all community metrics studied, Shannon-Wiener's diversity H' and Pielou's evenness J were the least variable and therefore most desirable ones (regardless of the method used to acquire them). Indices based on soft bottom biota such as the AZTI Marine Biotic Index or the Infaunal Quality Index have been successfully used in the past to detect anthropogenic impact (Borja *et al* 2009; Muniz *et al* 2005; Muxika *et al* 2005) and should be investigated further as possible metrics to establish baselines and detect change of *M. modiolus* communities.
i. Use of non-destructive *in situ* methods to determine *Modiolus modiolus* reef density and community indicators

a. Background

The method is a combination of *in situ* survey methodologies for subtidal biotopes adapted from Holt and Sanderson (2001)

b. Purpose

- Semi-quantitative estimation of epifaunal community indicators associated with *M. modiolus* reefs;
- Estimation of percentage cover for *M. modiolus* density indicators.

c. Advantages of non-destructive in situ estimates

- Allows for more accurate estimation of density and cover than remote methods;
- Divers able to brush off epifauna and determine if mussels are alive or dead;
- Can be used to cover smaller, more fragmented reefs in areas where coastal topography might preclude easy towed camera systems;
- Less destructive than other methods (clearance quadrats);
- Subject to less variability than other methods;
- Subject to less interoperator variability than other methods;
- Requires less replication to detect the same change than remote methods.

d. Disadvantages of non-destructive *in situ* estimates

- Survey time influenced by depth;
- Influenced by skill of divers, currents and visibility also factors;
- More costly than remote methods.

e. Procedures

General recommendations

The method is similar to that of Holt and Sanderson (2001). Divers descend the shotline and begin to survey a 50m transect as they reel out a measuring tape from a pole travelling in a straight line. Diver No.1 records all species along the transect and assigns a SACFOR scale score. Diver No.2 is in charge of acquiring *M. modiolus* density estimates as follows.

Cell frequency cover estimates by divers

- All ophiuroids are brushed-off by the diver prior to deploying the quadrat;
- The 0.25m² cross-stringed quadrat (total 25 square cells) is positioned on the *M. modiolus* bed every 5m to obtain a total of 10 replicates;
- All cells containing live *M. modiolus* are counted;
- If the same *M. modiolus* is present in more than one cell all cells are counted;
- On return to the laboratory percentage cover is estimated for each quadrat by dividing the counts by 25;
- Temporal and spatial differences are investigated by means of generalized linear mixed models (GLMMs) using binomial families.

Community indicators

- Multivariate biotic matrices are constructed by assigning numerical scores to each SACFORN abundance value: S=6; A=5; C=4; F=3; O=2; R=1; N=0;
- Data are incorporated into PRIMER; DIVERSE function is used to obtain univariate community indices;
- Temporal and spatial change between indices are investigated by means of mixed PERMANOVA models (or GLMMs);
- Shannon-Wiener H' and Pielou's J recommended over other indices;
- Bray-Curtis similarity matrices are constructed; CLUSTER and MDS multivariate analyses using SIMPROF will identify significant groupings in the community data;
- PERMANOVA models can be used to determine the categorical factors influencing variation in the community structure;
- SIMPER analyses on the significant groupings used to help identify the species responsible for the significant differences between the groups.

f. Procedural (fieldwork)

Logistics and operation follow standard Natura 2000 guidelines (Holt & Sanderson 2001).

g. Health and safety

• All appropriate requirements needed for *in situ* surveys using diving techniques (see e.g. Holt & Sanderson 2001).

h. QA/QC

- Field trials suggested cell frequency counts were not prone to significant differences between operators. However, it is important that all operators are familiar with the counting technique and able to differentiate between live and dead *M. Modiolus;*
- Occasional inter-operator variability tests are recommended to ensure quality in the estimations is maintained;
- Taxonomic expertise: guidance and training necessary for biotope/community identification;
- Repeat analysis with second or third operators to ensure accuracy of mussel counts/fauna identification;
- Procedural guidelines for photo still analyses as standard;
- Follow sprit of NMBAQC scheme recommendations.

ii. Use of remote methods to determine *Modiolus modiolus* reef density and community indicators

a. Background

High definition towed stills cameras enable rapid visual survey and monitoring over wide areas. Still camera technology varies widely and some systems have shown far better results than others. Service (2001) reviewed the use of towed video and still cameras and their application to the survey and monitoring of biotope extent, epifaunal richness and the abundance of associated species. Vertically mounted still cameras in drop down mode enable spot samples to be gathered over a wide area without time spent slowly motoring or

drifting along transects but can lack stability in tidal areas where a sled can sometimes be better.

b. Purpose

Remote cameras in a drop down mode can be successful methods for obtaining a density proxy (number of open *M. modiolus* showing mantle tissue or *A. digitatum* if present) for deep *Modiolus modiolus* reefs where diving is precluded by depth or extent of the bed. These methods are less useful for deriving measures or proxies for community diversity because the majority of the community is infaunal. However, all available data are from unprotected reefs (and therefore semi-impacted). It therefore remains possible that undisturbed reefs have a higher proportion of epifauna, in which case remote camera methods may be of use for monitoring community composition.

c. Advantages of still imagery

- Allows for density and community indicator estimations in beds that are too deep or too exposed for safe standard diving practises according to HSE Scientific and inshore commercial diving codes of practice;
- Able to survey large areas quickly;
- Can be used at numerous stations to evaluate small areas;
- Less destructive than other methods;
- Good reef detection compared to destructive methods;
- Can cover large areas as a reconnaissance tool to establish presence/absence of *M. Modiolus;*
- The image is a permanent monitoring data source that can be archived and fully revisited.

d. Disadvantages of still imagery

- Influenced by environmental conditions such as turbidity;
- Influenced by skill of camera/winch operator and sea state;
- Scale of camera field varies between available systems (if no reference is attached);
- Accurate estimates highly dependent on the presence of dense epifauna (ophiuroids, *A. digitatum*);
- Difficult to determine of *M. modiolus* are alive or dead if valves not open. Can lead to underestimation of cover/density where *M. modiolus* densities are high;
- Post-processing is slower than *in situ* counting methods by divers. Time to analyse one image ranges from 60 to 90 seconds depending on the density of the mussels and the presence of epifauna.

e. Procedures

General recommendations

- The system must be deployed in good visibility conditions;
- Standard drop cameras are adequate in good visibility but images must be of at least 180 dpi;
- Cameras should be mounted vertically and flashes positioned to avoid reflection into the images obtained;
- The number of replicates depends on both the power and resolution required (percentage change to be detected). Appropriate replication is highly dependent on the structure (fragmentation) of each reef to be surveyed as lack of homogeneity in density

influences the variance and hence the power. For example, dense beds (>100 mussel m⁻²) extending over 4km² off Noss Head would require 100 replicate photographs to detect 20% change in density at 80% power. These replicates should be located randomly across the known extension of the *M. modiolus* bed;

• Generalized linear mixed models (GLMMs) can satisfactorily deal with autocorrelation and unbalanced experimental designs. They are recommended to determine temporal and spatial change in *M. modiolus* density indicators.

Image analysis for M. modiolus density estimates from remote still photographs

- Image J is an appropriate example software package;
- Adjust settings: brightness, contrast (Image > Adjust > Brightness/Contrast). When images are grainy the function Smooth (Process>Smooth) reduces the noise. Sharpen (Process>Sharpen) also helps to increase the contrast of the tubes rim;
- Overlay guiding grip (Plugins>Analyze>Grid);
- Determine the area covered by the camera frame in advance (ideally a ruler should be on frame at ground level). Standardised area of coverage would be better for monitoring over time. Set the scale (Analyze > Set scale; choose global if reference scale is known to be constant);
- Counts are also possible for epifauna and vagile organisms such as *A. digitatum* spp., hydroids and ophiuroids;
- Only open *M. modiolus* with visible orange mantle should be counted;
- Carry out cell frequency counts using cell counter utility (Plugins > Analyze > Cell counter). Initialise and select counter type. Uncheck show numbers to avoid cluttering screen);
- To estimate bed density count all live mussels within the frame and repeat counts three times to calculate the average count and divide by the area covered by the photo frame;
- To estimate percentage cover all cells containing live mussels should be counted. If the same mussel occupies more than one cell, all cells should be counted. Percentage cover is calculated by dividing the number of cells with live *M. modiolus* by the total number of cells in the frame;
- If *M. modiolus* reefs are in association with dense cover of *A. digitatum* the latter can be used as proxy for *M. modiolus* density;

Image analysis for M. modiolus community estimates from remote imaging systems

- Use PRIMER[®] DIVERSE function to derive univariate diversity indexes from quantitative estimates of epifauna and epiflora extracted from high resolution imagery;
- The most sensitive and least variable community indexes are Shannon-Wiener's H' and Pielou's evenness J. They are also the recommended as the most cost effective metrics for the proposed community indicator as they require the least replication;
- PERMANOVA or GLMMs are the preferred statistical model designs to detect temporal and spatial change in community indicators.

f. Procedural (fieldwork)

- For logistics and operation follow standard Natura 2000 guidelines (Holt & Sanderson 2001);
- Standard drop cameras offer good results under normal visibility conditions;
- Scale must be established or standardised: a rule/scale must be positioned within the frame view and at seabed level;
- Camera should produce images of at least 180 dpi. However, a resolution of 250 or 300 dpi is recommended;

• Camera should be as vertical as possible to the reef.

g. Health and safety

• All appropriate requirements needed for boat based remote sampling (see e.g. Service 2001; Holt & Sanderson 2001).

h. QA/QC

- Taxonomic expertise: guidance and training necessary for biotope/community identification;
- Repeat analysis with second or third operators to ensure accuracy of mussel counts/fauna identification;
- Procedural guidelines for photo still analyses as standard;
- Follow sprit of NMBAQC scheme recommendations.

Appendix 1.2. Procedural guidelines for *Mytilus edulis* reef indicators

In 2001 JNCC published *Procedural Guidelines* (PGs) on marine monitoring methods as part of the UK national guidance on monitoring for marine nature conservation management and reporting (Davies *et al* 2001). The intention of the work by Davies *et al* (2001) was that PGs should remain 'live' documents and be supplemented and updated.

In the present study available data from a variety of methods have been reviewed for monitoring the density and community composition of *M. edulis* beds. Data originated primarily from published reports and datasets to which the authors had access to; there were no field trials undertaken as part of the present study. However, a workshop on mussel bed monitoring methodologies was organised and was held in Birmingham in November 2013. The workshop was intended for shellfish experts from the UK and Ireland to discuss mussel stock monitoring techniques (existing and potential) to ultimately help develop best monitoring practise for density and community indicators of GES, for both intertidal and subtidal mussel beds. The results have been incorporated into the final version of this study.

Most of the published research reviewed as part of this study indicates that: 1) mussel density metrics (coverage, density and total stocks) are temporally and spatially variable and dependant on a combination of different environmental parameters. The existing body of research, however, indicates declines in stocks can be unmistakably linked to excess fishing pressure; 2) epibiotic *M. edulis* beds substantially and significantly increase complexity if compared with surrounding substrata outside the mussel beds. The abundance and diversity of the epifaunal community is significantly enhanced by the presence of mussel aggregations. However, in some instances, diversity and abundance of, for example, infaunal polychaetes can be lower in mussel beds whereas infaunal deposit feeders are usually favoured. The metrics and methods used in the reviewed studies identified trends in density and diversity that, in most cases, were the result of environmental and human induced pressures.

Overall, for MSFD or MPAs, the most cost-effective *M. edulis* bed monitoring programme would involve:

• For intertidal beds: a methodology based on the Dutch MarinX approach modified to include biological analyses of the core samples (infauna and epifauna);

• For subtidal beds: following acoustic mapping of the beds, a systematic sampling using Day grabs to obtain both mussel density and biotic community metrics. Small, shallow subtidal beds on hard substratum (<30m) can be sampled by divers using destructive sampling methods, i.e. 0.25m² clearance quadrats.

The literature review indicated the most abundant and diverse component of the biotic community associated with *M. edulis* beds consists of crevice fauna and infaunal taxa. Conspicuous, sessile organisms (e.g. sponges, hydroids), although present in some mussel biotopes, might not be as representative of the *M. edulis* community as smaller taxa dwelling in microhabitats within the mussel matrix (Dittman 1990). Therefore, remote or *in situ* imaging methods are not considered adequate to capture representative community indicator metrics for subtidal and, particularly, intertidal *M. edulis* beds. Nonetheless, it would be advantageous to carry out fieldwork trials to determine the relationship between biodiversity indices derived from grab samples and those obtained from co-located photographs obtained from high definition cameras.

i. Methods to determine intertidal *Mytilus edulis* reef density and community indicators

a. Background

The method is a combination of *in situ* survey methodologies for intertidal biotopes adapted from Moore (2009); van Stralen and Boit (2004) and West *et al* (2004).

b. Purpose

- Estimation of percentage cover, density and total stocks for *M. edulis* density indicators;
- Quantitative estimation of community indicators associated with *M. edulis* reefs.

c. Advantages of destructive in situ walk surveys

- Only method for accurate estimation of mussel density, cover and biodiversity indices (existing remote technology still has high level of uncertainty when used to derive estimates of density and diversity);
- Offers increased replication compared to other methods resulting in less variability and higher statistical power;
- Subjective to less surveyor bias than other methods;
- Accurate representation of the full biotic assemblage.

d. Disadvantages of destructive in situ walk estimates

- Time consuming;
- Training required;
- Requirement of good tides to survey some beds in the lower intertidal;
- Destructive method.

e. Procedures

General recommendations

 Aerial photographs, LiDAR or side-scan sonar (the latter depending on depth at high water) are methods that can be used to locate and map mussel beds prior to the *in situ* surveys;

- Otherwise, bed position and perimeter are determined at low water: bed outline is determined by walking around bed and plotting representative waypoints into GPS;
- Mussel biomass is determined upon calculation of discrete bed area, density of mussels in patches within bed and percentage cover of mussels in patches;
- Transect lines determined to guarantee equal bed coverage;
- Percentage coverage is calculated as number of hits in relation to total number of presence/absence stations;
- Samples collected every 4-5 hits depending on the size of the bed but ensuring the number is within the recommended thresholds previously determined using power analyses;
- Samples are aggregated in 5L bucket. At the end of each track the samples are washed through a 0.5mm sieve;
- Retain macrofauna and mussels and preserve in 4% formaldehyde;
- Guidelines of the NMBAQC for sample processing and taxonomic conventions should be followed;
- The methodology needs to be adjusted in advance of each year's survey based on power calculations using existing datasets. An 80% power and a threshold of 40% change in the mean are appropriate when calculating sampling effort.

Density indicators

- Although there is co-linearity between all three parameters (coverage, density and total stocks) they are all important for conservation purposes;
- Generalized linear mixed models (GLMM) have the statistical power to determine temporal and spatial variation. Use of track and bed as random factors (within-bed variability) and year as fixed factor. Binomial and identity link functions should be respectively used for coverage and biomass analyses of variance.

Community indicators

- Univariate indices should be extracted using Primer;
- Spatial and temporal variability in univariate community indices can be investigated by means of PERMANOVA or GLMMs (using R or similar statistical packages);
- Multivariate community analyses require the use of Primer and PERMANOVA (or similar multivariate analytical tools, for example using R).

f. Procedural (fieldwork)

- Surveys to be undertaken in Autumn ensuring consistency with existing historical data;
- Logistics and operation follow methodology by Marnix van Stralen (Jessop *et al* 2012; van Stralen and Boit 2004) and standard Natura 2000 guidelines (Dalkin and Barnett 2001);
- Quads or other suitable All Terrain Vehicles can be used to cover long distances. Otherwise purpose built boats can be used to access the most remote beds (e.g. *ESF Three Counties*, see Jessop 2012).

g. Health and safety

• All appropriate requirements needed for *in situ* surveys using quantitative intertidal techniques (see e.g. Dalkin and Barnett 2001; Moore 2009; Walker and Nicholson 2004).

h. QA/QC

- Occasional inter-operator variability tests are recommended to ensure quality in the estimations is maintained;
- Taxonomic guidance and training necessary for identification of flora and fauna;
- Repeat analysis with second or third operators to ensure taxonomic consistency and accuracy;
- Follow sprit of NMBAQC scheme recommendations for taxonomic analyses of core samples.

ii. Use of combined acoustic mapping and grab sampling to determine *Mytilus edulis* reef density and community indicators

a. Background

Broad beam acoustic swath systems (e.g. side-scan sonar) produce high resolution images of the seafloor allowing for the distinction of mussel beds found on soft and mixed substrata. Benthic grab sampling has been a standard marine biological sampling tool since early studies by Peterson (1911) and the method has been described by various authors including Holme and McIntyre (1984) and Baker and Wolff (1987) and summarised by Thomas (2001). Benthic grab sampling has a dual purpose: 1) to ground-truth the acoustic map produced by the side-scan sonar; and 2) to obtain density and community indicator metrics useful for MSFD monitoring objectives.

b. Purpose

Side-scan sonar is preferred to other acoustic methods (i.e. Acoustic Ground Discrimination Systems) because it does not require constant calibration and it is less dependent on environmental conditions. Side-scan sonar is currently used with success in locating and mapping seed mussel beds (Mahuire *et al* 2007; <u>www.bim.ie</u>).

Grab samples are the most appropriate tool to accurately monitor the mostly infaunal *M. edulis* reef community. Grab samples are also an appropriate method to evaluate the density of mussels (adults and spat) on the reef. Concerns over damage to the reef using this technique are unnecessary given the overall small sampling footprint required and the capacity for subtidal *M. edulis* reefs to recover quickly.

c. Advantages of Grab Sampling

- Direct quantitative measurement of associated communities and *M. edulis* density;
- 'Industry standard' sampling tool facilitating combination with industry data for reporting purposes;
- Widely available type of sampling and analysis;
- Not as sensitive as seabed imagery to environmental conditions or skill of field operatives.

d. Disadvantages of Grab Sampling

- Full analysis of samples is time consuming and moderately expensive;
- Sampling can be perceived as destructive to a habitat that could be of moderate to high biodiversity conservation importance;
- Variability in the resulting data can make analysis difficult.

e. Procedural (fieldwork)

- The method is based on previous location and mapping of subtidal *M. edulis* beds using acoustic methods;
- High resolution side-scan sonar is the recommended technology to locate and map subtidal *M. edulis* beds. Procedural guidelines follow Kenny *et al* (2001);
- For logistics and operation follow standard Natura 2000 guidelines (Thomas 2001);
- Pilot surveys should be undertaken to the determine sample effort required to detect at least 40% changes in mean density and community indicators;
- Based on the results obtained from power analyses, a systematic coverage of the mussel bed area is recommended.

Density indicators

 Generalized linear mixed models (GLMM) have the statistical power to determine temporal and spatial variation in mussel density. Use of sampling station as a random factor and year as fixed factor. Poisson or quasi-Poisson GLMMs should be used for count data analyses.

Community indicators

- Univariate indices should be extracted using Primer;
- Spatial and temporal variability in univariate community indices can be investigated by means of PERMANOVA or GLMMs (using R or similar statistical packages);
- Multivariate community analyses require the use of Primer and PERMANOVA (or similar multivariate analytical tools, for example using R).

f. Health and safety

All appropriate requirements needed for boat based remote sampling (see e.g. Thomas 2001)

g. QA/QC

QA/QC procedures are well established with schemes such as the NMBAQC (see also Thomas 2001).

Appendix 1.3. Procedural guidance for *Sabellaria spinulosa* reef indicators

In 2001 JNCC published *Procedural Guidelines* (PGs) on methods as part of the UK national guidance on monitoring for marine nature conservation management and reporting (Davies *et al* 2001). Part of the intention of the work by Davies *et al* (2001) was that PGs should remain 'live' documents and be supplemented and updated. With the development of offshore renewable energy there has since been a steep rise in work to survey and otherwise investigated *Sabellaria spinulosa* reefs (e.g. Hendrick and Foster-Smith 2006; Limpenny *et al* 2010; Pearce 2008; Pearce *et al* 2008; 2011).

In the present study available data from a variety of methods have been reviewed for monitoring the density and community composition of *S. spinulosa* reefs. Data have primarily been sourced from the private sector and entirely from reefs that are subject to on-

going anthropogenic pressures, most notably from fishing. Appropriate studies from low pressure or protected sites have yet to emerge hence, the guidance that can be drawn for monitoring *S. spinulosa* reefs is not based on unimpacted baselines and there remains a small amount of uncertainty as to how exactly the proposed indicators would respond to anthropogenic impacts. Nevertheless, there is good supporting evidence about the response of *S. spinulosa* indicators based on comparisons between reefs and non-reefs.

i. Use of remote cameras to determine *Sabellaria spinulosa* reef density indicators

a. Background

Drop down stills cameras enable rapid visual survey and monitoring over wide areas. Still camera technology varies widely and some systems have shown far better results than others. Service (2001) reviewed the use of towed video and still cameras and their application to the survey and monitoring of biotope extent, epifaunal richness and the abundance of associated species. Vertically mounted still cameras in drop down mode enable spot samples to be gathered over a wide area without time spent slowly motoring or drifting along transects.

b. Purpose

Remote cameras in a drop down mode have been shown to be successful methods for obtaining a density proxy (tube counts) for *Sabellaria spinulosa* reefs. These methods are less useful for deriving measures or proxies for community diversity because the majority of the community is infaunal. However, all available data are from unprotected reefs (and therefore semi-impacted). It therefore remains possible that undisturbed reefs have a higher proportion of epifauna, in which case remote camera methods may be of use for monitoring community composition.

c. Advantages of still imagery

- Able to survey large areas quickly;
- Can be used at numerous stations to evaluate small areas;
- Less destructive than other methods;
- Tube counts significantly correlated to live worm densities recorded from grabs;
- Good reef detection compared to destructive methods;
- Detects wide range of other reefiness attributes as well as tube density i.e. patchiness, area, elevation, density, biotope type;
- Can cover large areas as a reconnaissance tool to establish presence/absence of *S. Spinulosa;*
- Post-processing is faster (estimate tube abundance) from photographs than counting worms from grabs. Time to analyse one image ranges from 5 to 20 minutes depending on the density of the reefs;
- The image is a permanent monitoring data source that can be archived and fully revisited.

d. Disadvantages of still imagery

- Influenced by environmental conditions such as turbidity;
- Influenced by skill of camera/winch operator and sea state;
- Scale of camera field varies between available systems (if no reference is attached).

e. Procedures

General recommendations

- The system must be deployed in good visibility conditions;
- Standard drop cameras are adequate in good visibility but images must be of at least 180dpi;
- Cameras with freshwater in front of the lens work in low visibility conditions but, as higher resolution settings result in less error in tube counting, a minimum of 180dpi is suggested and 270dpi is preferable;
- Cameras should be mounted vertically and flashes positioned to avoid reflection into the images obtained;
- Images that allow reefiness scores and fauna ID can be selected even if quality or conditions preclude tube counting;
- Select three replicate images for each sampling station.

Image analysis for tube density estimates from still photographs

- Image J is an appropriate example software package;
- Adjust settings: brightness, contrast (Image > Adjust > Brightness/Contrast). When images are grainy the function Smooth (Process>Smooth) reduces the noise. Sharpen (Process>Sharpen) also helps to increase the contrast of the tubes rim;
- Overlay guiding grip (Plugins>Analyze>Grid);
- Determine the area covered by the camera frame in advance (ideally a ruler should be on frame at ground level). Standardised area of coverage would be better for monitoring over time. Set the scale (Analyze > Set scale; choose global if reference scale is known to be constant);
- Determine the area covered by the wider reef in relation to the total area;
- Determine area covered by consolidated tubes in relation to reef area;
- Determine elevation (maximum/mean);
- Counts are also possible for some epifauna/vagile organisms such as *Sagartia* spp., hydroids and ophiuroids;
- Carry out tube opening counts using cell counter utility (Plugins > Analyze > Cell counter). Initialise and select counter type. Uncheck show numbers to avoid cluttering screen);
- Repeat counts three times and calculate the average count;
- Reefiness: assign scores based on Gubbay (2007) and Hendricks and Foster-Smith (2006):
 - Presence/absence of reef;
 - Reef coverage;
 - Reef patchiness;
 - Reef elevation;
 - Substrate type;
 - Elevation;
 - Associate community: mostly epifauna, sessile, crustose, vagile;
- It is easier to count tubes when the cephalic crown is exposed;
- Tube openings should be perpendicular to the field of view (upwards towards the camera).

f. Procedural (fieldwork)

- For logistics and operation follow standard Natura 2000 guidelines (Holt and Sanderson 2001);
- Standard drop cameras offer good results under normal visibility conditions;
- Scale must be established or standardised: a rule/scale must be positioned within the frame view and at seabed level;
- The closer the camera is to the seafloor the better for tube counts;
- Camera should produce images of at least 180 dpi, although 250 or 300 dpi are recommended;
- Camera should be as vertical as possible to the reef.

g. Health and safety

All appropriate requirements needed for boat based remote sampling (see e.g. Service 2001; Holt and Sanderson 2001).

h. QA/QC

- ii. Taxonomic expertise: guidance and training necessary for biotope/community identification;
- iii. Repeat analysis with second or third operators to ensure accuracy of tube counts/fauna identification;
- iv. Procedural guidelines for photo still analyses as standard;
- v. Follow sprit of NMBAQC scheme recommendations.

ii. Use of grab sampling to determine *Sabellaria spinulosa* reef density and community indicators

a. Background

Benthic grab sampling has been a standard marine biological sampling tool since early studies by Peterson (1911) and the method has been described by various authors including Holme and McIntyre (1984) and Baker and Wolff (1987) and summarised by Thomas (2001).

b. Purpose

Grab samples are the most appropriate tool to accurately monitor the majority of the *S. spinulosa* reef community. Grab samples are also an appropriate method to evaluate the density of living worms on the reef. Concerns over damage to the reef using this technique are unnecessary given the overall small sampling footprint required and the capacity for reefs to recover quickly.

c. Advantages of Grab Sampling

- Direct quantitative measurement of associated communities and *S. spinulosa* density;
- 'Industry standard' sampling tool facilitating combination with industry data for reporting purposes;
- Widely available type of sampling and analysis;
- Rapid assessment methods (*Pisidia longicornis* counts) are available to reduce analytical costs and time. Repeat elutriations (3+ may float-off *P. longicornis* but this requires testing);

- Not as sensitive as seabed imagery to environmental conditions or skill of field operatives;
- Other measures of reefiness can also be derived: maximum tube height, volume and weight of tubes and average tube aperture. These indices may help advance the understating of *S. spinulosa* reef ecology and prove useful tools at a later date.

d. Disadvantages of Grab Sampling

- Full analysis of samples is time consuming and moderately expensive;
- Sampling can be perceived as destructive to a habitat of high biodiversity conservation importance;
- Variability in the resulting data can make analysis difficult.

e. Procedural (fieldwork)

For logistics and operation follow standard Natura 2000 guidelines (Thomas 2001).

f. Health and safety

All appropriate requirements needed for boat based remote sampling (see e.g. Thomas 2001).

g. QA/QC

QA/QC procedures are well established with schemes such as the NMBAQC (see also Thomas 2001).

Appendix 2. References and Datasets

Appendix 2.1. Summary of references and datasets (Modiolus modiolus)

Table A2.1 Summary of the references and datasets used to inform the development of Descriptor 1 biodiversity indicators for *Modiolus modiolus* reefs. Site names with an asterisk indicate datasets incorporated in the statistical analyses used to define and validate the indicators. DDV=Drop down video; ROV=Remote Operated Vehicle; AGDS=Acoustic Ground Discrimination System; PMF=Priority Marine Feature; MNCR=Marine Nature Conservation Review.

Site	MNCR Sector	Month-Year	<i>M. modiolus</i> den data	sity indicators	<i>M. modiolus</i> community indicators		Marine Purpose of Data Collection Recorder		Source	
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key			
Basta Voe*	Shetland	September 2012	25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)	DDV tows	Replicate clumps (quantitative) 25m x 4m MNCR Phase II transect	DDV tows	Not in latest Marine Recorder snapshot (June 2013)	MPA search feature and quality assessment in suitable areas in Shetland (Fetla to Haroldswick)	Heriot Watt University Hirst <i>et al</i> (2013)	
Busta Voe*	Shetland	October 1999	In-situ: cross- hair and clearance quadrats. 25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)		In-situ: replicate <i>M.</i> modiolus clumps MNCR Phase II transects Video mosaics and camera stills		MRSNH012 00000002	Literature review and comparative study of <i>Modiolus</i> beds in Busta Voe, Loch Alsh and Loch Creran. Population structure, density and community composition	Heriot Watt University Mair <i>et al</i> (2000)	
Copinsay*	Orkney	September 2011	N/A	DDV tows. Biotope and SACFOR score estimations	N/A	DDV tows	MRSNH025 00000004	2011 Orkney PMF benthic survey	Heriot Watt University Hirst <i>et al</i> (2012b)	
Gutter Sound*	Orkney	September 2011	25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)	DDV tows	Replicate clumps (quantitative) 25m x 4m MNCR Phase II transect	DDV tows 0.1m ² Van Veen grabs	MRSNH025 00000004	Validation of MPA search features in Orkney waters.	Heriot Watt University Hirst <i>et al</i> (2012b)	

Site	MNCR Sector	Month-Year	<i>M. modiolus</i> den data	sity indicators	<i>M. modiolus</i> community indicators data		Marine Recorder	Purpose of Data Collection	Source
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key		
Hascosay Sound*	Shetland	September 2012	25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)	DDV tows	Replicate clumps (quantitative) 25m x 4m MNCR Phase II transect	DDV tows	Not in latest Marine Recorder snapshot (June 2013)	MPA search feature and quality assessment in suitable areas in Shetland (Fetla to Haroldswick)	Hirst <i>et al</i> (2012b)
Loch Alsh (String Rock*)	North-west Scotland	September 1999	In-situ: cross- hair and clearance quadrats. MNCR transects (SACFOR abundance for <i>M. modiolus</i>)		Replicate clumps (quantitative)		MRSNH012 00000002	Literature review and comparative study of <i>Modiolus</i> beds in Busta Voe, Loch Alsh and Loch Creran. Population structure, density and community composition	Mair <i>et al</i> (2000)
Loch Alsh (String Rock)*	North-west Scotland	June 2004	In-situ: cross- hair and clearance quadrats. Interoperator variability records. MNCR transects (SACFOR abundance for <i>M. modiolus</i>)	ROV transects for presence / absence, extent and abundance estimations	Replicate clumps (quantitative) MNCR transects (SACFOR abundance for <i>M.</i> <i>modiolus</i>)		MRSNH012 00000007	Repeat surveys at original sites surveyed by Mair <i>et al</i> in 1998. Assessment of Population structure and density and community change in the String Rock (Kyle Akin) <i>M. modiolus</i> beds. Investigation of interoperator variability and power analyses in density (cover) estimations of <i>M. modiolus</i>	Emu Ltd. (2006)
Loch Alsh (String Rock)*	North-west Scotland	2007	In-situ: cross- hair quadrats. 25m x 4m MNCR Phase II transect		Replicate clumps (quantitative)			Repeat surveys at original <i>Modiolus modiolus</i> sites SW of String Rock (Kyle Akin) surveyed by Mair <i>et al</i> in 1998. Assessment of decline in density and establish possible causes.	Marine Bio- images (2007)

Site	MNCR Sector	Month-Year	<i>M. modiolus</i> der data	density indicators <i>M. modiolus</i> community indicators data		Marine Recorder	Purpose of Data Collection	Source	
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key		
Loch Alsh (String Rock)*	North-west Scotland	2011	10 x 0.25m2 in situ: cross-hair quadrats at 14 sites. 25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>). Diver and photo stills along transects		Replicate clumps (quantitative) MNCR Phase II transect (SACFOR abundance for <i>M.</i> <i>modiolus</i>). Diver and photo stills along transects		Not in latest Marine Recorder snapshot (June 2013)	Validate records of MPA features in Loch Alsh, Duich, Creran and Fyne. Establish, extension and community of <i>Modiolus modiolus</i> beds in the Kyleakin and String Rock sites within Loch Alsh SAC. Carry out repeated <i>M. modiolus</i> density surveys at the historical String Rock site.	Moore <i>et al</i> (2013)
Loch Alsh (Kyleakin)*	North-west Scotland	2011	10 x 0.25m2 <i>in</i> <i>situ</i> : cross-hair quadrats at 14 sites. 25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>). Diver and photo stills along transects		Replicate clumps (quantitative) MNCR Phase II transect (SACFOR abundance for <i>M.</i> <i>modiolus</i>). Diver and photo stills along transects	DDV and Van Veen grabs only for Limaria MPA features	Not in latest Marine Recorder snapshot (June 2013)	Validate records of MPA features in Loch Alsh, Duich, Creran and Fyne. Establish, extension and community of <i>Modiolus modiolus</i> beds in the Kyleakin and String Rock sites within Loch Alsh SAC. Carry out repeated <i>M. modiolus</i> density surveys at the historical String Rock site. <i>Modiolus modiolus</i> surveys focussed on Lochs Alsh and Creran only although records exist for Loch Duich.	Moore <i>et al</i> (2013)
Loch Broom	North-west Scotland	August 2010		DDV and towed cameras (stills and video)	Spot dives for <i>M.</i> modiolus presence	DDV and towed cameras (stills and video) 0.1m ² Van Veen grabs	Not in latest Marine Recorder snapshot (June 2013)	Validate historical records of <i>Modiolus modiolus</i> as MPA search features in Loch Broom and Loch Ewe MPAs (negative for both).	Heriot Watt University Moore <i>et al</i> (2011a)

Site	MNCR Sector	Month-Year	<i>M. modiolus</i> den data	sity indicators	<i>M. modiolus</i> community indicators data		Marine Purpose of Data Collection Recorder		Source
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key		
Loch Creran*	South- west Scotland	August 1999	In situ: cross- hair and clearance quadrats. MNCR transects (SACFOR abundance for <i>M. modiolus</i>). Video mosaics of 5x5m ² / Video transects / camera stills		In-situ: replicate <i>M.</i> <i>modiolus</i> clumps MNCR Phase II transects Video mosaics and camera stills		MRSNH012 00000002	Literature review and comparative study of <i>Modiolus</i> beds in Busta Voe, Loch Alsh and Loch Creran. Population structure, density and community composition.	Mair <i>et al</i> (2000)
Loch Creran*	South- west Scotland	July 2005	In situ: cross- hair and clearance quadrats. MNCR transects (SACFOR abundance for <i>M. modiolus</i>)	Side-scan sonar used only for Serpulid reef detection.	Replicate clumps (quantitative) MNCR Phase II transects		MRSNH017 00000002	Mapping and characterisation of Serpula vermicularis and Modiolus modiolus reefs in Loch Creran. Distribution, density, community composition and population structure of Modiolus modiolus reefs.	Moore <i>et al</i> (2006)
Loch Creran *	South- west Scotland	August 2012	In situ: cross- hair and clearance quadrats. 25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)		Replicate clumps (quantitative)		Not in latest Marine Recorder snapshot (June 2013)	MPA feature characterisation survey at Loch Alsh, Creran, Fyne and Duich. <i>Modiolus modiolus</i> surveys focussed on Lochs Alsh and Creran only although records exist for Loch Duich.	Moore <i>et al</i> (2013)
Loch Duich	North-west Scotland	June 2004		ROV transects For biotope assignation only	MNCR Phase II and quadrat surveys	ROV transects For biotope assignation only	MRSNH012 00000007	Biotope assignation surveys at Loch Duich, Long and Alsh. Detailed repeated <i>Modiolus</i> <i>modiolus</i> surveys at String Rock (Kyleakin) site in Loch Alsh only.	Emu Ltd. (2006)
Loch Duich	North-west Scotland	August 2012	Diver video and photo stills at MNCR transects	DDV tows	Diver video and photo stills at MNCR transects	DDV tows	Not in latest Marine Recorder snapshot (June 2013)	Survey aimed exclusively at validating <i>Limaria hians</i> beds in Loch Duich.	Moore <i>et al</i> (2013)

Site	MNCR Month-Year M. modiolus density indicators M. modiolus community indicators Sector data data data		inity indicators	Marine Recorder	Purpose of Data Collection	Source			
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key		
Loch Ewe	North-west Scotland	August 2010		DDV and towed cameras (stills and video)	Spot dives for <i>M.</i> modiolus presence	DDV and towed cameras (stills and video) 0.1m ² Van Veen grabs	Not in latest Marine Recorder snapshot (June 2013)	Validate historical records of Modiolus modiolus as MPA search features in Loch Broom and Loch Ewe MPAs (negative for both).	Moore <i>et al</i> (2011a)
Loch Fyne	Clyde Sea	August 2012			Validation dive at Kilbride Island		Not in latest Marine Recorder snapshot (June 2013)	Survey to validate <i>Limaria</i> records using MNCR protocols. An exploratory dive for <i>Modiolus modiolus</i> beds was undertaken (negative results).	Moore <i>et al</i> (2013)
Loch Laxford	North-west Scotland	August 2009	MNCR surveys	DDV tows	100m 4m-wide MNCR transects or 30m deep Video and stills Spot dives	DDV (SACFOR and biotopes) 0.1m ² Van Veen grab samples	Not in latest Marine Recorder snapshot (June 2013)	Validation of MPA search features in Loch Laxford. <i>Modiolus</i> found west of Eilean a'Chadh-Fi density unclear. Some off Eilean Port a'Choit	Moore <i>et al</i> (2010)
Loch Leven*	South- west Scotland	August 2011	In situ: cross- hair and clearance quadrats. 25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)		Replicate clumps (quantitative) MNCR Phase II transects	DDV at 66 stations (SACFOR and biotopes)	MRSNH018 00000016	Validation of historical records of MPA search features including <i>Modiolus modiolus</i> , <i>Limaria hians</i> and <i>Ascophyllum nodosum</i> in Lochs Linnhe, Etive, Leven and Eil	Moore <i>et al</i> (2012b)
Loch Linnhe – Port Appin *	South- west Scotland	August 2011	In situ: cross- hair and clearance quadrats. 25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)		Replicate clumps (quantitative) MNCR Phase II transects	DDV at 41 stations (SACFOR and biotopes)	MRSNH018 00000016	Validation of historical records of MPA search features including <i>Modiolus modiolus</i> , <i>Limaria hians</i> and <i>Ascophyllum nodosum</i> in Lochs Linnhe, Etive, Leven and Eil	Moore <i>et al</i> (2012b)

Site MNCR Sector		Month-Year	<i>M. modiolus</i> den data	sity indicators	<i>M. modiolus</i> commu data	nity indicators	Marine Purpose of Data Collection Recorder		Source
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key		
Loch Linne – Corpach*	South- west Scotland	August 2011	In situ: cross- hair and clearance quadrats. 25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)		Replicate clumps (quantitative) MNCR Phase II transects	DDV at five stations; supplemented by <i>in situ</i> diver video (SACFOR and biotopes)	MRSNH018 00000016	Validation of historical records of MPA search features including <i>Modiolus modiolus</i> , <i>Limaria hians</i> and <i>Ascophyllum nodosum</i> in Lochs Linnhe, Etive, Leven and Eil	Moore <i>et al</i> (2012b)
Loch Long	North-west Scotland	June 2004		ROV transects For biotope assignation only	MNCR Phase II and quadrat surveys within 5m radius of shotline	ROV transects For biotope assignation only	MRSNH012 00000007	Biotope assignation surveys at Loch Duich, Long and Alsh. Detailed repeated <i>Modiolus</i> <i>modiolus</i> surveys at String Rock (Kyleakin) site in Loch Alsh only.	Emu Ltd. (2006)
Loch Sunart	South- west Scotland	July 2001	Diver video at 25 dive sites, ground-truthing for acoustic data (biotope confirmation) Ground-truthing MNCR transect to supplement	AGDS / Side- scan and ROV for ground- truthing	Diver video at 25 dive sites, ground- truthing for acoustic data (biotope confirmation) Ground-truthing MNCR transects to supplement acoustic data. Only biotopes.	AGDS / Side- scan and ROV for ground- truthing Six stations, 0.1m ² Van Veen grab. Sieved 0.5mm	MRSNH001 00000004	Comprehensive biotope mapping survey at Loch Sunart using ground-truthed rapid broad-scale acoustic survey methods. Loch Tecuis/Slaen to Laudel aras- Not dense	Bates <i>et al</i> (2004)
North Cava*	Orkney	September 2011	25m x 4m MNCR Phase II transect (SACFOR abundance for <i>M. modiolus</i>)	DDV tows	Replicate clumps (quantitative) 25m x 4m MNCR Phase II transect	DDV tows	MRSNH025 00000004	Validation of MPA search features in Orkney waters.	Hirst <i>et al</i> (2012b)
North Pen Llŷn*	Cardigan Bay and north Wales	July 1998			25m x 4m MNCR Phase II transects on 10 sites		JNCCMNCR 40000771	1998 NRW North Lleyn sublittoral survey	Bunker <i>et al</i> (1999)
North Pen Llŷn*	Cardigan Bay and north Wales	1999-2012	Video footage of 0.25m ² stringed quadrats.	Repeat DDV tows from 2008 to 2010	Video footage of 0.25m ² stringed quadrats.	Repeat DDV tows from 2008 to 2010		Repeat monitoring surveys of <i>M. modiolus</i> bioherms in Pen Llŷn SAC including impacted sites	NRW unpublished data
Point of Ayre*	Isle of Man	2007-2009; 2012	Video footage of 0.25m ² stringed quadrats.					Repeat monitoring surveys of <i>M. modiolus</i> beds and impact studies.	NRW unpublished data

Site MNCR Sector		Month-Year	<i>M. modiolus</i> der data	nsity indicators	<i>M. modiolus</i> commo data	unity indicators	Marine Recorder	Purpose of Data Collection	Source
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key		
Noss Head*	North-east Scotland	September 2011	N/A	51 DDV stations for biotope characterizati- on and SACFOR Multibeam used for <i>Modiolus</i> beds	N/A	51 DDV stations for biotope characterizati- on and SACFOR	Not in latest Marine Recorder snapshot (June 2013)	Confirmation of the presence of <i>Modiolus modiolus</i> PMFs in the Noss Head areas. Included estimation of the extension of the feature using acoustic methods.	Hirst <i>et al</i> (2012a)
				extension estimates					
Noss Head*	North-east Scotland	2009/10		DDV and ROV surveys for presence and biotope assignment		DDV and ROV surveys to validate PMFs	Not in latest Marine Recorder snapshot (June 2013)	Improve knowledge on the presence and distribution of PMFs in areas around Scotland.	Moore & Roberts (2011b)
Shapinsay*	Orkney	September 2011		33 DDV tows. Biotope and SACFOR		33 DDV tows. Biotope and SACFOR	MRSNH025 00000004	Validation of MPA search features in Orkney waters.	Hirst <i>et al</i> (2012b)
South of Canna	South- west Scotland	2009/10		DDV and ROV surveys for presence and biotope assignment only		DDV and ROV surveys to validate PMFs	Not in latest Marine Recorder snapshot (June 2013)	Improve knowledge on the presence and distribution of PMFs in areas around Scotland. Deep bed (120- 180m) of <i>Modiolus modiolus</i> in the Sound of Canna.	Moore & Roberts (2011b)
South Ronaldsay	Orkney	2009/10		DDV and ROV surveys for presence and biotope assignment only		DDV and ROV surveys to validate PMFs	Not in latest Marine Recorder snapshot (June 2013)	Improve knowledge on the presence and distribution of PMFs in areas around Scotland.	Moore & Roberts (2011b)
Strangford Lough*	Northern Ireland	2010	0.25m ² clearance quadrats	ROV surveys to validate extent	0.25m ² clearance quadrats / dive surveys along transects		Not in latest Marine Recorder snapshot (June 2013)	Determining if favourable conservation status was achieved following introduction of mobile fishing gear bans	Roberts <i>et al</i> (2011)

Site	MNCR Sector	Month-Year	<i>M. modiolus</i> den data	sity indicators	<i>M. modiolus</i> commu data	<i>M. modiolus</i> community indicators data		Purpose of Data Collection	Source
			<i>In situ</i> work	Remote work	<i>In situ</i> work	Remote work	survey key		
Sullom Voe: Calback Ness and Voster Ness	Shetland	September 2004		AGDS, swath bathymetry and video ground- truthing for biotope identification		AGDS, swath bathymetry and video ground- truthing	Not in latest Marine Recorder snapshot (June 2013)	Broad-scale intertidal and subtidal habitat and biotope mapping in the Sullom Voe SAC	Mair <i>et al</i> (2010)
Sullom Voe: Calback Ness*	Shetland	August 2004	MNCR Phase II surveys (SACFOR for Modiolus)		Replicate clumps (semi-quantitative) MNCR Phase II surveys		Not in latest Marine Recorder snapshot (June 2013)	Site condition monitoring in the Sullom Voe SAC <i>Modiolus</i> <i>modiolus</i> beds to establish baseline conditions.	Mair <i>et al</i> (2009)
Sullom Voe: Calback Ness*	Shetland	1979-2001			Replicate clumps (semi-quantitative)	Biotope identification		Bi-annual monitoring surveys at the Sullom Voe oil terminal effluent BP discharge pipe.	Confidential unpublished reports, ERT ltd. on behalf of British Petroleum
Uyea Sound*	Shetland	September 2012	25m x 4m MNCR Phase II transect	DDV tows	Replicate clumps (quantitative) 25m x 4m MNCR Phase II transect	DDV tows 0.1m2 Van Veen grabs	Not in latest Marine Recorder snapshot (June 2013)	MPA search feature and quality assessment in suitable areas in Shetland (Fetla to Haroldswick)	Hirst <i>et al</i> 2012b: SNH Commission-ed report No. 509

Appendix 2.2. Summary of references and datasets (Sabellaria spinulosa)

Table A2.2 Summary of all sources of possible *Sabellaria spinulosa* aggregation/reef data in the UK known to the authors of this work. Details of the data types collected, the ownership, vintage and the restrictions/availability of the data for use in this study are also provided where known. For clarity, raw data used in the current study are indicated with an asterisk. REC=Regional Environmental Characterisation; MALSF=Marine Aggregate Levy Sustainability Fund; AGDS=Acoustic Ground Discrimination System;

Data Source	Year(s)	Associated Reference(s)	Data Owner	Data Types Collected	Restrictions / Availability for this Study
Humber REC*	2011	Tappin <i>et al</i> 2011	Defra (MALSF)	Acoustic data (corridors only) Grab samples Seabed images	Publically available and used in this study http://www.marinealsf.org.uk/
East Coast REC*	2011	Pearce <i>et al</i> 2011; Limpenny <i>et al</i> 2011	Defra (MALSF)	Acoustic data Grab samples Seabed images Sabellaria polygons	Publically available and used in this study http://www.marinealsf.org.uk/
Thames REC*	2010	Emu Ltd 2009	Defra (MALSF)	Acoustic data (corridors only) Grab samples Seabed images	Publically available and used in this study http://www.marinealsf.org.uk/
South Coast REC*	2010	James <i>et al</i> 2011; James <i>et al</i> 201	Defra (MALSF)	Acoustic data (corridors only) Grab samples Seabed images	Publically available and used in this study http://www.marinealsf.org.uk/
Hastings Shingle Bank Research Study*	2007	Pearce <i>et al</i> 2007	The Crown Estate, Natural England and Defra	Grab samples Seabed images Sabellaria polygons	Publically available and used in this study http://www.cefas.defra.gov.uk/alsf/projects/natural-seabed- resources/mal0027.aspx
Rehabilitation of the Seabed Following Aggregate Extraction Part II	2001 2004	Cooper <i>et al</i> 2005	Cefas	Acoustic data Grab samples Seabed images	Data not requested as <i>S. spinulosa</i> habitats were only sampled incidentally and more recent and coherent data were available from the same area (Hastings Shingle Bank).
Cutline Research Study*	2008 2009 2010	Pearce <i>et al</i> 2011	Defra	Acoustics Grab samples Seabed images	Publically available and used in this study http://www.cefas.defra.gov.uk/alsf/projects/natural-seabed- resources/08p39.aspx
North Sea, English Channel and Irish Sea Beam Trawl Surveys	1992- 1996	N/A	Cefas	Beam trawl samples	Data not requested as type not compatible with the current study.
An ecological survey of the rocky coast adjacent to a Bromine extraction works	1974	Hoare & Hiscock 1974	N/A	Diver observations	Data not available. Authors no longer have a copy of the raw data.
Bristol Channel	1985	George & Warwick 1985	N/A	Grab samples	Data only kept as paper records which could not be located but the paper gives some indication of the diversity associated with these reefs. Qualitative comparisons are therefore made in the text.
Vicky Hendrick PhD Surveys of Area 107 (Box 1) and the outer Wash (Box 4)	2003 2004 (x3) 2005	Hendrick 2007	n/a	Grab samples	Data collected from Area 107 and the Outer Wash. Data requested but not provided.

Data Source	Year(s)	Associated Reference(s)	Data Owner	Data Types Collected	Restrictions / Availability for this Study
Sublittoral survey of the north coast of the outer Solway (Mull of Galloway to Auchencairn)	1992 2007	Covey 1992; ERT (Scotland) Ltd 2011	SNH	Grab samples	Most recent surveys of Luce Bay (2007) did not find any <i>S. spinulosa</i> aggregations. Historic images (from the early 90's) were not available for further analysis but the associated report gives some indication of the diversity associated with these reefs. Qualitative comparisons are therefore made in the text.
The Wash and North Norfolk cSAC surveys	1996-2001	Foster-Smith <i>et al</i> 1997; Foster-Smith & Sotheran 1999; Foster-Smith & White 2001; Foster-Smith & Hendrick 2003)	Natural England	AGDS data (RoxAnn) Grab samples Trawl samples Dredge samples Towed video	Requested but not provided – data currently sits with Bob Foster- Smith.
Baseline Survey of Saturn Reef cSAC	2003	N/A	Natural England	Swath Bathymetry Video	Data not requested. Data type not compatible with the current study. Note no well-developed reef was identified in this survey.
Baseline Survey of Haisborough, Hammond and Winterton cSAC	2011	N/A	Cefas	Grab samples Seabed images	Data was not made available in time for inclusion in the current study but would be suitable for any further work in this area.
Fauna associated with longitudinal furrows and sand ribbons in a tide-swept area in the English Channel	1985	Holme & Wilson 1985	N/A	Acoustic data Towed video	Data not requested as type not compatible with this study.
Lynn Knock and Area 107 Sabellaria surveys	2001 2003 2004 (x2) 2005	N/A	Natural England	Grab samples Video footage	Requested but not provided – data currently sits with Bob Foster- Smith.
Area 107, Saturn Reef, Hastings Shingle Bank, Swanage Bay Surveys to Evaluate Sampling methods	2005 2006	Limpenny <i>et al</i> 2010)	Defra (MALSF) and JNCC	Acoustic data Seabed images Grab samples Beam trawl samples	Data requested but not made available in time to be included in this study. Note seabed images are not of sufficient quality / type for density estimations or the assessment of faunal diversity but some of the grab data could be included if this work were to be expanded or repeated. Note significant reef structures were not identified at the Saturn Reef site during these surveys.
Sussex Seasearch Surveys	1996	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
Dorset Seasearch Surveys	1995-2004	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
Kent Seasearch Surveys	2005-2009	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
North Llyn Peninsula, North Wales Seasearch Surveys	2005	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
West Anglesey, North Wales Seasearch Surveys	2006	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
North East Durham Heritage Coast Seasearch Surveys	2009	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.

Data Source	Year(s)	Associated Reference(s)	Data Owner	Data Types Collected	Restrictions / Availability for this Study
Tynemouth Coast Seasearch Surveys	2009	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
Yorkshire Flamborough Seasearch Surveys	2009	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
Norfolk Coast Seasearch Surveys	2010	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
Lincolnshire Seasearch Surveys	2011	N/A	Marine Conservation Society	Diver observations Seasearch records	Data not requested. Data type not compatible with current study.
Dorset marine habitat surveys	2000- 2005	Collins 2003; Collins 2005; Collins 2005	University of Southampton	Acoustics Towed video Diver observations	Data not requested as data types not suitable for this study
The Wash	2005- ongoing	Jessop <i>et al</i> 2012	Eastern IFCA	AGDS data Grab samples and photographs of grab samples	Advised that grab samples were never fully analysed. A note of the presence or absence of <i>Sabellaria spinulosa</i> was noted and some aspects of the aggregations such as height were recorded where present. Data not compatible with the current study.
Aggregate Extraction Licence Area 106 East (Area 480)	2002 2008	Marine Ecological Surveys Ltd 2008	Hanson Aggregates Marine Ltd	Acoustic data Grab samples Trawl samples Seabed images	Data not requested as although well-developed S. <i>spinulosa</i> reefs were identified in the acoustic data there was limited ground truthing carried out and seabed images displayed in the associated report did not appear to be of a high enough resolution for automated image analysis. These data might be suitable for any further work in this area, especially where there is more time available for data assessment and collation.
Aggregate Extraction Licence Area 107	1994-1997	N/A	South Coast Shipping Co Ltd (now CEMEX UK Ltd)	Acoustic data Drop down video	Data not requested. Data type not compatible with the current study. Video was excluded ant an early stage of the project. These data may warrant further assessment where more time is available for data collation.
Aggregate Extraction Licence Areas 366-370 Hastings Shingle Bank	2005	Marine Ecological Surveys Ltd 2006	Resource Management Association (RMA)	Side-scan sonar Grab samples Seabed images	Data not requested as more comprehensive data available from the Hastings Shingle Bank research study.
Aggregate Extraction Licence Area 401	2008	Emu Ltd 2008; Marine Ecological Surveys Ltd 2001; Unicomarine Ltd 1993; Unicomarine Ltd 1995	Hanson Aggregates Marine Ltd	Grab samples Trawl samples Seabed images	Data not requested. Direct sampling of <i>S. spinulosa</i> avoided in recent years. More recent and comprehensive data available from the East Coast REC.
Aggregate Extraction Licence Area 430 Southwold	1997 2006	Marine Ecological Surveys Ltd 2007	Tarmac Ltd and CEMEX Marine UK Ltd	Acoustic Data Grab samples Trawl samples Seabed imagery	Data not requested. Sabellaria spinulosa aggregations only recorded from one station. More recent and comprehensive data available from the East Coast REC.
Aggregate Extraction Areas 432/1 & 432/2 West Varne	1996	Marine Ecological Surveys Ltd 1997	CEMEX Marine UK Ltd	Grab samples Trawl samples	Data not requested. Only a small number of incidental samples taken from S. spinulosa habitat.
Aggregate Extraction Area 447 Cutline	2008	Marine Ecological Surveys Ltd 2008	Resource Management Association (RMA)	Side-scan sonar Grab samples Seabed images	Data not requested. More comprehensive and recent data available from the Cutline Research study.

Data Source	Year(s)	Associated Reference(s)	Data Owner	Data Types Collected	Restrictions / Availability for this Study
Aggregate Extraction Licence Area 454 Lowestoft	1996	Marine Ecological Surveys Itd 1996	CEMEX UK Ltd	Grab samples Trawl samples	Data not requested. Only a small number of incidental samples taken from <i>S. spinulosa</i> habitat.
Aggregate Extraction Area 460 Hastings Shingle Bank	2005	Marine Ecological Surveys Ltd 2006	Resource Management Association (RMA)	Side-scan sonar Grab samples Seabed images	Data not requested as more comprehensive data available from the Hastings Shingle Bank research study .
Aggregate Extraction Area 481	2003 2008	Marine Ecological Surveys Ltd 2009	Tarmac Ltd and Van Oord UK Ltd	Acoustic data Grab samples Trawl samples	Data not requested. More recent and comprehensive data available from the Humber REC.
Proposed Aggregate Extraction Area Benacre	2001	Marine Ecological Surveys Ltd 2002	Sea Aggregates Ltd.	Grab samples Trawl samples	Data not requested as only a small number of samples taken of suspected <i>S. spinulosa</i> reefs and company is no longer trading in the UK.
Thanet Offshore Windfarm Licencing Surveys*	2005 2007 2012	Marine Ecological Surveys Ltd 2005; Marine Ecological Surveys Ltd 2007; Marine Ecological Surveys Ltd 2012	Vattenfall	Acoustic data Grab samples (2005 and 2007 only from <i>Sabellaria</i> habitats) Seabed images <i>Sabellaria</i> polygons "Before and After" data	Made available for this study through a data agreement with the data owner.
Lincs (LID6) Windfarm Export Cable Route Survey*	2008	Fugro Survey Ltd, 2008	Centrica Renewable Energy Ltd	Seabed images	Made available for this study through a data agreement with the owner. Seabed images could not be used in this study as they were not supplied as image files.
Lincs (LID6) Windfarm Licencing Surveys*	2004 2010	EGS International Ltd 2011; Envision Mapping Ltd 2005	Centrica Renewable Energy Ltd	Acoustic data Grab samples Seabed images	Made available for this study through a data agreement with the owner. Grab sample data could not be used as the taxonomic analysis and enumeration were inconsistent with all other datasets.
Race Bank and Docking Shoal Export cable Route Survey*	2006	Emu Ltd 2007	Centrica Renewable Energy Ltd	Grab samples Drop down video	Made available for this study through a data agreement with the owner. Data could not be used as the raw data and images were not provided (PDF reports only). Grab sampling were not taken where <i>S. spinulosa</i> habitats were observed in the seabed images.
Race Bank & Docking Shoal Surveys*	2004 2006	Institute of Estuarine and Coastal Studies 2007; Envision Mapping Ltd 2005	Centrica Renewable Energy Ltd	Grab samples Beam trawl samples Drop down video	Made available for this study through a data agreement with the owner. Data could not be used as the raw data and images were not provided (PDF reports only). Grab sampling were not taken where <i>S. spinulosa</i> habitats were observed in the seabed images.
Sheringham Shoal Offshore Windfarm Licenceing Surveys	2004	Envision Mapping Ltd 2005	Scira Offshore Energy Itd (Stakraft and Statoil)	Acoustic data Grab samples Video footage	Data not requested.
East Anglia Offshore Windfarm Licencing Surveys	2010	Marine Ecological Surveys Ltd 2010; Marine Ecological Surveys Ltd 2011	East Anglia Offshore Wind (Scottish Power Renewables and Vattenfall)	Acoustic data Grab samples Seabed images Trawl samples	Data not requested as more comprehensive data available from the East Coast REC.
Surveys of the Stour, Orwell and Harwich Approaches	1997 2003	Unicomarine Ltd 2005	Harwich Haven Authority	Grab samples Trawl samples Towed video	Data not requested.

Data Source	Year(s)	Associated Reference(s)	Data Owner	Data Types Collected	Restrictions / Availability for this Study
Saturn pipeline installation surveys	2003	BMT Cordah Ltd 2003	ConocoPhillips	Side-scan sonar ROV	Data not requested. Data types not compatible with the current study.
Wash Surveys	1991 1993 1999 2002 2003	Ecomaris Ltd 2001; Unicomarine Ltd 1994	National Rivers Authority (NRA) / Environment Agency	Grabs samples	Report sand data requested from a number of sources but could not be located.
South West Britain Sublittoral Surveys: Upper Bristol Channel sublittoral survey	1978-1979	Hiscock 1979	Joint Nature Conservation Committee (JNCC)	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
South West Britain Sublittoral Surveys: Lundy sublittoral survey	1978-1979	Hiscock 1981	Joint Nature Conservation Committee (JNCC)	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
Bardsey and the Lleyn Peninsula sublittoral survey	1983	Hiscock 1983	Joint Nature Conservation Committee (JNCC)	Diver observations	Data not requested. Data type not compatible with current study.
MNCR Berwick-on-Tweed to Newbiggin sublittoral survey	1992	N/A	Joint Nature Conservation Committee (JNCC)	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
MNCR Newbiggin to Saltburn sublittoral survey	1993	N/A	Joint Nature Conservation Committee (JNCC)	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
MNCR Saltburn to Flamborough Head sublittoral survey	1993	N/A	Joint Nature Conservation Committee (JNCC)	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
MNCR Blyth to Flamborough Head sublittoral sediment survey	1993	Cullinane 1974	Joint Nature Conservation Committee (JNCC)	Grab samples	Data not requested.
MNCR south Isle of Wight sublittoral survey	1997	N/A	Joint Nature Conservation Committee (JNCC)	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
MNCR Ceredigion coast sublittoral survey	1995-1997	Brazier <i>et al</i> 1999	Grab samples	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
MNCR west Anglesey sublittoral survey	1996	Brazier <i>et al</i> 1999	Joint Nature Conservation Committee (JNCC)	Diver observations Phase li Records	Data not requested. Data type not compatible with current study.
Blackwater Estuary sublittoral sediment survey	1997	N/A	Natural England	Grab samples	Data not requested. Two grab samples only.
South Wight Maritime cSAC sublittoral survey	1999	N/A	Natural England	Diver observations Phase II Records	Data not requested. Data type not compatible with current study.
Flamborough Head sublittoral survey	2002	N/A	Natural England	Diver observations Phase II Records and Quadrats	Data not requested. Data type not compatible with current study.

Data Source	Year(s)	Associated Reference(s)	Data Owner	Data Types Collected	Restrictions / Availability for this Study
St Brides Bay Sublittoral Sediment Benthic Survey	2000	Rostron 2001	Natural Resources Wales (NRW)	Grab samples	Data not requested.
Macrofaunal Survey of Welsh Sandbanks	2001	N/A	Natural Resources Wales (NRW)	Grab samples	Data not requested.
Across Wales towed video monitoring survey	2004-2005	Seastar Survey 2006	Natural Resources Wales (NRW)	Towed video	Data not requested. Data type not compatible with current study.
North Wales towed video survey trials	1999	Sanderson <i>et al</i> 1999	Natural Resources Wales (NRW)	Towed video	Data not requested. Data type not compatible with current study.
North Lleyn sublittoral survey	1998	Marine Seen 1998	Natural Resources Wales (NRW)	Diver observations Phase II records	Data not requested. Data type not compatible with current study.
Llyn Peninsula sublittoral monitoring trials	1998	Sanderson <i>et al</i> 1999	Natural Resources Wales (NRW)	Diver observations Phase II records	Data not requested. Data type not compatible with current study.
HABMAP sublittoral survey	2005	Robinson <i>et al</i> 2009	Natural Resources Wales (NRW)	Dredge samples	Data not requested. Data type not compatible with current study.
Skomer MNR sublittoral macrobenthos survey	1998	Barfield 1999	Natural Resources Wales (NRW)	Recording Phase I	Data not requested. Data type not compatible with current study.
Cardigan Bay sublittoral sediment survey	1997	Brazier <i>et al</i> 1999	Joint Nature Conservation Committee (JNCC)	Grab samples	Data not requested.
Lyme Bay sublittoral sediment survey	1994	Ambios Environmental Consultants 1995	Kerr-McGee Oil (UK)	Grab samples	Data not requested.
Sellafield sublittoral sediment survey	1992	N/A	British Nuclear Fuels	Pipe dredge samples	Data not requested. Data type not compatible with the current study.
Holderness Coast- Aldbrough sublittoral sediment survey	1998	N/A	Institute of Estuarine and Coastal Studies	Grab samples	Data not requested
Holderness Coast-Easington sublittoral sediment survey	1991	N/A	Institute of Estuarine and Coastal Studies	Grab samples	Data not requested
Race Bank sublittoral sediment survey	1993	N/A	Institute of Estuarine and Coastal Studies	Grab samples	Data not requested
Inner Dowsing sublittoral sediment survey	1999	N/A	Institute of Estuarine and Coastal Studies	Grab samples	Data not requested
Thames Estuary sublittoral sediment survey	1996	N/A	Institute of Estuarine and Coastal Studies	Grab samples	Data not requested
Selsey Bill National Marine Monitoring programme	N/A	N/A	Joint Nature Conservation Committee (JNCC)	n/a	Data not requested

Appendix 3. Statistical Methods

Appendix 3.1. Univariate Statistical Methods

i. Univariate community indices

The following diversity indices were calculated using the PRIMER v6 software package (Clarke and Gorley 2006; Clarke and Warwick 2001b):

a. Margalef's Species Richness (d')

Margalef's d index (Margalef 1958) is a widely used species richness index which compensates the effect of sampling size by dividing the number of species (S) by the total abundance of individuals sampled (N) as expressed by the formula:

$$d = \frac{S-1}{\log_2 N}$$

b. Shannon Diversity (H')

The Shannon information index (Shannon & Weaver 1949) is calculated by the formula:

$$H' = -\sum_{i=1}^{S} \sum p_i \ln p_i$$

where p_i is the proportion of the total count (N) accounted by the ith taxa. The Shannon diversity index is a measure of the entropy of the system, and was first used by Pielou (1969) as a measure of the diversity of a biological community. The values usually range from 1.5 to 4.5, a value higher than 5 is often rare and indicates a high number of species (> 10^5 ; Margalef 1975 cited in Magurran 2004).

c. Simpson's Diversity $(1-\lambda)$

Simpson's diversity index is derived from the number of species present as well as the relative abundance of each species. A high Simpson's diversity index (approaching 1) indicates a high number of species and an even spread of the abundance between those species (evenness). A low diversity index (approaching zero) indicates a low number of species or an uneven spread of the abundance between the species present.

d. Pielou's Evenness (J)

Evenness (Pielou 1977) is a measure of how similar species are in terms of their abundance. It is calculated by the formula:

$$J = \frac{H'_{observed}}{H'_{max}}$$

where H'_{max} is the maximum Shannon diversity that can be achieved if all species were equally abundant (=log₂S).

A high evenness value (approaching 1) indicates that the majority of species are present in equal abundance. Conversely, a low evenness value indicates that one or more species is numerically dominant.

ii. Correlation Indices

Least squares regression models and non-parametric Spearman's correlation indices (r_s) were calculated to respectively determine the relationship and the correlation strength between the associated macrofaunal community (described by abundance and diversity of taxa as well as community evenness) and the density of the reef forming species, if any. Correlation matrices were constructed using the R function scatterplot Matrix within the library car (Fox and Weisberg 2011). The output includes histograms and smooth curves showing the frequency distribution for each variable and scatterplots between the variables compared fitted with lowess and best fit lines.

iii. Approach to statistical modelling of univariate indices

Number of individuals (N), Shannon-Wiener's diversity (H'), Margalef's richness (d) and Pielou evenness (J) were imported into R (version 2.13.1) and tested for normality and heteroscedasticity. Spatial and temporal variation of diversity and evenness indices were tested (α of 0.05) by fitting linear mixed effects models (LMMs: lme4 package; [41]) with individual sampling sites and replicates as random factors to account for spatial and temporal pseudoreplication. Sampling year and broad location were the categorical predictors (fixed factor) in the mixed models. Generalized LMMs with Poisson error distribution and logit link function were fitted to the abundance data (N; M. modiolus) incorporating the same fixed and random factors as the LMMs to cope with non-normal data in unbalanced, mixed-effect experiments (see Bolker et al (2009) and references therein). Overdispersed Poisson models were refitted using Penalized Quasi Likelihood approximations (glmmPQL: MASS package (Venables and Ripley 2002). The Akaike Information Criterion (AIC) was used to assess the effect of the physical impact on the null model for PoA and nLP while controlling for the random effects. Model selection was based on the lowest AIC score (most parsimonious model once all parameters have been fitted). All models were tested using residual plots to confirm that the assumptions of normality and sphericity of the residuals were met. For statistical model fitting definitions see Crawley (2013; pp. 391-393). Venables and Dichmont (2004) describe the theoretical basis of GLMs and GLMMs in detail.

Appendix 3.2. Multivariate Statistical Methods

i. Multivariate statistical analysis

Quantitative species abundance data were subject to a meta-analysis using the Plymouth Routines for Multivariate Analysis in Ecology (PRIMER v6) software package (Clarke and Gorley 2006; Clarke and Warwick 2001). The following routines were employed in this investigation:

a. Multidimensional Scaling (MDS) Ordination

This technique allows the construction of a "map" or configuration of the samples in multidimensional space. This configuration attempts to position the samples as accurately as possible to reflect the similarity between the samples. For example, if sample 1 has a greater similarity to sample 2 than it does to sample 3 then sample 1 will be positioned more closely to sample 2 than it is to sample 3. This "map" of the relative similarities between samples is then plotted in two dimensions. It is important to remember that this two-dimensional plot is a representation of a multidimensional picture. When large numbers of samples are analysed, or datasets that include samples that are very different from one another, the accuracy of the two-dimensional plot may be reduced. A measure of the accuracy of the two-dimensional representation (stress) is given on the MDS plot. Stress values <0.1 correspond to a good

ordination; values <0.2 give a useful two-dimensional picture but one should not place too much reliance on the fine details of the plot; stress >0.2 indicates that the samples are close to being positioned in an arbitrary manner and should not be regarded as necessarily similar to one another, particularly in the upper half of this range.

b. The PERMANOVA test

The PERMANOVA test is a Permutational Multivariate ANOVA test based on distances or dissimilarities. This test allows us to identify differences which exist between groups of samples, where the groups have been determined according to some a-priori factor such as substrate type, depth zone, biotope class etc. Because this test is based on permutations of the actual data, no assumptions are made about the distribution of the data making it a robust test for faunal abundance type data. This test also allows for full partitioning of the variability so that interaction terms can be investigated and sources of variability more thoroughly explored.

c. Similarity of Percentages (SIMPER) Routine

The SIMPER routine allows for comparisons to be made between groups of samples. SIMPER analysis identifies species that typify a group as well as those that account for the dissimilarities between groups. The initial tables of the SIMPER analysis list the typical taxa (found in consistently high abundances) in each group while pair-wise comparisons of groups are also given, indicating where the differences in faunal assemblage composition lie for each community sampled.

d. The RELATE Test

The RELATE test provides a means of testing for correlations between two multivariate patterns. This is most often used to test for correlations between biological communities and sets of environmental variables, or between two different biological datasets such as predator and prey communities. The RELATE test is used here to test for correlations between *S. spinulosa* density and diversity, and gradients of anthropogenic disturbance using model distances between samples along the gradients (Somerfield *et al* 2002). As an example, this technique has been used to test for differences in the number of species recorded in *S. spinulosa* aggregations subjected to different levels of fishing pressure. The samples were categorised according to fishing pressure they were exposed to in terms of sightings per unit effort (SPUE) and then ranked. The sample ranks were then used to create a model matrix and corresponding resemblance matrix based on Euclidean distance, and this was used to test for a gradient response in the number of species recorded in the same samples.

Appendix 4. Statistical Analyses

Appendix 4.1. Summary of taxa contributing to 90% of the withingroup similarity and 60% between-group dissimilarities for SIMPROF Modiolus modiolus groupings

Table A4.1 Summary of taxa contributing to 90% of the within-group similarity and 60% between-group dissimilarities for SIMPROF *Modiolus modiolus* groupings. The data are derived from a SIMPER test carried out on square root transformed benthic abundance data from MNCR Phase II type dive surveys.

Group GI

Less than 2 samples in group

Group GII

Average similarity: 63.74

Species	Av.Abund	Av.Sim	Contrib%	Cum.%
Modiolus modiolus	2.24	5.73	8.99	8.99
Ophiopholis aculeata	2.24	5.73	8.99	17.98
Amphilectus fucorum	2.16	5.31	8.33	26.31
Chorda filum	1.91	4.65	7.29	33.61
Clausinella fasciata	1.82	4.44	6.96	40.57
Protula tubularia	1.8	4.09	6.41	46.98
Bugula avicularia	1.72	3.87	6.08	53.06
Pagurus	1.63	3.87	6.08	59.13
Spirobranchus lamarcki	1.72	3.87	6.08	65.21
Sagartia elegans	1.41	3.62	5.69	70.9
Ophiocomina nigra	1.28	2.89	4.53	75.43
Pagurus bernhardus	1.28	2.89	4.53	79.96
Pione vastifica	1.41	2.56	4.02	83.98
Ascidiella scabra	1.24	1.56	2.45	86.43
Plumularia setacea	1.15	1.56	2.45	88.89
Adamsia carciniopados	1.24	1.36	2.14	91.02

Group GIII

Average similarity: 42.71

Species	Av.Abund	Av.Sim	Contrib%	Cum.%
Modiolus modiolus	2.24	2.47	5.78	5.78
Liocarcinus depurator	1.72	1.71	4.01	9.78
Bugula avicularia	1.63	1.69	3.96	13.74
Asterias rubens	1.6	1.56	3.66	17.4
Ectocarpaceae	1.68	1.52	3.56	20.95
Hiatella arctica	1.35	1.41	3.31	24.27
Spirobranchus lamarcki	1.75	1.4	3.27	27.54
Ophiopholis aculeata	1.72	1.35	3.16	30.7
Balanus crenatus	1.52	1.29	3.02	33.72
Corella parallelogramma	1.31	1.21	2.83	36.55
Kirchenpaueria pinnata	1.37	1.12	2.62	39.17
Verruca stroemia	1.41	1.11	2.6	41.77
Dendronotus frondosus	1.36	1.03	2.4	44.17
Pomatoschistus minutus	1.22	0.97	2.28	46.46

Coryphella	1.22	0.97	2.26	48.72
Clausinella fasciata	1.26	0.96	2.26	50.98
Protula tubularia	1.21	0.92	2.16	53.13
Pagurus	1.33	0.92	2.15	55.28
Plumularia setacea	1.33	0.9	2.11	57.39
Chorda filum	1.19	0.87	2.03	59.42

Group GIV

Average similarity: 39.56

Species	Av.Abund	Av.Sim	Contrib%	Cum.%
Modiolus modiolus	2.24	4.19	10.59	10.59
Protula tubularia	2.12	3.75	9.47	20.06
Laminaria hyperborea	1.98	3.24	8.2	28.27
Pagurus	1.87	3.24	8.2	36.47
Bugula avicularia	1.57	2.65	6.7	43.17
Amphilectus fucorum	1	1.87	4.74	47.9
Antedon bifida	1.21	1.87	4.74	52.64
Balanus crenatus	1	1.87	4.74	57.38
Caridea	1.37	1.87	4.74	62.11
Clausinella fasciata	1.21	1.87	4.74	66.85
Corella parallelogramma	1.21	1.87	4.74	71.58
Cryptopleura ramosa	1	1.87	4.74	76.32
Gibbula umbilicalis	1.21	1.87	4.74	81.06
Leptochiton asellus	1.21	1.87	4.74	85.79
Phrynorhombus regius	1.5	1.87	4.74	90.53

Group GV

Average similarity: 39.49

Species	Av.Abund	Av.Sim	Contrib	% Cum.%
Modiolus modiolus	2.23	3.97	10.04	10.04
Ectocarpaceae	1.91	3.47	8.79	18.83
Ascidiella scabra	1.82	3.31	8.39	27.22
Balanus crenatus	1.8	3.05	7.73	34.95
Amphilectus fucorum	1.52	2.7	6.85	41.8
Cryptopleura ramosa	1.49	2.4	6.07	47.87
Ophiopholis aculeata	1.66	2.35	5.94	53.81
Antedon bifida	1.38	2.18	5.53	59.35
Liocarcinus depurator	1.14	1.91	4.84	64.19
Necora puber	1.14	1.91	4.84	69.03
Pagurus	1.14	1.91	4.84	73.88
Galathea intermedia	1.33	1.32	3.35	77.23
Munida sarsi	1.05	0.94	2.37	79.6
Chlamys distorta	1.05	0.93	2.35	81.95
Ascidiella aspersa	0.8	0.66	1.68	83.63
Galathea strigosa	0.91	0.66	1.68	85.31
Henricia	0.8	0.66	1.68	86.99
Pandalus	0.8	0.66	1.68	88.66
Spirobranchus	0.67	0.66	1.68	90.34

Group GVI

Average similarity: 44.17

Species	Av.Abund	Av.Sim	Contril	o% Cum.%
Limaria hians	2.24	3.01	6.82	6.82
Ophiopholis aculeata	2.34	3.01	6.82	13.65
Modiolus modiolus	2	2.7	6.1	19.75
Balanus balanus	1.73	2.34	5.29	25.04
Ectocarpaceae	1.87	2.34	5.29	30.33
Bugula avicularia	1.57	1.91	4.32	34.64
Munida rugosa	1.71	1.91	4.32	38.96
Aglaothamnion priceanum	1	1.35	3.05	42.01
Botryllus schlosseri	1	1.35	3.05	45.06
Brosme brosme	1	1.35	3.05	48.11
Capulus ungaricus	1.37	1.35	3.05	51.17
Corella parallelogramma	1	1.35	3.05	54.22
Cryptopleura ramosa	1	1.35	3.05	57.27
Diaphorodoris luteocincta	1	1.35	3.05	60.32

Group GVII

Average similarity: 47.94	
Species	ł

Species	Av.Abund	Av.Sim	Contrib%	Cum.%
Modiolus modiolus	2.3	4.12	8.59	8.59
Ophiopholis aculeata	2.21	3.8	7.93	16.52
Pagurus	1.73	3.29	6.86	23.39
Bugula avicularia	1.88	3.1	6.48	29.86
Ophiocomina nigra	1.88	3.1	6.48	36.34
Antedon bifida	1.72	2.87	5.99	42.33
Corella parallelogramma	1.41	2.69	5.6	47.93
Ascidiella scabra	1.55	2.2	4.58	52.51
Rhodophyceae crusts	1.38	2.15	4.48	56.99

Groups GII & GV

Average dissimilarity = 77.05

	Group Gll	Group GV			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Chorda filum	1.91	0	14.7	2.72	2.72
Ectocarpaceae	0	1.91	17.67	2.71	5.43
Protula tubularia	1.8	0	7.6	2.55	7.98
Balanus crenatus	0	1.8	8.38	2.55	10.53
Clausinella fasciata	1.82	0.33	2.66	2.14	12.67
Cryptopleura ramosa	0	1.49	3.49	2.14	14.81
Sagartia elegans	1.41	0	11.63	2.02	16.83
Antedon bifida	0	1.38	3.41	1.99	18.82
Pione vastifica	1.41	0	2.48	1.99	20.81
Bugula avicularia	1.72	0.33	2.39	1.98	22.79
Galathea intermedia	0	1.33	1.32	1.95	24.73
Pagurus bernhardus	1.28	0	7.6	1.81	26.54
Spirobranchus lamarcki	1.72	0.58	1.53	1.75	28.29
Adamsia carciniopados	1.24	0	1.32	1.71	30
Plumularia setacea	1.15	0	1.32	1.69	31.68

34

Group Gll	Group GV			
Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
0	1.14	3.86	1.64	33.32
0	1.14	3.86	1.64	34.96
0	1.05	1.28	1.54	36.5
0	1.05	1.33	1.51	38.02
1.05	0	1.3	1.44	39.46
0	1	1.2	1.42	40.88
0.94	0	1.32	1.36	42.24
0	0.91	1.16	1.35	43.6
1.05	0.33	1.33	1.35	44.94
1.28	0.47	1.5	1.3	46.24
0	0.8	1.23	1.18	47.42
0.94	0.33	1.37	1.18	48.6
0.8	0	1.3	1.17	49.77
0	0.8	1.3	1.16	50.93
0	0.8	1.3	1.16	52.1
0	0.8	1.27	1.1	53.19
0.47	0.8	1.18	1.07	54.26
0	0.67	0.67	1.03	55.29
0.47	0.67	1.39	0.99	56.29
0	0.67	1.32	0.97	57.26
0	0.67	1.32	0.97	58.23
1.24	1.82	0.82	0.95	59.18
0	0.67	1.33	0.91	60.09
	Group Gli Av.Abund 0 0 1.05 0 0.94 0 1.05 1.28 0 0.94 0.94 0.94 0.94 0.8 0 0.94 0.8 0 0 0.94 0.8 0 0.94 0.8 0 0 0.47 0 0 0.47 0 0 1.24 0	Group GIIGroup GVAv.AbundAv.Abund01.1401.1401.0501.051.0501.050010.940.911.050.331.280.470.940.330.940.330.940.330.940.330.940.330.940.330.940.80.940.80.470.80.470.670.470.670.470.671.241.8200.67	Group GIIGroup GVAv.AbundDiss/SD01.143.8601.143.8601.051.2801.051.331.0501.331.0501.320.940.911.320.940.911.321.050.331.331.280.471.50.940.331.370.940.81.320.940.811.320.940.811.320.940.811.320.940.811.320.470.811.320.470.671.320.470.671.320.470.671.320.470.671.320.470.671.320.470.671.320.470.671.320.470.671.320.470.671.32	Group GIIGroup GVAv.AbundDiss/SDContrib%01.143.861.6401.143.861.6401.051.281.5401.051.331.511.0501.331.44011.21.420.9401.321.360.940.911.161.351.050.331.331.351.050.471.51.300.81.231.180.940.331.371.180.940.81.31.160.940.81.31.160.940.81.31.160.940.81.31.160.940.81.31.160.940.81.321.160.940.81.321.160.940.81.321.070.940.671.320.970.940.671.320.971.241.820.820.950.950.671.330.91

Groups Gll & GIV

Average dissimilarity = 75.54

	Group GII	Group GIV			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib% Cum.%	
Laminaria hyperborea	0	1.98	12.76	2.84	2.84
Chorda filum	1.91	0	11.31	2.75	5.58
Spirobranchus lamarcki	1.72	0	6.9	2.46	8.05
Phrynorhombus regius	0	1.5	3.42	2.11	10.15
Sagartia elegans	1.41	0	9.64	2.04	12.19
Caridea	0	1.37	2.65	2.01	14.21
Pione vastifica	1.41	0	2.38	2.01	16.22
Ophiocomina nigra	1.28	0	6.84	1.83	18.04
Pagurus bernhardus	1.28	0	6.84	1.83	19.87
Corella parallelogramma	0	1.21	3.61	1.77	21.64
Gibbula umbilicalis	0	1.21	3.61	1.77	23.4
Adamsia carciniopados	1.24	0	1.27	1.73	25.13
Antedon bifida	0	1.21	8.47	1.72	26.85
Leptochiton asellus	0	1.21	8.47	1.72	28.57
Plumularia setacea	1.15	0	1.27	1.71	30.28
Amphilectus fucorum	2.16	1	9.88	1.66	31.94
Ophiopholis aculeata	2.24	1.22	1.14	1.65	33.59
Ascidiella scabra	1.24	0.5	1.36	1.59	35.18
Limaria hians	0	1	0.91	1.56	36.74
Bispira volutacornis	1.05	0	1.28	1.52	38.26

	Group GII	Group GIV			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib% Cum.%	
Hinia incrassata	1.05	0	1.26	1.46	39.72
Balanus crenatus	0	1	9.64	1.44	41.17
Cryptopleura ramosa	0	1	9.64	1.44	42.61
Porella concinna	0	1	9.64	1.44	44.05
Spirobranchus	0	1	9.64	1.44	45.5
Inachus phalangium	0.94	0	1.27	1.38	46.87
Urticina eques	0.94	0	1.27	1.38	48.25
Balanus balanus	0	0.87	0.91	1.35	49.6
Chaetopterus variopedatus	0	1	0.91	1.33	50.93
Porifera	0	1	0.91	1.33	52.26
Scrupocellaria scruposa	0.8	0	1.25	1.18	53.44
Testudinalia testudinalis	0	0.87	0.91	1.15	54.59
Lithothamnion glaciale	0	0.71	0.91	1.1	55.69
Necora puber	0	0.71	0.91	1.1	56.79
Saccharina latissima	0	0.71	0.91	1.1	57.89
Tectura virginea	0	0.71	0.91	1.1	58.99
Eledone cirrhosa	0	0.71	0.91	0.94	59.93

Groups GV & GIV

Average dissimilarity = 71.70

Group GV Group GIV

Species	Av.Abund	Av.Abund	Diss/SD	Contrib% Cum.%	
Protula tubularia	0	2.12	6.76	2.82	2.82
Laminaria hyperborea	0	1.98	11.57	2.61	5.44
Ectocarpaceae	1.91	0	13	2.53	7.96
Phrynorhombus regius	0	1.5	3.31	1.95	9.91
Galathea intermedia	1.33	0	1.28	1.81	11.72
Ascidiella scabra	1.82	0.5	2.05	1.8	13.51
Bugula avicularia	0.33	1.57	2.23	1.65	15.16
Gibbula umbilicalis	0	1.21	3.74	1.62	16.79
Ophiopholis aculeata	1.66	1.22	1.8	1.59	18.38
Leptochiton asellus	0	1.21	7.83	1.58	19.96
Liocarcinus depurator	1.14	0	3.81	1.53	21.49
Munida sarsi	1.05	0	1.24	1.43	22.92
Limaria hians	0	1	0.91	1.42	24.33
Chlamys distorta	1.05	0	1.28	1.41	25.74
Caridea	0.58	1.37	1.43	1.4	27.14
Porifera	0.33	1	1.12	1.27	28.41
Galathea strigosa	0.91	0	1.12	1.25	29.66
Chaetopterus variopedatus	0	1	0.91	1.24	30.9
Balanus balanus	0	0.87	0.91	1.23	32.12
Clausinella fasciata	0.33	1.21	1.47	1.2	33.32
Hydroidolina	0.67	0.5	1	1.13	34.45
Callophyllis cristata	0.8	0	1.19	1.1	35.55
Ascidiella aspersa	0.8	0	1.26	1.08	36.63
Pandalus	0.8	0	1.26	1.08	37.71
Testudinalia testudinalis	0	0.87	0.91	1.07	38.78
Balanus crenatus	1.8	1	2.88	1.05	39.83

	Group GV	Group GIV			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib% Cum.%	
Lithothamnion glaciale	0	0.71	0.91	1	40.83
Tectura virginea	0	0.71	0.91	1	41.84
Saccharina latissima	0.33	0.71	1.04	0.98	42.81
Pagurus	1.14	1.87	2.86	0.96	43.77
Corella parallelogramma	1	1.21	1.47	0.96	44.73
Hyas araneus	0.8	0.71	1.05	0.94	45.68
Porella concinna	0.33	1	1.28	0.9	46.58
Ascidia mentula	0.67	0	1.28	0.9	47.48
Colaconema daviesii	0.67	0	1.28	0.9	48.38
Necora puber	1.14	0.71	1.37	0.9	49.28
Eledone cirrhosa	0	0.71	0.91	0.87	50.15
Henricia	0.8	0.5	1.05	0.86	51.01
Botryllus schlosseri	0.67	0	1.28	0.85	51.86
Eupolymnia nebulosa	0.47	0.5	1.05	0.82	52.69
Rhizocaulus verticillatus	0.58	0	0.64	0.74	53.43
Halecium halecinum	0.58	0	0.64	0.74	54.16
Spirobranchus lamarcki	0.58	0	0.64	0.74	54.9
Chlamys	0	0.5	0.91	0.71	55.61
Hydractinia echinata	0	0.5	0.91	0.71	56.32
Hydrallmania falcata	0	0.5	0.91	0.71	57.03
Mimachlamys varia nivea	0	0.5	0.91	0.71	57.74
Monia patelliformis	0	0.5	0.91	0.71	58.44
Pandalus montagui	0	0.5	0.91	0.71	59.15
Pilayella littoralis	0	0.5	0.91	0.71	59.86

Groups Gll & Glll

Average dissimilarity = 74.23

	Group GII	Group GIII			
Species	Av.Abund	Av.Abund Diss/SD		Contrib% Cum.%	
Liocarcinus depurator	0	1.72	7.59	1.78	1.78
Ectocarpaceae	0	1.68	4.83	1.72	3.5
Asterias rubens	0	1.6	3.7	1.69	5.19
Balanus crenatus	0	1.52	1.95	1.64	6.83
Amphilectus fucorum	2.16	0.64	2.03	1.6	8.43
Verruca stroemia	0	1.41	1.83	1.51	9.93
Sagartia elegans	1.41	0	7.98	1.48	11.41
Kirchenpaueria pinnata	0	1.37	2	1.46	12.87
Dendronotus frondosus	0	1.36	1.96	1.43	14.3
Hiatella arctica	0	1.35	5.19	1.41	15.71
Corella parallelogramma	0	1.31	5	1.35	17.06
Distomus variolosus	0	1.33	1.36	1.34	18.4
Pagurus bernhardus	1.28	0	5.81	1.33	19.72
Pomatoschistus minutus	0	1.22	1.95	1.3	21.02
Coryphella	0	1.22	1.9	1.29	22.31
Adamsia carciniopados	1.24	0	1.34	1.27	23.58
Aglaothamnion priceanum	0	1.09	1.26	1.22	24.8
Munida rugosa	0	1.2	1.8	1.2	26
Pione vastifica	1.41	0.46	1.73	1.14	27.14
	Group GII	Group GIII			
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Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Ophiocomina nigra	1.28	0.64	2.17	1.13	28.27
Cryptopleura ramosa	0	1.15	1.97	1.13	29.4
Inachus phalangium	0.94	0.33	1.37	1.01	30.41
Gibbula magus	0	0.97	1.25	1.01	31.42
Urticina eques	0.94	0	1.34	0.99	32.41
Eurynome aspera	0.47	0.91	1.09	0.94	33.35
Solaster endeca	0	0.9	2.01	0.94	34.29
Serpula vermicularis	0	0.9	0.94	0.93	35.22
Ascidiella scabra	1.24	1.15	1.06	0.93	36.15
Caridea	0	0.87	1.3	0.93	37.08
Hyas araneus	0	0.86	1.24	0.92	38
Plumularia setacea	1.15	1.33	1.28	0.92	38.92
Disporella hispida	0	0.91	1.24	0.91	39.83
Hinia incrassata	1.05	0.57	1.23	0.9	40.74
Halecium muricatum	0	0.91	0.96	0.89	41.63
Bispira volutacornis	1.05	1.15	1.05	0.87	42.5
Scrupocellaria scruposa	0.8	0	1.31	0.85	43.36
Colaconema daviesii	0	0.83	0.94	0.84	44.2
Myxicola infundibulum	0	0.81	0.96	0.84	45.03
Mya truncata	0	0.83	1.2	0.83	45.86
Phrynorhombus regius	0	0.78	0.91	0.81	46.67
Pagurus	1.63	1.33	1.33	0.8	47.47
Chorda filum	1.91	1.19	1.22	0.77	48.24
Ascidia mentula	0.47	0.58	0.92	0.77	49
Inachus dorsettensis	0.58	0.4	0.94	0.75	49.76
Balanus balanus	0	0.71	0.68	0.75	50.51
Trisopterus minutus	0	0.79	0.91	0.75	51.26
Lithothamnion glaciale	0	0.76	0.97	0.74	52
Ophiothrix fragilis	0	0.78	0.69	0.74	52.74
Halecium halecinum	0	0.62	0.84	0.71	53.45
Scrupocellaria	0	0.69	0.96	0.71	54.16
Capulus ungaricus	0.33	0.69	1.07	0.71	54.87
Pholis gunnellus	0	0.67	1.34	0.71	55.57
Anemonia viridis	0	0.71	0.97	0.69	56.27
Protula tubularia	1.8	1.21	1.09	0.69	56.95
Spirobranchus lamarcki	1.72	1.75	0.91	0.68	57.64
Henricia	0.47	0.57	1.05	0.68	58.32
Antedon bifida	0	0.62	0.68	0.66	58.98
Pyura microcosmus	0	0.62	0.68	0.66	59.64
Pagurus prideaux	0	0.67	0.68	0.66	60.29

Groups GV & GIII

Average dissimilarity = 71.80

	Group GV Group GIII						
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%		
Asterias rubens	0	1.6	3.81	1.58	1.58		
Galathea intermedia	1.33	0	1.35	1.32	2.91		
Hiatella arctica	0	1.35	5.45	1.32	4.23		

	Group GV	Group GIII			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Spirobranchus lamarcki	0.58	1.75	1.46	1.31	5.53
Plumularia setacea	0	1.33	1.76	1.28	6.81
Bugula avicularia	0.33	1.63	2.47	1.26	8.08
Distomus variolosus	0	1.33	1.37	1.26	9.34
Pomatoschistus minutus	0	1.22	1.95	1.21	10.55
Coryphella	0	1.22	1.91	1.21	11.76
Protula tubularia	0	1.21	1.92	1.18	12.94
Chorda filum	0	1.19	1.79	1.18	14.12
Kirchenpaueria pinnata	0.33	1.37	1.68	1.14	15.26
Verruca stroemia	0.47	1.41	1.45	1.14	16.4
Munida rugosa	0	1.2	1.8	1.13	17.53
Necora puber	1.14	0	4.06	1.12	18.65
Dendronotus frondosus	0.33	1.36	1.6	1.11	19.77
Antedon bifida	1.38	0.62	1.87	1.06	20.83
Munida sarsi	1.05	0	1.31	1.05	21.87
Chlamys distorta	1.05	0	1.35	1.03	22.91
Clausinella fasciata	0.33	1.26	1.55	1.03	23.93
Aglaothamnion priceanum	0.33	1.09	1.28	1.01	24.94
Bispira volutacornis	0.33	1.15	1.42	0.99	25.93
Gibbula magus	0	0.97	1.25	0.94	26.87
Pagurus	1.14	1.33	3.88	0.93	27.8
Serpula vermicularis	0.33	0.9	1.16	0.88	28.68
Eurynome aspera	0.33	0.91	1.22	0.88	29.57
Amphilectus fucorum	1.52	0.64	1.23	0.87	30.44
Galathea strigosa	0.91	0.24	1.24	0.87	31.31
Halecium muricatum	0.47	0.91	1.09	0.87	32.17
Disporella hispida	0	0.91	1.24	0.86	33.03
Caridea	0.58	0.87	1.31	0.85	33.89
Pandalus	0.8	0	1.32	0.79	34.68
Ascidia mentula	0.67	0.58	1.52	0.79	35.47
Colaconema daviesii	0.67	0.83	1.4	0.78	36.25
Myxicola infundibulum	0	0.81	0.96	0.78	37.04
Halecium halecinum	0.58	0.62	1.04	0.78	37.82
Mya truncata	0	0.83	1.2	0.78	38.6
Phrynorhombus regius	0	0.78	0.91	0.76	39.35
Callophyllis cristata	0.8	0.17	1.2	0.75	40.11
Ophiopholis aculeata	1.66	1.72	1.3	0.75	40.85
Ophiocomina nigra	0.47	0.64	0.91	0.75	41.6
Trisopterus minutus	0.33	0.79	1.1	0.74	42.33
Ascidiella scabra	1.82	1.15	0.92	0.74	43.07
Corella parallelogramma	1	1.31	1.45	0.73	43.8
Scrupocellaria	0.47	0.69	1.07	0.71	44.52
Ascidiella aspersa	0.8	0.62	1.2	0.7	45.22
Balanus balanus	0	0.71	0.68	0.7	45.92
Ophiothrix fragilis	0	0.78	0.69	0.7	46.62
Lithothamnion glaciale	0	0.76	0.97	0.7	47.32
Hydroidolina	0.67	0	0.68	0.69	48.01
Hyas araneus	0.8	0.86	1.13	0.67	48.68

	Group GV	Group GIII			
Species	Av.Abund	Av.Abund Diss/SD		Contrib% Cum.%	
Solaster endeca	0.33	0.9	1.26	0.67	49.34
Capulus ungaricus	0.33	0.69	1.07	0.66	50.01
Spirobranchus	0.67	0	1.35	0.66	50.67
Bougainvillia/Eudendrium sp.	0.47	0.64	1.01	0.66	51.33
Anemonia viridis	0	0.71	0.97	0.65	51.98
Henricia	0.8	0.57	1.13	0.65	52.63
Botryllus schlosseri	0.67	0	1.35	0.63	53.26
Pagurus prideaux	0	0.67	0.68	0.62	53.88
Pyura microcosmus	0	0.62	0.68	0.62	54.5
Eubranchus pallidus	0	0.62	0.9	0.61	55.1
Gobiusculus flavescens	0	0.62	0.69	0.59	55.7
Macropodia rostrata	0	0.57	0.69	0.59	56.28
Cryptopleura ramosa	1.49	1.15	0.9	0.58	56.87
Protanthea simplex	0.33	0.37	0.8	0.58	57.45
Parasmittina trispinosa	0.47	0.33	0.93	0.55	58
Rhizocaulus verticillatus	0.58	0	0.68	0.55	58.55
Liocarcinus depurator	1.14	1.72	1.92	0.55	59.1
Pholis gunnellus	0.33	0.67	1.07	0.54	59.64
Hinia incrassata	0	0.57	0.94	0.51	60.15

Groups GIV & GIII

Average dissimilarity = 75.45

	Group GIV	Group GIII				
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%	
Laminaria hyperborea	1.98	0	7.48	1.83	1.83	
Liocarcinus depurator	0	1.72	7.33	1.58	3.4	
Spirobranchus lamarcki	0	1.75	2.1	1.55	4.95	
Ectocarpaceae	0	1.68	4.61	1.53	6.48	
Asterias rubens	0	1.6	3.73	1.5	7.97	
Verruca stroemia	0	1.41	1.81	1.33	9.3	
Kirchenpaueria pinnata	0	1.37	1.97	1.29	10.6	
Dendronotus frondosus	0	1.36	1.92	1.26	11.86	
Hiatella arctica	0	1.35	5.28	1.25	13.11	
Plumularia setacea	0	1.33	1.73	1.21	14.32	
Distomus variolosus	0	1.33	1.34	1.19	15.52	
Pomatoschistus minutus	0	1.22	1.92	1.15	16.66	
Coryphella	0	1.22	1.88	1.14	17.81	
Gibbula umbilicalis	1.21	0	3.9	1.13	18.93	
Chorda filum	0	1.19	1.76	1.11	20.05	
Ophiopholis aculeata	1.22	1.72	1.27	1.11	21.16	
Aglaothamnion priceanum	0	1.09	1.24	1.07	22.23	
Munida rugosa	0	1.2	1.77	1.07	23.3	
Bispira volutacornis	0	1.15	1.32	1.02	24.32	
Limaria hians	1	0.17	1.01	0.97	25.29	
Antedon bifida	1.21	0.62	2.91	0.94	26.23	
Balanus balanus	0.87	0.71	1.07	0.94	27.17	
Phrynorhombus regius	1.5	0.78	1.58	0.93	28.1	
Spirobranchus	1	0	8.28	0.93	29.03	

	Group GIV Group GIII					
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%	
Leptochiton asellus	1.21	0.57	2.11	0.91	29.94	
Ascidiella scabra	0.5	1.15	1.45	0.9	30.84	
Chaetopterus variopedatus	1	0.17	1.05	0.89	31.73	
Porifera	1	0	0.95	0.88	32.6	
Protula tubularia	2.12	1.21	1.4	0.85	33.45	
Eurynome aspera	0.5	0.91	1.4	0.84	34.29	
Solaster endeca	0	0.9	1.99	0.83	35.12	
Serpula vermicularis	0	0.9	0.92	0.83	35.95	
Disporella hispida	0	0.91	1.22	0.81	36.76	
Balanus crenatus	1	1.52	3.19	0.8	37.56	
Halecium muricatum	0	0.91	0.95	0.8	38.36	
Porella concinna	1	0.17	2.07	0.79	39.15	
Testudinalia testudinalis	0.87	0	0.95	0.76	39.91	
Gibbula magus	0.5	0.97	1.21	0.75	40.66	
Colaconema daviesii	0	0.83	0.93	0.75	41.41	
Myxicola infundibulum	0	0.81	0.95	0.74	42.15	
Mya truncata	0	0.83	1.18	0.74	42.89	
Tectura virginea	0.71	0.46	1.07	0.72	43.61	
Hyas araneus	0.71	0.86	1.14	0.71	44.32	
Lithothamnion glaciale	0.71	0.76	1	0.7	45.02	
Necora puber	0.71	0	0.95	0.69	45.71	
Saccharina latissima	0.71	0	0.95	0.69	46.4	
Trisopterus minutus	0	0.79	0.9	0.67	47.07	
Ophiothrix fragilis	0	0.78	0.68	0.66	47.73	
Caridea	1.37	0.87	1.22	0.65	48.38	
Eledone cirrhosa	0.71	0.5	1.25	0.65	49.02	
Pagurus	1.87	1.33	0.85	0.63	49.66	
Capulus ungaricus	0	0.69	0.94	0.63	50.29	
Scrupocellaria	0	0.69	0.94	0.63	50.92	
Halecium halecinum	0	0.62	0.84	0.63	51.55	
Protanthea simplex	0.5	0.37	0.98	0.63	52.17	
Anemonia viridis	0	0.71	0.95	0.62	52.79	
Amphilectus fucorum	1	0.64	1.52	0.61	53.39	
Pagurus prideaux	0	0.67	0.67	0.59	53.98	
Pyura microcosmus	0	0.62	0.67	0.58	54.56	
Eubranchus pallidus	0	0.62	0.88	0.57	55.14	
Ascidiella aspersa	0	0.62	0.87	0.57	55.7	
Bougainvillia/Eudendrium sp.	0	0.64	0.94	0.56	56.27	
Gobiusculus flavescens	0	0.62	0.68	0.56	56.83	
Macropodia rostrata	0	0.57	0.67	0.55	57.38	
Obelia geniculata	0.5	0.29	1.04	0.55	57.93	
Ascidia mentula	0	0.58	0.68	0.54	58.47	
Ophiocomina nigra	0	0.64	0.65	0.54	59.01	
Callionymus reticulatus	0.5	0.24	1.04	0.54	59.55	
Henricia	0.5	0.57	1.02	0.53	60.08	

Groups GII & GVI

Average dissimilarity = 80.39

	Group GII	Group GVI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Limaria hians	0	2.24	5.43	2.52	2.52
Chorda filum	1.91	0	5.6	2.15	4.68
Ectocarpaceae	0	1.87	9.01	2.08	6.76
Protula tubularia	1.8	0	4.61	2.02	8.78
Balanus balanus	0	1.73	5.43	1.96	10.74
Munida rugosa	0	1.71	20.31	1.87	12.61
Amphilectus fucorum	2.16	0.5	5.97	1.77	14.39
Ophiothrix fragilis	0	1.37	2.2	1.61	15.99
Sagartia elegans	1.41	0	5.43	1.6	17.59
Pione vastifica	1.41	0	2.19	1.57	19.17
Pagurus bernhardus	1.28	0	4.61	1.43	20.6
Ascidiella scabra	1.24	0	1.24	1.43	22.03
Pholis gunnellus	0	1.21	2.78	1.4	23.43
Adamsia carciniopados	1.24	0	1.24	1.36	24.79
Plumularia setacea	1.15	0	1.24	1.33	26.12
Heterosiphonia japonica	0	1.21	20.31	1.33	27.44
Phrynorhombus regius	0	1.21	20.31	1.33	28.77
Polymastia penicillus	0	1.21	20.31	1.33	30.09
Rhizocaulus verticillatus	0	1	0.91	1.31	31.4
Spirobranchus lamarcki	1.72	0.87	1.06	1.21	32.62
Bispira volutacornis	1.05	0	1.24	1.19	33.81
Hinia incrassata	1.05	0	1.22	1.15	34.95
Aglaothamnion priceanum	0	1	5.43	1.13	36.08
Botryllus schlosseri	0	1	5.43	1.13	37.21
Brosme brosme	0	1	5.43	1.13	38.34
Corella parallelogramma	0	1	5.43	1.13	39.47
Cryptopleura ramosa	0	1	5.43	1.13	40.6
Diaphorodoris luteocincta	0	1	5.43	1.13	41.73
Eledone cirrhosa	0	1	5.43	1.13	42.86
Gibbula umbilicalis	0	1	5.43	1.13	43.99
Lissoclinum	0	1	5.43	1.13	45.12
Pandalus montagui	0	1	5.43	1.13	46.24
Pleuronectes platessa	0	1	5.43	1.13	47.37
Clausinella fasciata	1.82	0.71	1.68	1.13	48.5
Capulus ungaricus	0.33	1.37	1.69	1.11	49.62
Pagurus	1.63	0.87	1	1.11	50.73
Inachus phalangium	0.94	0	1.24	1.08	51.8
Urticina eques	0.94	0	1.24	1.08	52.88
Pollachius pollachius	0.47	1.21	1.67	0.99	53.87
Antedon bifida	0	1	0.91	0.95	54.81
Hyas	0	0.71	0.91	0.93	55.74
Scrupocellaria scruposa	0.8	0	1.21	0.92	56.66
Henricia	0.47	1	2.26	0.92	57.58
Balanus crenatus	0	0.87	0.91	0.82	58.4
Ciona intestinalis	0	0.87	0.91	0.82	59.22
Inachus	0	0.87	0.91	0.82	60.04

Groups GV & GVI

Average dissimilarity = 71.12

	Group GV	Group GVI				
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%	
Limaria hians	0	2.24	6	2.54	2.54	
Ascidiella scabra	1.82	0	5.8	2.07	4.61	
Balanus balanus	0	1.73	6	1.97	6.58	
Munida rugosa	0	1.71	17.38	1.89	8.47	
Ophiothrix fragilis	0	1.37	2.28	1.61	10.08	
Galathea intermedia	1.33	0	1.25	1.54	11.62	
Ophiocomina nigra	0.47	1.5	1.4	1.4	13.02	
Bugula avicularia	0.33	1.57	2.23	1.4	14.42	
Phrynorhombus regius	0	1.21	17.38	1.34	15.76	
Polymastia penicillus	0	1.21	17.38	1.34	17.1	
Balanus crenatus	1.8	0.87	1.14	1.3	18.4	
Munida sarsi	1.05	0	1.21	1.22	19.62	
Rhizocaulus verticillatus	0.58	1	0.96	1.21	20.83	
Chlamys distorta	1.05	0	1.25	1.2	22.03	
Antedon bifida	1.38	1	1.52	1.2	23.24	
Brosme brosme	0	1	6	1.14	24.37	
Diaphorodoris luteocincta	0	1	6	1.14	25.51	
Eledone cirrhosa	0	1	6	1.14	26.64	
Gibbula umbilicalis	0	1	6	1.14	27.78	
Lissoclinum	0	1	6	1.14	28.92	
Pandalus montagui	0	1	6	1.14	30.05	
Capulus ungaricus	0.33	1.37	1.69	1.12	31.18	
Amphilectus fucorum	1.52	0.5	2.28	1.07	32.25	
Pagurus	1.14	0.87	1.91	1.03	33.28	
Pholis gunnellus	0.33	1.21	1.45	1	34.28	
Heterosiphonia japonica	0.33	1.21	1.58	0.97	35.25	
Pollachius pollachius	0.33	1.21	1.58	0.97	36.22	
Spirobranchus lamarcki	0.58	0.87	0.9	0.93	37.15	
Ascidiella aspersa	0.8	0	1.22	0.92	38.08	
Pandalus	0.8	0	1.22	0.92	39	
Hyas	0	0.71	0.91	0.92	39.92	
Saccharina latissima	0.33	0.87	1.14	0.89	40.81	
Luidia ciliaris	0.33	0.87	1.13	0.89	41.71	
Hyas araneus	0.8	0	1.2	0.89	42.59	
Clausinella fasciata	0.33	0.71	1.01	0.87	43.46	
Ciona intestinalis	0	0.87	0.91	0.84	44.3	
Inachus	0	0.87	0.91	0.84	45.14	
Kallymenia reniformis	0	0.87	0.91	0.84	45.99	
Marthasterias glacialis	0	0.87	0.91	0.84	46.83	
Galathea strigosa	0.91	0.5	1.18	0.83	47.66	
Callophyllis cristata	0.8	0.71	1.01	0.83	48.49	
Ophiopholis aculeata	1.66	2.34	1.11	0.82	49.31	
Liocarcinus depurator	1.14	0.5	1.02	0.82	50.13	
Necora puber	1.14	0.5	1.02	0.82	50.94	
Caridea	0.58	0.5	1	0.81	51.75	

	Group GV	Group GVI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Hydroidolina	0.67	0	0.63	0.81	52.56
Spirobranchus	0.67	0	1.25	0.77	53.33
Aglaothamnion priceanum	0.33	1	1.25	0.77	54.1
Ascidia mentula	0.67	0	1.25	0.77	54.87
Nemertesia ramosa	0.47	0.71	0.9	0.76	55.63
Bougainvillia/Eudendrium sp.	0.47	0.5	1.13	0.75	56.38
Scrupocellaria	0.47	0.5	1.13	0.75	57.13
Sabella pavonina	0.33	0.71	1.08	0.74	57.87
Corella parallelogramma	1	1	1.25	0.73	58.6
Pleuronectes platessa	0.33	1	1.25	0.73	59.33
Monia patelliformis	0	0.71	0.91	0.69	60.02

Groups GIV & GVI

Groups Grv & Gvr									
Average dissimilarity = 71.22									
Group GIV Group GVI									
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%				
Protula tubularia	2.12	0	4.66	2.4	2.4				
Laminaria hyperborea	1.98	0	5.47	2.22	4.62				
Ectocarpaceae	0	1.87	9.2	2.08	6.7				
Munida rugosa	0	1.71	13.42	1.88	8.58				
Ophiocomina nigra	0	1.5	1.88	1.77	10.35				
Ophiothrix fragilis	0	1.37	2.15	1.6	11.95				
Capulus ungaricus	0	1.37	6.14	1.48	13.43				
Leptochiton asellus	1.21	0	4.75	1.35	14.78				
Heterosiphonia japonica	0	1.21	13.42	1.33	16.11				
Pollachius pollachius	0	1.21	13.42	1.33	17.43				
Polymastia penicillus	0	1.21	13.42	1.33	18.76				
Limaria hians	1	2.24	1.07	1.33	20.09				
Ophiopholis aculeata	1.22	2.34	0.91	1.3	21.39				
Rhizocaulus verticillatus	0	1	0.86	1.29	22.68				
Pagurus	1.87	0.87	0.96	1.26	23.94				
Antedon bifida	1.21	1	2.31	1.16	25.1				
Aglaothamnion priceanum	0	1	5.54	1.13	26.23				
Botryllus schlosseri	0	1	5.54	1.13	27.35				
Brosme brosme	0	1	5.54	1.13	28.48				
Diaphorodoris luteocincta	0	1	5.54	1.13	29.61				
Lissoclinum	0	1	5.54	1.13	30.74				
Pleuronectes platessa	0	1	5.54	1.13	31.86				
Porella concinna	1	0	5.54	1.13	32.99				
Spirobranchus	1	0	5.54	1.13	34.12				
Caridea	1.37	0.5	1.08	1.08	35.2				
Chaetopterus variopedatus	1	0	0.85	1.06	36.26				
Porifera	1	0	0.85	1.06	37.32				
Balanus crenatus	1	0.87	2.89	1	38.32				
Saccharina latissima	0.71	0.87	1.02	0.96	39.29				
Testudinalia testudinalis	0.87	0.5	1.36	0.96	40.25				
Balanus balanus	0.87	1.73	0.85	0.92	41.17				
Hyas	0	0.71	0.86	0.91	42.08				

	Group GIV Group GVI						
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%		
Lithothamnion glaciale	0.71	0	0.85	0.84	42.92		
Tectura virginea	0.71	0	0.85	0.84	43.77		
Ciona intestinalis	0	0.87	0.86	0.84	44.6		
Inachus	0	0.87	0.86	0.84	45.44		
Kallymenia reniformis	0	0.87	0.86	0.84	46.28		
Luidia ciliaris	0	0.87	0.86	0.84	47.11		
Marthasterias glacialis	0	0.87	0.86	0.84	47.95		
Spirobranchus lamarcki	0	0.87	0.86	0.84	48.78		
Necora puber	0.71	0.5	0.98	0.82	49.61		
Eledone cirrhosa	0.71	1	1.77	0.82	50.42		
Pholis gunnellus	0.5	1.21	1.12	0.8	51.22		
Monia patelliformis	0.5	0.71	1.15	0.77	51.99		
Hyas araneus	0.71	0	0.85	0.75	52.74		
Clausinella fasciata	1.21	0.71	1.17	0.71	53.46		
Callophyllis cristata	0	0.71	0.86	0.68	54.14		
Nemertesia ramosa	0	0.71	0.86	0.68	54.82		
Pecten maximus	0	0.71	0.86	0.68	55.5		
Sabella pavonina	0	0.71	0.86	0.68	56.19		
Ulva compressa	0	0.71	0.86	0.68	56.87		
Anomiidae	0	0.5	0.86	0.64	57.51		
Bonnemaisonia hamifera	0	0.5	0.86	0.64	58.16		
Bougainvillia/Eudendrium sp.	0	0.5	0.86	0.64	58.8		
Cyclostomatida	0	0.5	0.86	0.64	59.45		
Diplecogaster bimaculata	0	0.5	0.86	0.64	60.09		

Groups GIII & GVI

Average dissimilarity = 72.57

Group GIII Group GVI									
Species	Av.Abu	nd Av.Abund	Diss/SD Contrib%		ն Cum.%				
Limaria hians	0.17	2.24	3.77	1.77	1.77				
Asterias rubens	1.6	0	3.54	1.37	3.14				
Verruca stroemia	1.41	0	1.78	1.22	4.36				
Kirchenpaueria pinnata	1.37	0	1.92	1.18	5.54				
Dendronotus frondosus	1.36	0	1.87	1.16	6.7				
Hiatella arctica	1.35	0	4.77	1.15	7.84				
Plumularia setacea	1.33	0	1.69	1.11	8.95				
Ophiocomina nigra	0.64	1.5	1.61	1.1	10.05				
Distomus variolosus	1.33	0	1.33	1.1	11.15				
Balanus balanus	0.71	1.73	1.83	1.08	12.23				
Ophiothrix fragilis	0.78	1.37	2.17	1.07	13.31				
Liocarcinus depurator	1.72	0.5	1.81	1.07	14.38				
Pomatoschistus minutus	1.22	0	1.88	1.05	15.43				
Protula tubularia	1.21	0	1.84	1.03	16.45				
Spirobranchus lamarcki	1.75	0.87	1.2	1.02	17.48				
Chorda filum	1.19	0	1.72	1.02	18.5				
Heterosiphonia japonica	0	1.21	8.85	1	19.5				
Polymastia penicillus	0	1.21	8.85	1	20.51				
Ascidiella scabra	1.15	0	1.33	0.95	21.46				

Group GIII Group GVI								
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%			
Rhizocaulus verticillatus	0	1	0.95	0.94	22.4			
Bispira volutacornis	1.15	0	1.3	0.94	23.34			
Pagurus	1.33	0.87	1.07	0.92	24.26			
Balanus crenatus	1.52	0.87	1.07	0.91	25.17			
Botryllus schlosseri	0	1	6.57	0.85	26.02			
Brosme brosme	0	1	6.57	0.85	26.87			
Diaphorodoris luteocincta	0	1	6.57	0.85	27.72			
Gibbula umbilicalis	0	1	6.57	0.85	28.57			
Lissoclinum	0	1	6.57	0.85	29.42			
Pandalus montagui	0	1	6.57	0.85	30.27			
Gibbula magus	0.97	0	1.22	0.82	31.08			
Antedon bifida	0.62	1	0.98	0.81	31.9			
Coryphella	1.22	0.5	1.3	0.79	32.69			
Eurynome aspera	0.91	0	0.95	0.76	33.45			
Serpula vermicularis	0.9	0	0.92	0.76	34.21			
Disporella hispida	0.91	0	1.2	0.75	34.96			
Hyas araneus	0.86	0	1.22	0.74	35.7			
Halecium muricatum	0.91	0	0.94	0.74	36.44			
Pollachius pollachius	0.33	1.21	1.63	0.73	37.17			
Pleuronectes platessa	0.17	1	2.02	0.72	37.89			
Capulus ungaricus	0.69	1.37	1.47	0.71	38.6			
Phrynorhombus regius	0.78	1.21	1.67	0.69	39.3			
Colaconema daviesii	0.83	0.5	1.1	0.69	39.98			
Myxicola infundibulum	0.81	0	0.94	0.68	40.66			
Mya truncata	0.83	0	1.16	0.68	41.34			
Clausinella fasciata	1.26	0.71	1.23	0.68	42.02			
Luidia ciliaris	0.24	0.87	1	0.67	42.69			
Marthasterias glacialis	0.24	0.87	1	0.67	43.36			
Ciona intestinalis	0.17	0.87	1.04	0.67	44.03			
Inachus	0	0.87	0.95	0.65	44.68			
Kallymenia reniformis	0	0.87	0.95	0.65	45.33			
Saccharina latissima	0	0.87	0.95	0.65	45.99			
Hyas	0.24	0.71	0.94	0.65	46.64			
Aglaothamnion priceanum	1.09	1	2.46	0.65	47.28			
Gobiusculus flavescens	0.62	0.5	1.06	0.65	47.93			
Trisopterus minutus	0.79	0	0.89	0.62	48.55			
Caridea	0.87	0.5	1.14	0.62	49.17			
Lithothamnion glaciale	0.76	0	0.94	0.61	49.78			
Pecten maximus	0.46	0.71	1.06	0.61	50.39			
Munida rugosa	1.2	1.71	1.08	0.58	50.97			
Scrupocellaria	0.69	0.5	1.21	0.57	51.54			
Ophiopholis aculeata	1.72	2.34	0.79	0.57	52.11			
Anemonia viridis	0.71	0	0.94	0.57	52.68			
Halecium halecinum	0.62	0	0.84	0.57	53.25			
Protanthea simplex	0.37	0.5	0.93	0.56	53.81			
Callophyllis cristata	0.17	0.71	1.02	0.55	54.35			
Nemertesia ramosa	0.17	0.71	1.02	0.55	54.9			
Ulva compressa	0.17	0.71	1.02	0.55	55.44			
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Group GIII Group GVI								
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%			
Amphilectus fucorum	0.64	0.5	1.17	0.55	55.99			
Pagurus prideaux	0.67	0	0.67	0.54	56.53			
Bougainvillia/Eudendrium sp.	0.64	0.5	1.17	0.54	57.07			
Pyura microcosmus	0.62	0	0.67	0.54	57.6			
Monia patelliformis	0	0.71	0.95	0.53	58.14			
Sabella pavonina	0	0.71	0.95	0.53	58.67			
Eubranchus pallidus	0.62	0	0.88	0.53	59.19			
Ascidiella aspersa	0.62	0	0.86	0.52	59.71			
Galathea strigosa	0.24	0.5	1.06	0.52	60.23			

Groups GII & GI

Average dissimilarity = 88.79

	Group GII	Group GI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Ophiopholis aculeata	2.24	0	17.23	2.5	2.5
Amphilectus fucorum	2.16	0	70.09	2.4	4.9
Golfingia (Golfingia) vulgaris	0	2	17.23	2.23	7.14
Liocarcinus depurator	0	2	17.23	2.23	9.37
Macropodia rostrata	0	2	17.23	2.23	11.61
Pomatoschistus minutus	0	2	17.23	2.23	13.84
Chorda filum	1.91	0	34.23	2.13	15.97
Clausinella fasciata	1.82	0	11.96	2.03	18
Protula tubularia	1.8	0	7.24	2	20.01
Balanus crenatus	0	1.73	17.23	1.94	21.94
Caryophyllia (Caryophyllia) smithii	0	1.73	17.23	1.94	23.88
Cerianthus Iloydii	0	1.73	17.23	1.94	25.81
Corella parallelogramma	0	1.73	17.23	1.94	27.75
Galathea intermedia	0	1.73	17.23	1.94	29.68
Phycodrys rubens	0	1.73	17.23	1.94	31.62
Rhodomenia	0	1.73	17.23	1.94	33.55
Rhodophyceae crusts	0	1.73	17.23	1.94	35.49
Terebellidae	0	1.73	17.23	1.94	37.42
Bugula avicularia	1.72	0	7.45	1.91	39.33
Aequipecten opercularis	0	1.41	17.23	1.58	40.91
Botryllus schlosseri	0	1.41	17.23	1.58	42.49
Brosme brosme	0	1.41	17.23	1.58	44.07
Callophyllis laciniata	0	1.41	17.23	1.58	45.65
Crossaster papposus	0	1.41	17.23	1.58	47.23
Didemnum maculosum	0	1.41	17.23	1.58	48.81
Eledone cirrhosa	0	1.41	17.23	1.58	50.39
Eupolymnia nebulosa	0	1.41	17.23	1.58	51.97
Halecium halecinum	0	1.41	17.23	1.58	53.55
Halecium muricatum	0	1.41	17.23	1.58	55.13
Heterosiphonia plumosa	0	1.41	17.23	1.58	56.71
Mycale	0	1.41	17.23	1.58	58.29
Pecten maximus	0	1.41	17.23	1.58	59.87

Groups GV & GI

Average dissimilarity = 73.66

	Group GV	Group GI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Golfingia (Golfingia) vulgaris	0	2	16.9	2.38	2.38
Macropodia rostrata	0	2	16.9	2.38	4.76
Pomatoschistus minutus	0	2	16.9	2.38	7.14
Ectocarpaceae	1.91	0	40.09	2.27	9.41
Ascidiella scabra	1.82	0	13.16	2.17	11.58
Caryophyllia (Caryophyllia) smithii	0	1.73	16.9	2.06	13.64
Cerianthus lloydii	0	1.73	16.9	2.06	15.7
Phycodrys rubens	0	1.73	16.9	2.06	17.76
Rhodomenia	0	1.73	16.9	2.06	19.83
Terebellidae	0	1.73	16.9	2.06	21.89
Ophiopholis aculeata	1.66	0	3	1.94	23.83
Amphilectus fucorum	1.52	0	9.56	1.81	25.64
Cryptopleura ramosa	1.49	0	3.18	1.78	27.42
Aequipecten opercularis	0	1.41	16.9	1.68	29.1
Brosme brosme	0	1.41	16.9	1.68	30.79
Callophyllis laciniata	0	1.41	16.9	1.68	32.47
Eledone cirrhosa	0	1.41	16.9	1.68	34.16
Heterosiphonia plumosa	0	1.41	16.9	1.68	35.84
Mycale	0	1.41	16.9	1.68	37.52
Pecten maximus	0	1.41	16.9	1.68	39.21
Rhodophyllis divaricata	0	1.41	16.9	1.68	40.89
Rhodophyceae crusts	0.33	1.73	2.28	1.68	42.57
Necora puber	1.14	0	3.65	1.37	43.94
Crossaster papposus	0.33	1.41	1.81	1.3	45.24
Munida sarsi	1.05	0	1.12	1.28	46.51
Halecium halecinum	0.58	1.41	1.61	1.26	47.78
Spirobranchus lamarcki	0.58	1.41	1.61	1.26	49.04
Chlamys distorta	1.05	0	1.15	1.26	50.31
Pleuronectes platessa	0.33	1.41	1.98	1.26	51.56
Chirolophis ascanii	0	1	16.9	1.19	52.76
Cyclostomatida	0	1	16.9	1.19	53.95
Desmarestia ligulata	0	1	16.9	1.19	55.14
Holothuroidea	0	1	16.9	1.19	56.33
Leucosolenida	0	1	16.9	1.19	57.52
Pagurus bernhardus	0	1	16.9	1.19	58.71
Phrynorhombus regius	0	1	16.9	1.19	59.9

Groups GIV & GI

Average dissimilarity = 82.59

Group GIV Group GI							
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	۵ Cum.%		
Protula tubularia	2.12	0	6	2.24	2.24		
Liocarcinus depurator	0	2	11.29	2.11	4.35		
Macropodia rostrata	0	2	11.29	2.11	6.46		
Pomatoschistus minutus	0	2	11.29	2.11	8.57		
Laminaria hyperborea	1.98	0	10.89	2.08	10.65		

	Group GIV	Group GI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Caryophyllia (Caryophyllia) smithii	0	1.73	11.29	1.83	12.47
Cerianthus Iloydii	0	1.73	11.29	1.83	14.3
Galathea intermedia	0	1.73	11.29	1.83	16.13
Phycodrys rubens	0	1.73	11.29	1.83	17.95
Rhodomenia	0	1.73	11.29	1.83	19.78
Terebellidae	0	1.73	11.29	1.83	21.61
Bugula avicularia	1.57	0	18.29	1.65	23.26
Golfingia (Golfingia) vulgaris	0.5	2	1.82	1.62	24.87
Aequipecten opercularis	0	1.41	11.29	1.49	26.36
Botryllus schlosseri	0	1.41	11.29	1.49	27.86
Brosme brosme	0	1.41	11.29	1.49	29.35
Callophyllis laciniata	0	1.41	11.29	1.49	30.84
Crossaster papposus	0	1.41	11.29	1.49	32.33
Didemnum maculosum	0	1.41	11.29	1.49	33.82
Halecium halecinum	0	1.41	11.29	1.49	35.31
Halecium muricatum	0	1.41	11.29	1.49	36.81
Heterosiphonia plumosa	0	1.41	11.29	1.49	38.3
Mycale	0	1.41	11.29	1.49	39.79
Pecten maximus	0	1.41	11.29	1.49	41.28
Pleuronectes platessa	0	1.41	11.29	1.49	42.77
Rhodophyllis divaricata	0	1.41	11.29	1.49	44.26
Scrupocellaria	0	1.41	11.29	1.49	45.76
Spirobranchus lamarcki	0	1.41	11.29	1.49	47.25
Caridea	1.37	0	2.18	1.47	48.71
Ophiopholis aculeata	1.22	0	0.71	1.37	50.09
Rhodophyceae crusts	0.5	1.73	1.55	1.33	51.42
Clausinella fasciata	1.21	0	3.05	1.29	52.71
Gibbula umbilicalis	1.21	0	3.05	1.29	53.99
Leptochiton asellus	1.21	0	6.42	1.26	55.25
Limaria hians	1	0	0.71	1.12	56.37
Amphilectus fucorum	1	0	11.29	1.05	57.43
Chirolophis ascanii	0	1	11.29	1.05	58.48
Colaconema daviesii	0	1	11.29	1.05	59.54

Groups GIII & GI

Average dissimilarity = 76.75

Group GIII Group GI									
Species	Av.Abı	und Av.Abund	Diss/SD	Contrib	% Cum.%				
Golfingia (Golfingia) vulgaris	0	2	9.47	1.71	1.71				
Cerianthus Iloydii	0	1.73	9.47	1.48	3.2				
Galathea intermedia	0	1.73	9.47	1.48	4.68				
Phycodrys rubens	0	1.73	9.47	1.48	6.17				
Rhodomenia	0	1.73	9.47	1.48	7.65				
Rhodophyceae crusts	0	1.73	9.47	1.48	9.13				
Ophiopholis aculeata	1.72	0	1.98	1.47	10.6				
Ectocarpaceae	1.68	0	4.48	1.41	12.02				
Bugula avicularia	1.63	0	13.44	1.38	13.4				
Asterias rubens	1.6	0	3.7	1.38	14.78				

	Group GIII	Group GI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Caryophyllia (Caryophyllia) smithii	0.29	1.73	1.98	1.25	16.04
Terebellidae	0.29	1.73	1.98	1.25	17.29
Botryllus schlosseri	0	1.41	9.47	1.21	18.5
Brosme brosme	0	1.41	9.47	1.21	19.71
Callophyllis laciniata	0	1.41	9.47	1.21	20.92
Didemnum maculosum	0	1.41	9.47	1.21	22.13
Eupolymnia nebulosa	0	1.41	9.47	1.21	23.35
Heterosiphonia plumosa	0	1.41	9.47	1.21	24.56
Rhodophyllis divaricata	0	1.41	9.47	1.21	25.77
Macropodia rostrata	0.57	2	1.65	1.2	26.97
Kirchenpaueria pinnata	1.37	0	1.89	1.2	28.17
Dendronotus frondosus	1.36	0	1.84	1.17	29.34
Hiatella arctica	1.35	0	5.43	1.16	30.49
Plumularia setacea	1.33	0	1.65	1.12	31.61
Distomus variolosus	1.33	0	1.29	1.11	32.72
Clausinella fasciata	1.26	0	1.82	1.09	33.81
Pleuronectes platessa	0.17	1.41	2.77	1.08	34.89
Coryphella	1.22	0	1.81	1.06	35.95
Protula tubularia	1.21	0	1.81	1.04	36.99
Chorda filum	1.19	0	1.69	1.03	38.02
Aequipecten opercularis	0.24	1.41	1.98	1.02	39.04
Crossaster papposus	0.24	1.41	1.98	1.02	40.06
Munida rugosa	1.2	0	1.7	0.99	41.05
Adlaothamnion priceanum	1.09	0	1.2	0.99	42.04
Ascidiella scabra	1.15	0	1.29	0.96	43
Bispira volutacornis	1 15	0	1 26	0.95	43 95
Cryptopleura ramosa	1 15	0	1.84	0.93	44 89
Pecten maximus	0.46	1 41	1.84	0.93	45 82
Mycale	0.4	1 41	1.56	0.86	46 68
Chirolophis ascanii	0	1	9 47	0.86	47 53
Cvclostomatida	0	1	9.47	0.86	48.39
Desmarestia ligulata	0	1	9.47	0.86	49 25
Leucosolenida	0	1	9.47	0.86	50 11
Pagurus	1 33	1	9.47	0.86	50.96
Pagurus bernhardus	0	1	9.47	0.86	51 82
Gibbula magus	0.97	0	1 18	0.83	52.65
Antedon bifida	0.62	1	6 58	0.82	53.46
Halecium muricatum	0.02	1 41	1 51	0.8	54 27
Eledone cirrhosa	0.5	1.41	1.61	0.78	55.05
Solaster endeca	0.0	0	1.00	0.70	55.82
	0.0	0	0.01	0.77	56 58
Sernula vermicularis	0.9	0	0.88	0.77	57 35
Caridea	0.87	0	1.23	0.76	58 11
Disnorella hisnida	0.01	0	1 16	0.75	58.86
Halecium halecinum	0.62	1 41	1.68	0.73	50.00
Holothuroidee	0.17	1	1.00	0.72	60 33
าเอเอแานเอเนซิล	0.17	1	1.50	0.12	00.00

Groups GVI & GI

Average dissimilarity = 78.63

	Group GVI	Group GI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Ophiopholis aculeata	2.34	0	3.96	2.24	2.24
Limaria hians	2.24	0	5.28	2.13	4.37
Golfingia (Golfingia) vulgaris	0	2	5.28	1.9	6.28
Macropodia rostrata	0	2	5.28	1.9	8.18
Pomatoschistus minutus	0	2	5.28	1.9	10.08
Ectocarpaceae	1.87	0	11.27	1.76	11.84
Balanus balanus	1.73	0	5.28	1.65	13.49
Caryophyllia (Caryophyllia) smithii	0	1.73	5.28	1.65	15.14
Cerianthus Iloydii	0	1.73	5.28	1.65	16.79
Galathea intermedia	0	1.73	5.28	1.65	18.44
Phycodrys rubens	0	1.73	5.28	1.65	20.09
Rhodomenia	0	1.73	5.28	1.65	21.74
Rhodophyceae crusts	0	1.73	5.28	1.65	23.38
Terebellidae	0	1.73	5.28	1.65	25.03
Munida rugosa	1.71	0	18.36	1.59	26.62
Liocarcinus depurator	0.5	2	1.58	1.49	28.11
Ophiocomina nigra	1.5	0	1.58	1.49	29.6
Bugula avicularia	1.57	0	21.18	1.48	31.08
Ophiothrix fragilis	1.37	0	1.82	1.35	32.43
Aequipecten opercularis	0	1.41	5.28	1.35	33.77
Callophyllis laciniata	0	1.41	5.28	1.35	35.12
Crossaster papposus	0	1.41	5.28	1.35	36.47
Didemnum maculosum	0	1.41	5.28	1.35	37.81
Eupolymnia nebulosa	0	1.41	5.28	1.35	39.16
Halecium halecinum	0	1.41	5.28	1.35	40.5
Halecium muricatum	0	1.41	5.28	1.35	41.85
Mycale	0	1.41	5.28	1.35	43.2
Capulus ungaricus	1.37	0	5.09	1.25	44.45
Pholis gunnellus	1.21	0	2.37	1.18	45.63
Heterosiphonia japonica	1.21	0	18.36	1.12	46.75
Pollachius pollachius	1.21	0	18.36	1.12	47.87
Polymastia penicillus	1.21	0	18.36	1.12	48.99
Rhizocaulus verticillatus	1	0	0.71	1.08	50.07
Aglaothamnion priceanum	1	0	5.28	0.95	51.03
Antedon bifida	1	1	5.28	0.95	51.98
Chirolophis ascanii	0	1	5.28	0.95	52.93
Cryptopleura ramosa	1	0	5.28	0.95	53.88
Diaphorodoris luteocincta	1	0	5.28	0.95	54.83
Gibbula umbilicalis	1	0	5.28	0.95	55.79
Henricia	1	0	5.28	0.95	56.74
Holothuroidea	0	1	5.28	0.95	57.69
Hyas araneus	0	1	5.28	0.95	58.64
Leucosolenida	0	1	5.28	0.95	59.59

Groups GII & GVII

Average dissimilarity = 73.61

Group GII Group GVII

Species	Av.Abund	Av.Abund Diss/SD		Contrib% Cum.%	
Amphilectus fucorum	2.16	0	9.98	3.21	3.21
Chorda filum	1.91	0	9.85	2.84	6.05
Protula tubularia	1.8	0	6.6	2.67	8.72
Antedon bifida	0	1.72	5.63	2.55	11.27
Spirobranchus lamarcki	1.72	0	6.65	2.54	13.81
Ectocarpaceae	0	1.41	1.32	2.24	16.05
Ophiothrix fragilis	0	1.39	1.28	2.21	18.26
Corella parallelogramma	0	1.41	8.7	2.11	20.37
Sagartia elegans	1.41	0	8.7	2.11	22.47
Pione vastifica	1.41	0	2.44	2.07	24.55
Rhodophyceae crusts	0	1.38	5.48	2.03	26.58
Sycon ciliatum	0	1.38	5.48	2.03	28.61
Carcinus maenas	0	1.24	1.32	1.97	30.58
Gibbula umbilicalis	0	1.24	1.28	1.79	32.37
Adamsia carciniopados	1.24	0	1.31	1.78	34.16
Plumularia setacea	1.15	0	1.31	1.76	35.92
Phyllophora crispa	0	1.14	3.76	1.71	37.63
Cryptopleura ramosa	0	1.14	4.13	1.71	39.34
Aglaothamnion priceanum	0	1.14	10.78	1.67	41.01
Cucumaria frondosa	0	1	1.16	1.58	42.59
Bispira volutacornis	1.05	0	1.31	1.57	44.16
Pecten maximus	0	1.05	1.25	1.54	45.71
Hinia incrassata	1.05	0	1.29	1.51	47.21
Inachus phalangium	0.94	0	1.31	1.42	48.64
Urticina eques	0.94	0	1.31	1.42	50.06
Clausinella fasciata	1.82	1.22	1.15	1.37	51.43
Pagurus bernhardus	1.28	0.33	1.76	1.37	52.8
Ascidiella scabra	1.24	1.55	1.42	1.3	54.1
Hydractinia echinata	0	0.8	1.21	1.17	55.26
Strongylocentrotus droebachiensis	0	0.75	0.67	1.16	56.43
Balanus crenatus	0	0.8	1.32	1.15	57.58
Lanice conchilega	0	0.8	1.32	1.13	58.71
Neptunea antiqua	0	0.67	1.3	0.97	59.68

Groups GV & GVII

Average dissimilarity = 69.61

	Group GV	Group GVII				
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%	
Bugula avicularia	0.33	1.88	2.19	2.19	2.19	
Amphilectus fucorum	1.52	0	7.92	2.09	4.27	
Ophiothrix fragilis	0	1.39	1.28	2.02	6.29	
Ophiocomina nigra	0.47	1.88	1.66	2	8.3	
Sycon ciliatum	0	1.38	5.37	1.88	10.17	
Carcinus maenas	0	1.24	1.32	1.81	11.98	
Galathea intermedia	1.33	0.33	1.5	1.69	13.68	
Gibbula umbilicalis	0	1.24	1.28	1.66	15.33	
Necora puber	1.14	0	3.85	1.58	16.91	
Phyllophora crispa	0	1.14	3.9	1.58	18.49	
Clausinella fasciata	0.33	1.22	1.23	1.52	20.01	

Munida sarsi	1.05	0	1.28	1.48	21.49
Chlamys distorta	1.05	0	1.32	1.46	22.94
Cucumaria frondosa	0	1	1.16	1.45	24.39
Rhodophyceae crusts	0.33	1.38	1.79	1.44	25.83
Pecten maximus	0	1.05	1.26	1.43	27.25
Balanus crenatus	1.8	0.8	1.41	1.4	28.65
Galathea strigosa	0.91	0.33	1.08	1.15	29.8
Ascidiella aspersa	0.8	0	1.29	1.12	30.92
Pandalus	0.8	0	1.29	1.12	32.04
Liocarcinus depurator	1.14	0.33	1.43	1.1	33.14
Aglaothamnion priceanum	0.33	1.14	1.51	1.1	34.24
Corella parallelogramma	1	1.41	1.79	1.09	35.33
Hydractinia echinata	0	0.8	1.22	1.08	36.41
Strongylocentrotus droebachiensis	0	0.75	0.67	1.07	37.48
Hyas araneus	0.8	0	1.26	1.07	38.55
Callophyllis cristata	0.8	0.47	1.12	1.04	39.59
Hydroidolina	0.67	0	0.66	0.99	40.57
Ectocarpaceae	1.91	1.41	0.95	0.98	41.55
Henricia	0.8	0.33	1.14	0.97	42.53
Ophiopholis aculeata	1.66	2.21	1.18	0.97	43.49
Spirobranchus	0.67	0	1.31	0.94	44.43
Ascidia mentula	0.67	0	1.31	0.93	45.36
Colaconema daviesii	0.67	0	1.31	0.93	46.29
Lanice conchilega	0.33	0.8	1.18	0.92	47.22
Neptunea antiqua	0	0.67	1.31	0.9	48.12
Pomatoschistus pictus	0	0.67	1.31	0.9	49.01
Scrupocellaria scruposa	0	0.67	1.31	0.9	49.91
Suberites carnosus	0	0.67	1.31	0.9	50.81
Botryllus schlosseri	0.67	0	1.32	0.88	51.69
Kirchenpaueria pinnata	0.33	0.58	0.95	0.88	52.57
Pagurus	1.14	1.73	2.6	0.82	53.4
Capulus ungaricus	0.33	0.67	1.04	0.79	54.19
Pholis gunnellus	0.33	0.47	0.96	0.78	54.96
Caridea	0.58	0	0.66	0.77	55.73
Rhizocaulus verticillatus	0.58	0	0.66	0.77	56.5
Nemertesia ramosa	0.47	0.33	0.9	0.77	57.27
Ophiura albida	0.47	0.33	0.9	0.77	58.03
Halecium halecinum	0.58	0	0.66	0.76	58.79
Spirobranchus lamarcki	0.58	0	0.66	0.76	59.55

Groups GIV & GVII

Average dissimilarity = 72.24

	Group GIV	Group	GVII		
Species	Av.Abund	Av.Ab	und Diss/SD	Contri	b% Cum.%
Protula tubularia	2.12	0	6.09	2.8	2.8
Laminaria hyperborea	1.98	0	9.07	2.59	5.39
Ophiocomina nigra	0	1.88	3.55	2.51	7.9
Ectocarpaceae	0	1.41	1.28	1.96	9.86

Ophiothrix fragilis	0	1.39	1.24	1.93	11.8
Phrynorhombus regius	1.5	0	3.23	1.93	13.72
Caridea	1.37	0	2.66	1.83	15.55
Sycon ciliatum	0	1.38	5.04	1.8	17.35
Carcinus maenas	0	1.24	1.28	1.73	19.08
Leptochiton asellus	1.21	0	6.91	1.57	20.65
Ophiopholis aculeata	1.22	2.21	1.04	1.52	22.17
Phyllophora crispa	0	1.14	3.72	1.51	23.68
Aglaothamnion priceanum	0	1.14	8.55	1.48	25.15
Ascidiella scabra	0.5	1.55	1.25	1.46	26.62
Limaria hians	1	0	0.91	1.41	28.02
Cucumaria frondosa	0	1	1.12	1.38	29.41
Pecten maximus	0	1.05	1.21	1.36	30.77
Amphilectus fucorum	1	0	8.47	1.32	32.09
Porella concinna	1	0	8.47	1.32	33.4
Spirobranchus	1	0	8.47	1.32	34.72
Chaetopterus variopedatus	1	0	0.91	1.23	35.94
Porifera	1	0	0.91	1.23	37.17
Balanus balanus	0.87	0	0.91	1.22	38.39
Rhodophyceae crusts	0.5	1.38	1.34	1.18	39.57
Gibbula umbilicalis	1.21	1.24	1.88	1.13	40.7
Clausinella fasciata	1.21	1.22	1.44	1.11	41.81
Testudinalia testudinalis	0.87	0	0.91	1.06	42.88
Strongylocentrotus droebachiensis	0	0.75	0.64	1.02	43.9
Lanice conchilega	0	0.8	1.27	1.01	44.9
Lithothamnion glaciale	0.71	0	0.91	0.99	45.9
Necora puber	0.71	0	0.91	0.99	46.89
Saccharina latissima	0.71	0	0.91	0.99	47.89
Tectura virginea	0.71	0	0.91	0.99	48.88
Capulus ungaricus	0	0.67	1.28	0.93	49.81
Eledone cirrhosa	0.71	0	0.91	0.87	50.68
Hyas araneus	0.71	0	0.91	0.87	51.54
Pomatoschistus pictus	0	0.67	1.26	0.86	52.4
Scrupocellaria scruposa	0	0.67	1.26	0.86	53.26
Suberites carnosus	0	0.67	1.26	0.86	54.12
Solaster endeca	0	0.67	1.27	0.84	54.96
Pholis gunnellus	0.5	0.47	1.11	0.84	55.8
Hydractinia echinata	0.5	0.8	1.1	0.84	56.64
Obelia geniculata	0.5	0.47	1.07	0.8	57.44
Amphipholis squamata	0.5	0	0.91	0.7	58.14
Eurynome aspera	0.5	0	0.91	0.7	58.84
Hydrallmania falcata	0.5	0	0.91	0.7	59.55
Mimachlamys varia nivea	0.5	0	0.91	0.7	60.25
Groups GIII & GVII					

Average dissimilarity = 72.30

Group GIII Group GVII

Species	Av.Abı	und Av.Ab	und Diss/SD	Contril	b% Cum.%
Spirobranchus lamarcki	1.75	0	2.13	1.62	1.62
Asterias rubens	1.6	0	3.74	1.57	3.19
Ophiocomina nigra	0.64	1.88	1.71	1.44	4.63

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Verruca stroemia	1.41	0	1.83	1.4	6.02
Rhodophyceae crusts	0	1.38	4.75	1.33	7.35
Sycon ciliatum	0	1.38	4.75	1.33	8.68
Dendronotus frondosus	1.36	0	1.94	1.32	10
Liocarcinus depurator	1.72	0.33	2.62	1.32	11.32
Hiatella arctica	1.35	0	5.24	1.31	12.63
Plumularia setacea	1.33	0	1.75	1.27	13.9
Ophiothrix fragilis	0.78	1.39	1.18	1.27	15.16
Antedon bifida	0.62	1.72	1.59	1.18	16.34
Gibbula umbilicalis	0	1.24	1.32	1.18	17.52
Protula tubularia	1.21	0	1.91	1.17	18.69
Chorda filum	1.19	0	1.78	1.17	19.86
Distomus variolosus	1.33	0.33	1.45	1.16	21.02
Carcinus maenas	0.33	1.24	1.37	1.14	22.17
Munida rugosa	1.2	0	1.79	1.12	23.29
Phyllophora crispa	0	1.14	3.99	1.11	24.4
Kirchenpaueria pinnata	1.37	0.58	1.41	1.09	25.49
Bispira volutacornis	1.15	0	1.34	1.07	26.56
Cucumaria frondosa	0	1	1.18	1.01	27.57
Coryphella	1.22	0.33	1.49	1	28.57
Balanus crenatus	1.52	0.8	1.42	0.99	29.55
Gibbula magus	0.97	0	1.25	0.94	30.49
Pomatoschistus minutus	1.22	0.47	1.36	0.91	31.4
Pecten maximus	0.46	1.05	1.2	0.91	32.31
Clausinella fasciata	1.26	1.22	1.27	0.88	33.19
Serpula vermicularis	0.9	0	0.94	0.87	34.06
Eurvnome aspera	0.91	0	0.97	0.87	34.93
Caridea	0.87	0	1.3	0.86	35.78
Hvas araneus	0.86	0	1 24	0.85	36 64
Halecium muricatum	0.91	0	0.96	0.84	37 47
Ascidiella scabra	1 15	1 55	1 28	0.83	38.31
Ectocarpaceae	1.68	1 41	1.34	0.83	39.13
Colaconema daviesii	0.83	0	0.94	0.78	30 02
Myxicola infundibulum	0.00	0	0.04	0.78	40.69
	0.01	0	1 10	0.70	40.03
	0.05	0.8	1.19	0.77	41.47
Disperalla hispida	0 01	0.0	1.20	0.77	42.23
	0.91	0.55	0.01	0.77	40 75
Philynombus regius	0.70	0 75	0.91	0.75	43.75
	1 22	0.75	0.00	0.75	44.49
Pagurus	1.33	1.73	1.09	0.71	45.2
i risopterus minutus	0.79	0	0.91	0.7	45.9
Macropodia rostrata	0.57	0.47	0.89	0.7	46.61
Balanus balanus	0.71	0	0.68	0.7	47.3
Aglaothamnion priceanum	1.09	1.14	1.75	0.69	47.99
Litnothamnion glaciale	0.76	U	0.96	0.69	48.69
Ophiopholis aculeata	1.72	2.21	0.94	0.69	49.38
Pholis gunnellus	0.67	0.47	1.4	0.67	50.05
Lanice conchilega	0.33	0.8	1.19	0.66	50.71
Scrupocellaria	0.69	0	0.95	0.66	51.37
Halecium halecinum	0.62	0	0.85	0.66	52.03

Capulus ungaricus	0.69	0.67	1.34	0.65	52.68
lophon nigricans	0.46	0.47	0.93	0.65	53.34
Anemonia viridis	0.71	0	0.97	0.65	53.98
Pomatoschistus pictus	0	0.67	1.34	0.64	54.62
Scrupocellaria scruposa	0	0.67	1.34	0.64	55.25
Pagurus prideaux	0.67	0	0.68	0.61	55.87
Pyura microcosmus	0.62	0	0.68	0.61	56.48
Amphilectus fucorum	0.64	0	0.93	0.61	57.09
Neptunea antiqua	0.4	0.67	1.21	0.61	57.69
Eubranchus pallidus	0.62	0	0.9	0.6	58.29
Ascidiella aspersa	0.62	0	0.88	0.59	58.89
Bougainvillia/Eudendrium sp.	0.64	0	0.95	0.59	59.48
Gobiusculus flavescens	0.62	0	0.69	0.59	60.06

Groups GVI & GVII

Average dissimilarity = 68.58

	Group GVI	Group GVI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Limaria hians	2.24	0	5.61	2.63	2.63
Balanus balanus	1.73	0	5.61	2.04	4.67
Munida rugosa	1.71	0	12.08	1.96	6.63
Ascidiella scabra	0	1.55	2.27	1.85	8.48
Rhodophyceae crusts	0	1.38	4.06	1.61	10.09
Carcinus maenas	0	1.24	1.24	1.54	11.62
Heterosiphonia japonica	1.21	0	12.08	1.38	13.01
Phrynorhombus regius	1.21	0	12.08	1.38	14.39
Pollachius pollachius	1.21	0	12.08	1.38	15.78
Polymastia penicillus	1.21	0	12.08	1.38	17.16
Phyllophora crispa	0	1.14	3.36	1.35	18.51
Rhizocaulus verticillatus	1	0	0.91	1.35	19.86
Antedon bifida	1	1.72	1.12	1.3	21.15
Cucumaria frondosa	0	1	1.1	1.23	22.38
Botryllus schlosseri	1	0	5.61	1.18	23.56
Brosme brosme	1	0	5.61	1.18	24.74
Diaphorodoris luteocincta	1	0	5.61	1.18	25.91
Eledone cirrhosa	1	0	5.61	1.18	27.09
Lissoclinum	1	0	5.61	1.18	28.26
Pandalus montagui	1	0	5.61	1.18	29.44
Pleuronectes platessa	1	0	5.61	1.18	30.62
Pagurus	0.87	1.73	0.91	1.17	31.78
Clausinella fasciata	0.71	1.22	1.16	1.14	32.92
Pholis gunnellus	1.21	0.47	1.32	1.08	34
Gibbula umbilicalis	1	1.24	3.64	1.08	35.08
Ophiothrix fragilis	1.37	1.39	1.5	1.05	36.13
Balanus crenatus	0.87	0.8	1.32	1	37.13
Pecten maximus	0.71	1.05	0.96	1	38.13
Hyas	0.71	0	0.91	0.95	39.08
Luidia ciliaris	0.87	0.33	1.17	0.94	40.02
Sycon ciliatum	0.5	1.38	1.58	0.93	40.95
Hydractinia echinata	0	0.8	1.16	0.93	41.88

Strongylocentrotus droebachien	sis 0	0.75	0.64	0.91	42.79
Lanice conchilega	0	0.8	1.24	0.91	43.69
Ectocarpaceae	1.87	1.41	1	0.88	44.57
Ciona intestinalis	0.87	0	0.91	0.87	45.44
Inachus	0.87	0	0.91	0.87	46.31
Kallymenia reniformis	0.87	0	0.91	0.87	47.18
Marthasterias glacialis	0.87	0	0.91	0.87	48.05
Saccharina latissima	0.87	0	0.91	0.87	48.93
Spirobranchus lamarcki	0.87	0	0.91	0.87	49.8
Henricia	1	0.33	1.25	0.82	50.62
Callophyllis cristata	0.71	0.47	0.91	0.78	51.4
Neptunea antiqua	0	0.67	1.24	0.77	52.17
Pomatoschistus pictus	0	0.67	1.24	0.77	52.94
Scrupocellaria scruposa	0	0.67	1.24	0.77	53.71
Suberites carnosus	0	0.67	1.24	0.77	54.48
Nemertesia ramosa	0.71	0.33	1.06	0.76	55.23
Capulus ungaricus	1.37	0.67	1.13	0.72	55.96
Monia patelliformis	0.71	0	0.91	0.71	56.67
Sabella pavonina	0.71	0	0.91	0.71	57.38
Ulva compressa	0.71	0	0.91	0.71	58.09
Amphilectus fucorum	0.5	0	0.91	0.67	58.76
Anomiidae	0.5	0	0.91	0.67	59.44
Bonnemaisonia hamifera	0.5	0	0.91	0.67	60.11

Groups GI & GVII

Average dissimilarity = 79.84

	Group GI	Group GVI			
Species	Av.Abund	Av.Abund	Diss/SD	Contrib%	Cum.%
Ophiopholis aculeata	0	2.21	3.75	2.45	2.45
Golfingia (Golfingia) vulgaris	2	0	10.76	2.19	4.64
Bugula avicularia	0	1.88	3.38	2.09	6.73
Ophiocomina nigra	0	1.88	3.38	2.09	8.83
Caryophyllia (Caryophyllia) smithii	1.73	0	10.76	1.9	10.72
Cerianthus Iloydii	1.73	0	10.76	1.9	12.62
Phycodrys rubens	1.73	0	10.76	1.9	14.52
Rhodomenia	1.73	0	10.76	1.9	16.42
Liocarcinus depurator	2	0.33	2.95	1.81	18.23
Macropodia rostrata	2	0.47	1.73	1.73	19.96
Ascidiella scabra	0	1.55	2.2	1.73	21.69
Pomatoschistus minutus	2	0.47	1.95	1.64	23.33
Ectocarpaceae	0	1.41	1.15	1.63	24.96
Ophiothrix fragilis	0	1.39	1.11	1.61	26.57
Terebellidae	1.73	0.33	2.12	1.57	28.14
Botryllus schlosseri	1.41	0	10.76	1.55	29.69
Brosme brosme	1.41	0	10.76	1.55	31.24
Callophyllis laciniata	1.41	0	10.76	1.55	32.79
Crossaster papposus	1.41	0	10.76	1.55	34.34
Didemnum maculosum	1.41	0	10.76	1.55	35.89
Eledone cirrhosa	1.41	0	10.76	1.55	37.44
Eupolymnia nebulosa	1.41	0	10.76	1.55	38.99
Halecium halecinum	1.41	0	10.76	1.55	40.54

Halecium muricatum	1.41	0	10.76	1.55	42.09
Heterosiphonia plumosa	1.41	0	10.76	1.55	43.64
Mycale	1.41	0	10.76	1.55	45.19
Pleuronectes platessa	1.41	0	10.76	1.55	46.74
Rhodophyllis divaricata	1.41	0	10.76	1.55	48.29
Scrupocellaria	1.41	0	10.76	1.55	49.84
Spirobranchus lamarcki	1.41	0	10.76	1.55	51.4
Galathea intermedia	1.73	0.33	2.6	1.51	52.9
Sycon ciliatum	0	1.38	4.81	1.5	54.4
Carcinus maenas	0	1.24	1.15	1.43	55.84
Gibbula umbilicalis	0	1.24	1.12	1.32	57.16
Clausinella fasciata	0	1.22	1.03	1.31	58.47
Phyllophora crispa	0	1.14	3.56	1.26	59.73

Appendix 4.2. Summary of taxa contributing to 30% of the withingroup similarity and between-group dissimilarities for Modiolus modiolus reef communities

Table A4.2. Summary of taxa contributing to 30% of the within-group similarity and betweengroup dissimilarities for *Modiolus modiolus* reef communities sampled across the U.K. distributional range for the habitat. The data are derived from a SIMPER test carried out on square root transformed benthic abundance data from replicated clump removal samples.

Group Orkney

Average similarity: 54.55

Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Nereimyra punctata	2.3	1.7	6.72	3.11	3.11
Spirobranchus	1.88	1.61	5.3	2.94	6.05
Pisidia longicornis	1.97	1.53	7.4	2.81	8.86
Pholoe baltica	1.68	1.42	7.28	2.6	11.46
Sphaerosyllis taylori	1.78	1.38	6.92	2.53	13.99
Kefersteinia cirrata	1.67	1.36	8.4	2.5	16.49
Harmothoe	1.75	1.36	7.98	2.5	18.99
Nucula nucleus	1.49	1.32	6.3	2.42	21.41
Polycirrus norvegicus	1.59	1.3	5.46	2.38	23.79
Modiolus modiolus	1.42	1.27	5.35	2.33	26.12
Hiatella arctica	1.49	1.26	8.68	2.31	28.43
Pholoe inornata	1.51	1.23	8.82	2.25	30.68

Group Loch Linnhe

Average similarity: 49.37

Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Nereimyra punctata	2.47	2.24	6.8	4.53	4.53
Spirobranchus	2.26	2.17	8.81	4.39	8.92
Balanus	2.91	1.94	2.05	3.92	12.84
Myrianida	1.99	1.79	11.79	3.62	16.46
Modiolus modiolus	1.89	1.78	10.32	3.61	20.07
Harmothoe	1.82	1.68	11.09	3.4	23.47
Leptochiton asellus	1.72	1.6	7.55	3.24	26.71
Glycera lapidum	1.42	1.32	10.75	2.67	29.38
Paradialychone filicaudata	1.47	1.31	4.56	2.65	32.03

Group Loch Alsh

Average similarity: 47.23

	Av Abund	Av Sim	Sim/SD	Contrib%	Cum %
Species	700,0000		0	0011011070	eann /
Pholoe inornata	2.57	1.72	5.41	3.64	3.64
Nereimyra punctata	2.33	1.56	3.26	3.31	6.95
Ophiopholis aculeata	2	1.47	4.45	3.11	10.05
Spirobranchus	2.09	1.4	2.37	2.95	13.01
Polynoidae	1.98	1.36	2.44	2.89	15.89
Ophiothrix fragilis	1.98	1.34	4.05	2.85	18.74
Nucula nucleus	1.81	1.33	6.79	2.82	21.57
Eumida sanguinea	1.82	1.29	6.8	2.73	24.3
Anomiidae	2.14	1.28	2.62	2.71	27
Ophiocomina nigra	1.79	1.22	5.03	2.59	29.59
Jasmineira elegans	2.15	1.22	1.68	2.58	32.17

Group Loch Creran

Average similarity: 55.26

Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Balanus	2.64	2.26	5.82	4.1	4.1
Spirobranchus	2.3	2.13	8.61	3.85	7.94
Monia patelliformis	2.2	2.04	9.9	3.69	11.63
Dendrodoa grossularia	2.1	2.02	12.6	3.65	15.28
Pisidia longicornis	2.12	1.85	5.96	3.35	18.63
Lepidonotus squamatus	1.88	1.76	6.63	3.18	21.81
Phtisica marina	1.96	1.72	6.25	3.1	24.91
Mytilus edulis	2.03	1.71	4.81	3.09	28
Nereimyra punctata	1.92	1.69	5.13	3.06	31.06

Group Shetland

Average similarity: 42.80

Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Hiatella arctica	1.94	2.18	5.19	5.09	5.09
Nereimyra punctata	1.82	2.02	4.93	4.71	9.8
Modiolus modiolus	1.61	1.98	6.27	4.64	14.44
Polynoidae	1.68	1.84	2.28	4.3	18.74
Ophiopholis aculeata	2.24	1.83	0.97	4.28	23.02
Ostracoda	2.05	1.74	1.05	4.07	27.09
Anomiidae	1.78	1.68	2.07	3.92	31.01

Group Pen Llyn

Average similarity: 61.04

Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Scalibregma inflatum	2.93	2.39	6.05	3.91	3.91
Pisidia longicornis	2.68	2.01	6.93	3.3	7.21
Abra	2.7	1.96	6.86	3.2	10.41
Nucula nucleus	2.5	1.91	7.72	3.13	13.54
Aphelochaeta	2.64	1.9	7.55	3.12	16.66
Exogone (Exogone) naidina	2.15	1.75	7.41	2.86	19.53
Caulleriella	2.13	1.71	6.6	2.81	22.33
Pholoe	2.22	1.67	9.03	2.73	25.06
Polycirrus	1.99	1.56	8.61	2.55	27.61
Sphaerosyllis hystrix	1.98	1.54	5.84	2.53	30.14

Groups Orkney & Loch Linnhe

Average dissimilarity = 65.48

	Group Orkney	Group Loch Linnhe				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Balanus	0	2.91	1.38	1.92	2.11	2.11
Onoba semicostata	2.07	0.38	0.82	1.75	1.25	3.37
Paradialychone filicaudata	0	1.47	0.71	4.33	1.09	4.45
Myrianida	0.45	1.99	0.71	2.59	1.08	5.54
Pisidia longicornis	1.97	0.54	0.7	1.81	1.06	6.6
Crassicorophium bonellii	2.14	0.9	0.69	1.48	1.05	7.65
Jugaria granulata	0	1.48	0.69	1.32	1.05	8.7
Ophiothrix fragilis	1.55	0.38	0.65	1.4	1	9.7
Glycera lapidum	0.16	1.42	0.61	2.66	0.94	10.63
Dipolydora coeca	0.16	1.31	0.58	1.97	0.89	11.52
Exogone (Exogone) naidina	1.33	0.18	0.57	1.62	0.88	12.39
Modiolus modiolus	1.38	0.19	0.57	2.5	0.87	13.27
Gitana sarsi	1.23	0.11	0.57	1.56	0.87	14.13
Ophiocomina nigra	1.12	0	0.56	1.54	0.86	15
Amphipholis squamata	1.23	0	0.56	1.58	0.86	15.85
Oxydromus pallidus	1.16	0	0.56	2.33	0.85	16.71
Phyllodoce	0	1.1	0.53	2	0.81	17.51
Prionospio cirrifera	0	1.13	0.53	2.02	0.81	18.32
Phtisica marina	1.19	0.1	0.52	1.25	0.79	19.12
Harmothoe fragilis	1.33	0.25	0.52	2.16	0.79	19.91
Kefersteinia cirrata	1.67	0.63	0.51	1.53	0.78	20.69
Proceraea	0	1.04	0.51	1.94	0.78	21.47

Aonides oxycephala	0.16	1.14	0.51	1.7	0.78	22.25
Sphaerosyllis taylori	1.78	0.78	0.5	1.41	0.77	23.01
Copepoda	1.13	0.08	0.5	2.14	0.77	23.78
Janira maculosa	1.01	1.2	0.5	1.85	0.76	24.54
Nucula nucleus	1.49	0.52	0.48	1.69	0.73	25.27
Trichobranchus glacialis	1.3	0.67	0.46	1.45	0.7	25.98
Amphipholis squamata	0	0.93	0.45	1.58	0.68	26.66
Paradoneis lyra	1.25	0.63	0.44	1.42	0.67	27.33
Dexamine	1.04	0.17	0.44	1.42	0.67	28
Ascidiacea	0	0.86	0.42	1.27	0.65	28.65
Eualus	1.03	0.74	0.42	1.19	0.64	29.29
Pseudoparatanais batei	0.91	0	0.41	1.25	0.63	29.92
Hiatella arctica	1.49	0.69	0.41	1.33	0.62	30.54

Groups Orkney & Loch Alsh

Average dissimilarity = 67.49

	Group Orkney	Group Loch Alsh				
Species	Av.Abund	Av.Abund	Av.Dis	s Diss/SD	Contrib%	Cum.%
Anomiidae	0	2.14	0.94	1.92	1.39	1.39
Jasmineira elegans	0.18	2.15	0.88	1.73	1.3	2.68
Polynoidae	0.13	1.98	0.81	2.79	1.2	3.89
Crassicorophium bonellii	2.14	0.27	0.79	1.71	1.17	5.06
Harmothoe	1.75	0.08	0.71	3.51	1.06	6.12
Balanus	0	1.54	0.69	1	1.02	7.14
Polycirrus norvegicus	1.59	0	0.68	4.56	1.01	8.15
Onoba semicostata	2.07	0.98	0.66	1.5	0.97	9.13
Ophiopholis aculeata	0.49	2	0.64	2.09	0.95	10.08
Pisidia longicornis	1.97	0.49	0.63	2.04	0.94	11.02
Aonides oxycephala	0.16	1.51	0.62	1.48	0.91	11.94
Flabelligera affinis	0	1.39	0.61	1.72	0.9	12.84
Ascidiacea	0	1.43	0.61	2.22	0.9	13.74
Sphaerosyllis hystrix	0	1.4	0.59	1.27	0.88	14.61
Sphaerosyllis taylori	1.78	0.43	0.58	1.96	0.87	15.48
Ostracoda	0.96	1.99	0.57	1.46	0.84	16.32
Kefersteinia cirrata	1.67	0.77	0.54	1.97	0.8	17.13
Harmothoe fragilis	1.33	0.06	0.54	3.82	0.8	17.92
Exogone (Exogone) naidina	1.33	0.26	0.5	1.63	0.74	18.67
Gitana sarsi	1.23	0.16	0.5	1.54	0.74	19.41
Limaria hians	0.34	1.42	0.5	1.86	0.74	20.14
Eumida sanguinea	0.67	1.82	0.49	1.8	0.73	20.87
Paradoneis lyra	1.25	0.21	0.49	1.69	0.72	21.6
Oxydromus pallidus	1.16	0.2	0.47	2.05	0.7	22.3

Phtisica marina	1.19	0.07	0.47	1.25	0.7	23
Polycirrus	0.76	1.27	0.47	1.53	0.7	23.7
Mytilus edulis	0	1.09	0.47	1.25	0.69	24.39
Pholoe inornata	1.51	2.57	0.47	1.73	0.69	25.08
Amphipholis squamata	1.23	0.4	0.47	1.42	0.69	25.77
Psammechinus miliaris	0.69	1.63	0.46	1.42	0.67	26.45
Janira maculosa	1.01	0.57	0.45	1.05	0.66	27.11
Pholoe baltica	1.68	0.67	0.45	1.55	0.66	27.77
Copepoda	1.13	0.42	0.44	1.85	0.65	28.42
Golfingia	1.1	0.55	0.42	1.76	0.63	29.05
Dexamine	1.04	0.06	0.42	1.47	0.62	29.67
Pseudoparatanais batei	0.91	1.02	0.42	1.26	0.62	30.29

Groups Loch Linnhe & Loch Alsh

Average dissimilarity = 69.93

	Group Loch Linnhe	Group Loch Alsh				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Jasmineira elegans	0	2.15	0.96	1.95	1.38	1.38
Balanus	2.91	1.54	0.9	1.42	1.29	2.66
Ophiopholis aculeata	0	2	0.9	4.37	1.28	3.95
Polynoidae	0	1.98	0.88	3.34	1.26	5.21
Myrianida	1.99	0	0.88	6.98	1.26	6.47
Pholoe inornata	0.74	2.57	0.8	2.23	1.15	7.61
Ophiocomina nigra	0	1.79	0.79	4.92	1.12	8.74
Harmothoe	1.82	0.08	0.78	4.18	1.11	9.85
Anomiidae	0.53	2.14	0.77	1.41	1.09	10.94
Ophiothrix fragilis	0.38	1.98	0.71	2.19	1.02	11.96
Jugaria granulata	1.48	0.22	0.61	1.35	0.87	12.83
Sphaerosyllis hystrix	0	1.4	0.61	1.28	0.87	13.7
Paradialychone filicaudata	1.47	0.13	0.6	2.85	0.86	14.57
Nucula nucleus	0.52	1.81	0.58	2.1	0.83	15.4
Modiolus modiolus	0.19	1.39	0.57	1.62	0.81	16.21
Polycirrus norvegicus	1.26	0	0.55	2.07	0.79	17
Ostracoda	1.16	1.99	0.55	1.41	0.78	17.78
Polycirrus	0.53	1.27	0.51	1.4	0.73	18.5
Psammechinus miliaris	0.55	1.63	0.51	1.57	0.73	19.23
Nereididae	1.11	0	0.5	1.93	0.72	19.95
Limaria hians	0.33	1.42	0.5	1.93	0.72	20.67
Eusyllis blomstrandi	1.11	0	0.5	1.53	0.71	21.39
Dipolydora coeca	1.31	0.26	0.5	1.79	0.71	22.09
Hiatella arctica	0.69	1.64	0.47	1.53	0.68	22.77
Flabelligera affinis	0.63	1.39	0.47	1.37	0.67	23.44

Proceraea	1.04	0	0.47	1.95	0.67	24.12
Janira maculosa	1.2	0.57	0.47	2.05	0.67	24.79
Mytilus edulis	0.29	1.09	0.45	1.29	0.64	25.43
Glycera lapidum	1.42	0.42	0.44	1.73	0.64	26.07
Aonides oxycephala	1.14	1.51	0.44	1.55	0.62	26.69
Ophiuroidea	0.32	1.09	0.43	1.28	0.62	27.31
Kurtiella bidentata	0.74	1.13	0.43	1.32	0.62	27.93
Sipuncula	0	0.96	0.43	1.18	0.61	28.54
Trichobranchus glacialis	0.67	1.6	0.42	1.39	0.61	29.15
Aurospio banyulensis	0	0.93	0.42	0.98	0.6	29.75
Serpulidae	1.36	0.56	0.42	1.52	0.6	30.35

Groups Orkney & Loch Creran

Average dissimilarity = 67.91

	Group Orkney	Group Loch Crera	n			
Species	Av.Abund	Av.Abund	Av.Dis	s Diss/SD	Contrib%	Cum.%
Balanus	0	2.64	1.28	4.09	1.88	1.88
Dendrodoa grossularia	0	2.1	1.02	6.63	1.51	3.39
Mytilus edulis	0	2.03	0.98	3.87	1.44	4.83
Crassicorophium bonellii	2.14	0	0.98	1.92	1.44	6.28
Onoba semicostata	2.07	0	0.94	1.92	1.39	7.67
Sphaerosyllis taylori	1.78	0	0.84	6.2	1.24	8.91
Pyura microcosmus	0	1.72	0.83	6.1	1.23	10.14
Polycirrus norvegicus	1.59	0.16	0.69	2.76	1.01	11.15
Modiolus modiolus	1.38	0	0.66	5.43	0.98	12.13
Harmothoe fragilis	1.33	0	0.63	7.75	0.93	13.06
Exogone (Exogone) naidina	1.33	0	0.61	1.69	0.9	13.97
Tritaeta gibbosa	0	1.27	0.61	1.38	0.9	14.87
Nucula nucleus	1.49	0.25	0.6	2.59	0.89	15.76
Monia patelliformis	1.02	2.2	0.6	1.52	0.88	16.63
Gitana sarsi	1.23	0	0.59	1.65	0.87	17.51
Lepidonotus squamatus	0.69	1.88	0.57	2.06	0.84	18.35
Ophiocomina nigra	1.12	0	0.57	1.55	0.83	19.18
Amphipholis squamata	1.23	0	0.56	1.59	0.83	20.02
Testudinalia testudinalis	1.15	0	0.56	2.28	0.82	20.84
Liljeborgia kinahani	0	1.16	0.56	1.49	0.82	21.66
Liljeborgia pallida	0	1.13	0.56	2.29	0.82	22.48
Aoridae	0.67	1.14	0.55	1.17	0.82	23.29
Perrierella audouiniana	0	1.13	0.55	1.56	0.8	24.1
Flabelligera affinis	0	1.12	0.54	2.24	0.8	24.9
Paradoneis lyra	1.25	0.25	0.52	1.74	0.77	25.67
Phtisica marina	1.19	1.96	0.52	1.16	0.77	26.45

Golfingia	1.1	0	0.52	2.34	0.77	27.21
Harmothoe	1.75	0.85	0.5	1.5	0.74	27.96
Janira maculosa	1.01	0.91	0.5	1.32	0.74	28.7
Stenosemus albus	0	1.03	0.5	2.22	0.74	29.44
Galathea	1.01	0	0.49	2.44	0.72	30.16

Groups Loch Linnhe & Loch Creran

Average dissimilarity = 68.22

	Group Loch Linnhe	Group Loch Creran				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Dendrodoa grossularia	0	2.1	1.05	8.1	1.54	1.54
Myrianida	1.99	0	0.98	10.35	1.44	2.99
Phtisica marina	0.1	1.96	0.93	3.44	1.37	4.36
Mytilus edulis	0.29	2.03	0.87	2.44	1.27	5.62
Pyura microcosmus	0	1.72	0.86	7.2	1.26	6.88
Pisidia longicornis	0.54	2.12	0.81	1.87	1.19	8.07
Ophiothrix fragilis	0.38	1.85	0.78	1.96	1.14	9.21
Jugaria granulata	1.48	0	0.71	1.34	1.04	10.25
Glycera lapidum	1.42	0	0.71	9.94	1.03	11.29
Balanus	2.91	2.64	0.68	1.48	1	12.28
Paradialychone filicaudata	1.47	0.13	0.67	3.03	0.99	13.27
Eupolymnia nebulosa	0	1.35	0.67	4.75	0.98	14.25
Tritaeta gibbosa	0	1.27	0.63	1.4	0.93	15.18
Oxydromus pallidus	0	1.27	0.63	1.6	0.92	16.1
Terebellides stroemii	0.08	1.27	0.59	3.4	0.87	16.97
Liljeborgia pallida	0	1.13	0.57	2.35	0.84	17.81
Nereididae	1.11	0	0.57	1.96	0.83	18.64
Polycirrus norvegicus	1.26	0.16	0.57	1.78	0.83	19.47
Aoridae	0	1.14	0.56	0.95	0.83	20.29
Perrierella audouiniana	0	1.13	0.56	1.58	0.82	21.12
Eusyllis blomstrandi	1.11	0	0.56	1.55	0.82	21.94
Phyllodoce	1.1	0	0.55	2.06	0.8	22.74
Proceraea	1.04	0	0.53	1.99	0.77	23.52
Aonides oxycephala	1.14	0.13	0.52	1.88	0.77	24.28
Pholoe inornata	0.74	1.76	0.52	1.44	0.76	25.05
Corophiidae	0.76	0.99	0.5	1.2	0.74	25.78
Harmothoe	1.82	0.85	0.5	1.21	0.73	26.51
Hiatella arctica	0.69	1.63	0.5	1.45	0.73	27.24
Liljeborgia kinahani	0.36	1.16	0.49	1.38	0.72	27.96
Monia patelliformis	1.27	2.2	0.47	1.71	0.69	28.65
Chlamys	0	0.93	0.47	1.63	0.69	29.34
Lepidonotus squamatus	0.97	1.88	0.46	1.7	0.67	30.01

Groups Loch Alsh & Loch Creran

Average dissimilarity = 65.84

	Group Loch Alsh	Group Loch Creran				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Jasmineira elegans	2.15	0	0.97	1.97	1.47	1.47
Dendrodoa grossularia	0	2.1	0.94	6.8	1.43	2.9
Ophiopholis aculeata	2	0	0.9	4.51	1.37	4.27
Phtisica marina	0.07	1.96	0.84	3.58	1.28	5.55
Ophiocomina nigra	1.79	0	0.79	5.11	1.2	6.75
Pyura microcosmus	0	1.72	0.77	6.24	1.17	7.92
Polynoidae	1.98	0.33	0.75	2.31	1.14	9.07
Anomiidae	2.14	0.57	0.75	1.33	1.13	10.2
Pisidia longicornis	0.49	2.12	0.74	2.15	1.12	11.32
Balanus	1.54	2.64	0.73	1.61	1.1	12.43
Nucula nucleus	1.81	0.25	0.7	3.07	1.06	13.48
Aonides oxycephala	1.51	0.13	0.64	1.58	0.97	14.45
Limaria hians	1.42	0	0.63	5.31	0.96	15.41
Sphaerosyllis hystrix	1.4	0	0.61	1.29	0.93	16.34
Modiolus modiolus	1.39	0	0.61	1.67	0.92	17.27
Metaphoxus fultoni	1.44	0.13	0.59	2.46	0.9	18.16
Tritaeta gibbosa	0	1.27	0.57	1.39	0.86	19.02
Ostracoda	1.99	1.03	0.56	1.52	0.85	19.87
Oxydromus pallidus	0.2	1.27	0.54	1.53	0.82	20.69
Eupolymnia nebulosa	0.21	1.35	0.52	2.21	0.79	21.47
Liljeborgia kinahani	0	1.16	0.52	1.5	0.78	22.26
Aoridae	0.57	1.14	0.51	1.14	0.78	23.04
Liljeborgia pallida	0	1.13	0.51	2.3	0.78	23.82
Mytilus edulis	1.09	2.03	0.49	1.31	0.75	24.57
Kefersteinia cirrata	0.77	1.15	0.48	1.99	0.72	25.29
Polycirrus	1.27	1.09	0.47	1.47	0.71	25.99
Ophiuroidea	1.09	0.37	0.45	1.31	0.68	26.67
Corophiidae	0.4	0.99	0.45	1.07	0.68	27.35
Perrierella audouiniana	0.3	1.13	0.45	1.41	0.68	28.03
Protodorvillea kefersteini	0.06	1.01	0.44	1.59	0.67	28.69
Janira maculosa	0.57	0.91	0.44	1.21	0.66	29.35
Sipuncula	0.96	0	0.43	1.19	0.65	30

Groups Orkney & Shetland

Average dissimilarity = 69.30

	Group Orkney	Group Shetland				
Species	Av.Abund	Av.Abund	Av.Dis	s Diss/SD	Contrib%	Cum.%
Ophiopholis aculeata	0.49	2.24	1.08	1.63	1.55	1.55
Anomiidae	0	1.78	0.95	2.22	1.38	2.93
Harmothoe	1.75	0	0.93	5.63	1.34	4.27
Crassicorophium bonellii	2.14	0.71	0.9	1.51	1.29	5.56
Ostracoda	0.96	2.05	0.86	1.76	1.25	6.8
Polycirrus norvegicus	1.59	0	0.86	4.37	1.24	8.04
Polynoidae	0.13	1.68	0.85	2.49	1.23	9.27
Pisidia longicornis	1.97	0.4	0.85	2.11	1.23	10.5
Kefersteinia cirrata	1.67	0.16	0.8	2.93	1.16	11.66
Onoba semicostata	2.07	1.09	0.77	1.6	1.1	12.76
Paradoneis Iyra	1.25	0.06	0.68	1.93	0.98	13.74
Harmothoe fragilis	1.33	0.06	0.67	3.85	0.97	14.72
Amphipholis squamata	1.23	0	0.63	1.58	0.91	15.62
Mya truncata	1.46	0.31	0.61	2.14	0.88	16.51
Pholoe inornata	1.51	0.49	0.61	1.93	0.88	17.39
Copepoda	1.13	0	0.59	2.4	0.85	18.24
Oxydromus pallidus	1.16	0.18	0.59	2.01	0.85	19.09
Sphaerosyllis taylori	1.78	0.78	0.59	1.44	0.85	19.94
Golfingia	1.1	0	0.58	2.26	0.84	20.79
Trichobranchus glacialis	1.3	0.38	0.58	1.47	0.84	21.62
Amphipholis squamata	0	1.09	0.58	1.87	0.84	22.46
Exogone (Exogone) naidina	1.33	0.46	0.58	1.57	0.84	23.3
Phtisica marina	1.19	0.14	0.58	1.25	0.83	24.13
Harpinia crenulata	0.6	1.31	0.57	1.29	0.82	24.95
Scalibregma inflatum	1.32	0.38	0.57	1.59	0.82	25.76
Janira maculosa	1.01	0.74	0.53	1.33	0.77	26.53
Chaetozone	0	1.03	0.52	1.25	0.75	27.28
Eusyllis blomstrandi	0.96	0	0.52	1.66	0.75	28.03
Balanus	0	0.91	0.52	0.56	0.75	28.78
Dexamine	1.04	0.06	0.51	1.49	0.74	29.52
Ophiothrix fragilis	1.55	1.18	0.5	1.34	0.72	30.25

Groups Loch Linnhe & Shetland

Average dissimilarity = 73.94

	Group Loch Linnhe	Group Shetland				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Balanus	2.91	0.91	1.41	1.73	1.91	1.91
Ophiopholis aculeata	0	2.24	1.26	1.5	1.7	3.61
Myrianida	1.99	0	1.11	7.08	1.49	5.11
Harmothoe	1.82	0	1.02	7.07	1.38	6.48
Polynoidae	0	1.68	0.94	3.1	1.27	7.75
Ophiocomina nigra	0	1.49	0.84	1.95	1.13	8.89
Ostracoda	1.16	2.05	0.84	1.68	1.13	10.02
Paradialychone filicaudata	1.47	0.07	0.79	3.12	1.07	11.09
Jugaria granulata	1.48	0	0.79	1.32	1.07	12.16
Anomiidae	0.53	1.78	0.78	1.59	1.05	13.22
Glycera lapidum	1.42	0.06	0.75	4.5	1.02	14.23
Serpulidae	1.36	0.06	0.73	3.1	0.99	15.22
Hiatella arctica	0.69	1.94	0.71	1.75	0.97	16.19
Polycirrus norvegicus	1.26	0	0.69	2.05	0.94	17.12
Harpinia crenulata	0.36	1.31	0.65	1.32	0.88	18
Dipolydora coeca	1.31	0.41	0.64	1.82	0.87	18.87
Nereididae	1.11	0	0.64	1.9	0.86	19.73
Eusyllis blomstrandi	1.11	0	0.63	1.52	0.85	20.58
Proceraea	1.04	0	0.59	1.93	0.8	21.39
Phyllodoce	1.1	0.06	0.59	1.88	0.8	22.19
Ophiothrix fragilis	0.38	1.18	0.58	1.37	0.79	22.97
Gitana sarsi	0.11	1.07	0.57	1.34	0.78	23.75
Chaetozone	0	1.03	0.54	1.27	0.73	24.47
Ophiuroidea	0.32	1.09	0.53	1.37	0.72	25.19
Onoba semicostata	0.38	1.09	0.53	1.31	0.71	25.91
Mya truncata	1.19	0.31	0.52	1.56	0.7	26.61
Monia patelliformis	1.27	0.44	0.52	1.5	0.7	27.31
Lepidonotus squamatus	0.97	0.06	0.52	1.82	0.7	28.01
Crassicorophium bonellii	0.9	0.71	0.5	1.27	0.67	28.68
Leptochiton asellus	1.72	0.85	0.48	1.46	0.65	29.33
Eumida sanguinea	1	0.41	0.46	1.3	0.63	29.96
Nucula nucleus	0.52	1.08	0.46	1.35	0.62	30.59

Groups Loch Alsh & Shetland

Average dissimilarity = 66.06

	Group Loch Alsh	Group Shetland				
Species	Av.Abund	Av.Abund	Av.Dis	s Diss/SD	Contrib%	Cum.%
Jasmineira elegans	2.15	0	1.07	1.92	1.62	1.62
Pholoe inornata	2.57	0.49	1.01	2.29	1.53	3.15
Balanus	1.54	0.91	0.87	1.14	1.32	4.48
Psammechinus miliaris	1.63	0.19	0.73	1.98	1.1	5.58
Limaria hians	1.42	0	0.7	4.58	1.06	6.64
Eumida sanguinea	1.82	0.41	0.69	2.29	1.05	7.69
Lepidonotus squamatus	1.47	0.06	0.69	2.59	1.04	8.73
Flabelligera affinis	1.39	0.13	0.66	1.64	1	9.73
Ophiopholis aculeata	2	2.24	0.66	1.8	1	10.73
Sphaerosyllis hystrix	1.4	0.14	0.65	1.28	0.99	11.72
Ostracoda	1.99	2.05	0.65	1.31	0.99	12.71
Trichobranchus glacialis	1.6	0.38	0.62	1.84	0.93	13.64
Aonides oxycephala	1.51	0.94	0.55	1.3	0.83	14.46
Polycirrus	1.27	0.88	0.54	1.42	0.81	15.28
Ascidiacea	1.43	0.52	0.53	1.51	0.81	16.08
Monia patelliformis	1.37	0.44	0.52	1.47	0.78	16.87
Anomiidae	2.14	1.78	0.51	1.1	0.77	17.64
Gitana sarsi	0.16	1.07	0.5	1.32	0.76	18.4
Mytilus edulis	1.09	0.69	0.49	1.26	0.75	19.14
Onoba semicostata	0.98	1.09	0.49	1.27	0.75	19.89
Harpinia crenulata	0.8	1.31	0.49	1.3	0.74	20.63
Modiolus modiolus	1.39	0.89	0.48	1.28	0.73	21.36
Chaetozone	0	1.03	0.48	1.25	0.73	22.09
Metaphoxus fultoni	1.44	0.56	0.48	1.52	0.72	22.81
Aurospio banyulensis	0.93	0.14	0.47	1.02	0.71	23.52
Pseudoparatanais batei	1.02	0	0.46	1.03	0.7	24.22
Sipuncula	0.96	0.14	0.46	1.17	0.7	24.92
Terebellides stroemii	0.94	0.06	0.45	1.55	0.68	25.6
Janira maculosa	0.57	0.74	0.44	1.11	0.66	26.26
Kurtiella bidentata	1.13	0.71	0.44	1.31	0.66	26.92
Ophiothrix fragilis	1.98	1.18	0.43	1.29	0.66	27.58
Ophiuroidea	1.09	1.09	0.43	1.27	0.65	28.23
Leptochiton asellus	1	0.85	0.41	1.33	0.62	28.84
Spirobranchus	2.09	1.66	0.4	1.06	0.6	29.45
Dendrodoa grossularia	0	0.77	0.39	0.6	0.59	30.04

Groups Loch Creran & Shetland

Average dissimilarity = 72.59

	Group Loch Creran	Group Shetland				
Species	Av.Abund	Av.Abund	Av.Diss	B Diss/SD	Contrib%	Cum.%
Ophiopholis aculeata	0	2.24	1.26	1.51	1.74	1.74
Balanus	2.64	0.91	1.26	2.44	1.73	3.47
Phtisica marina	1.96	0.14	1.04	2.97	1.43	4.9
Lepidonotus squamatus	1.88	0.06	1.03	4.36	1.42	6.31
Monia patelliformis	2.2	0.44	1	2.53	1.38	7.69
Pisidia longicornis	2.12	0.4	0.99	2.12	1.36	9.06
Pyura microcosmus	1.72	0	0.97	5.94	1.34	10.39
Dendrodoa grossularia	2.1	0.77	0.96	2.47	1.33	11.72
Ostracoda	1.03	2.05	0.86	2	1.19	12.91
Ophiocomina nigra	0	1.49	0.84	1.97	1.16	14.06
Mytilus edulis	2.03	0.69	0.8	1.71	1.1	15.17
Polynoidae	0.33	1.68	0.78	2.04	1.08	16.25
Anomiidae	0.57	1.78	0.76	1.53	1.05	17.3
Pholoe inornata	1.76	0.49	0.75	1.76	1.04	18.34
Tritaeta gibbosa	1.27	0	0.71	1.38	0.98	19.32
Terebellides stroemii	1.27	0.06	0.68	3.55	0.94	20.26
Oxydromus pallidus	1.27	0.18	0.68	1.52	0.93	21.19
Liljeborgia kinahani	1.16	0	0.65	1.49	0.89	22.08
Liljeborgia pallida	1.13	0	0.65	2.29	0.89	22.98
Aoridae	1.14	0.69	0.64	1.2	0.89	23.86
Perrierella audouiniana	1.13	0	0.63	1.57	0.87	24.74
Harpinia crenulata	0.4	1.31	0.63	1.39	0.87	25.61
Eupolymnia nebulosa	1.35	0.36	0.6	2.03	0.83	26.44
Gitana sarsi	0	1.07	0.59	1.38	0.82	27.26
Onoba semicostata	0	1.09	0.59	1.32	0.82	28.08
Kefersteinia cirrata	1.15	0.16	0.59	1.9	0.82	28.89
Flabelligera affinis	1.12	0.13	0.58	1.89	0.8	29.69
Corophiidae	0.99	0	0.56	0.97	0.76	30.46

Groups Orkney & Pen Llyn

Average dissimilarity = 72.02

	Group Orkney	Group Pen Llyn				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Aphelochaeta	0.13	2.64	1.12	3.15	1.55	1.55

Abra	0.27	2.7	1.05	3.81	1.46	3.01
Nereimyra punctata	2.3	0	1	4.32	1.39	4.4
Pholoe	0	2.22	0.98	7.8	1.36	5.76
Crassicorophium bonellii	2.14	0	0.91	1.87	1.26	7.02
Sphaerosyllis hystrix	0	1.98	0.88	5.2	1.23	8.24
Onoba semicostata	2.07	0	0.88	1.87	1.22	9.46
Caulleriella	0.25	2.13	0.84	3.48	1.17	10.63
Amphipholis squamata	0	1.86	0.82	6.95	1.14	11.77
Sphaerosyllis taylori	1.78	0	0.78	5.16	1.08	12.85
Pholoe baltica	1.68	0	0.75	6.38	1.04	13.89
Kefersteinia cirrata	1.67	0	0.74	6.26	1.02	14.91
Scalibregma inflatum	1.32	2.93	0.72	4.1	1	15.91
Polycirrus norvegicus	1.59	0	0.71	4.54	0.98	16.89
Pholoe inornata	1.51	0	0.66	6.15	0.92	17.81
Sabellaria spinulosa	0	1.53	0.66	3.18	0.92	18.73
Cirriformia tentaculata	0	1.49	0.66	5.83	0.91	19.65
Mya truncata	1.46	0	0.65	5.3	0.9	20.55
Dipolydora caulleryi	0.13	1.52	0.63	3.08	0.87	21.42
Scalibregma celticum	0.25	1.66	0.62	2.39	0.86	22.28
Balanus	0	1.51	0.62	1.01	0.86	23.14
Kurtiella bidentata	1.06	2.11	0.59	1.72	0.82	23.96
Harmothoe fragilis	1.33	0	0.59	6.02	0.81	24.77
Autolytinae	0.13	1.39	0.56	3.09	0.78	25.55
Polycirrus	0.76	1.99	0.53	2	0.74	26.29
Amphipholis squamata	1.23	0	0.52	1.55	0.73	27.02
Ophiocomina nigra	1.12	0	0.52	1.53	0.72	27.74
Oxydromus pallidus	1.16	0	0.52	2.28	0.72	28.46
Testudinalia testudinalis	1.15	0	0.52	2.22	0.72	29.18
Bradypontius magniceps	0	1.18	0.51	2.18	0.71	29.89
Eunereis longissima	0	1.13	0.51	4.96	0.7	30.59

Groups Loch Linnhe & Pen Llyn

Average dissimilarity = 75.59

Group Loch Linnhe Group Pen Llyn						
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Nereimyra punctata	2.47	0	1.14	5.1	1.51	1.51
Scalibregma inflatum	0.68	2.93	1.04	3.04	1.38	2.89
Aphelochaeta	0.38	2.64	1.03	2.6	1.36	4.25
Abra	0.44	2.7	1.02	3.06	1.35	5.6
Pholoe	0	2.22	1	10.11	1.33	6.93
Pisidia longicornis	0.54	2.68	1	2.36	1.32	8.25
Balanus	2.91	1.51	0.99	1.39	1.31	9.56

Sphaerosyllis hystrix	0	1.98	0.91	5.73	1.2	10.76
Myrianida	1.99	0	0.91	7.05	1.2	11.96
Nucula nucleus	0.52	2.5	0.91	2.98	1.2	13.16
Exogone (Exogone) naidina	0.18	2.15	0.91	4.02	1.2	14.36
Caulleriella	0.18	2.13	0.9	3.83	1.19	15.54
Modiolus modiolus	0.19	2.04	0.83	2.99	1.09	16.64
Kurtiella bidentata	0.74	2.11	0.71	1.65	0.94	17.58
Exogone (Parexogone) hebes	0.18	1.75	0.71	2.9	0.93	18.51
Sabellaria spinulosa	0	1.53	0.68	3.32	0.9	19.41
Nephtys kersivalensis	0.08	1.56	0.68	3.77	0.9	20.31
Polycirrus	0.53	1.99	0.66	2.52	0.88	21.19
Jugaria granulata	1.48	0	0.66	1.32	0.87	22.06
Serpulidae	1.36	0	0.63	4.06	0.83	22.89
Pholoe baltica	1.34	0	0.62	5.22	0.82	23.7
Scalibregma celticum	0.39	1.66	0.59	1.92	0.78	24.48
Polycirrus norvegicus	1.26	0	0.57	2.06	0.75	25.24
Prosphaerosyllis tetralix	0.18	1.32	0.56	1.81	0.74	25.98
Perioculodes longimanus	0	1.21	0.55	1.86	0.73	26.71
Janira maculosa	1.2	0	0.55	2.89	0.73	27.44
Mya truncata	1.19	0	0.55	2.4	0.73	28.16
Harmothoe	1.82	0.69	0.55	1.62	0.72	28.88
Polydora	0	1.21	0.54	2.17	0.71	29.6
Bradypontius magniceps	0	1.18	0.52	2.23	0.69	30.29

Groups Loch Alsh & Pen Llyn

Average dissimilarity = 72.02

	Group Loch Alsh	Group Pen Llyn				
Species	Av.Abund	Av.Abund	Av.Dis	Av.Diss Diss/SD		Cum.%
Pholoe inornata	2.57	0	1.05	4.63	1.45	1.45
Aphelochaeta	0.29	2.64	0.98	2.43	1.36	2.82
Nereimyra punctata	2.33	0	0.97	3.11	1.35	4.16
Pisidia longicornis	0.49	2.68	0.91	2.69	1.27	5.43
Scalibregma inflatum	0.78	2.93	0.91	2.56	1.26	6.7
Pholoe	0	2.22	0.91	7.7	1.26	7.96
Abra	0.51	2.7	0.91	2.42	1.26	9.22
Anomiidae	2.14	0	0.9	1.93	1.24	10.46
Ophiopholis aculeata	2	0	0.84	4.21	1.16	11.62
Caulleriella	0.25	2.13	0.79	3.27	1.09	12.72
Exogone (Exogone) naidina	0.26	2.15	0.78	3.62	1.08	13.8
Jasmineira elegans	2.15	0.46	0.75	1.48	1.03	14.84
Ophiocomina nigra	1.79	0	0.73	4.59	1.02	15.85
Balanus	1.54	1.51	0.69	1.24	0.96	16.82

Polynoidae	1.98	0.43	0.68	2.2	0.94	17.76
Exogone (Parexogone) hebes	0.16	1.75	0.65	2.91	0.91	18.66
Nephtys kersivalensis	0	1.56	0.65	5.44	0.9	19.56
Sabellaria spinulosa	0	1.53	0.62	3.15	0.86	20.42
Limaria hians	1.42	0	0.59	4.8	0.82	21.24
Flabelligera affinis	1.39	0	0.58	1.72	0.81	22.05
Kurtiella bidentata	1.13	2.11	0.58	1.44	0.8	22.85
Trichobranchus glacialis	1.6	0.29	0.55	2.29	0.76	23.61
Ascidiacea	1.43	0.14	0.54	1.95	0.75	24.36
Ostracoda	1.99	1.03	0.53	1.49	0.73	25.1
Metaphoxus fultoni	1.44	0.19	0.52	2.11	0.72	25.82
Aonides oxycephala	1.51	0.59	0.52	1.37	0.72	26.54
Prosphaerosyllis tetralix	0.2	1.32	0.5	1.78	0.7	27.24
Polydora	0	1.21	0.49	2.14	0.68	27.91
Cirriformia tentaculata	0.35	1.49	0.48	1.88	0.67	28.58
Amphipholis squamata	0.7	1.86	0.48	1.68	0.67	29.25
Bradypontius magniceps	0	1.18	0.47	2.19	0.66	29.91
Hilbigneris gracilis	0.41	1.45	0.47	1.9	0.65	30.55

Groups Loch Creran & Pen Llyn

Average dissimilarity = 71.68

	Group Loch Crera	n Group Pen Llyn				
Species	Av.Abund	Av.Abund	Av.Dis	s Diss/SD	Contrib%	Cum.%
Aphelochaeta	0	2.64	1.21	4.1	1.68	1.68
Nucula nucleus	0.25	2.5	1.03	4.25	1.43	3.11
Pholoe	0	2.22	1.01	12.32	1.41	4.52
Exogone (Exogone) naidina	0	2.15	0.99	12.19	1.38	5.9
Scalibregma inflatum	0.93	2.93	0.94	2.86	1.3	7.21
Abra	0.65	2.7	0.92	2.51	1.29	8.49
Modiolus modiolus	0	2.04	0.92	5.03	1.28	9.77
Sphaerosyllis hystrix	0	1.98	0.91	6.07	1.27	11.05
Phtisica marina	1.96	0	0.91	4.37	1.27	12.31
Dendrodoa grossularia	2.1	0.14	0.9	4.44	1.26	13.58
Nereimyra punctata	1.92	0	0.89	4.43	1.24	14.81
Pholoe inornata	1.76	0	0.81	3.85	1.14	15.95
Balanus	2.64	1.51	0.81	1.48	1.13	17.08
Pyura microcosmus	1.72	0	0.8	6.24	1.11	18.19
Caulleriella	0.41	2.13	0.79	2.8	1.11	19.29
Scalibregma celticum	0	1.66	0.77	3.93	1.07	20.37
Mytilus edulis	2.03	0.46	0.74	2.03	1.03	21.39
Exogone (Parexogone) hebes	0.21	1.75	0.71	2.79	0.99	22.39
Sabellaria spinulosa	0	1.53	0.68	3.37	0.95	23.34
Amphipholis squamata	0.43	1.86	0.65	2.33	0.91	24.25
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Kurtiella bidentata	1	2.11	0.63	1.64	0.87	25.13
Cirriformia tentaculata	0.13	1.49	0.62	3.4	0.87	26
Prosphaerosyllis tetralix	0	1.32	0.62	2.16	0.87	26.86
Oxydromus pallidus	1.27	0	0.58	1.58	0.81	27.68
Syllis	0.13	1.36	0.57	3.21	0.79	28.47
Nephtys kersivalensis	0.38	1.56	0.54	2.21	0.76	29.23
Kefersteinia cirrata	1.15	0	0.53	2.39	0.74	29.97
Liljeborgia kinahani	1.16	0	0.53	1.49	0.74	30.72

Groups Shetland & Pen Llyn

Average dissimilarity = 77.17

	Group Shetland	Group Pen Llyn				
Species	Av.Abund	Av.Abund	Av.Dis	s Diss/SD	Contrib%	Cum.%
Scalibregma inflatum	0.38	2.93	1.33	3.03	1.73	1.73
Aphelochaeta	0.13	2.64	1.28	3.31	1.65	3.38
Abra	0.26	2.7	1.24	3.42	1.6	4.98
Pisidia longicornis	0.4	2.68	1.18	2.58	1.53	6.52
Ophiopholis aculeata	2.24	0	1.15	1.48	1.49	8.01
Pholoe	0	2.22	1.12	8.14	1.45	9.47
Caulleriella	0.13	2.13	1.04	3.94	1.34	10.81
Sphaerosyllis hystrix	0.14	1.98	0.95	3.39	1.23	12.04
Nereimyra punctata	1.82	0	0.93	4.61	1.2	13.23
Anomiidae	1.78	0	0.9	2.23	1.17	14.41
Exogone (Exogone) naidina	0.46	2.15	0.88	2.6	1.14	15.54
Balanus	0.91	1.51	0.83	1.05	1.07	16.62
Scalibregma celticum	0.13	1.66	0.8	2.68	1.03	17.65
Kurtiella bidentata	0.71	2.11	0.8	1.7	1.03	18.68
Ostracoda	2.05	1.03	0.79	1.85	1.03	19.71
Ophiocomina nigra	1.49	0	0.77	1.92	0.99	20.7
Nephtys kersivalensis	0.07	1.56	0.77	3.51	0.99	21.69
Sabellaria spinulosa	0	1.53	0.76	3.29	0.98	22.68
Hilbigneris gracilis	0	1.45	0.74	5.4	0.96	23.63
Nucula nucleus	1.08	2.5	0.72	1.86	0.93	24.57
Monia patelliformis	0.44	1.82	0.7	1.78	0.9	25.47
Prosphaerosyllis tetralix	0	1.32	0.69	2.08	0.9	26.37
Polynoidae	1.68	0.43	0.69	1.89	0.89	27.26
Exogone (Parexogone) hebes	0.44	1.75	0.67	1.84	0.87	28.13
Cirriformia tentaculata	0.19	1.49	0.66	2.71	0.86	28.99
Syllis	0.13	1.36	0.63	3	0.82	29.81
Paradoneis lyra	0.06	1.29	0.62	3.58	0.81	30.62

Appendix 4.3. Summary of species contributing to 90% of the within-group similarity and 60% of the dissimilarity between group similarities for SIMPROF Modiolus modiolus groupings

Table A3.3. Summary of species contributing to 90% of the within-group similarity and 60% of the dissimilarity between group similarities for SIMPROF *Modiolus modiolus* groupings. The data are derived from a SIMPER test carried out on squareroot transformed benthic abundance data from DDV footage.

Group d

Average similarity: 70.27

species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Alcyonium digitatum	2.31	13.47	26.43	19.17	19.17
Modiolus modiolus	2.16	12.52	15.27	17.81	36.98
Asterias rubens	2	12.05	26.43	17.14	54.12
Abietinaria abietina	1.82	10.43	26.43	14.85	68.97
Ophiothrix fragilis	1.88	9.66	5.48	13.75	82.72
Sagartia elegans	1.52	8.52	26.43	12.12	94.84

Group i

Average similarity: 37.98

species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Echinus esculentus	1.83	8.09	16.1	21.29	21.29
Pomatoceros spp.	1.72	6.82	3.39	17.95	39.24
Modiolus modiolus	1.1	5.2	6.81	13.71	52.94
Pagurus bernhadus	1.06	4	0.9	10.55	63.49
Red Calcareous Encrusting Algae	1.3	3.27	0.83	8.62	72.11
Encrusting sp.onge sp.	1.14	3.23	0.91	8.5	80.61
Flustra foliacea	1	1.62	0.41	4.26	84.87
Calliostoma zizyphinum	0.71	1.14	0.41	3.01	87.89
Asterias rubens	0.71	1.03	0.41	2.7	90.59

Group c

Average similarity: 75.88

species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Ophiothrix fragilis	2.34	22.98	#######	30.29	30.29
Modiolus modiolus	2.12	20.56	#######	27.09	57.38
Echinus esculentus	1.73	17.8	#######	23.46	80.84
Asterias rubens	1.41	14.54	#######	19.16	100

Group e

Average similarity: 74.34

species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Echinus esculentus	2.24	5.71	13.1	7.68	7.68
Modiolus modiolus	2.31	5.71	13.1	7.68	15.35

Alcyonium digitatum	2.16	5.32	7.82	7.15	22.5
Barnacle	2.08	5.1	13.1	6.87	29.37
Pomatoceros spp.	2.08	5.1	13.1	6.87	36.23
Ophiothrix fragilis	2.14	4.81	13.87	6.47	42.71
Encrusting sp.onge	1.91	4.66	7.52	6.26	48.97
Sertularia spp.	1.91	4.66	7.52	6.26	55.23
Nemertesia antennina	1.88	4.12	4.01	5.55	60.78
Bryozoa	1.8	4.06	7.87	5.47	66.25
Asterias rubens	1.63	3.86	24.2	5.19	71.43
Flustra foliacea	1.72	3.86	24.2	5.19	76.62
<i>Henricia</i> sp.	1.41	3.61	13.1	4.85	81.47
Halecium sp.	1.55	2.92	4.01	3.92	85.4
Pagurus bernhadus	1.61	1.55	0.58	2.09	87.49
Crossaster papposus	1.05	1.27	0.58	1.71	89.19
Ascidia	0.94	1.24	0.58	1.67	90.86

Group f

Average similarity: 63.54

species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Ophiothrix fragilis	2.34	7.66	5.18	12.05	12.05
Echinus esculentus	2.09	6.81	7.84	10.72	22.77
Modiolus modiolus	2.11	6.59	4.04	10.36	33.13
Asterias rubens	1.78	5.61	6.63	8.83	41.96
Henricia sp.	1.6	5.17	8.54	8.13	50.09
Sertularia spp.	1.83	4.82	1.81	7.59	57.68
Myoxocephalus scorpius	1.3	3.8	1.88	5.98	63.66
Kirchenpaueria pinnata	1.5	3.54	1.24	5.57	69.22
Cancer pagurus	1.32	3.25	1.24	5.11	74.33
Encrusting sponge	1.28	3.03	1.72	4.77	79.1
Crossaster papposus	1.16	2.9	1.25	4.56	83.66
Pagurus bernhadus	0.99	2.07	0.92	3.26	86.92
Halecium sp.	1.14	2	0.87	3.14	90.06

Group h

Average similarity: 55.70

species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Modiolus modiolus	1.99	7.71	7.33	13.83	13.83
Echinus esculentus	1.92	6.83	5.98	12.26	26.1
Ophiothrix fragilis	1.87	5.79	1.16	10.4	36.5
Crossaster papposus	1.48	5.73	14.47	10.29	46.79
Ophiopholis albida	1.83	5.7	1.16	10.23	57.02
Corallinaceae	1.52	5.41	4.53	9.72	66.74
Asterias rubens	1.25	3.43	1.15	6.16	72.9
Porania pulvillus	1.13	3.43	1.15	6.16	79.05
Pomatoceros spp.	0.95	2.38	1.16	4.27	83.32
Ophiocomina nigra	0.83	1.42	0.61	2.56	85.87
<i>Bryozoan</i> sp.	0.6	1.14	0.62	2.04	87.92
Halecium sp.	0.75	1.14	0.62	2.04	89.96
Hydroid	0.69	0.74	0.32	1.33	91.29

Group a

Average similarity: 49.57

species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Echinus esculentus	1.73	6.63	#######	13.38	13.38
Pagurus bernhadus	1.98	6.63	#######	13.38	26.75
Aequipecten opercularis	1.57	5.41	#######	10.92	37.67
Asterias rubens	1.41	5.41	#######	10.92	48.59
Carcinus maenas	1.41	5.41	#######	10.92	59.51
Liocarcinus depurator	1.41	5.41	#######	10.92	70.44
Modiolus modiolus	1.57	5.41	#######	10.92	81.36
Pecten maximus	1.41	5.41	#######	10.92	92.28

Groups b & d

Average dissimilarity = 59.90

	Group b	Group d				
sp.ecies	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Cirripedia	2	0	3.07	53.06	5.12	5.12
Ophiothrix fragilis	0	1.88	2.88	4.78	4.81	9.93
Maja squinado	1.73	0	2.66	53.06	4.43	14.36
Pagurus bernhadus	1.73	0	2.66	53.06	4.43	18.79
Scyliorhinus canicula	1.73	0	2.66	53.06	4.43	23.23
Sertularia spp.	1.73	0	2.66	53.06	4.43	27.66
Hydroid	2	0.47	2.34	1.88	3.91	31.57
Aequipecten opercularis	1.41	0	2.17	53.06	3.62	35.19
Alcyonidium diaphanum	1.41	0	2.17	53.06	3.62	38.81
Aspitrigla cuculus	1.41	0	2.17	53.06	3.62	42.43
Clavelina lepadiformis	1.41	0	2.17	53.06	3.62	46.05
Ctenolabrus rupestris	1.41	0	2.17	53.06	3.62	49.67
Gadoid sp.	1.41	0	2.17	53.06	3.62	53.29
Liocarcinus depurator	1.41	0	2.17	53.06	3.62	56.91

Groups b & i

Average dissimilarity = 79.74

	Group b	Group i				
sp.ecies	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.45	0	3.61	12.55	4.53	4.53
Cirripedia	2	0	2.95	12.55	3.7	8.22
Hydroid sp	2	0	2.95	12.55	3.7	11.92
Echinus esculentus	0	1.83	2.68	4.49	3.37	15.28
Abietinaria abietina	2	0.25	2.58	3.29	3.24	18.52
Maja squinado	1.73	0	2.55	12.55	3.2	21.72
Sertularia spp.	1.73	0	2.55	12.55	3.2	24.92
Asterias rubens	2.24	0.71	2.31	1.71	2.89	27.81
Alcyonidium diaphanum	1.41	0	2.08	12.55	2.61	30.42
Aspitrigla cuculus	1.41	0	2.08	12.55	2.61	33.04

Callionymus lyra	1.41	0	2.08	12.55	2.61	35.65	
Clavelina lepadiformis	1.41	0	2.08	12.55	2.61	38.26	
Ctenolabrus rupestris	1.41	0	2.08	12.55	2.61	40.88	
Gadoid sp.	1.41	0	2.08	12.55	2.61	43.49	
Neopentadactyla mixta	1.41	0	2.08	12.55	2.61	46.1	
Pentapora foliacea	1.41	0	2.08	12.55	2.61	48.71	
Pholis gunnellus	1.41	0	2.08	12.55	2.61	51.33	
Sagartia elegans	1.41	0	2.08	12.55	2.61	53.94	
Suberites ficus	1.41	0	2.08	12.55	2.61	56.55	
Taurulus bubalis	1.41	0	2.08	12.55	2.61	59.17	

Groups d & i

Average dissimilarity = 84.05

	Group d	Group i				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.31	0	6.49	6.94	7.72	7.72
Ophiothrix fragilis	1.88	0	5.28	4.53	6.28	13.99
Echinus esculentus	0	1.83	5.09	4.99	6.06	20.05
Pomatoceros spp.	0	1.72	4.73	3.49	5.63	25.68
Abietinaria abietina	1.82	0.25	4.43	2.9	5.27	30.95
Sagartia elegans	1.52	0	4.28	5.45	5.09	36.04
Asterias rubens	2	0.71	3.84	1.52	4.57	40.61
Red Calcareous Encrusting Algae	0	1.3	3.64	1.39	4.33	44.94
Pagurus bernhadus	0	1.06	3.12	1.61	3.72	48.65
Encrusting sponge	0	1.14	2.98	1.59	3.54	52.19
Modiolus modiolus	2.16	1.1	2.97	3.72	3.54	55.73
Flustra foliacea	1.15	1	2.84	1.12	3.38	59.11

Groups b & c

Average dissimilarity = 83.22

	Group b	Group c				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	0	2.34	4.03	8.63	4.84	4.84
Abietinaria abietina	2	0	3.43	19.35	4.12	8.96
Cirripedia	2	0	3.43	19.35	4.12	13.09
Hydroid sp	2	0	3.43	19.35	4.12	17.21
Alcyonium digitatum	2.45	0.71	3.03	1.62	3.65	20.86
Echinus esculentus	0	1.73	2.97	19.35	3.57	24.43
Flustra foliacea	1.73	0	2.97	19.35	3.57	28
Maja squinado	1.73	0	2.97	19.35	3.57	31.57
Scyliorhinus canicula	1.73	0	2.97	19.35	3.57	35.15
Sertularia spp.	1.73	0	2.97	19.35	3.57	38.72
Aequipecten opercularis	1.41	0	2.43	19.35	2.92	41.63
Alcyonidium diaphanum	1.41	0	2.43	19.35	2.92	44.55
Aspitrigla cuculus	1.41	0	2.43	19.35	2.92	47.47
Callionymus lyra	1.41	0	2.43	19.35	2.92	50.38
Clavelina lepadiformis	1.41	0	2.43	19.35	2.92	53.3
Ctenolabrus rupestris	1.41	0	2.43	19.35	2.92	56.22
Gadoid sp.	1.41	0	2.43	19.35	2.92	59.13

Groups d & c

Average dissimilarity = 54.02

	Group d	Group c				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Abietinaria abietina	1.82	0	6.95	10.44	12.87	12.87
Echinus esculentus	0	1.73	6.63	10.15	12.27	25.14
Alcyonium digitatum	2.31	0.71	6.33	1.8	11.72	36.85
Sagartia elegans	1.52	0	5.82	6.53	10.78	47.63
Flustra foliacea	1.15	0	4.53	1.28	8.39	56.02
Encrusting sponge	0	0.71	2.49	0.91	4.6	60.62

Groupsi& c

Average dissimilarity = 69.60

	Group i	Group c				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	0	2.34	8.3	4.29	11.92	11.92
Pomatoceros spp.	1.72	0	5.88	3.41	8.45	20.37
Red Calcareous Encrusting Algae	1.3	0	4.57	1.33	6.57	26.94
Modiolus modiolus	1.1	2.12	3.56	3.39	5.12	32.06
Flustra foliacea	1	0	3.32	0.93	4.77	36.82
Barnacle	0.91	0	2.84	0.87	4.09	40.91
Asterias rubens	0.71	1.41	2.81	0.91	4.04	44.95
Encrusting sponge	1.14	0.71	2.8	1	4.03	48.98
Pagurus bernhadus	1.06	0.71	2.62	0.89	3.77	52.75
Calliostoma zizyphinum	0.71	0	2.35	0.93	3.37	56.12
Alcyonium digitatum	0	0.71	2.3	0.91	3.31	59.42

Groups b & e

Average dissimilarity = 68.83

	Group b	Group e				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Echinus esculentus	0	2.24	2.55	13.51	3.71	3.71
Ophiothrix fragilis	0	2.14	2.42	8.74	3.52	7.22
Barnacle sp.	0	2.08	2.37	16.38	3.44	10.66
Cirripedia	2	0	2.28	13.51	3.31	13.98
Hydroid	2	0	2.28	13.51	3.31	17.29
Encrusting sponge.	0	1.91	2.18	7.68	3.17	20.46
Bryozoa	0	1.8	2.04	8.12	2.96	23.43
Maja squinado	1.73	0	1.98	13.51	2.87	26.3
Scyliorhinus canicula	1.73	0	1.98	13.51	2.87	29.17
Abietinaria abietina	2	0.33	1.91	2.66	2.78	31.95
<i>Halecium</i> sp.	0	1.55	1.76	2.66	2.55	34.5
Aequipecten opercularis	1.41	0	1.61	13.51	2.34	36.85

Alcyonidium diaphanum	1.41	0	1.61	13.51	2.34	39.19
Aspitrigla cuculus	1.41	0	1.61	13.51	2.34	41.54
Callionymus lyra	1.41	0	1.61	13.51	2.34	43.88
Clavelina lepadiformis	1.41	0	1.61	13.51	2.34	46.22
Ctenolabrus rupestris	1.41	0	1.61	13.51	2.34	48.57
Gadoid sp.	1.41	0	1.61	13.51	2.34	50.91
Henricia sp.	0	1.41	1.61	13.51	2.34	53.26
Liocarcinus depurator	1.41	0	1.61	13.51	2.34	55.6
Macropodia sp.	1.41	0	1.61	13.51	2.34	57.94

Groups d & e

Average dissimilarity = 67.11

	Group d	Group e				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Echinus esculentus	0	2.24	4.03	9.51	6.01	6.01
Barnacle.	0	2.08	3.74	11.99	5.57	11.58
Pomatoceros spp.	0	2.08	3.74	11.99	5.57	17.15
Encrusting sponge	0	1.91	3.46	6.71	5.15	22.3
Sertularia spp.	0	1.91	3.46	6.71	5.15	27.45
Nemertesia antennina	0	1.88	3.41	4.41	5.07	32.52
Bryozoan	0	1.8	3.21	13.31	4.78	37.3
Halecium sp.	0	1.55	2.77	3.16	4.13	41.43
Sagartia elegans	1.52	0	2.74	6.58	4.09	45.51
Abietinaria abietina	1.82	0.33	2.71	2.58	4.04	49.56
Pagurus bernhadus	0	1.61	2.71	1.27	4.04	53.59
Henricia sp.	0	1.41	2.55	9.51	3.8	57.4

Groupsi& e

Average dissimilarity = 65.33

	Group i	Group e				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	0	2.16	3.73	6.17	5.7	5.7
Ophiothrix fragilis	0	2.14	3.64	8.88	5.57	11.28
Sertularia spp.	0	1.91	3.3	5.99	5.05	16.33
Nemertesia antennina	0.25	1.88	2.84	2.55	4.34	20.67
Halecium sp.	0	1.55	2.65	3.09	4.05	24.72
Bryozoan	0.5	1.8	2.45	1.85	3.75	28.47
Red Calcareous Encrusting Algae	1.3	0	2.22	1.38	3.41	31.87
Barnacle	0.91	2.08	2.22	1.31	3.4	35.27
Pagurus bernhadus	1.06	1.61	2.14	1.79	3.27	38.55
Schizotricha frutescens	0	1.22	2.1	1.31	3.22	41.77
Modiolus modiolus	1.1	2.31	2.07	4.75	3.17	44.93
Crossaster papposus	0	1.05	1.86	1.34	2.85	47.78
<i>Henricia</i> sp.	0.35	1.41	1.83	1.6	2.81	50.59
Flustra foliacea	1	1.72	1.75	1.28	2.68	53.26
Necora puber	0	0.94	1.67	1.32	2.55	55.82
Asterias rubens	0.71	1.63	1.63	1.18	2.5	58.32

Groupsc & e

Average dissimilarity = 62.39

	Group c	Group e				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Barnacle	0	2.08	4.28	9.11	6.86	6.86
Pomatoceros spp.	0	2.08	4.28	9.11	6.86	13.72
Sertularia spp.	0	1.91	3.96	5.7	6.35	20.07
Nemertesia antennina	0	1.88	3.9	4.03	6.26	26.32
Bryozoan sp.	0	1.8	3.66	12.91	5.87	32.2
Flustra foliacea	0	1.72	3.49	13.35	5.6	37.8
Halecium sp.	0	1.55	3.17	3.05	5.08	42.88
Alcyonium digitatum	0.71	2.16	3.08	1.66	4.93	47.81
<i>Henricia</i> sp.	0	1.41	2.92	7.52	4.68	52.49
Pagurus bernhadus	0.71	1.61	2.74	1.35	4.4	56.89

Groups b & f

Average dissimilarity = 76.88

	Group b	Group f				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	0	2.34	3	7.62	3.9	3.9
Alcyonium digitatum	2.45	0.24	2.85	3.76	3.71	7.61
Echinus esculentus	0	2.09	2.68	11.43	3.48	11.09
Cirripedia	2	0	2.57	12.04	3.34	14.43
Hydroid	2	0.1	2.43	5.7	3.16	17.59
Maja squinado	1.73	0	2.22	12.04	2.89	20.48
Scyliorhinus canicula	1.73	0	2.22	12.04	2.89	23.37
Abietinaria abietina	2	0.3	2.21	3.08	2.87	26.25
Henricia sp.	0	1.6	2.05	13.19	2.67	28.91
Kirchenpaueria pinnata	0	1.5	1.91	1.79	2.49	31.4
Alcyonidium diaphanum	1.41	0	1.82	12.04	2.36	33.76
Aspitrigla cuculus	1.41	0	1.82	12.04	2.36	36.12
Callionymus lyra	1.41	0	1.82	12.04	2.36	38.48
Clavelina lepadiformis	1.41	0	1.82	12.04	2.36	40.84
Ctenolabrus rupestris	1.41	0	1.82	12.04	2.36	43.2
Gadoid.	1.41	0	1.82	12.04	2.36	45.56
Liocarcinus depurator	1.41	0	1.82	12.04	2.36	47.92
<i>Macropodi</i> a sp.	1.41	0	1.82	12.04	2.36	50.28
Pentapora foliacea	1.41	0	1.82	12.04	2.36	52.65
Sagartia elegans	1.41	0	1.82	12.04	2.36	55.01
Suberites ficus	1.41	0	1.82	12.04	2.36	57.37
Taurulus bubalis	1.41	0	1.82	12.04	2.36	59.73

Groups d & f

Average dissimilarity = 71.44

	Group d	Group f				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.31	0.24	4.59	3.26	6.43	6.43
Echinus esculentus	0	2.09	4.57	7.92	6.4	12.82
Sertularia spp.	0	1.83	3.89	2.55	5.45	18.27

<i>Henricia</i> sp.	0	1.6	3.49	10.11	4.89	23.16
Abietinaria abietina	1.82	0.3	3.41	2.6	4.77	27.93
Sagartia elegans	1.52	0	3.34	5.59	4.68	32.61
Kirchenpaueria pinnata	0	1.5	3.25	1.84	4.55	37.16
Myoxocephalus scorpius	0	1.3	2.83	2.67	3.96	41.12
Cancer pagurus	0	1.32	2.83	1.89	3.96	45.07
Encrusting sponge	0	1.28	2.71	2.15	3.79	48.86
Crossaster papposus	0	1.16	2.48	1.91	3.47	52.34
Ascidia virginea	0	1.14	2.45	1.14	3.43	55.77
Halecium sp.	0	1.14	2.4	1.34	3.36	59.13

Groupsi & f

Average dissimilarity = 72.37

	Group i	Group f				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	0	2.34	4.86	5.01	6.71	6.71
Sertularia spp.	0	1.83	3.69	2.48	5.1	11.81
Kirchenpaueria pinnata	0	1.5	3.08	1.81	4.26	16.07
Red Calcareous Encrusting Algae	1.3	0	2.69	1.4	3.71	19.78
Cancer pagurus	0	1.32	2.68	1.87	3.7	23.49
Myoxocephalus scorpius	0	1.3	2.68	2.59	3.7	27.19
Henricia sp.	0.35	1.6	2.59	1.88	3.57	30.76
Crossaster papposus	0	1.16	2.35	1.88	3.25	34.01
Pomatoceros spp.	1.72	0.62	2.35	1.57	3.25	37.26
Ascidia virginea	0	1.14	2.32	1.13	3.21	40.47
Asterias rubens	0.71	1.78	2.31	1.32	3.19	43.66
Halecium sp.	0	1.14	2.28	1.33	3.15	46.81
Modiolus modiolus	1.1	2.11	2.1	2.16	2.91	49.71
Flustra foliacea	1	0.44	2.04	1.05	2.81	52.53
Barnacle	0.91	0	1.77	0.92	2.44	54.97
Bryozoa	0.5	0.54	1.57	1.15	2.16	57.14
Encrusting sponge	1.14	1.28	1.56	1.13	2.16	59.29

Groups c & f

Average dissimilarity = 56.14

	Group c Group f							
species	Av.Abu	nd Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%		
Sertularia spp.	0	1.83	4.58	2.47	8.16	8.16		
<i>Henricia</i> sp.	0	1.6	4.14	7.62	7.37	15.53		
Kirchenpaueria pinnata	0	1.5	3.85	1.8	6.86	22.38		
Myoxocephalus scorpius	0	1.3	3.35	2.53	5.96	28.34		
Cancer pagurus	0	1.32	3.33	1.85	5.93	34.27		
Crossaster papposus	0	1.16	2.93	1.86	5.21	39.48		
Ascidia virginea	0	1.14	2.89	1.13	5.15	44.63		
Halecium sp.	0	1.14	2.83	1.32	5.03	49.67		
Encrusting sponge	0.71	1.28	2.31	1.3	4.11	53.78		
Pagurus bernhadus	0.71	0.99	1.86	0.96	3.32	57.1		
Alcyonium digitatum	0.71	0.24	1.76	0.99	3.14	60.24		

Groupse & f

Average dissimilarity = 47.48

	Group	e Group f				
species	Av.Abu	nd Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Barnacle	2.08	0	3.04	8.87	6.41	6.41
Alcyonium digitatum	2.16	0.24	2.85	3.12	5.99	12.4
Nemertesia antennina	1.88	0.2	2.47	2.98	5.2	17.6
Pomatoceros spp.	2.08	0.62	2.13	2.23	4.48	22.08
Flustra foliacea	1.72	0.44	1.98	2.01	4.17	26.25
Myoxocephalus scorpius	0	1.3	1.9	2.72	3.99	30.24
Pagurus bernhadus	1.61	0.99	1.87	1.56	3.93	34.18
Bryozoan sp.	1.8	0.54	1.81	2.08	3.81	37.99
Schizotricha frutescens	1.22	0.39	1.71	1.31	3.6	41.59
Ascidia virginea	0	1.14	1.65	1.12	3.47	45.06
Kirchenpaueria pinnata	0.67	1.5	1.63	1.65	3.44	48.5
Ascidia sp.	0.94	0	1.41	1.36	2.98	51.48
Necora puber	0.94	0	1.41	1.36	2.98	54.46
Halecium sp.	1.55	1.14	1.26	1.29	2.65	57.11
Pholis gunnellus	0.75	0.28	1.19	0.84	2.5	59.61

Groups b & h

Average dissimilarity = 78.31

	Group b Group h							
species	Av.Abund Av.Abund		Av.Diss	Diss/SD	Contrib%	Cum.%		
Cirripedia	2	0	2.73	25.22	3.48	3.48		
Echinus esculentus	0	1.92	2.62	4.65	3.35	6.83		
Ophiothrix fragilis	0	1.87	2.6	1.78	3.32	10.15		
Ophiopholis albida	0	1.83	2.54	1.79	3.24	13.39		
Flustra foliacea	1.73	0	2.36	25.22	3.02	16.41		
Maja squinado	1.73	0	2.36	25.22	3.02	19.42		
Alcyonium digitatum	2.45	0.79	2.3	1.49	2.93	22.35		
Sertularia spp.	1.73	0.2	2.11	3.21	2.69	25.04		
Corallinaceae	0	1.52	2.07	4.94	2.64	27.69		
Crossaster papposus	0	1.48	2.02	9.45	2.57	30.26		
Abietinaria abietina	2	0.57	1.98	1.8	2.53	32.79		
Aequipecten opercularis	1.41	0	1.93	25.22	2.46	35.25		
Alcyonidium diaphanum	1.41	0	1.93	25.22	2.46	37.72		
Aspitrigla cuculus	1.41	0	1.93	25.22	2.46	40.18		
Callionymus lyra	1.41	0	1.93	25.22	2.46	42.65		
Clavelina lepadiformis	1.41	0	1.93	25.22	2.46	45.11		
Ctenolabrus rupestris	1.41	0	1.93	25.22	2.46	47.57		
Gadoid sp.	1.41	0	1.93	25.22	2.46	50.04		
Macropodia sp.	1.41	0	1.93	25.22	2.46	52.5		
Nemertesia antennina	1.41	0	1.93	25.22	2.46	54.96		
Neopentadactyla mixta	1.41	0	1.93	25.22	2.46	57.43		
Pentapora foliacea	1.41	0	1.93	25.22	2.46	59.89		

Groups d & h

Average dissimilarity = 68.50

	Group d	Group h				
species	Av.Abund Av.Abund		Av.Diss	Diss/SD	Contrib%	Cum.%
Echinus esculentus	0	1.92	4.67	4.63	6.82	6.82
Ophiopholis albida	0	1.83	4.56	1.93	6.66	13.48
Alcyonium digitatum	2.31	0.79	3.78	1.46	5.52	19
Sagartia elegans	1.52	0	3.69	7.81	5.39	24.39
Corallinaceae	0	1.52	3.67	5.33	5.36	29.76
Crossaster papposus	0	1.48	3.59	8.81	5.24	34.99
Abietinaria abietina	1.82	0.57	3.11	1.67	4.54	39.54
Flustra foliacea	1.15	0	2.85	1.36	4.16	43.7
Porania pulvillus	0	1.13	2.74	1.91	4	47.7
Pomatoceros spp.	0	0.95	2.23	1.76	3.26	50.96
Hydroid sp	0.47	0.69	1.94	0.98	2.83	53.78
Ophiocomina nigra	0.47	0.83	1.93	1.12	2.82	56.6
Asterias rubens	2	1.25	1.86	1.11	2.71	59.32

Groupsi & h

Average dissimilarity = 76.91

	Group i	Group h				
species	Av.Abun	d Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	0	1.87	4.4	1.88	5.72	5.72
Ophiopholis albida	0	1.83	4.3	1.89	5.59	11.31
Corallinaceae	0	1.52	3.47	4.61	4.51	15.82
Crossaster papposus	0	1.48	3.38	6.36	4.4	20.22
Red Calcareous Encrusting Algae	1.3	0	2.96	1.42	3.85	24.07
Porania pulvillus	0	1.13	2.59	1.87	3.36	27.43
Encrusting sponge sp.	1.14	0	2.46	1.62	3.2	30.63
Flustra foliacea	1	0	2.21	0.97	2.88	33.51
Modiolus modiolus	1.1	1.99	2.05	3.08	2.67	36.18
Pomatoceros spp.	1.72	0.95	1.99	1.28	2.59	38.77
Asterias rubens	0.71	1.25	1.94	1.06	2.52	41.29
Barnacle sp.	0.91	0	1.94	0.91	2.52	43.8
Ophiocomina nigra	0	0.83	1.92	1.12	2.5	46.3
Pagurus bernhadus	1.06	0.57	1.85	1.06	2.4	48.7
Bryozoan sp.	0.5	0.6	1.75	1.3	2.27	50.98
Scyliorhinus canicula	0.35	0.69	1.72	0.91	2.24	53.22
Alcyonium digitatum	0	0.79	1.72	0.79	2.23	55.45
Hydroid sp	0	0.69	1.63	0.79	2.12	57.57
Halecium sp.	0	0.75	1.62	1.11	2.11	59.68

Groupsc & h

Average dissimilarity = 58.07

	Group c	Group h	Group h					
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%		
Ophiopholis albida	0	1.83	5.53	1.87	9.52	9.52		
Corallinaceae	0	1.52	4.43	4.97	7.62	17.14		
Crossaster papposus	0	1.48	4.33	7.29	7.45	24.59		
Porania pulvillus	0	1.13	3.31	1.85	5.69	30.28		
Pomatoceros spp.	0	0.95	2.68	1.74	4.61	34.89		

Alcyonium digitatum	0.71	0.79	2.64	1.13	4.54	39.43
Ophiocomina nigra	0	0.83	2.47	1.1	4.25	43.68
Hydroid sp	0	0.69	2.1	0.77	3.62	47.29
Halecium sp.	0	0.75	2.05	1.1	3.52	50.82
Strongylocentrotus droebachiensis	0	0.73	2.03	0.76	3.5	54.31
Pagurus bernhadus	0.71	0.57	2.03	0.95	3.49	57.81

Groupse & h

Average dissimilarity = 62.56

	Group e	Group h	Group h					
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%		
Barnacle sp.	2.08	0	3.26	12.4	5.21	5.21		
Encrusting sp.onge sp.	1.91	0	3.01	7.2	4.82	10.03		
Nemertesia antennina	1.88	0	2.97	4.59	4.75	14.78		
Ophiopholis albida	0	1.83	2.93	1.89	4.69	19.46		
Sertularia spp.	1.91	0.2	2.72	3.34	4.35	23.81		
Flustra foliacea	1.72	0	2.67	11.96	4.27	28.08		
Corallinaceae	0	1.52	2.39	4.84	3.81	31.89		
Alcyonium digitatum	2.16	0.79	2.25	1.39	3.59	35.48		
Henricia sp.	1.41	0	2.22	10.26	3.56	39.04		
Pagurus bernhadus	1.61	0.57	2.17	1.39	3.46	42.5		
Schizotricha frutescens	1.22	0	1.92	1.33	3.07	45.57		
Bryozoan sp.	1.8	0.6	1.88	2.04	3.01	48.58		
Pomatoceros spp.	2.08	0.95	1.8	1.87	2.88	51.46		
Porania pulvillus	0	1.13	1.78	1.89	2.84	54.3		
Ascidia sp.	0.94	0	1.52	1.35	2.43	56.73		
Necora puber	0.94	0	1.52	1.35	2.43	59.16		

Groupsf&h

Average dissimilarity = 59.02

	Group f	Group h				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiopholis albida	0.17	1.83	3.3	1.75	5.59	5.59
Sertularia spp.	1.83	0.2	3.06	2.24	5.18	10.77
<i>Henricia</i> sp.	1.6	0	2.97	10.32	5.02	15.8
Corallinaceae	0	1.52	2.82	4.6	4.78	20.58
Kirchenpaueria pinnata	1.5	0.2	2.53	1.81	4.29	24.87
Myoxocephalus scorpius	1.3	0	2.4	2.73	4.07	28.94
Encrusting sponge sp.	1.28	0	2.31	2.16	3.92	32.86
Porania pulvillus	0	1.13	2.1	1.9	3.56	36.42
Ascidia virginea	1.14	0	2.08	1.14	3.53	39.95
Ophiocomina nigra	0.73	0.83	1.95	1.34	3.3	43.25
Cancer pagurus	1.32	0.57	1.84	1.29	3.11	46.37
<i>Halecium</i> sp.	1.14	0.75	1.68	1.28	2.84	49.2
Alcyonium digitatum	0.24	0.79	1.5	0.92	2.55	51.75
Pagurus bernhadus	0.99	0.57	1.42	1.06	2.4	54.15
Hydroid sp	0.1	0.69	1.36	0.87	2.3	56.45
Strongylocentrotus droebachiensis	0	0.73	1.31	0.78	2.23	58.68

Groups b & a

Average dissimilarity = 76.68

	Group b	Group a				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.45	0	3.28	17.55	4.28	4.28
Abietinaria abietina	2	0	2.68	17.55	3.49	7.78
Cirripedia	2	0	2.68	17.55	3.49	11.27
Hydroid sp	2	0	2.68	17.55	3.49	14.77
Echinus esculentus	0	1.73	2.32	17.55	3.03	17.79
Flustra foliacea	1.73	0	2.32	17.55	3.03	20.82
Maja squinado	1.73	0	2.32	17.55	3.03	23.85
Scyliorhinus canicula	1.73	0	2.32	17.55	3.03	26.87
Sertularia spp.	1.73	0	2.32	17.55	3.03	29.9
Hydractinia echinata	0	1.5	2.04	1.92	2.66	32.56
Alcyonidium diaphanum	1.41	0	1.9	17.55	2.47	35.03
Aspitrigla cuculus	1.41	0	1.9	17.55	2.47	37.5
Callionymus lyra	1.41	0	1.9	17.55	2.47	39.97
Carcinus maenas	0	1.41	1.9	17.55	2.47	42.44
Clavelina lepadiformis	1.41	0	1.9	17.55	2.47	44.91
Ctenolabrus rupestris	1.41	0	1.9	17.55	2.47	47.38
Gadoid sp.	1.41	0	1.9	17.55	2.47	49.85
Macropodia sp.	1.41	0	1.9	17.55	2.47	52.33
Nemertesia antennina	1.41	0	1.9	17.55	2.47	54.8
Neopentadactyla mixta	1.41	0	1.9	17.55	2.47	57.27
Pecten maximus	0	1.41	1.9	17.55	2.47	59.74

Groups d & a

Average dissimilarity = 86.00

	Group d	Group a				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.31	0	5.42	12.4	6.31	6.31
Pagurus bernhadus	0	1.98	4.71	4.63	5.48	11.78
Ophiothrix fragilis	1.88	0	4.41	5.1	5.13	16.92
Abietinaria abietina	1.82	0	4.28	10.84	4.98	21.89
Echinus esculentus	0	1.73	4.08	12.26	4.74	26.63
Aequipecten opercularis	0	1.57	3.67	23.63	4.27	30.9
Hydractinia echinata	0	1.5	3.61	2.31	4.2	35.11
Sagartia elegans	1.52	0	3.58	7.12	4.16	39.27
Carcinus maenas	0	1.41	3.33	12.26	3.87	43.14
Liocarcinus depurator	0	1.41	3.33	12.26	3.87	47.01
Pecten maximus	0	1.41	3.33	12.26	3.87	50.87
Flustra foliacea	1.15	0	2.76	1.28	3.21	54.08
Tunicata	0	1	2.52	0.91	2.93	57.01
Rhodophyta sp.	0	1.12	2.44	0.91	2.84	59.86

Groupsi & a

Average dissimilarity = 75.39

	Group i	Group a				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Pomatoceros spp.	1.72	0	3.76	3.23	4.98	4.98
Hydractinia echinata	0	1.5	3.41	2.28	4.52	9.5
Carcinus maenas	0	1.41	3.14	7.35	4.17	13.67
Aequipecten opercularis	0.25	1.57	2.98	2.47	3.95	17.62
Red Calcareous Encrusting Algae	1.3	0	2.87	1.36	3.81	21.43
Pecten maximus	0.25	1.41	2.65	2.23	3.51	24.94
Encrusting sp.onge sp.	1.14	0	2.39	1.55	3.17	28.12
Tunicata	0	1	2.37	0.92	3.15	31.26
Rhodophyta sp.	0	1.12	2.32	0.93	3.07	34.33
Liocarcinus depurator	0.35	1.41	2.22	1.6	2.94	37.28
Flustra foliacea	1	0	2.15	0.93	2.85	40.13
Pagurus bernhadus	1.06	1.98	2	1.41	2.66	42.79
Barnacle sp.	0.91	0	1.88	0.88	2.5	45.28
Asterias rubens	0.71	1.41	1.7	0.92	2.25	47.53
Arenicola marina	0	0.71	1.68	0.92	2.22	49.76
Diatom Layer	0	0.71	1.68	0.92	2.22	51.98
Virgularia mirabilis	0	0.71	1.68	0.92	2.22	54.21
<i>Henricia</i> sp.	0.35	0.71	1.63	0.92	2.16	56.36
Calliostoma zizyphinum	0.71	0	1.52	0.93	2.02	58.38
Buccinum undatum	0	0.71	1.47	0.93	1.94	60.32

Groupsc & a

Average dissimilarity = 69.72

	Group c	Group a				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	2.34	0	6.62	6.36	9.5	9.5
Aequipecten opercularis	0	1.57	4.4	13.92	6.31	15.81
Hydractinia echinata	0	1.5	4.35	2.1	6.24	22.05
Carcinus maenas	0	1.41	3.99	8.29	5.72	27.77
Liocarcinus depurator	0	1.41	3.99	8.29	5.72	33.48
Pecten maximus	0	1.41	3.99	8.29	5.72	39.2
Pagurus bernhadus	0.71	1.98	3.78	1.33	5.42	44.63
Tunicata	0	1	3.06	0.86	4.39	49.01
Rhodophyta sp.	0	1.12	2.89	0.86	4.14	53.15
Arenicola marina	0	0.71	2.16	0.86	3.1	56.25
Diatom Layer	0	0.71	2.16	0.86	3.1	59.36

Groupse & a

Average dissimilarity = 79.46

	Group e	Group a				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.16	0	3.33	7.06	4.19	4.19
Ophiothrix fragilis	2.14	0	3.26	10.09	4.1	8.3
Barnacle sp.	2.08	0	3.19	11.42	4.02	12.32
Pomatoceros spp.	2.08	0	3.19	11.42	4.02	16.34

Encrusting sp.onge sp.	1.91	0	2.95	6.78	3.71	20.05
Sertularia spp.	1.91	0	2.95	6.78	3.71	23.77
Nemertesia antennina	1.88	0	2.91	4.33	3.66	27.43
Bryozoan sp.	1.8	0	2.75	9.78	3.46	30.88
Flustra foliacea	1.72	0	2.61	10.77	3.29	34.17
Aequipecten opercularis	0	1.57	2.41	9.24	3.04	37.21
Halecium sp.	1.55	0	2.37	3	2.98	40.19
Hydractinia echinata	0	1.5	2.35	2.37	2.95	43.14
Carcinus maenas	0	1.41	2.18	9.58	2.74	45.89
Liocarcinus depurator	0	1.41	2.18	9.58	2.74	48.63
Pecten maximus	0	1.41	2.18	9.58	2.74	51.37
Schizotricha frutescens	1.22	0	1.88	1.25	2.37	53.74
Pagurus bernhadus	1.61	1.98	1.67	1.13	2.11	55.84
Crossaster papposus	1.05	0	1.66	1.29	2.09	57.93
Rhodophyta sp.	0	1.12	1.64	0.91	2.07	60

Groupsf&a

Average dissimilarity = 74.18

	Group f	Group a				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	2.34	0	4.24	6.05	5.72	5.72
Sertularia spp.	1.83	0	3.24	2.53	4.37	10.09
Hydractinia echinata	0	1.5	2.78	2.42	3.74	13.83
Kirchenpaueria pinnata	1.5	0	2.7	1.82	3.64	17.46
Carcinus maenas	0	1.41	2.57	7.65	3.46	20.93
Liocarcinus depurator	0	1.41	2.57	7.65	3.46	24.39
Cancer pagurus	1.32	0	2.35	1.88	3.17	27.56
Pecten maximus	0.14	1.41	2.35	2.73	3.17	30.73
Myoxocephalus scorpius	1.3	0	2.34	2.69	3.16	33.89
Encrusting sponge	1.28	0	2.26	2.12	3.05	36.93
Aequipecten opercularis	0.38	1.57	2.15	1.86	2.89	39.82
Crossaster papposus	1.16	0	2.06	1.89	2.78	42.61
Ascidia virginea	1.14	0	2.03	1.12	2.74	45.35
Halecium sp.	1.14	0	2	1.32	2.7	48.04
Rhodophyta sp.	0.1	1.12	1.93	1.01	2.6	50.64
Tunicata	0	1	1.92	0.96	2.58	53.22
Pagurus bernhadus	0.99	1.98	1.92	1.23	2.58	55.81
Henricia sp.	1.6	0.71	1.54	1.23	2.07	57.88
Arenicola marina	0	0.71	1.36	0.96	1.83	59.71

Groups h & a

Average dissimilarity = 76.04

	Group h	Group a				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Ophiothrix fragilis	1.87	0	3.79	1.87	4.98	4.98
Ophiopholis albida	1.83	0	3.7	1.88	4.87	9.85
Aequipecten opercularis	0	1.57	3.09	14.46	4.07	13.91
Hydractinia echinata	0	1.5	3.02	2.44	3.98	17.89

Crossaster papposus	1.48	0	2.92	8.06	3.84	21.73
Pagurus bernhadus	0.57	1.98	2.89	1.7	3.8	25.54
Carcinus maenas	0	1.41	2.8	12.09	3.68	29.21
Pecten maximus	0	1.41	2.8	12.09	3.68	32.89
Liocarcinus depurator	0.28	1.41	2.29	1.88	3.01	35.9
Porania pulvillus	1.13	0	2.23	1.87	2.94	38.84
Rhodophyta sp.	0.4	1.12	2.13	1.29	2.8	41.64
Tunicata	0	1	2.09	0.95	2.75	44.39
Corallinaceae	1.52	0.5	2.07	1.58	2.72	47.11
Pomatoceros spp.	0.95	0	1.83	1.7	2.4	49.51
Ophiocomina nigra	0.83	0	1.66	1.1	2.18	51.69
Alcyonium digitatum	0.79	0	1.49	0.77	1.96	53.66
Arenicola marina	0	0.71	1.48	0.95	1.95	55.6
Diatom Layer	0	0.71	1.48	0.95	1.95	57.55
<i>Henricia</i> sp.	0	0.71	1.48	0.95	1.95	59.5

Groups b & g

Average dissimilarity = 80.55

	Group b	Group g				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.45	0	3.49	Undefined!	4.33	4.33
Antedon bifida	0	2.24	3.18	Undefined!	3.95	8.28
Asterias rubens	2.24	0	3.18	Undefined!	3.95	12.24
Abietinaria abietina	2	0	2.85	Undefined!	3.54	15.77
Cirripedia	2	0	2.85	Undefined!	3.54	19.31
Hydroid sp	2	0	2.85	Undefined!	3.54	22.84
Ophiopholis albida	0	2	2.85	Undefined!	3.54	26.38
Corallinaceae	0	1.73	2.47	Undefined!	3.06	29.44
Flustra foliacea	1.73	0	2.47	Undefined!	3.06	32.5
Maja squinado	1.73	0	2.47	Undefined!	3.06	35.56
Ophiothrix fragilis	0	1.73	2.47	Undefined!	3.06	38.63
Pagurus bernhadus	1.73	0	2.47	Undefined!	3.06	41.69
Scyliorhinus canicula	1.73	0	2.47	Undefined!	3.06	44.75
Sertularia spp.	1.73	0	2.47	Undefined!	3.06	47.81
Alcyonidium diaphanum	1.41	0	2.01	Undefined!	2.5	50.31
Aspitrigla cuculus	1.41	0	2.01	Undefined!	2.5	52.81
Bryozoan sp.	0	1.41	2.01	Undefined!	2.5	55.31
Callionymus lyra	1.41	0	2.01	Undefined!	2.5	57.81
Clavelina lepadiformis	1.41	0	2.01	Undefined!	2.5	60.31

Groups d & g

Average dissimilarity = 78.49

	Group d	Group g				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Alcyonium digitatum	2.31	0	6.03	42.58	7.69	7.69
Antedon bifida	0	2.24	5.86	31.24	7.46	15.15
Asterias rubens	2	0	5.24	31.24	6.67	21.82
Ophiopholis albida	0	2	5.24	31.24	6.67	28.49

Abietinaria abietina	1.82	0	4.76	18.68	6.07	34.56
Corallinaceae	0	1.73	4.54	31.24	5.78	40.34
Sagartia elegans	1.52	0	3.98	7.58	5.07	45.41
Aequipecten opercularis	0	1.41	3.7	31.24	4.72	50.13
Bryozoan sp.	0	1.41	3.7	31.24	4.72	54.85
Buccinum undatum	0	1.41	3.7	31.24	4.72	59.57

Groupsi & g

Average dissimilarity = 90.89

	Group i	Group g				Cum.%
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	
Antedon bifida	0	2.24	5.5	7.31	6.06	6.06
Ophiopholis albida	0	2	4.92	7.31	5.42	11.47
Echinus esculentus	1.83	0	4.46	4.57	4.91	16.38
Corallinaceae	0	1.73	4.26	7.31	4.69	21.07
Ophiothrix fragilis	0	1.73	4.26	7.31	4.69	25.76
Pomatoceros spp.	1.72	0	4.15	3.12	4.57	30.33
Buccinum undatum	0	1.41	3.48	7.31	3.83	34.16
Crossaster papposus	0	1.41	3.48	7.31	3.83	37.99
Pomatoschistus pictus	0	1.41	3.48	7.31	3.83	41.82
Solaster endeca	0	1.41	3.48	7.31	3.83	45.65
Red Calcareous Encrusting Algae	1.3	0	3.18	1.26	3.5	49.15
Bryozoan sp.	0.5	1.41	3.03	2.45	3.34	52.48
Aequipecten opercularis	0.25	1.41	2.94	2.08	3.24	55.72
<i>Macropodia</i> sp.	0.25	1.41	2.94	2.08	3.24	58.96

Groupsc & g

Average dissimilarity = 76.06

	Group c	Group g				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Antedon bifida	0	2.24	7.17	10.39	9.43	9.43
Ophiopholis albida	0	2	6.41	10.39	8.43	17.86
Corallinaceae	0	1.73	5.55	10.39	7.3	25.16
Echinus esculentus	1.73	0	5.55	10.39	7.3	32.46
Aequipecten opercularis	0	1.41	4.54	10.39	5.96	38.43
Asterias rubens	1.41	0	4.54	10.39	5.96	44.39
Bryozoan	0	1.41	4.54	10.39	5.96	50.35
Buccinum undatum	0	1.41	4.54	10.39	5.96	56.31

Groupse & g

Average dissimilarity = 76.45

	Group e	Group g				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%

Antedon bifida	0	2.24	3.7	9.19	4.84	4.84
Echinus esculentus	2.24	0	3.7	9.19	4.84	9.67
Alcyonium digitatum	2.16	0	3.58	6.49	4.68	14.35
Barnacle sp.	2.08	0	3.43	11.75	4.48	18.83
Pomatoceros spp.	2.08	0	3.43	11.75	4.48	23.32
Ophiopholis albida	0	2	3.31	9.19	4.33	27.64
Encrusting sponge.	1.91	0	3.17	6.21	4.14	31.78
Sertularia spp.	1.91	0	3.17	6.21	4.14	35.92
Nemertesia antennina	1.88	0	3.12	3.92	4.08	40.01
Corallinaceae	0	1.73	2.86	9.19	3.75	43.75
Flustra foliacea	1.72	0	2.8	12.23	3.67	47.42
Asterias rubens	1.63	0	2.67	59.79	3.49	50.91
Pagurus bernhadus	1.61	0	2.5	1.1	3.27	54.18
Aequipecten opercularis	0	1.41	2.34	9.19	3.06	57.24

Groupsf & g

Average dissimilarity = 72.77

	Group f	Group g				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Antedon bifida	0	2.24	4.42	7.64	6.07	6.07
Echinus esculentus	2.09	0	4.11	8.56	5.65	11.72
Ophiopholis albida	0.17	2	3.65	2.99	5.01	16.73
Sertularia spp.	1.83	0	3.51	2.48	4.83	21.56
Asterias rubens	1.78	0	3.49	6.92	4.79	26.35
Corallinaceae	0	1.73	3.42	7.64	4.7	31.05
Henricia sp.	1.6	0	3.14	11.01	4.32	35.37
Kirchenpaueria pinnata	1.5	0	2.93	1.79	4.03	39.4
<i>Macropodia</i> sp.	0	1.41	2.79	7.64	3.84	43.24
Pomatoschistus pictus	0	1.41	2.79	7.64	3.84	47.08
Cancer pagurus	1.32	0	2.55	1.84	3.5	50.58
Myoxocephalus scorpius	1.3	0	2.55	2.63	3.5	54.08
Solaster endeca	0.14	1.41	2.53	2.64	3.48	57.56

Groupsh & g

Average dissimilarity = 59.35

species	Group h	Group g				
	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Antedon bifida	0	2.24	4.84	16.19	8.15	8.15
Echinus esculentus	1.92	0	4.16	4.4	7.01	15.16
Aequipecten opercularis	0	1.41	3.06	16.19	5.16	20.32
<i>Macropodia</i> sp.	0	1.41	3.06	16.19	5.16	25.48
Pomatoschistus pictus	0	1.41	3.06	16.19	5.16	30.63
Solaster endeca	0	1.41	3.06	16.19	5.16	35.79
Asterias rubens	1.25	0	2.67	1.75	4.5	40.29
Porania pulvillus	1.13	0	2.44	1.77	4.12	44.41
Buccinum undatum	0.28	1.41	2.41	1.78	4.07	48.48
Suberites ficus	0	1	2.16	16.19	3.65	52.12
Pomatoceros spp.	0.95	0	2	1.62	3.36	55.48

Ophiocomina nigra	0.83	0	1.82	1.04	3.06	58.55	
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Groups a & g

Average dissimilarity = 77.96

	Group a	Group g				
species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Antedon bifida	0	2.24	4.7	11.2	6.03	6.03
Ophiopholis albida	0	2	4.21	11.2	5.4	11.43
Pagurus bernhadus	1.98	0	4.21	3.75	5.4	16.83
Echinus esculentus	1.73	0	3.64	11.2	4.67	21.5
Ophiothrix fragilis	0	1.73	3.64	11.2	4.67	26.18
Hydractinia echinata	1.5	0	3.22	1.82	4.13	30.31
Asterias rubens	1.41	0	2.98	11.2	3.82	34.13
Bryozoan sp.	0	1.41	2.98	11.2	3.82	37.95
Carcinus maenas	1.41	0	2.98	11.2	3.82	41.76
Crossaster papposus	0	1.41	2.98	11.2	3.82	45.58
Liocarcinus depurator	1.41	0	2.98	11.2	3.82	49.4
Macropodia sp.	0	1.41	2.98	11.2	3.82	53.21
Pecten maximus	1.41	0	2.98	11.2	3.82	57.03