Title	Impact of kelp cultivation on the Ecological Status of benthic habitats and Zostera marina seagrass biomass
Author(s)	Walls, Aimee W.; Kennedy, Robert; Edwards, Maeve D.; Johnson, Mark P.
Publication Date	2017-07-25
Publication Information	Walls, A. M., Kennedy, R., Edwards, M. D., & Johnson, M. P. (2017). Impact of kelp cultivation on the Ecological Status of benthic habitats and Zostera marina seagrass biomass. Marine Pollution Bulletin, 123(1), 19-27. doi: https://doi.org/10.1016/j.marpolbul.2017.07.048
Publisher	Elsevier
Link to publisher's version	https://doi.org/10.1016/j.marpolbul.2017.07.048
Item record	http://hdl.handle.net/10379/7493
DOI	http://dx.doi.org/10.1016/j.marpolbul.2017.07.048

Impact of kelp cultivation on the Ecological Status of benthic habitats and *Zostera marina* seagrass biomass

Walls, A.M., [a*]Kennedy, R.,[b] Edwards, M.D.,[a] Johnson, M.P.[a]

[a] Irish Seaweed Research Group, Ryan Institute, National University of Ireland, Galway, Ireland.

[b] Marine Ecosystem Research Laboratory, Zoology, Ryan Institute, National University of Ireland, Galway, Ireland

[*]Corresponding author: email: a.walls2@nuigalway.ie

Abstract

The Ecological Status of subtidal benthic communities within a commercial kelp farm on the SW coast of Ireland was not impacted by macroalgal cultivation. Additionally, there was no effect on the biomass of *Zostera marina*, a key habitat under the EU Habitats Directive and OSPAR Commission. However, sediment grain size and total organic matter were influenced by abiotic and biotic aspects of the farm. A temporal effect on univariate and multivariate species data, Infaunal Quality Index (IQI) and *Z. marina* biomass was observed. This effect was likely a community response to high storm disturbance in winter 2013/14.

The use of IQI to assess the impact of macroalgal cultivation on benthic communities is a novel approach. This study supports a view that environmental impacts of macroalgal cultivation are relatively benign compared to other forms of aquaculture. Further research must be conducted to understand all interactions between aquaculture activities and the environment.

Keywords

Kelp cultivation; Ecological impact; Macrobenthic community composition; Ecological Status assessment; Infaunal Quality Index (IQI); *Zoster marina* biomass;

Introduction

World aquaculture production continues to grow year on year with approximately 131.4 million tonnes of fish, aquatic animals and plants produced in 2014 (FAO, 2016). It has long been established that cultivation methods can impact on the benthic environment; these impacts include organic loading of the sediments and associated biogeochemical changes caused by the bio-deposition of faeces and pseudofaeces at culture sites (Crawford et al., 2003; Forde et al., 2015; Kalantzi and Karakassis, 2006; O'Carroll et al., 2016). However, many of these studies have focused on finfish (Kalantzi and Karakassis, 2006; Silvert and Sowles, 1996) and shellfish (Crawford et al., 2003; Dubois et al., 2007; O'Carroll et al., 2016; Stenton-Dozey et al., 1999) aquaculture. Assessments of the impacts of macroalgal cultivation has so far focused on tropical macroalgal species (Eklöf et al., 2005; Johnstone and Olafsson, 1995; Ólafsson et al., 1995) or their impact when combined with shellfish cultivation in integrated multi-trophic aquaculture (IMTA) systems (Ning et al., 2016; Ren et al., 2014; Zhang et al., 2009).

Seaweed aquaculture farms are generally situated in nearshore coastal environments with average water depth ranging from 6 – 20 m. Semi-exposed sites with good current flow and shelter from the open ocean are ideal to provide the nutrients required for biomass growth without damage of the crop and infrastructure during storms. Typical farm set-up consists of a header ropes suspended approx. 1 m below the surface by buoys and kept in position by anchor ropes and weights, vertical ropes called dropper ropes (approx. 3 m in length) are sometimes added to increase the surface area of the farm (Edwards and Watson, 2011; Peteiro et al., 2016; Walls et al., 2017, 2016). Seaweed cultivation is an extractive cultivation method meaning it assimilates nutrients required for growth from the environment with no need for the addition of supplementary feed or nutrients (Chopin and Sawhney, 2009). As a consequence seaweed farms are assumed to have a more benign impact on the benthos when compared to finfish or shellfish aquaculture (Roberts and Upham, 2012; Soto, 2009). However, possible impacts include organic enrichment from loss of kelp biomass to the seabed and surrounding environment (Zhang et al., 2012 and discussed in more detail below) and from faeces and pseudofaeces released from fouling organisms (e.g. bivalves, polychaetes and amphipods) which use kelp as a habitat (Walls et al., 2017, 2016). In addition, the infrastructure of the farm and the biomass could have baffling effects and possible wave attenuation altering local hydrodynamics similar to that of wild kelp forests (Lovas and Torum, 2001; Mork, 1996; Rosman et al., 2007).

Over 33% of the 27.3 million tonnes of global annual aquatic plant production in 2014 came from just 2 kelp species *Laminaria japonica* and *Undaria pinnatifida* (FAO, 2016). Kelps are among the largest sources of primary productivity in marine habitats (Mann, 1973; Reed et al., 2008) and this primary productivity enters the food chain through two routes; direct grazing on kelp tissue or detrital pathways. Much of the standing stock in temperate kelp beds is released either as particulate organic matter (POM) also called detritus or as dissolved organic matter (DOM). Krumhansl and Scheibling (2012) estimate that > 80% of kelp production enters the carbon cycle as POM or DOM. Kelp detritus can range in size from small particles to whole thalli depending on how the biomass was removed. There are three main ways tissue can be lost. 1-Whole thalli are removed from breakage at the stipe or when the holdfast becomes detached from its substratum, either rocks or boulders in wild kelp forests or suspended rope substratum at cultivated sites. 2- Parts of the frond can break off removing large pieces from the frond. 3- Erosion of the distal ends of fronds can occur as tissue is continually lost through decay and natural senescence (Krumhansl and Scheibling, 2012; Zhang et al., 2012). The impacts of detrital deposition from macroalgal cultivation on the benthos could be analogous to the impacts caused by the bio-deposition of faeces and pseudofaeces from finfish and shellfish aquaculture on benthic communities.

Over the last few years, interest in kelp cultivation in Europe has increased, supported by feasibility studies (e.g. Bruton et al. 2009) and experimental farms which are being set up to begin to industrialise the industry and advance the cultivation of kelps native to this region This interest includes Ireland, with the establishment of Dingle Bay Seaweed in Ventry Harbour, County Kerry in 2011 as one of the larger commercial kelp farms (18 ha) in Europe (M.D. Edwards pers. comm.). With an increase in demand for kelp biomass to supply traditional (e.g. food) and expanding uses (e.g. biofuels) of kelp (Guiry, 1989; Walls et al., 2016), the industry is set to expand and investigation into the possible impacts of this cultivation method on the local environment is essential.

Aims of this study

The aim of this study was to assess any potential impacts on infaunal community structure at a commercial macroalgal farm at Ventry Harbour, County Kerry on the south west coast of Ireland over a 2-year period. This was conducted by using an asymmetrical before after control impact (BACI) design to test for differences between control and impact stations in terms of univariate and multivariate faunal distributions and biotic indices including Infaunal Quality Index (IQI). IQI has been used to successfully discriminate the responses of macrobenthic communities to a wide range of natural and anthropogenic environmental impacts including aquaculture, in both coastal and transitional waters. However, many of the studies investigating aquaculture impacts using AMBI (part of IQI) based indicators have only focused on finfish and shellfish aquaculture and not macroalgal cultivation. Additionally, we assessed particle grain size and total organic matter to investigate if the kelp farm had an impact on sediment characteristics. Lastly, the farm site in Ventry Harbour is located above a *Zostera marina* seagrass bed, which is recognised as an important habitat under the EU habitats Directive and as a threatened or declining habitat under OSPAR (OSPAR Commission, 2008). Given the importance of *Zostera* habitats we conducted analyses to test the trends of *Z. marina* biomass at our impacted and control sites over the duration of the study.

Materials and Methods

Study Site

This study was conducted in the south-west coast of Ireland in Ventry Harbour, County Kerry (52° 06' 49.45” N, -10° 21'20.17” W; Fig. 1) at the largest operating commercial seaweed farm in Ireland (18 ha site). Ventry Harbour is a moderately sheltered and shallow embayment orientated towards the south-east, approximately 2.5 x 1.5 km (3.75 km^2) with a wide mouth opening into Dingle Bay. *Zostera marina* (seagrass) is extensively distributed throughout the sandy seabed, leading to a rocky boulder reef towards the mouth of the bay. The licensed seaweed farm is orientated north-west to south-east, and located to the westerly side of Ventry Harbour (Fig. 1). The depth underneath the farm is approximately 6 m at the north-western end before gently sloping to 20 m at the eastern edge of the farm at mean low water spring tide (MLWS). The tidal range in Ventry Harbour is between 0.6 and 4.0 m. Irradiance values obtained from nearby Valentia weather observatory (51° 56' 23” N, -10° 14' 40” W) ranged from 5,447 to 63,823 J cm^{-2} (mean daily maximum value per month) for 2014. Sea surface temperature data was obtained from the M3 offshore weather buoy located approximately 56 km southwest of Mizen head (51° 13' 0” N, -10° 33' 0” W), and ranged from 10.1 to 17.6 ºC for 2014. Although offshore values are less extreme than inshore values, Ventry Harbour is a well flushed bay so values are broadly representative. The longline structure is similar to the set-up in Walls et al (2016) their Fig. 3); however, the farm in Ventry consists of 3 parallel units of 280 m linear longlines suspended approximately 1.5 m below the sea surface, and the dropper ropes used in this study were 1 m in length. The longlines were kept in position by buoys attached to the header rope and by 1500 kg anchor blocks at either end of the lines. The farm cultivates the kelps *Alaria esculenta* and *Saccharina latissima* for human consumption, animal feed and use in cosmetic products.

Sampling Design

In this study an asymmetrical distribution of control versus impact stations was used in a BACI (before after control impact) experiment *sensu* (Underwood, 1994). Sampling was conducted during 6 dates over a 2-year period between 2014 and 2016 (including May ‘14 and ‘15, September ‘14 and ‘15 and February ‘15 and ’16). Sampling in September was before any impact from deployment of seaweed lines and February and May were after impact i.e. seaweed was deployed and growing at the farm. 4 sites were sampled during each sampling date; 1 treatment site corresponded to potential impacts associated with cultivation activities underneath the farm (designated the Impacted Treatment, 52° 06' 54.418”N, -10° 21'23.724”W) and 3 treatment sites located outside of the farms footprint and not subject to any known anthropogenic activity were selected as control sites (designated as Control Treatment 1; 52° 06' 56.459”N, -10° 21'27.719”W, Control Treatment 2; 52° 06' 59.46”N, -10° 21'25.499”W and Control Treatment 3; 52° 06' 59.339”N, -10° 21'21.24”W) (Fig.1). The sites were selected as homogeneous patches of sandy sediment, at a depth of 7 to 10 m, with *Zostera marina* distributed throughout all sites.

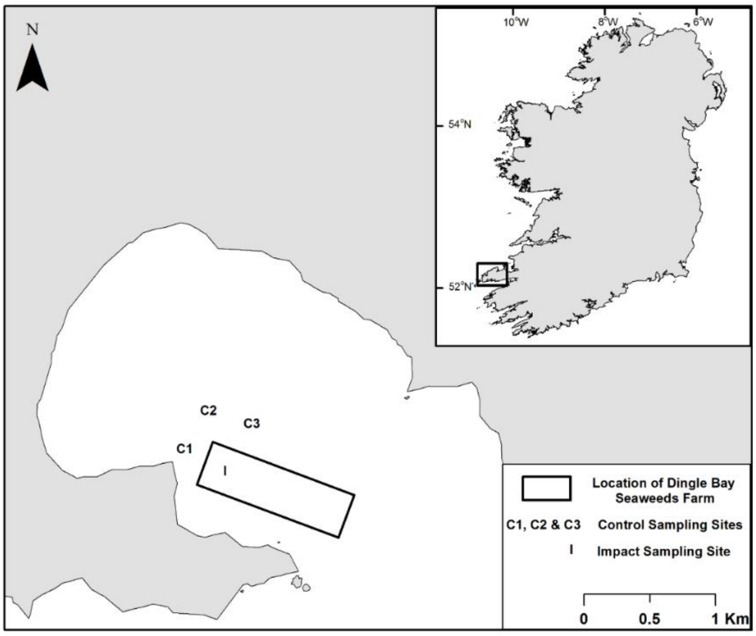

Fig. 1. Dingle Bay Seaweeds farm and sampling sites at Ventry Harbour, County Kerry, Ireland. I = Impacted Treatment Site; C1 = Control Treatment Site 1; C2 = Control Treatment Site 2; C3 = Control Treatment Site 3.

Within each treatment site a 20 x 20 m area was identified and 5 cores were sampled using a 0.01 m^2 diameter corer modified for use by SCUBA divers. The sampling locations were chosen using predetermined random numbers within each site. Once all 5 samples were collected the cores were raised to the boat and the samples were transferred to labelled buckets. One additional sediment sample was collected for sediment analysis in a labelled Ziploc® bag at each of the 4 treatment sites. The samples were transferred back to the laboratory in cooler boxes and sediment samples were frozen at -20 °C pending analyses. Faunal core samples were sieved through a 500 μm mesh and any retained material was stored separately in labelled buckets and fixed in buffered formalin for 48 h pending laboratory analyses.

Sample Processing

In the laboratory, formalin fixed macrofaunal samples were washed in running freshwater over a 500 μm sieve to remove formalin and excess sediment and the retained material was stored in 70% ethanol. The macrofauna were stained using Eosin-Biebrich scarlet dye, sorted, enumerated and identified to species level using standard keys. Fauna were checked for nomenclatural inconsistencies and synonyms using online Taxon Match tool in the World Register of Marine Species (WoRMS Editorial Board, 2016; http://www.marinespecies.org/aphia.php?p=match)

Sediment samples from each sampling site were removed from the freezer and allowed to defrost. Sediment granulometry was determined for each sample using laser particle sizing (LPS). It was not necessary to carry out wet-dry sieving as none of the sediment samples were seen to contain particles > 2 mm upon visual inspection. For each sample, 3 replicate aliquots of material were added to the Hydro-G dispersion unit of a Malvern Mastersizer 2000 until obscuration reached between 15% and 18% (Forde et al., 2012). For each aliquot the measurement cycle was 5 x 30,000 scans. The LPS distribution data was expressed as percentage weight within full Phi classes ranging between 4 and -2 Phi units. These data were processed using GRADISTAT (Blott and Pye, 2001) software to derive sediment type classification, distribution modality and sediment particle graphic mean (Mz; Folk & Ward 1957). Mz is a parameter used to describe the mean particle size of a distribution and is analogous to the graphic mean employed with the normal distribution in conventional statistics (Forde et al., 2012). Here, Mz values can be used with confidence as 95% of sediment samples exhibited unimodal distributions.

Sediment samples from each sampling station were dried to constant weight at 100 ^{o}C. For each dried sample, total organic material (TOM) was measured by loss of ignition (LOI). LOI allows TOM to be calculated through the combustion of 5 g of sediment in a furnace at 450 ^{o}C after 6 h (McIntyre and Eleftheriou, 2005). Any organic material will have been oxidised within this time. TOM values were determined by expressing as a percentage the sediment weight loss following combustion over the initial weight of the dried sediment (Dean, 1974) .

The ash-free dry weight (AFDW) of *Zostera marina* within each core sample was determined. *Z. marina* was removed from the core samples and added to a pre-weighed labelled tin container. The samples were dried in the oven at 105 ^{o}C for 24 h (or until constant weight was attained). Each dried sample was then added to pre-weighed labelled crucibles and combusted in a muffle furnace at 450 ^{o}C for 16 h to oxidise all organic material. Samples were left to cool and were weighed again. AFDW was calculated as dry weight minus the weight of the inorganic material which remained after combustion. Note there were no samples for all treatments in May 2014 and only 4 replicates for February 2015 Control 3 treatment.

Data Analysis

Statistical analyses was carried out using the PRIMER v6 (Clarke and Gorley, 2006) with PERMANOVA+ add-on (Anderson et al., 2008) packages and MINITAB v 17.

One-way similarity percentages (SIMPER) analysis in PRIMER v6 (Clarke, 1993) was used to determine the major taxa characterising the replicate cores within each treatment for each sampling date. The analysis was carried out on square-root transformed faunal abundance data. The SIMPER routine listed taxa characterising the faunal community within each treatment area in decreasing order of contribution to similarity between replicate cores (n = 5) (Clarke and Gorley, 2006).

Using untransformed faunal data, the DIVERSE routine in PRIMER was used to calculate a range of diversity measures for each replicate faunal core. The diversity measures calculated included the total number of taxa (*S*), total number of individuals (*N*), Shannon diversity index (H' (Log_e) and Simpson's evenness diversity index (1 – λ'). Replicate core diversity measures were used to calculate mean values for each treatment area. Changes in faunal distributions in relation to natural or anthropogenic disturbances can be effectively assessed using multiple metrics to describe different aspects of community structure (Borja et al., 2011; Reiss and Kröncke, 2005).

A multidimensional scaling (MDS) (Kruskal, 1964a, 1964b; Shepard, 1962) ordination was carried out using PRIMER v6, giving the position of each core sample in 2-dimensional spaces based on Bray-Curtis similarity matrix (Bray and Curtis, 1957) of square-root transformed species composition data.

PERMDISP routine was conducted to observe if any significant variations in multivariate dispersion (around the centroid) were present among treatments. Permutational analysis of variance (PERMANOVA (Anderson et al., 2008)) was used to analyse community structure. An asymmetrical BACI design was employed (Underwood, 1994, 1991). The fixed factor treatment had 2 levels: control (C) and impact (I). The fixed factor before/after had 2 levels: before (B) and after (A). The random factor sampling date was nested within before/after and had 6 levels: May-14, September-14, February-15, May-15, September-15 and February-16. The PERMANOVA design was applied to zero adjusted Bray-Curtis similarity matrix (Bray and Curtis, 1957) using square-root transformed faunal abundance data. P-values were determined by 9999 permutations of raw data.

To test the effect of factors treatment, before-after and sampling date on total organic carbon levels an analysis of covariance (ANCOVA) was run using MINITAB v17 to assess if an interaction is present. TOM correlates with sediment particle size with fine-grained sediments typically containing higher levels of organic matter than coarse sediments. To overcome any potential confounding effects of variation in particle size distributions between samples Mz was added as a covariate.

Infaunal Quality Index (IQI) was calculated for each faunal core using IQI version 4 (freely available at: http://www.wfduk.org/resources%20/coastal-and-transitional-waters-benthic-invertebrate-fauna) of the propriety tool in Microsoft Excel developed by the UK Environment Agency (Phillips et al., 2014). The IQI calculation involves truncation of the species list, spelling and synonym standardisation. The IQI EQR, a continuous variable between 0 and 1, is calculated by Eq. (1).

$$\mathrm{IQI} = \frac{\left(\left(0.38\times\frac{(1\text{-AMBI}/7)}{\left((1\text{-AMBI}/7)_{\mathrm{ref}}\right)}\right)+\left(0.08\times\left(\frac{(1\text{-}\lambda')}{(1\text{-}\lambda')_{\mathrm{ref}}}\right)\right)+\left(0.54\times\left(\frac{S^{0.1}}{S^{0.1}{}_{\mathrm{ref}}}\right)\right)\text{-}0.4\right)}{0.6} \qquad (1)$$

Where:

- AMBI is the AZTI Marine Biotic Index (Borja et al., 2000),
- 1-λ’ is Simpson’s Evenness Index,
- $S^{0.1}$ is number of taxa (S) raised to the power of 0.1,
- Ref. parameters are the maximum reference values for the habitat

The IQI tool sets reference conditions for each component which must be described regarding the physicochemical and hydromorphological quality elements of the metric based on local environmental parameters including sediment particle distribution and salinity classification. Environmental parameters can be entered into the tool as continuous or categorical variables. For each core, salinity classification was entered as “coastal” while sediment parameters were entered as wt% values across full Phi classes ranging between 4 and -2 units. Within the IQI tool AMBI value is a continuous variable based on the proportions of five ecological groups to which the species are allocated depending on their tolerance to disturbance (Borja et al., 2000; Muxika et al., 2007). Group allocation per animal is based on extensive literature describing North Atlantic species in relation to disturbance and expert knowledge (Teixeira et al., 2010). Based on AMBI index values benthic communities are classified as undisturbed, slightly disturbed, moderately disturbed, heavily disturbed or extremely disturbed. Other metrics used in the calculation of IQI include Simpson's evenness diversity index ($1 - \lambda'$) and the number of invertebrate taxa (S). IQI EQR values are converted to ES classes using the following class boundary values; Good–High, 0.75; Moderate–Good, 0.64; Poor–Moderate, 0.44; Bad–Poor, 0.24. These indices have been developed in response to the European Water Framework Directive (WFD; (Directive, 2000)). The WFD provides a framework for the improvement and protection of inland ground and surface waters as well as transitional coastal waters within all EU member states. The final objective was for all water bodies to achieve at least a good Ecological Status (ES) by 2015 (now extended to 2025). Under the Water Framework Directive (WFD) model management of ES focuses on the "Moderate-Good" critical boundary with remedial management actions required to restore areas classified as Moderate (or worse) to Good (or better).

A general linear model was run using MINITAB v17 on the IQI EQR values to assess if an interaction exists between the fixed factors treatment (I & C), before/after (B & A) and the random factor sampling date nested within before-after.

To test for the effect of factors treatment, before-after and sampling date on *Zostera marina* biomass an analysis of variance (ANOVA) general linear model was run using MINITAB v17 on the ash-free dry weight of *Z. marina* data to assess if an interaction is present.

Results

Community characteristics

A total of 131 benthic invertebrate species were sampled during this survey from May 2014 to February 2016. See Appendix 1 for full list of species sampled and their abundances. In general, the benthic communities present at each treatment area were comprised of polychaetes, amphipods, bivalves and gastropods. The biotope that the communities present conform to is a sublittoral seagrass *Zostera marina* dominated community on medium to fine sediment (SS.SMP.SSgr) (Connor et al., 2004). These communities are generally found in shallow sublittoral sediments, depth range 0 – 10 m, in sheltered to extremely sheltered embayments, marine inlets, estuaries and lagoons, with very weak tidal currents and variable salinity. While commonly found on mud and muddy sands this biotope may also occur in coarser sediments, in particular marine examples of *Zostera* communities similar to the communities present at our site (Connor et al., 2004).

The characterising species of each site remained largely similar over sampling dates. The polychaete *Chaetozone gibber, Owenia fusiformis,* nematodes and the bivalves *Tellina tenuis* and *Kurtiella bidentata* were generally the species contributing the highest percentage to group similarity within treatment sites. SIMPER analysis output of the taxa characterising the communities at each treatment site during each sampling date are included in Appendix 2. Average between group dissimilarity across all sampling dates for impact vs. control (1, 2 & 3) sites was 43.64% (± 1.73) and within control (1, 2 & 3) sites was 42.68% (± 1.47).

Mean species richness and mean number of individuals follows a pattern of low values in May 2014 which steadily increase over the duration of the study for all treatment sites, with February 2016 having the highest diversity and abundance. However, mean Simpson's evenness index and the mean Shannon diversity index was consistent across sampling dates (Table 1). This biotope is very spatially and temporally variable which is also reflected in the multivariate results below.

Table 1: Summary of mean community diversity, sediment characteristics and *Zostera marina* biomass at treatment sites (Impacted, Control 1, 2 & 3) sampled over 6 sampling dates at Ventry Harbour, County Kerry. Community characteristics include: S: total number of taxa; N: total number of individuals; H' Loge: Shannon diversity index; 1– λ': Simpson's evenness index. Mean values are based on replicate cores (n = 5) recovered within each Treatment site (standard error of means are included in parenthesis)

Sediment characteristics include; TOM%: total organic matter; Mz: graphic mean. Only one sample was taken per site for sediment and organic content analysis (n = 1). * No data for February 2016 Control 2

Zostera marina biomass data includes Ash-free dry weight (g) **No data for May 2014 all treatments; *** Only 4 replicates for February 2015 Control 3

Sampling Date	**Treatment**	**Community characteristics**				**Sediment characteristics**		***Zostera marina* biomass**
		S	**N**	**H' (Log_e)**	**1– λ'**	**TOM %**	**Mz**	**Ash-free dry weight**
May 2014	Impacted	19.8 (1.59)	44.6 (3.54)	2.65 (0.11)	0.92 (0.02)	3.22	1.85	**
	Control 1	16.8 (2.01)	43.8 (3.31)	2.36 (0.22)	0.85 (0.06)	3.49	2.14	**
	Control 2	19.6 (1.25)	59.2 (6.22)	2.53 (0.11)	0.89 (0.03)	3.34	2.04	**
	Control 3	23.6 (1.69)	72.4 (8.33)	2.76 (0.05)	0.92 (0.01)	3.28	1.72	**
September 2014	Impacted	30.8 (2.82)	155.8 (21.98)	2.80 (0.07)	0.91 (0.01)	2.76	1.80	0.36 (0.10)
	Control 1	24.2 (0.86)	99 (3.62)	2.57 (0.05)	0.88 (0.01)	3.27	2.23	0.28 (0.02)
	Control 2	27.8 (1.85)	94.6 (16.48)	2.81 (0.09)	0.91 (0.02)	3.2	1.83	0.24 (0.03)
	Control 3	28.8 (1.28)	93.6 (5.35)	2.85 (0.11)	0.91 (0.02)	3.18	1.74	0.22 (0.02)
February 2015	Impacted	23.6 (1.72)	94.8 (16.33)	2.41 (0.11)	0.84 (0.02)	2.56	1.56	0.30 (0.09)
	Control 1	20.8 (1.39)	102.2 (18.06)	2.28 (0.11)	0.83 (0.02)	2.81	2.58	0.15 (0.07)
	Control 2	29.2 (2.52)	170.4 (33.08)	2.63 (0.09)	0.88 (0.01)	3.18	1.95	0.39 (0.06)
	Control 3	25.6 (3.71)	143 (29.57)	2.63 (0.07)	0.91 (0.01)	3.26	1.87	0.59 (0.15) ***
May 2015	Impacted	29.4 (2.04)	170.6 (14.31)	2.4 (0.07)	0.83 (0.02)	2.93	1.82	0.96 (0.16)
	Control 1	27.2 (3.06)	190.6 (32.96)	2.25 (0.17)	0.8 (0.06)	3.47	2.64	0.52 (0.21)
	Control 2	29.6 (2.29)	185.4 (11.79)	2.6 (0.08)	0.88 (0.01)	3.42	2.18	0.60 (0.12)
	Control 3	24 (1.55)	133.4 (16.96)	2.54 (0.07)	0.89 (0.01)	3.09	1.77	0.50 (0.06)
September 2015	Impacted	37.6 (2.32)	253 (55.55)	2.88 (0.04)	0.92 (0.01)	2.59	1.63	0.54 (0.03)
	Control 1	33.2 (3.06)	198.8 (26.95)	2.69 (0.14)	0.89 (0.02)	3.94	2.77	0.44 (0.04)
	Control 2	32.8 (2.52)	196 (17.46)	2.74 (0.13)	0.9 (0.02)	3.10	2.01	0.36 (0.04)
	Control 3	31.2 (1.66)	152.6 (18.07)	2.71 (0.1)	0.89 (0.02)	2.68	1.59	0.28 (0.06)
February 2016	Impacted	30.6 (2.18)	164.2 (21.97)	2.7 (0.08)	0.89 (0.01)	2.70	1.76	0.54 (0.09)
	Control 1	33.4 (2.27)	220.0 (25.4)	2.53 (0.13)	0.85 (0.03)	*	*	0.52 (0.07)
	Control 2	29.6 (1.57)	176.8 (24.16)	2.58 (0.05)	0.87 (0.01)	2.91	2.24	0.39 (0.03)
	Control 3	31.2 (2.2)	143.4 (6.85)	2.8 (0.09)	0.91 (0.01)	2.66	2.71	0.44 (0.04)

Effect of treatment on community structure

Some separation and clustering of benthic invertebrates can be seen between sampling dates and before/after factor in the MDS plot (Fig. 2), however little separation can be seen between treatments (I & C). A stress value of 0.24 indicates the data are only partially represented by the 2-dimensional plot and little reliance should be placed on the finer detail of the plot (Clarke and Warwick, 2001). However, the broad scale pattern shows little separation between treatments and some separation of May 2014, September 2014 and February 2015 and clustering of the remaining sampling dates May 2015, September 2015 and February 2016.

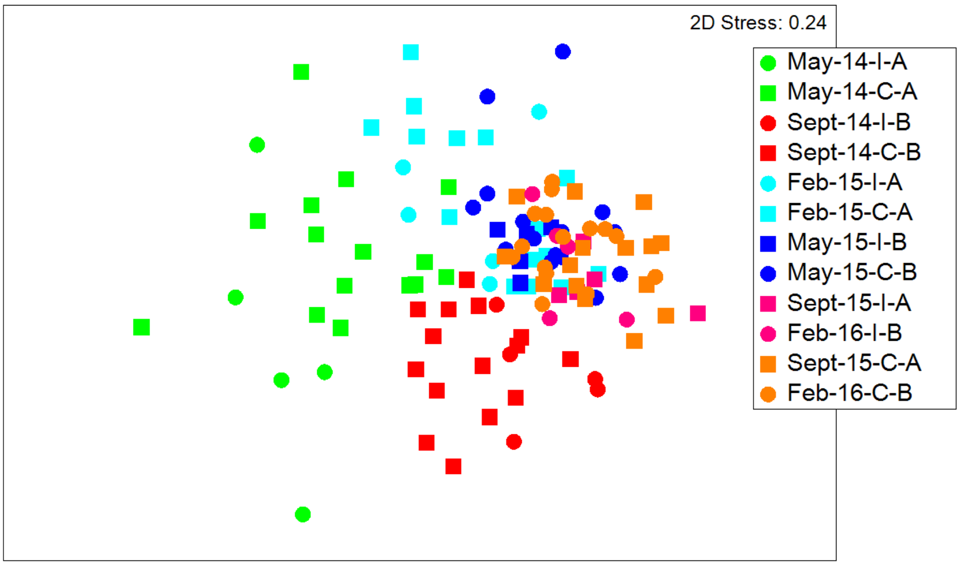

Fig. 2. 2-dimensional multidimensional scaling (MDS) plot of 120 core samples: n = 5 cores from each treatment site (I = Impacted- circles and C= Control 1- squares, A= After and B= Before) during each sampling month (May 2014- Green, September 2014- Red, February 2015- Light Blue, May 2015- Dark Blue, September 2015- Pink, February 2016- Orange), based on square-root of species abundance Bray Curtis similarity matrix of species sampled from each core (stress = 0.24)

PERMDISP routine revealed that the variation in multivariate dispersion (around the centroid) was significant ($P < 0.01$) for all factors (Sampling date $P = 0.001$; Treatment $P = 0.014$; and Sampling Date*Treatment $P = 0.002$). From the PERMDISP output significant result for sampling date can be attributed to higher dispersion early in sampling dates (May 2014), which decreases as sampling dates continue to lowest dispersion values for February 2016. The significant results for differences between treatment seem to be random and are not consistent with the impact versus control results. All PERMDISP mean and standard error values for within group dispersion can be found in Appendix 3.

Results from the PERMANOVA analysis on community structure are shown in Table 2. P (perm) results showed no significant difference on community structure between control and impact treatment sites. Pairwise test show that within and between group similarity between Impact and Control treatments are very similar (I-I: 51.26%; C-C: 52.59% and I-C: 51.3%). There was no significant effect of time before or after seaweed farming activities. Sampling date nested within before/after factor was significant ($P < 0.05$). Pairwise tests indicate that within and between sampling date before and after factors were always significant. Average similarity between- and within-groups increases with time, e.g. within group similarity for May-14 was 47% whereas for February-16 within-group similarity was 64.5%. This result is visible in the MDS (Fig. 2.) and the diversity and abundance univariate data in Table 1 and confirms a change in species composition over time.

Table 2: Permutational multivariate analyses of variance based on Bray Curtis similarity matrix based on square-root transformed abundance data for benthic invertebrates sampled during 6 sampling dates at 2 treatments. All tests were conducted using unrestricted permutation of raw data with 9999 permutations. *df*: *d*egrees of freedom; SS: sum of squares; MS: mean squares; F: ratio of within-group variation to between-group variation; P (perm): permutational probability value, * $p < 0.05$

Source	***df***	**SS**	**MS**	**Pseudo-F**	**P (perm)**	**Unique Perms**
Treatment	1	2391.4	2391.4	2.160	0.067	9926
Before/After	1	5514.6	5514.6	0.982	0.334	15
Sampling Date (Before/After)	4	22470	5617.5	6.118	< 0.001*	9842
Treatment x Before/After	1	1873	1873	1.691	0.135	9935
Treatment x Sampling Date (Before/After)	4	4429.4	1107.3	1.206	0.126	9826
Residuals	108	99159	918.14			
Total	119	1.4269 E5				

Effect of treatment on sediment characteristics

Sediment TOM values at the treatment sites were very similar ranging between 2.56% and 3.94% (Table 1). Similarly, sediment Mz (Phi) values at the treatment sites did not vary widely, ranging between a minimum of 1.56 and maximum of 2.77. Results from the analysis of covariance (ANCOVA) general linear model with Mz as a covariate indicated that TOM was significantly affected by treatment. However, before/after, sampling date nested within before/after and treatment interactions were not significant. From analysis of the data, TOM% values for impact treatment are consistently lower or equated to the lowest value in the range of values for the control treatment. The impacted site also has low Mz values, this does not conform with the typical pattern observed in the literature that fine-grained sediments contain higher levels of organic matter than coarse sediments. Samples from the control treatment did adhere to the correlation of TOM% with particle size, e.g. samples with higher Mz had lower TOM% values. These differences between treatment sites are reflected in the ANCOVA (Table 3).

Table 3. ANCOVA of TOM% with treatment, before/after and sampling date factors and Mz as a covariate. *df*: *d*egrees of freedom; SS: sum of squares; MS: mean squares; F: ratio of within-group variation to between-group variation; * p < 0.05. Note there was no data from February 2016 control 2 treatment.

Source	***df***	**SS**	**MS**	**F**	**p**
Treatment	1	0.1.9790	0.197899	5.32	0.043*
Before/After	1	0.00003	0.000034	0.00	0.989
Sampling Date(Before/After)	4	0.67359	0.168398	6.52	0.053
Treatment*Before/After	1	0.10205	0.102048	4.09	0.125
Treatment*Sampling Date (Before/After)	4	0.10510	0.026276	0.35	0.841
Mz	1	0.32190	0.321897	4.23	0.067
Error	11	0.76095	0.076095		
Total	22	2.62226			

Effect of treatment on IQI ES classification

All sites had an Ecological Status of either 'good' or 'high' from the IQI results, the Ecological Status and mean IQI EQR values for each site are given in Appendix 4. Using ANOVA general linear model, we tested the effect of treatment, before/after and sampling date on IQI EQR values. Sampling date nested within before/after had a significant effect on IQI EQR values (Table 4). Analysis of the mean IQI EQR values suggest a general trend of increasing values as the study continued, e.g. IQI EQR values at start of the study in May 2014 for Impacted site was 0.713 and Control sites 1,2 & 3 were 0.69-0.722 and at end of the study in February 2016 for Impacted site was 0.755 and Control sites 1,2 & 3 were 0.75-0.803. This long-term pattern has been seen above in our species diversity and abundances (Table 1) and the multivariate PERMANOVA results (Table 2).

Table 4. General linear model of IQI EQR values with treatment, before/after and sampling date factors. *df*: *d*egrees of freedom; SS: sum of squares; MS: mean squares; F: ratio of within-group variation to between-group variation; P (perm): permutational probability value, * p < 0.05

Source	***df***	**SS**	**MS**	**F**	**p**
Treatment	1	0.000001	0.000001	0.00	0.978
Before/After	1	0.003481	0.003481	0.30	0.613
Sampling Date (Before/After)	4	0.046337	0.011584	17.00	0.009*
Treatment*Before/After	1	0.000516	0.000516	0.76	0.433
Treatment*Sampling Date (Before/After)	4	0.002725	0.000681	0.82	0.515
Error	108	0.089712	0.000831		
Total	119	0.171754			

Effect of treatment on Zostera marina *biomass*

Observations of the *Z. marina* ash-free dry weight (AFDW) data show a slight increase with sampling data with a peak in May 2014 at the impacted site (Fig. 3). Generally, impact and control sites weight are quite similar. Results from the analysis of variance (ANOVA) general linear model indicate no significant effect of treatment, before/after or sampling date on *Z. marina* AFDW samples (Table 5).

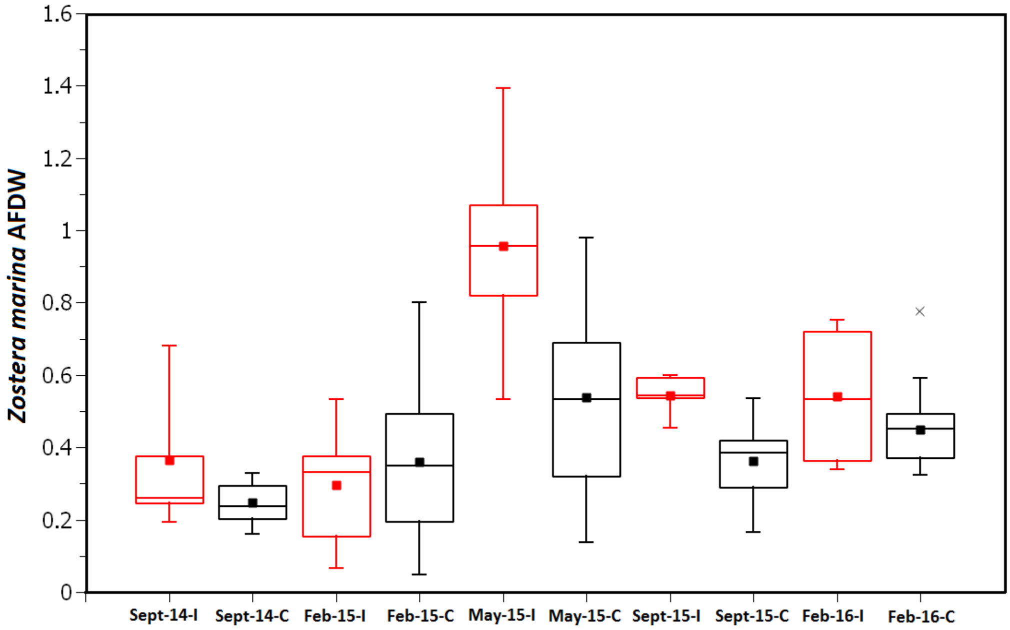

Fig. 3. Box plot showing ash free dry weight (AFDW) of *Zostera marina* at impacted (I = red) and control (C = black) treatment sites over sampling dates. The horizontal line and solid square within each box mark the median and means of the data, respectively. The boxes encompass 50% of the data, and whiskers show the range between the 5 % (bottom) and 95 % (top) portions of the data. Outliers are represented by crosses (*n*=5 samples for the impacted sites and *n*=15 samples for the control site; note *n*=14 for February)

Table 5. General linear model of *Zostera marina* ash free dry weight with treatment, before/after and sampling date factors. *df*: *d*egrees of freedom; SS: sum of squares; MS: mean squares; F: ratio of within-group variation to between-group variation; P (perm): permutational probability value, * $p < 0.05$
Note there was no data for May 2014 sampling date and only 14 replicates for February 2015 control treatment.

Source	***df***	**SS**	**MS**	**F**	**p**
Treatment	1	0.00280	0.00280	1.98	0.254
Before/After	1	0.00928	0.00928	2.55	0.208
Sampling Date (Before/After)	3	0.01091	0.00364	2.57	0.229
Treatment*Before/After	1	0.00002	0.00002	0.01	0.911
Treatment*Sampling Date (Before/After)	3	0.00424	0.00141	0.83	0.507
Error	10	0.01703	0.00170		
Total	19	0.05025			

Discussion

Given the predicted increase in demand for kelp biomass and the expansion of the industry to novel waters in Europe and Ireland, there is significant potential for environmental impacts. Although, macroalgal cultivation is an extractive form of aquaculture and not a fed form such as finfish aquaculture, it still provides a source of organic matter to the marine environment through the release of particulate organic matter (POM) detritus (Zhang et al., 2009). By increasing the natural levels of organic material to benthic habitats, kelp aquaculture could cause significant organic enrichment of benthic habitats and changes in macrofaunal assemblages.

In the current study, we observed a general trend of long-term change evident from our univariate and multivariate results attributed to sampling date and not any impact versus control treatment effect. Species richness and diversity increased temporally, however Shannon diversity and Simpson’s evenness indices remained relatively constant. Thus, the increase in species and abundance was not due to influx of opportunistic or stress tolerant species as described by Pearson and Rosenberg’s (1978) succession model which outlines how benthic infaunal community structure changes along a gradient of increasing organic enrichment and oxygen depletion. The main characterising species were similar for each site and no highly opportunistic stress tolerant species were sampled (SIMPER; Appendix 2). The polychaete *Owenia fusiformis* contributed a high percentage to overall similarity within treatment sites and is a disturbance-sensitive species from the AMBI classification species list (Borja et al., 2000). This temporal shift in community structure is also obvious from our multivariate results including the MDS ordination plot (Fig. 2) and PERMDISP results (Appendix 3). The higher dispersion rates early in sampling dates (May-14, September-14 and February-15) are confirmed by the spread of the same sampling dates from the MDS with a clustering of samples and a decrease in dispersion for later sampling dates (May-15, September-15 and February-16). Also, the PERMANOVA returned a significant result for sampling date nested within before/after (Table 2), with within-group similarity increasing temporally for sampling dates.

This temporal shift in community structure can be explained through analysis of the properties of the biotope and the biotic environment. The biotope present at all our treatment sites is a sublittoral *Zostera marina* dominated community (Connor et al., 2004). This biotope is highly variable both spatially and temporally (Davidson and Hughes, 1998; Unsworth et al., 2014). The use of an asymmetrical BACI (before after control impact) design (Underwood, 1994, 1991) here was essential and provided a robust design with multiple controls to handle the inherent variability within sites. In addition to being highly variable this *Zostera* biotope is also very sensitive to disturbance, in particular storm disturbance (Davidson and Hughes, 1998). During the period of December 2013 to February 2014, Ireland and the UK were subject to a number of winter storms as a consequence of low pressure, tidal surges and record wave heights. These events resulted in considerable damage to coastal infrastructure, caused persistent flooding and significant erosion events (Kendon and McCarthy, 2015). Our study site at Ventry Harbour was also subject to these storms with large (> 1 tonne) blocks pulled from the harbour wall during this period (pers. obs.). Storms and high wave activity have been observed to remove large amounts of *Zostera marina* (Den Hartog, 1987; Olesen and Sandjensen, 1994; Orth and Moore, 1983). Sediments and infaunal communities are likely to be affected by this disturbance as *Zostera* rhizomes and root networks bind together the substratum reducing erosion and allowing oxygen to penetrate the sediment (Davidson and Hughes, 1998; Herkül and Kotta, 2009). It is likely that the temporal changes we observed were a response to the severe storm activity experienced at this site. Again, the use of the asymmetrical BACI design allowed us to account for the natural variability and other studies should employ this design when conducting environmental impact studies of this nature.

Zostera beds are important habitats as they provide ecosystem services such as substratum stabilisation, shelter and substrate for associated organisms, nursery grounds for fish, and are hugely productive (Bertelli and Unsworth, 2014; Davidson and Hughes, 1998; Herkül and Kotta, 2009; OSPAR Commission, 2008). As a result of the supply these important ecosystem services Z*ostera marina* beds are recognised as a characteristic component of five Annex I habitats in the EU habitats Directive (92/43/EEC). Additionally in 2004, OSPAR produced descriptions of habitats on the Initial List of OSPAR Threatened and/or Declining Species and Habitats, which outlined 14 habitat types considered to be a cause for concern and included *Zostera* seagrass beds (OSPAR Commission, 2008). Therefore, the evaluation of the impact of a macroalgal cultivation site positioned above a *Zostera* bed was important. We observed a slight trend in the data of increasing *Z. marina* biomass temporally which was evident in both the impact and control treatment sites which again could be in response to the winter storms on 2013/14, as mentioned above. However, despite this trend and the larger weights recorded for May 2015 at the impacted site, none of the factors tested in this study influenced *Z. marina* ash-free dry weight values. Numerous factors are likely to affect the degree of sensitivity of *Zostera* habitats to physical disturbances such as storms. Rasheed et al (2014) found that seagrass at deeper depths recovered quicker than shallower species. Although the Rasheed et al (2014) study was conducted on tropical species it is possible that the depth of our *Z. marina* beds (7-10 m) lessened the degree of disturbance experienced at the site and increased the rate of recovery.

In the current study, significant effects of kelp aquaculture were detected on sediment total organic matter (TOM) across treatment sites. Also, observations of the TOM% and Mz values show that values were lower at the impacted site, this pattern is opposite to the pattern typically observed in the literature that fine-grained sediments contain higher levels of organic matter than coarse sediments. Potential baffling effects of the seaweed farm could be the cause of the small particle size of the sediments at the impacted site due to larger particles being inhibited from settling. Little is known about the baffling effects of cultivated seaweed structures; the role of wild kelp forests in coastal protection has been investigated (Firth et al., 2016). Kelp forests protect coastlines and adjacent sedimentary habitats by attenuating wave energy, buffering against storm surges, and preventing the movement of sediment from adjacent beaches (Lovas and Torum, 2001; Mork, 1996; Rosman et al., 2007). Baffling from the cultivated kelp may cause large particles of sediment and detritus to be exported away from the farm allowing only the settlement of smaller particles underneath the farm. Another explanation for the low TOM% at the impact site could be the consumption of detritus by fouling organisms attached to the kelp which already acts as a habitat for these species (Walls et al., 2017, 2016). Stable isotope studies have shown the importance of kelp detritus in marine food webs (Fredriksen, 2003; Leclerc et al., 2013; Schaal et al., 2012). Fouling organisms such as filter-feeding bivalve molluscs and deposit feeding polychaetes may consume detritus at the farm site and thus less organic matter is deposited to the seabed. However, there are many factors that need to be considered, such as quality of organic matter, alternative food sources, organism selectivity and bio-deposition of faeces and pseudofaeces before this hypothesis can be properly tested. Few studies have assessed TOM% underneath macroalgal farm, yet, a preliminary environmental study at a 21 ha pilot farm site of *Macrocystis pyrifera*, in Chile indicated organic matter under the culture site did not show significant trends of increase over time (Buschmann et al., 2014).

Infaunal Quality Index (IQI) was identified as an appropriate tool, as like most Water Framework Directive (WFD) benthic multimetrics, it has been developed in subtidal systems as a means of assessing, and comparing anthropogenic impacts across a range of sediment types and in different locations (Borja et al., 2009). Ecological Quality Ratio (EQR) values have been shown to be suitable monitoring tools within highly variable sedimentary habitats (Forde et al., 2015). The IQI classification of the sites in this study revealed that the ES of all our treatment sites were either 'good' or 'high' status. A significant effect of sampling date nested within before/after on EQR values was identified. Detailed analysis of the EQR values revealed that the ecological classification of sites was improving as the study continued. This pattern parallels with the temporal change we identified for the univariate and multivariate species data, which reflects the macrofaunal data which is incorporated in the calculation of IQI EQR values. The significant result from the ANOVA detected the long-term temporal response to the storm disturbance during winter 2013/14.

Conclusions

From this primary study, we found that the impact of macroalgal kelp cultivation on the benthic environment studied over a 2-year period was minimal. The farm created a baffling effect within the water column which was detected by a response of sediment TOM% and Mz. Additionally, the wider ecosystem services of the farm i.e. the provision of food to attached fauna, could be an explanation for lower TOM% values at the impacted site. If this process is found to occur and cultivated kelp detritus is incorporated into the food web, then kelp farms provide additional ecosystem benefits beyond the supply of commercial crop. However, the quality of the organic material supplied needs to be measured (e.g. sediment traps) and the selectivity and preference of the fouling organisms for kelp detritus needs to be understood.

An intriguing result which was not anticipated, was the influence of disturbance from storm activity in winter 2013/14 (Kendon and McCarthy, 2015) which was detected on benthic communities, the Ecological Status and *Zostera marina* biomass. We do not know if the detected change is a return to pre-disturbance levels or if it is to a new altered state. An understanding of the functioning of the benthic environment pre-disturbance events (storm and aquaculture) would be optimal, but, in a dynamic and spatially and temporally variable environment this would require an extensive time series to be established.

This is the one of the first studies to assess the impacts on kelp cultivation on the benthic environment. The data collected suggest that seaweed farms may be a benign form of aquaculture and have little impact of the local environment. Further research is required over larger spatial scales, varying locations and longer temporal scales to properly understand the interactions between seaweed farms and the benthos.

With demand for cultivated kelp set to continue and a focus on government bodies and stakeholders to develop the industry, the licensing of new sites for seaweed aquaculture is ongoing. The apparent resilience of *Zostera* beds to severe storm disturbance and the minimal impact of kelp farming identified in this study may indicate that subtidal seagrass habitats situated in semi-sheltered embayments represent favourable habitats for the positioning of new kelp farms. This point should be taken with caution as research into this area is only beginning and other characteristics of *Zostera* beds such as shoot and root density, reproductive rates and epiphytes may be impacted and need to be studied.

Acknowledgements

The authors acknowledge Mr. David Moran and Mr. Brendan Walls for their assistance in field sampling. In addition, we thank Dr. Adrian Patterson and Mr. Jack O'Carroll from NUI Galway's Benthic Ecology Laboratory for taxonomic support, Mr. Michael Murphy and Mr. Paul Flannery of Dingle Bay Seaweed for boat work and assistance at Ventry Harbour. This work was supported by the Energetic Algae project (EU Interreg IVB NWE Strategic Initiative; www.enalgae.eu). A.M.W. is currently funded by the Dr. Tony Ryan Research Trust, NUI Galway.

References

Anderson, M.J., Gorley, R.N., Clarke, K.R., 2008. PERMANOVA+ for PRIMER: guide to software and statistical methods., PRIMER-E, Plymouth, UK.

Bertelli, C.M., Unsworth, R.K.F., 2014. Protecting the hand that feeds us: Seagrass (*Zostera marina*) serves as commercial juvenile fish habitat. Mar. Pollut. Bull. 83, 425–429. doi:10.1016/j.marpolbul.2013.08.011

Blott, S.J., Pye, K., 2001. Gradistat: a grain size distribution and statistics package for the analysis of unconsolidated sediments. Earth Surf. Process. Landforms 26, 1237–1248. doi:10.1002/esp.261

Borja, A., Barbone, E., Basset, A., Borgersen, G., Brkljacic, M., Elliott, M., Garmendia, J.M., Marques, J.C., Mazik, K., Muxika, I., Neto, J.M., Norling, K., Rodríguez, J.G., Rosati, I., Rygg, B., Teixeira, H., Trayanova, A., 2011. Response of single benthic metrics and multi-metric methods to anthropogenic pressure gradients, in five distinct European coastal and transitional ecosystems. Mar. Pollut. Bull. 62, 499–513. doi:10.1016/j.marpolbul.2010.12.009

Borja, A., Franco, J., Pérez, V., 2000. A marine biotic index to establish the ecological quality of soft-bottom benthos within European estuarine and coastal environments. Mar. Pollut. Bull. 40, 1100–1114. doi:10.1016/S0025-326X(00)00061-8

Borja, Á., Rodríguez, J.G., Black, K., Bodoy, A., Emblow, C., Fernandes, T.F., Forte, J., Karakassis, I., Muxika, I., Nickell, T.D., Papageorgiou, N., Pranovi, F., Sevastou, K., Tomassetti, P., Angel, D., 2009. Assessing the suitability of a range of benthic indices in the evaluation of environmental impact of fin and shellfish aquaculture located in sites across Europe. Aquaculture 293, 231–240. doi:10.1016/j.aquaculture.2009.04.037

Bray, J.R., Curtis, J.T., 1957. An ordination of the upland forest communities of southern Wisconsin. Ecol. Monogr. 27, 325–349. doi:10.2307/1942268

Bruton, T., Lyons, H., Lerat, Y., Stanley, M., Rasmussen, M.B., 2009. A review of the potential of marine algae as a source of biofuel in Ireland, Sustainable Energy Ireland Dublin. Dublin, Ireland.

Buschmann, A.H., Prescott, S., Potin, P., Faugeron, S., Vásquez, J.A., Carolina, C., Infante, J., Hernández-González, M.C., Gutíerrez, A., A.Varela, D., 2014. The status of kelp exploitation and marine agronomy, with emphasis on *Macrocystis pyrifera*, in Chile, in: Advances in Botanical Research. pp. 161–188. doi:10.1016/j.jpsychires.2009.09.011

Chopin, T., Sawhney, M., 2009. Seaweeds and their Mariculture, in: Encyclopedia of Ocean Sciences. Elsevier, pp. 317–326. doi:10.1016/B978-012374473-9.00757-8

Clarke, K.R., 1993. Non-parametric multivariate analyses of changes in community structure. Aust. J. Ecol. 18, 117–143.

Clarke, K.R., Gorley, R.N., 2006. PRIMER v6: User manual/Tutorial. Primer-E, Plymouth.

Clarke, K.R., Warwick, R.M., 2001. Change in Marine Communities: An approach to statistical analysis and interpretation, PRIMER-E Ltd. Plymouth.

Connor, D.W., Allen, J.H., Golding, N., Howell, K.L., Lieberknecht, L.M., Northen, K.O., Reker, J.B., 2004. The Marine Habitat Classification for Britain and Ireland Version 04.05, JNCC. Peterborough (ISBN 1 861 07561 8).

Crawford, C.M., Macleod, C.K.A., Mitchell, I.M., 2003. Effects of shellfish farming on the benthic environment. Aquaculture 224, 117–140. doi:10.1016/S0044-8486(03)00210-2

Davidson, D.M., Hughes, D.J., 1998. Zostera Biotopes (volume I). An overview of dynamics and sensitivity characteristics for conservation management of marine SACs., Scottish Association for Marine Science.

Dean, W.E., 1974. Determination of carbonate and organic matter in calcareous sediments and sedimentary rocks by loss on ignition: comparison with other methods. J. Sediment. Petrol. 242–248.

Den Hartog, C., 1987. "Wasting disease" and other dynamic phenomena in *Zostera* beds. Aquat. Bot. 27, 3–14. doi:10.1016/0304-3770(87)90082-9

Directive, 2000. /60/EC of the European Parliament and of the Council of 23rd October 2000 Establishing a Framework for Community Action in the Field of Water Policy.

Dubois, S., Marin-Léal, J.C., Ropert, M., Lefebvre, S., 2007. Effects of oyster farming on macrofaunal assemblages associated with *Lanice conchilega* tubeworm populations: A trophic analysis using natural stable isotopes. Aquaculture 271, 336–349. doi:10.1016/j.aquaculture.2007.03.023

Edwards, M., Watson, L., 2011. Cultivating *Laminaria digitata* Aquaculture Explained No. 26 Bord Iscaigh Mhara. Dublin.

Eklöf, J.S., de la Torre Castro, M., Adelsköld, L., Jiddawi, N.S., Kautsky, N., 2005. Differences in macrofaunal and seagrass assemblages in seagrass beds with and without seaweed farms. Estuar. Coast. Shelf Sci. 63, 385–396.

FAO, 2016. The State of World Fisheries and Aquaculture 2016. ROME.

Firth, L.B., Knights, A.M., Bridger, D., Evans, A.J., Mieszkowska, N., Moore, P.J., O 'connor, N.E., Sheehan, E.V., Thompson, R.C., Hawkins, S.J., 2016. Ocean Sprawl: Challenges and opportunities for biodiversity

management in a changing world. Oceanogr. Mar. Biol. An Annu. Rev. 54, 189–262.
Folk, R.R.L., Ward, W.W.C., 1957. Brazos River Bar: A study in the significance of grain size parameters. SEPM J. Sediment. Res. doi:10.1306/74D70646-2B21-11D7-8648000102C1865D
Forde, J., Collins, P.C., Patterson, A., Kennedy, R., 2012. Comparison of granulometric methods and sampling strategies used in marine habitat classification and Ecological Status assessment. Mar. Pollut. Bull. 64, 1018–1028. doi:10.1016/j.marpolbul.2012.01.036
Forde, J., O'Beirn, F.X., O'Carroll, J.P.J., Patterson, A., Kennedy, R., 2015. Impact of intertidal oyster trestle cultivation on the Ecological Status of benthic habitats. Mar. Pollut. Bull. 95, 223–233. doi:10.1016/j.marpolbul.2015.04.013
Fredriksen, S., 2003. Food web studies in a Norwegian kelp forest based on stable isotope (d13C and d15N) analysis. Mar. Ecol. Prog. Ser. 260, 71–81.
Guiry, M.D., 1989. Uses and cultivation of seaweed, Camera di Commercio Industria Artigiantoe Agricoltura; Università Degli Studi.
Herkül, K., Kotta, J., 2009. Effects of eelgrass (*Zostera marina*) canopy removal and sediment addition on sediment characteristics and benthic communities in the Northern Baltic Sea. Mar. Ecol. 30, 74–82. doi:10.1111/j.1439-0485.2009.00307.x
Johnstone, R.W., Olafsson, E., 1995. Some environmental aspects of open water algal cultivation: Zanzibar, Tanzania. Ambio 24, 465–469.
Kalantzi, I., Karakassis, I., 2006. Benthic impacts of fish farming: Meta-analysis of community and geochemical data. Mar. Pollut. Bull. 52, 484–493. doi:10.1016/j.marpolbul.2005.09.034
Kendon, M., McCarthy, M., 2015. The UK's wet and stormy winter of 2013/2014. Weather 70, 40–47. doi:10.1002/wea.2474
Krumhansl, K., Scheibling, R., 2012. Production and fate of kelp detritus. Mar. Ecol. Prog. Ser. 467, 281–302. doi:10.3354/meps09940
Kruskal, J.B., 1964a. Multidimensional scaling by optimizing goodness-of-fit to a nonmetric hypothesis. Psychometrika 29, 115–129.
Kruskal, J.B., 1964b. Nonmetric multidimensional scaling: A numerical method. Psychometrika 29, 115–129. doi:10.1007/bf02289694
Leclerc, J., Riera, P., Leroux, C., Lévêque, L., Davoult, D., 2013. Temporal variation in organic matter supply in kelp forests: linking structure to trophic functioning. Mar. Ecol. Prog. Ser. 494, 87–105. doi:10.3354/meps10564
Lovas, S.M., Torum, A., 2001. Effect of the kelp *Laminaria hyperborea* upon sand dune erosion and water particle velocities. Coast. Eng. 44, 37–63. doi:10.1016/S0378-3839(01)00021-7
Mann, K.H., 1973. Seaweeds: Their productivity and strategy for growth. Science (80-.). 182, 975–981.
McIntyre, A.D., Eleftheriou, A., 2005. Methods for the study of marine benthos, third ed. ed. Blackwell scientific publications, Oxford, UK.
Mork, M., 1996. Wave attenuation due to bottom vegetation, in: Grue, J., Gjevik, B., Weber, J.E. (Eds.), Waves and Nonlinear Processes in Hydrodynamics. Kluwer Academic Publishing, Oslo, pp. 371–382.
Muxika, I., Borja, Á., Bald, J., 2007. Using historical data, expert judgement and multivariate analysis in assessing reference conditions and benthic ecological status, according to the European Water Framework Directive. Mar. Pollut. Bull. 55, 16–29. doi:10.1016/j.marpolbul.2006.05.025
Ning, Z., Liu, S., Zhang, G., Ning, X., Li, R., Jiang, Z., 2016. Impacts of an integrated multi-trophic aquaculture system on benthic nutrient fluxes: a case study in Sanggou Bay, China. Aquac. Environ. Interact. 8, 221–232. doi:10.3354/aei00144
O'Carroll, J.P.J., Quinn, C., Forde, J., Patterson, A., O'Beirn, F.X., Kennedy, R., 2016. Impact of prolonged storm activity on the Ecological Status of intertidal benthic habitats within oyster (*Crassostrea gigas*) trestle cultivation sites. Mar. Pollut. Bull. 110, 460–469. doi:10.1016/j.marpolbul.2016.06.014
Ólafsson, E., Johnstone, R., Ndaro, S., 1995. Effects of intensive seaweed farming on the meiobenthos in a tropical lagoon. J. Exp. Mar. Bio. Ecol. 191, 101–117.
Olesen, B., Sandjensen, K., 1994. Patch dynamics of eelgrass *Zostera marina*. Mar. Ecol. Prog. Ser. 106, 147–156. doi:10.3354/meps106147
Orth, R.J., Moore, K.A., 1983. Chesapeake bay: an unprecedented decline in submerged aquatic vegetation. Science (80-.). 222, 51–53. doi:10.1126/science.222.4619.51
OSPAR Commission, 2008. Case Reports for the OSPAR List of threatened and/or declining species and habitats.
Pearson, T.H., Rosenberg, R., 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. Oceanogr. Mar. Biol. Annu. Rev. 16, 229–311. doi:10.1111/j.1540-5834.2012.00707.x
Peteiro, C., Sánchez, N., Martínez, B., 2016. Mariculture of the Asian kelp *Undaria pinnatifida* and the native kelp *Saccharina latissima* along the Atlantic coast of Southern Europe: An overview. Algal Res. 15, 9–23.

doi:10.1016/j.algal.2016.01.012
Phillips, G.R., Anwar, A., Brooks, L., Martina, L.J., Miles, A.C., Prior, A., 2014. Infaunal quality index : Water Framework Directive classification scheme for marine benthic invertebrates. Environment Agency, United Kingdom.
Rasheed, M.A., McKenna, S.A., Carter, A.B., Coles, R.G., 2014. Contrasting recovery of shallow and deep water seagrass communities following climate associated losses in tropical north Queensland, Australia. Mar. Pollut. Bull. 83, 491–499. doi:10.1016/j.marpolbul.2014.02.013
Reed, D.C., Rassweiler, A., Arkema, K.K., 2008. Biomass rather than growth rate determines variation in net primary production by giant kelp. Ecology 89, 2493–2505.
Reiss, H., Kröncke, I., 2005. Seasonal variability of benthic indices: An approach to test the applicability of different indices for ecosystem quality assessment. Mar. Pollut. Bull. 50, 1490–1499. doi:10.1016/j.marpolbul.2005.06.017
Ren, L., Zhang, J., Fang, J., Tang, Q., Zhang, M., Du, M., 2014. Impact of shellfish biodeposits and rotten seaweed on the sediments of Ailian Bay, China. Aquac. Int. 22, 811–819. doi:10.1007/s10499-013-9709-7
Roberts, T., Upham, P., 2012. Prospects for the use of macro-algae for fuel in Ireland and the UK: An overview of marine management issues. Mar. Policy 36, 1047–1053. doi:10.1016/j.marpol.2012.03.001
Rosman, J.H., Koseff, J.R., Monismith, S.G., Grover, J., 2007. A field investigation into the effects of a kelp forest (*Macrocystis pyrifera*) on coastal hydrodynamics and transport. J. Geophys. Res. 112, C02016. doi:10.1029/2005JC003430
Schaal, G., Riera, P., Leroux, C., 2012. Food web structure within kelp holdfasts (*Laminaria*): a stable isotope study. J. Mar. Ecol. 33, 370–376.
Shepard, R., 1962. The analysis of proximities: Multidimensional scaling with an unknown distance function. II. Psychometrika 27, 219–246. doi:10.1007/bf02289621
Silvert, W., Sowles, J.W., 1996. Modelling environmental impacts of marine finfish aquaculture. J. Appl. Ichthyol. 12, 75–81. doi:10.1111/j.1439-0426.1996.tb00066.x
Soto, D., 2009. Integrated mariculture: A global review. FAO Fish. Aquac. Tech. Pap. FAO, Rome No. 529, 183.
Stenton-Dozey, J.M.E., Jackson, L.F., Busby, A.J., 1999. Impact of mussel culture on macrobenthic community structure in Saldanha Bay, South Africa, in: Marine Pollution Bulletin. pp. 357–366. doi:10.1016/S0025-326X(98)00180-5
Teixeira, H., Borja, Á., Weisberg, S.B., Ananda Ranasinghe, J., Cadien, D.B., Dauer, D.M., Dauvin, J.C., Degraer, S., Diaz, R.J., Grémare, A., Karakassis, I., Llansó, R.J., Lovell, L.L., Marques, J.C., Montagne, D.E., Occhipinti-Ambrogi, A., Rosenberg, R., Sardá, R., Schaffner, L.C., Velarde, R.G., 2010. Assessing coastal benthic macrofauna community condition using best professional judgement - Developing consensus across North America and Europe. Mar. Pollut. Bull. 60, 589–600. doi:10.1016/j.marpolbul.2009.11.005
Underwood, A.J., 1994. On beyond BACI: sampling designs that might reliably detect environmental disturbances. Ecol. Appl. 4, 3–15.
Underwood, A.J., 1991. Beyond baci: Experimental designs for detecting human environmental impacts on temporal variations in natural populations. Mar. Freshw. Res. 42, 569–587. doi:10.1071/MF9910569
Unsworth, R.K.F., van Keulen, M., Coles, R.G., 2014. Seagrass meadows in a globally changing environment. Mar. Pollut. Bull. 83, 383–386. doi:10.1016/j.marpolbul.2014.02.026
Walls, A.M., Edwards, M.D., Firth, L.B., Johnson, M.P., 2017. Successional changes of epibiont fouling communities of the cultivated kelp *Alaria esculenta*: predictability and influences. Aquac. Environ. Interact. 9, 55–69. doi:10.3354/aei00215
Walls, A.M., Kennedy, R., Fitzgerald, R.D., Blight, A.J., Johnson, M.P., Edwards, M.D., 2016. Potential novel habitat created by holdfasts from cultivated *Laminaria digitata* : assessing the macroinvertebrate assemblages. Aquac. Environ. Interact. 8, 157–169. doi:10.3354/aei00170
WoRMS Editorial Board, 2016. World register of marine species. [WWW Document]. URL www.marinespecies.org (accessed 7.17.16).
Zhang, J., Fang, J., Wang, W., Du, M., Gao, Y., Zhang, M., 2012. Growth and loss of mariculture kelp *Saccharina japonica* in Sungo Bay, China. J. Appl. Phycol. 24, 1209–1216. doi:10.1007/s10811-011-9762-4
Zhang, J., Hansen, P.K., Fang, J., Wang, W., Jiang, Z., 2009. Assessment of the local environmental impact of intensive marine shellfish and seaweed farming—Application of the MOM system in the Sungo Bay, China. Aquaculture 287, 304–310. doi:10.1016/j.aquaculture.2008.10.008