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Effects of coexistence between the blue mussel and eelgrass on sediment biogeochemistry and plant performance

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ABSTRACT: The habitat-modifying suspension-feeding mussel *Mytilus edulis* may have facilitating or inhibiting effects on seagrass meadows depending on the environmental conditions. We investigated the effects of *M. edulis* on sediment biogeochemistry in *Zostera marina* meadows under eutrophic conditions in Flensborg fjord, Denmark. Sediment and plant samples were collected at 5 stations with *Z. marina* (Eelgrass), 5 with *Z. marina* and *M. edulis* (Mixed), and at 2 unvegetated ones, 1 with mussels (Mussel) and 1 with sand (Sand). The Mixed sediment was enriched in fine particles (2 to 3 times), nutrients and sulphides compared to Eelgrass stations. Increased sediment nutrient availability at the Mixed stations was reflected in increased N and P content in eelgrass. However, the plant biomass did not differ significantly between stations, while shoot features (number of leaves and leaf area) were significantly reduced at Mixed stations, suggesting an inhibiting effect of *M. edulis* on *Z. marina*. Negative correlations between eelgrass measures and sediment sulphide at Mixed stations indicate that the presence of mussels increases sulphide invasion in the plants. A survey of 318 stations in Danish fjords suggests a threshold of 1.6 kg *M. edulis* m^{-2} beyond which no coexistence between *Z. marina* and *M. edulis* was found.

KEY WORDS: *Mytilus edulis* · *Zostera marina* · Coexistence · Eutrophication · Organic content · Total reducible sulphide · Seagrass · Suspension feeders

INTRODUCTION

Seagrass meadows are important as feeding and nursery grounds and as habitats for fishes, benthic organisms and birds, and they are vital for nutrient cycling and sediment stabilisation in the coastal zone (Boström & Bonsdorff 1997, Duarte et al. 2005, Heck & Valentine 2006). However, seagrass habitats are highly dynamic, and a dramatic decrease in the distribution of seagrass has been documented worldwide over the last centuries, mainly caused by human activities (Baden et al. 2003, Waycott et al. 2009). The decline has been attributed primarily to changes in abiotic factors, like increased nutrient load, turbidity and erosion (Orth et al. 2006, De Boer 2007). However, biotic disturbances such as overgrowth with epiphytes, excessive grazing and negative impacts from mussels may also affect seagrass abundance (Delgado et al. 1999, Holmer et al. 2008, Vinther et al. 2008).

Previous studies have indicated that the coexistence of habitat-modifying species such as mussels can generate either facilitating or inhibiting effects on seagrasses, depending on the status of the seagrass meadow, the abundance of mussels and environmental conditions (Reusch et al. 1994, Vinther et al. 2008, Wall et al. 2008).

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In coastal areas of the northern temperate zone, such as the Baltic Sea or the North American east coast, eelgrass *Zostera marina* coexists with blue mussels *Mytilus edulis* (Reusch 1998, Bologna et al. 2005). Both seagrasses and mussels act as 'ecosystem engineers', modifying their environment and thereby influencing other species (Jones et al. 1994). *Z. marina* beds are suitable habitats for *M. edulis*, providing substrate for settlement, shelter from predators and high sedimentation of particles (Reusch 1998, Gacia et al. 2002, Bologna et al. 2005). Being an efficient suspension feeder, *M. edulis* is among the most important regulators of suspended materials in the coastal zone, and is able to filter the water up to 1 m above the mussel bed (Prins et al. 1998, Dolmer 2000, Lassen et al. 2006). Furthermore, *M. edulis* excretes a large part of ingested material as faeces or pseudofaeces (Kautsky & Evans 1987, Hartstein & Rowden 2004), which can enhance nutrient availability in the sediments. Biodeposits generally consist of small particles, decreasing the mean grain size of the sediments by increasing the silt and mud fraction (Stoeck & Albers 2000). Biodeposits are high-quality organic matter, which are favourable substrates for sediment bacteria, and increased mineralisation and regeneration of nutrients have been found in mussel beds (Stoeck & Albers 2000). Biodepostion may thus facilitate the uptake of nutrients and stimulate growth of seagrasses through increased nutrient availability (Reusch et al. 1994, Peterson & Heck 2001a, Carroll et al. 2008). On the other hand, biodeposits from the mussels may negatively affect *Z. marina* by turning the sediments sulphidic due to enhanced sulphate reduction rates (Vinther et al. 2008). Several studies have found high sulphate reduction rates in sediments in mussel beds (Sorokin et al. 1999, Stenton-Dozey et al. 2001, Vinther et al. 2008), and as sulphide is toxic to plants, enhanced sulphide pools in the sediments may result in reduced photosynthetic activity and growth (Holmer & Bondgaard 2001), degeneration of meristems (Greve et al. 2003) and die-off of shoots in seagrass beds (Borum et al. 2005). Sulphide can invade from the sediment through the roots, and is indicated by decreasing values of stable sulphur isotopes ($\delta^{34}S$) and increasing total sulphur (TS) content in plant tissues (Frederiksen et al. 2006).

Since most previous studies on the effects of coexistence between mussels and seagrasses have been conducted under oligotrophic conditions, limited information is available from eutrophic environments, where there is increased risk of hypoxia and high pools of sediment sulphides. Furthermore, most studies have focused on effects on water column and plant nutrients (e.g. Wall et al. 2008), whereas impacts on sediment biogeochemistry and possible relationships with plant performance are less explored.

The present *in situ* study investigated whether *Mytulis edulis* affects sediment biogeochemistry in *Zostera marina* beds when the 2 species coexist under eutrophic conditions. The biogeochemical conditions in sediments were examined with particular focus on organic enrichment and sulphide pools. Possible relationships between sediment biogeochemical conditions and plant biomass, morphology, nutrient and TS content were explored through correlation analysis.

MATERIALS AND METHODS

Study site and field work

The study was conducted in Flensborg fjord, Denmark, a 48 km long fjord with an area of 308 km^2 and an average depth of 14.5 m. The fjord is divided into an inner and outer part, and consists of several basins with a water depth of 20 to 45 m with shallow areas in between, which limit the circulation of water (Laursen & Bruntse 2004). Flensborg fjord is eutrophic, receiving nitrogen and phosphorus from rivers and wastewater treatment plants in the area. In the outer part, the average summer chlorophyll concentration is 4 to 6 µg l^{-1}, and the average winter NO_3 concentrations are 100 to 150 µg l^{-1} (Laursen & Bruntse 2004). Sampling was conducted at 10 + 2 stations in the outer part of the fjord in April 2006 (Fig. 1): 5 stations with *Zostera marina* beds (Eelgrass) and 5 stations with coexisting *Z. marina* and *Mytilus edulis* (Mixed). In addition, we sampled 1 station with only mussels (Mussel) and 1 with only sand (Sand). The Mussel and Sand stations were included to measure sediment characteristics at stations unaffected by coexistence or by eelgrass. The coverage of *M. edulis* in the investigated area is highly variable (0 to 100%) and the Mixed stations were placed in areas with high coverage (90 to 100%) of both *M. edulis* and *Z. marina*, which corresponds to a mussel biomass of ~510 ± 60 g dry weight (DW) m^{-2} minus shells and an eelgrass biomass of ~550 ± 40 g DW m^{-2} (mean ± SE) (Vinther et al. 2008). Both species lived in close connection with each other, with *M. edulis* covering the sediment surface beneath the *Z. marina* shoots. The water depth at the stations ranged between 0.9 and 1.8 m. The salinity was 16 to 18‰, and water temperature was 6 to 8°C during sampling. Secchi depth in the study area is >2 m throughout the year according to data from the Danish Ministry of Environment, so

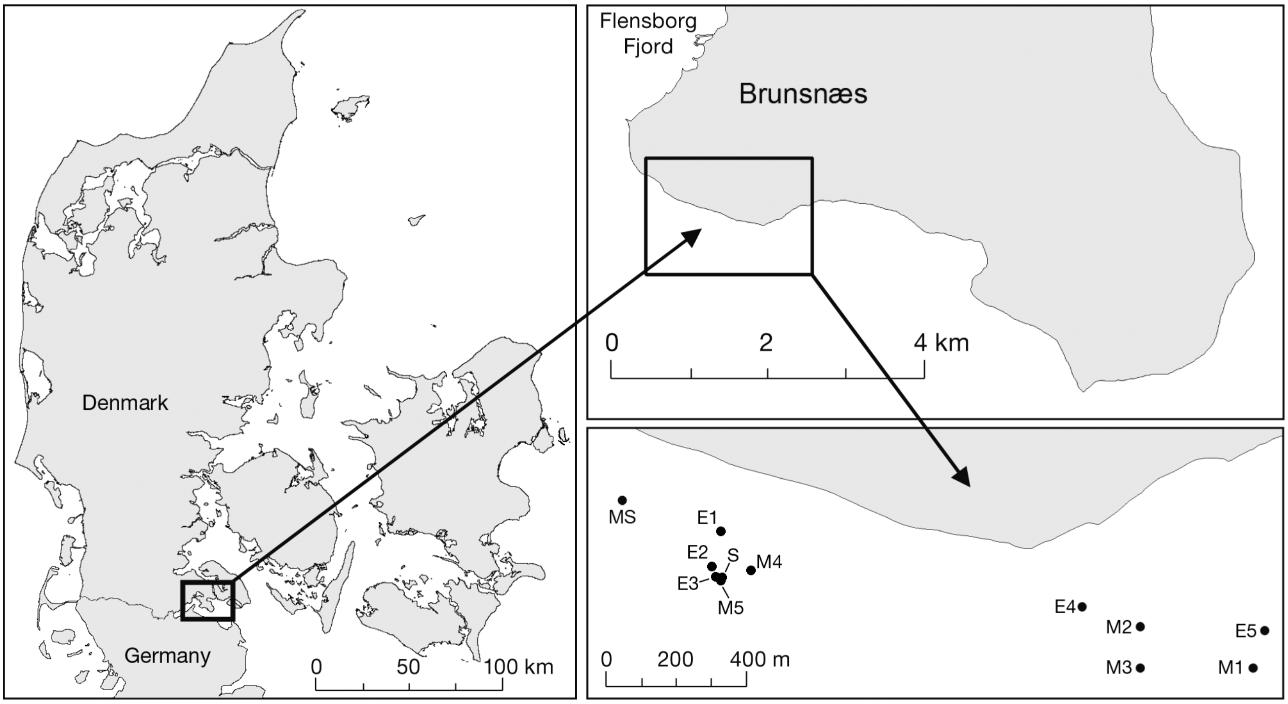

Fig. 1. Sampling stations in Flensborg fjord, Denmark. Stations: E1 to E5 = pure eelgrass, M1 to M5 = mixed, MS = pure mussel, and S = bare sediment (sand)

effects of light limitation on *Z. marina* were therefore not considered in this study.

Samples of sediment and plants were collected with cores (i.d. 2.6 cm, n = 3) by SCUBA diving. The upper 5 cm of each sediment core sample was homogenised and split into sub-samples for determination of organic content, grain size and total reducible sulphide (TRS) concentration. Sediment was stored frozen, and samples for TRS were preserved in 10 ml 1M ZnAC and stored frozen. Three cores (i.d. 8 cm) were sampled at each Eelgrass and Mixed station to determine biomass and seed density and were transferred to the laboratory and kept aerated for a maximum of 4 d until processing. Shoots were collected, and the upper 5 cm of the sediment was rinsed in demineralised water and sieved (500 µm) to determine the seed density and root and rhizome biomass. All plant parts were frozen for later analysis.

Finally, eelgrass shoots were collected during diving for determination of epiphyte load at 3 Eelgrass and 3 Mixed stations. Three samples containing at least 10 shoots were cut at the sediment surface with scissors and gently transferred to a plastic bag and stored at 5°C for maximum of 2 d before epiphytes were scraped off the leaves with a razor blade. Epiphyte biomass and leaves were frozen for later analysis.

Sediment analysis

Sediment samples were thawed and dried overnight at 105°C for determination of dry weight (DW). Loss on ignition (LOI) was determined by combustion (6 h at 520°C), and particulate organic carbon (POC) and nitrogen (PON) were analysed in dried sediment by elemental analysis using a Carlo Erba EA1108 elemental analyser. Total phosphorus (TP) in sediments was determined after boiling combusted sediment in 1 M HCl for 1 h followed by spectrophotometric measurements following the method of Koroleff (1983). TRSs in the sediment were determined according to the 2-step procedure of Fossing & Jørgensen (1989), where the first step extracts the acid volatile sulphides (AVS) consisting of FeS and porewater sulphides. The second step extracts chromium reducible sulphur (CRS) consisting of FeS_2 and S^0. The concentration of reduced inorganic sulphides was determined according to the method of Cline (1969), and TRSs were calculated by adding the AVS and CRS fractions.

Sediment grain size distribution was obtained by sieving wet sediment samples through a series of sieves (1000, 500, 250, 125 and 63 µm mesh size) and carefully transferring every fraction to preweighed aluminium trays. The water used during sieving was collected in a glass container and left overnight for sedimentation of fine particles (<63 µm). All fractions were dried for 24 h at 105°C to obtain DW.

Plant analysis

Eelgrass plants were thawed and divided into aboveground, rhizome and root biomasses and freeze dried for DW determination. The different plant fractions were homogenised and kept for analysis of TP, POC and PON, stable sulphur isotope ratio ($\delta^{34}S$) and TS content in each plant compartment. TP content was measured after acid digestion (1 M HCl for 30 min) of combusted samples as inorganic phosphate as described for sediments. POC and PON were measured by elemental analysis as described for the sediments. Epiphyte biomass was determined by drying the scraped material for 24 h at 105°C. Length and width of each leaf from the epiphyte scraped leaves were measured to determine the area.

For sulphur isotope analysis, 9 mg vanadium oxide were added to samples of dried plant tissue (5 mg) and packed in tin capsules and analysed at Isoanalytical Ltd. (UK). TS content was obtained during the analysis of $\delta^{34}S$. The sulphur isotopic composition of a sample is expressed in the standard δ notation given by $\delta^{34}S = [(R_{sample}/R_{standard}) - 1] \times 1000$, where $R = {}^{34}S/{}^{32}S$. Values are expressed in per thousand (‰) bases and are calibrated to Canyon Diablo Troilite (CDT). Precision of the analysis was better than 0.4‰ based on internal standards.

Survey of coexistence of eelgrass and blue mussels

A survey of eelgrass and blue mussel biomass was conducted at 318 stations (1994 to 2002) on the east coast of Denmark (see Fig. 5) at 3 to 6 m of water depth (Dolmer et al. 2009). At 3 to 4 m depth, samples were taken by a 0.1 m^2 van Veen grab, and at 5 to 6 m, samples were collected by a 1 m wide mussel dredge (Dolmer et al. 1998). The catch efficiency of the grab is assumed to be 100% for eelgrass and mussels. The catch efficiency of the dredge for blue mussels was corrected according to (Dolmer et al. 1998), and for eelgrass it was assumed to be 100%.

Statistical analysis

One-way analysis of variance (ANOVA) and Tukey test ($\alpha = 0.05$) were used to test for significant differences between Eelgrass and Mixed stations. Mussel and Sand stations were not included due to lack of replication (n = 1). Log transformations were used to normalise data when these were not normally distributed. When data did not meet the conditions for ANOVA, a non-parametric test was applied (Kruskal-Wallis 1-way ANOVA on rank). Biomass of plants, $\delta^{34}S$ and TS in plants, TP, PON and POC in plants and sediment together with LOI and grain size were tested for differences between Eelgrass and Mixed stations using ANOVA. Leaf number, leaf area, epiphyte loading, seed density and reducible sulphides (AVS, CRS and TRS) were tested using the Kruskal-Wallis test. Linear regressions were used to analyse for relationships between all measured parameters, and *t*-tests were used to test for differences in slopes. The software used was Sigma Stat®, version 2.03 SPSS Inc.

RESULTS

Sediment parameters

Visual inspection of the sediments revealed a clear difference in colouration between the Eelgrass and Mixed stations. Eelgrass sediments were light brown, whereas Mixed sediments were darkish and smelled of hydrogen sulphide during handling. The sediment at all stations was sandy with a dominating grain size fraction of 125 to 250 µm, which constituted from 79 to 89% of the total (Table 1). The fine particle fractions 63 to 125 µm and <63 µm (silt) were 2 and 3 times higher, respectively, at the Mixed compared to the Eelgrass stations (1-way ANOVA and Tukey test, $p < 0.05$). The Mussel station resembled the Mixed stations by its high fraction (9%) of fine particles (63 to 125 µm), while the Sand station was more similar to the Eelgrass stations. The LOI and contents of

Table 1. Grain size fraction in percent for the 4 types of stations (Eelgrass, Mixed, Sand and Mussel). Means ± SE, n = 5, except for Sand and Mussel, where values are an average of 3 subsamples taken at 1 station. *Significant differences between Eelgrass and Mixed stations ($p < 0.05$, 1-way analysis of variance)

Grain size fraction (µm)	Stations			
	Eelgrass	Mixed	Sand	Mussel
500–1000	0.33 ± 0.18	0.94 ± 0.59	0.47	0.39
250–500	6.06 ± 2.01	7.94 ± 5.50	2.86	1.94
125–250	89.40 ± 3.22	79.72 ± 5.49	92.52	86.83
63–125	3.45 ± 0.90	8.92 ± 2.02*	2.37	9.11
<63	0.76 ± .020	2.47 ± 0.42*	1.78	1.74

PON, POC and TP in the sediments were significantly higher (POC up to 270 %) at the Mixed stations compared to the Eelgrass stations (Fig. 2, 1-way ANOVA and Tukey test, $p < 0.05$). At the Mussel station, values of POC and TP were in the same range as at the Mixed stations, while PON and LOI were ~27 % lower. At the Sand station, values of LOI and TP were in the same range as at the Eelgrass stations, while PON was 66 % lower and POC was 350 % higher. The average AVS pool was 4 times higher, and the CRS and TRS (AVS+CRS) were up to 2 times higher at Mixed compared to Eelgrass stations (Fig. 3, 1-way ANOVA and Tukey test, $p < 0.05$). TRS was higher at the vegetated sites, as TRS at the Mussel station was 2.99 µmol cm^{-3} compared to 4.68 µmol cm^{-3} at the Mixed station, and at the Sand station TRS was 1.55 µmol cm^{-3} compared to 2.14 µmol cm^{-3} at the Eelgrass station.

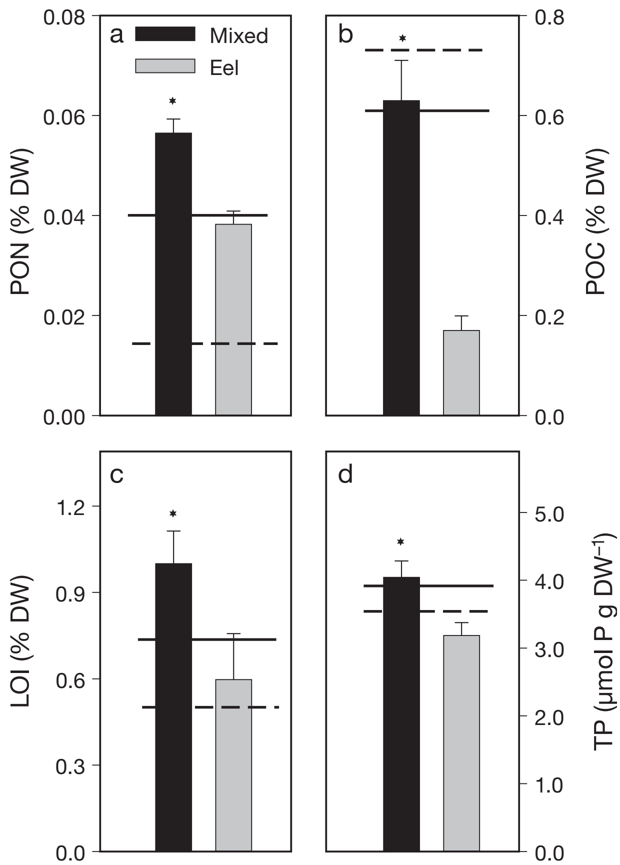

Fig. 2. (a) Particulate organic nitrogen (PON), (b) carbon (POC) content, (c) loss on ignition (LOI) and (d) total phosphorus (TP) content in the sediment of Eelgrass (Eel) and Mixed stations (mean ± SE, n = 5). *Significant differences between stations ($p < 0.05$, 1-way ANOVA). Values from the Sand and Mussel station are indicated by dashed (Sand) and solid (Mussel) lines (averages of 3 subsamples taken at each station)

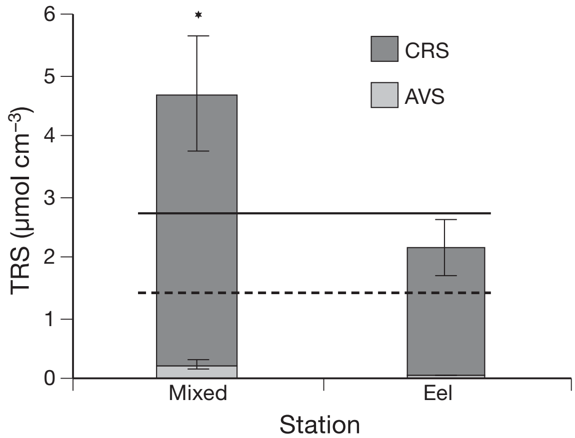

Fig. 3. Total reducible sulphide (TRS, µmol cm^{-3}) content, divided into the acid volatile sulphide (AVS) and chromium reducible sulphur (CRS) fraction, in the sediment for Eelgrass (Eel) and Mixed stations. *Significant differences between stations ($p < 0.05$, 1-way ANOVA). TRS values from the Sand and Mussel station are indicated by dashed (Sand) and solid (Mussel) lines (averages of 3 subsamples taken at each station)

Plant parameters

Plants at the Eelgrass stations had on average 8 % more leaves per shoots, and the surface area of the leaves was 19 % larger than at the Mixed station (1-way ANOVA and Tukey test, $p < 0.05$; Table 2). The above- and belowground biomass showed no differences between station types, and all stations had a higher below- than aboveground biomass (Table 2). During sampling, leaves were in some cases accidentally cut off during coring, and the aboveground biomass may therefore have been underestimated. There were no significant differences in epiphyte loading and seed density between Eelgrass and Mixed stations (Table 2).

The PON content in leaves, rhizomes and roots was significantly higher at Mixed compared to Eelgrass stations (Fig. 4, 1-way ANOVA and Tukey test, $p < 0.05$). The same was observed for TP except for rhizomes (Fig. 4). No differences were observed for POC content in plants between stations (data not shown).

The $\delta^{34}S$ of leaves, roots and rhizomes showed no differences between stations, while TS content showed a trend with higher values at the Mixed compared to the Eelgrass stations although the differences were not significant (Table 3). The $\delta^{34}S$ was highest in the leaves, and decreased in the roots and rhizomes, while the opposite was observed for the TS content.

Table 2. *Zostera marina*. Plant parameters for Eelgrass and Mixed stations. Mean ± SE (n = 5) except for leaf number and area, and epiphyte biomass (n = 3). *Significant differences between stations (p < 0.05). DW: dry weight

Station	Biomass (g DW m^{-2}) Aboveground	Rhizomes	Roots	Leaves (no. $shoot^{-1}$)	Leaf area (cm^2)	Epiphytes (mg cm^{-2})	Seed density (no. m^{-2})
Eelgrass	90.8 ± 16.3	118.8 ± 24.9	83.3 ± 14.6	5.6 ± 0.09	99.6 ± 5.2	0.10 ± 0.04	1207 ± 348
Mixed	96.4 ± 15.4	178.9 ± 51.8	88.1 ± 20.7	5.2 ± 0.09*	83.3 ± 4.3*	0.06 ± 0.02	1167 ± 402

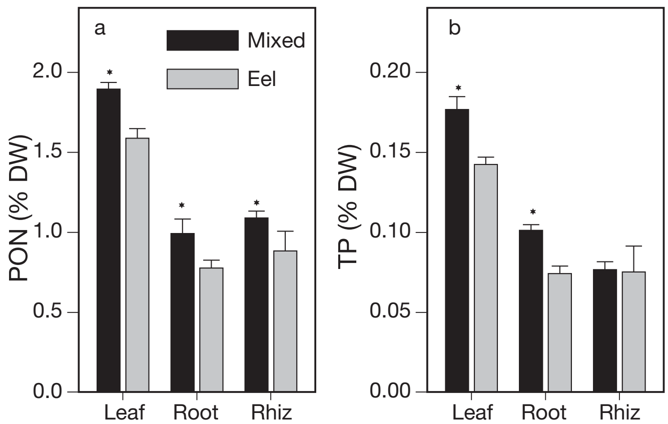

Fig. 4. *Zostera marina*. (a) Particulate organic nitrogen (PON) and (b) total phosphorus (TP) content in leaves (Leaf), roots (Root) and rhizomes (Rhiz) of plants from Eelgrass and Mixed stations. Data are mean ± SE, n = 5. *Significant differences between stations (p < 0.05, 1-way ANOVA)

Correlation between sediment biogeochemistry and plant parameters

At the Mixed stations, LOI showed a positive correlation with TP and TRS in sediment (Table 4, $p < 0.05$), but this was not observed for Eelgrass stations. At both station types, LOI correlated positively with PON, while a correlation between LOI and POC was only found at the Eelgrass stations (Table 4, $p < 0.001$). A negative correlation between LOI and TRS with leaf + root biomass was only found at the Mixed stations (Table 4, $p < 0.05$), and in addition, TRS in the sediment correlated positively with TS in plants (Table 4, $p < 0.05$). Other correlations between LOI and TRS and POC, PON, TP, leaf number and leaf area in plants were not significant for any of the station types.

Survey of coexistence of *Zostera marina* and *Mytilus edulis*

The survey of the biomass of *Z. marina* and *M. edulis* at 318 stations at 3 to 6 m depth showed no presence of *Z. marina* at an *M. edulis* biomass higher than 1.6 kg m^{-2} (Fig. 5). The maximum observed biomass of *Z. marina* at stations where both species coexisted was 4.0 kg m^{-2} with a corresponding *M. edulis* biomass of 1.5 kg m^{-2}.

DISCUSSION

Previous studies have indicated that mussels have facilitating effects on seagrasses (Reusch et al. 1994, Peterson & Heck 1999, 2001a,b), except when seagrasses grow under eutrophic conditions (Vinther et al. 2008, Table 5). The results from our study of seagrasses growing under eutrophic conditions show that the presence of mussels modifies the sediment biogeochemistry by enriching the sediments with nutrients. The sediments at Mixed stations were more fine-grained (Table 1) and had higher pools of nutrients (Fig. 2) and sulphides than the Eelgrass stations (Fig. 3). At the same time, negative correlations were found between sediment sulphide pools and plant biomass at the Mixed stations, as well as a positive relationship between sediment sulphide pools and sulphur

Table 3. *Zostera marina*. $\delta^{34}S$ (‰) and total sulphur (TS) in leaf, root and rhizome material from Eelgrass and Mixed stations. Means ± SE, n = 5. No significant differences were found between stations

Station	$\delta^{34}S$ (‰) Leaves	Roots	Rhizomes	TS Leaves	Roots	Rhizomes
Eelgrass	2.25 ± 1.06	−6.04 ± 1.09	−7.52 ± 0.61	111.9 ± 4.0	117.2 ± 4.7	182.3 ± 16.4
Mixed	1.88 ± 0.71	−6.01 ± 1.18	−6.12 ± 0.50	123.1 ± 14.7	135.7 ± 21.8	186.2 ± 26.9

Table 4. Linear regressions between sediment LOI (loss on ignition, organic content %) or total reducible sulphides (TRS; µmol cm^{-3}) and sediment or plant parameters at the Eelgrass and Mixed stations. Equations and values for R^2 and p are only given for significant regressions. TP: total phosphorus, PON: particulate organic nitrogen, POC: particulate organic carbon, TS: total sulphur, ns: not significant, DW: dry weight

Parameters	Station	Equation (*y*)	R^2	p
Sediment parameters — LOI (%)				
TP	Mixed – ns for Eelgrass	$1.864x + 2.11$	0.51	<0.05
PON	Mixed	$0.031x + 0.03$	0.59	<0.001
	Eelgrass	$0.015x + 0.03$	0.67	<0.001
POC	Eelgrass – ns for Mixed	$0.143x + 0.08$	0.64	<0.001
TRS (µmol cm^{-3})	Mixed – ns for Eelgrass	$5.514x - 0.84$	0.63	<0.001
Plant parameters — LOI (%)				
PON in leaves, roots and rhizomes	Both Eelgrass and Mixed	–	–	ns
POC in leaves, roots and rhizomes	Both Eelgrass and Mixed	–	–	ns
TP in leaves, roots and rhizomes	Both Eelgrass and Mixed	–	–	ns
Leaf number	Both Eelgrass and Mixed	–	–	ns
Leaf area	Both Eelgrass and Mixed	–	–	ns
TS in leaves roots and rhizomes	Both Eelgrass and Mixed	–	–	ns
Leaf+Root biomass (g DW m^{-2})	Mixed – ns for Eelgrass	$-97.2x + 281.8$	0.40	0.01
Plant parameters — TRS (µmol cm^{-3})				
PON in leaves, roots and rhizomes	Both Eelgrass and Mixed	–	–	ns
POC in leaves, roots and rhizomes	Both Eelgrass and Mixed	–	–	ns
TP in leaves, roots and rhizomes	Both Eelgrass and Mixed	–	–	ns
Leaf number	Both Eelgrass and Mixed	–	–	ns
Leaf area	Both Eelgrass and Mixed	–	–	ns
Leaf+Root biomass (g DW m^{-2})	Mixed – ns for Eelgrass	$-16.08x + 259.7$	0.52	<0.05
TS_{leaves} (µmol g DW^{-1})	Mixed – ns for Eelgrass	$9.37x + 79.2$	0.49	<0.05
TS_{roots} (µmol g DW^{-1})	Mixed – ns for Eelgrass	$16.85x + 56.9$	0.64	<0.001
$TS_{rhizomes}$ (µmol g DW^{-1})	Mixed – ns for Eelgrass	$17.86x + 102.7$	0.47	<0.05

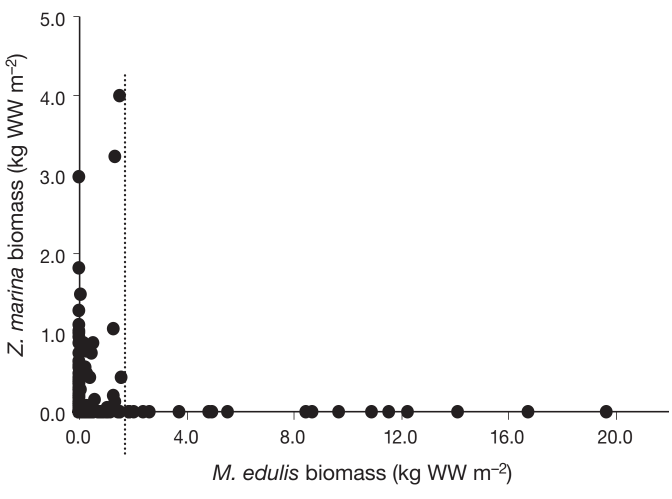

Fig. 5. *Zostera marina* and *Mytilus edulis*. Biomasses of eelgrass and mussels at 318 stations at 3 to 6 m water depth. Dotted line = 'threshold' of 1.6 kg WW m^{-2} where *Z. marina* is no longer found. Modified from Dolmer et al. (2009)

accumulation in plants (Table 4), indicating a relationship between the sediment biogeochemistry and plant measures.

Effects of coexistence on sediment characteristics

The sediments in Flensborg fjord in general were fine grained. The fine-grained fraction, however, was increased 3 times at the Mixed and Mussel stations (Table 1). Similar enrichment of the fine-grained fractions in mussel bed sediments was also found by Stoeck & Albers (2000), where biodeposits from *Mytilus edulis* decreased the grain size by increasing the silt and mud fraction. The presence of *Zostera marina* increased the accumulation of the organic matter in excess of the mussel bed itself, probably as the leaf canopy additionally reduces water flow, turbulence and resuspension (Petersen et al. 1997, Gacia et al. 2002, Allen & Williams 2003), and this is consistent with higher accumulation of organic matter and nutrients in seagrass sediments compared to unvegetated sediments (Duarte et al. 2005). Furthermore, the positive relationship between sediment LOI and sediment TP at the Mixed stations (Table 4) suggests direct coupling of TP to biodeposits. Peterson & Heck (1999) found higher TP pools in seagrass and mussel sediment compared to seagrass sediment and related this to the accumulation of P-enriched biodeposits from mussels. A positive relationship was also found between sediment LOI and sediment PON but for both Mixed and Eelgrass stations (Table 4), indicating that increased N accumulation was related to *Z. marina* rather than *M. edulis*. N pools were higher than in the bare sand, showing the enriching effect of seagrasses on sediment organic matter (Duarte et al. 2005)

The organic and nutrient enrichment of the Mixed stations increased the pools of reduced sulphides, as reflected in a positive correlation between LOI and TRS (Table 4). Higher pools of sulphides are usually correlated with enhanced sulphate reduction rates (Holmer & Frederiksen 2007), and several studies have found high sulphate reduction rates in sediments with mussel biodeposits (Sorokin et al. 1999, Stenton-Dozey et al. 2001). Stimulated sulphate reduction rates in *Zostera marina* beds in the presence of *Mytilus edulis* have been attributed to enrichment with labile organic material from biodeposits (Vinther et al. 2008). Vinther et al. (2008) also found enhanced TRS pools in Mixed sediments.

Plant responses to coexistence

The plants responded to the sediment organic enrichment by increasing nutrient contents (N and P, Fig. 4) and decreasing leaf number and leaf area, whereas the biomass was not significantly different between stations (Table 2), indicating a variable response of *Zostera marina* to the mussel biodeposits. In comparison, others observed mainly positive effects of suspension-feeding mussels and clams on seagrass (Reusch et al. 1994, Peterson & Heck 1999, 2001a,b, Carroll et al. 2008). Peterson & Heck (1999, 2001a,b) and Reusch et al. (1994) described their seagrass communities as nutrient limited and found positive effects of coexistence due to the fertilising effect of biodeposits (Table 5). Studying the effects of hard

Table 5. Comparison of studies on mixed mussel and seagrass meadows. LOI: loss on ignition, SRR: sulfate reduction rate

Characteristics of the study area	Studied species Mussel / seagrass	Effects of mussels on — Sediment	Effects of mussels on — Seagrass	Field study
No data on trophic condition Sandy sediment (LOI = 1 to 1.6%)	*Mytilus edulis* / *Zostera marina*	Increased porewater concentrations of ammonium and phosphate Increased LOI	Increased leaf area No data on plant nutrient content Facilitation of *Z. marina* due to biodeposition	Reusch et al. (1994)
Oligotrophic (N < 3 µM) No data on LOI	*Modiolus americanus* / *Thalassia testudinum*	Increased porewater concentrations of ammonium and phosphate	Reduced C:N and C:P ratios in leaves No data on plant morphology Increased nutrient content in *T. testudinum* due to biodeposition	Peterson & Heck (1999)
Oligotrophic (N < 3 µM) No data on LOI	*M. americanus* / *T. testudinum*	Increased total N and P pools	Increased leaf width and lengt Reduced C:N, C:P and N:P in leaves Increased production; fewer epiphytes Facilitation of *T. testudinum* due to biodeposition and/or reduced epiphyte load	Peterson & Heck (2001a,b)
Eutrophic (N = 7 to 10 µM) Sandy sediment (LOI = 0.3 to 0.75)	*M. edulis* / *Z. marina*	Increased LOI High NH_4^+ efflux Increased P pools Increased SRR	Reduced C:N in leaves Decreased production; more epiphytes *Z. marina* growth reduced due to possible sulphide stress and increased epiphyte load	Vinther et al. (2008)
Eutrophic (N = 7 to 10 µM) Sandy sediment (LOI = 0.6 to 1.0)	*M. edulis* / *Z. marina*	Increased fine-grained fraction Increased LOI Increased N, C and P pools Increased sulphide pools	Reduced C:N, C:P and N:P in leaves Decreased leaf area and length No positive leaf morphology response to increased nutrient content Risk of sulphide stress	This study

clams on *Z. marina*, Carroll et al. (2008) did not find correlations between sediment characteristics and the presence of hard clams. Leaf N content in *Z. marina*, however, increased when hard clams were present, and contributed to higher leaf production. Since N and P contents in *Z. marina* in our study were higher at the Mixed stations (Figs. 2 & 4), an increase in plant biomass stimulated by higher nutrient availability in the sediments could be expected, but this was not observed (Table 2). In fact, leaf area was 19% and leaf number was 8% lower at the Mixed stations, which is in contrast to Reusch et al. (1994), who found 36% higher leaf area (Table 5), and Carroll et al. (2008), who found an increased number of leaves. However, lower leaf number in *Z. marina* growing in coexistence with *Mytilus edulis* has been found in the field (Vinther et al. 2008) and in a laboratory experiment (Vinther & Holmer 2008). The positive correlation between TRS in the sediment and TS in the plants at the Mixed stations (Table 4) indicates an accumulation of sulphides in the form of S^0 or other reoxidation products in the plants at increasing sulphide pools in the sediment. Sulphide can cause detrimental effects on seagrasses, resulting in reduced photosynthetic activity and growth together with increased mortality (Holmer & Bondgaard 2001, Koch et al. 2007) and could account for the reduced seagrass performance at Mixed stations.

Implications of coexistence under eutrophic conditions

A key factor for the different outcome of the coexistence between *Mytilus edulis* and *Zostera marina* in this study compared to others (Table 5) could be eutrophication. Norkko et al. (2006) found that the facilitation effect of one species on another can shift from positive to none or even negative along environmental gradients. In our case, *Z. marina* may grow at its limits of sulphide pressure and anoxia in the sediments, and the organic enrichment by biodeposits pushes the community over a threshold value beyond which degradation is initiated. Clearance and improvement of light climate was not considered as an environmental factor in our study, as the meadows were located at shallow depths (<2 m), well above the depth limit of *Z. marina* in the area (4.6 m). Carroll et al. (2008) found that both improved light and sediment nutrient conditions contributed to increased leaf growth of *Z. marina*. However, the density of hard clams was low compared to our study of *M. edulis*, and as the 2 species have different life strategies, where the hard clams burrow in the sediment, while *M. edulis* lives on top of the sediment, this may affect the deposition of faeces. Biodeposition on the surface may increase sediment respiration and lower oxygen levels near the plant meristems (Valdemarsen et al. 2009), which is a critical factor for sulphide invasion into *Z. marina*. (Pedersen et al. 2004). A possible threshold for coexistence of *Z. marina* and *M. edulis* in Danish coastal waters was indicated by a survey of 318 stations in the depth interval 3 to 6 m (Dolmer et al. 2009). The study suggests that the 2 species coexist up to a threshold value of 1.6 kg mussel m^{-2}, whereafter *Z. marina* is absent (Fig. 5). Although it was conducted at depth intervals different from our stations, the results support that *M. edulis* may stress *Z. marina* under eutrophic conditions. The complete absence of *Z. marina* may be due to a negative feedback loop at Mixed stations, where decreasing biomass of *Z. marina* creates space for more mussels, which in turn increases the negative effects on *Z. marina*. Many years of eutrophication have favoured the growth of *M. edulis* in Danish coastal areas, and mussel beds have established in previous eelgrass habitats in Flensborg fjord. This shift in benthic communities may in some cases be irreversible, as dense mussel beds are persistent and inhibit natural recolonisation by eelgrass. The balance between the 2 types of benthic communities thus seems to be delicate, and the outcome of coexistence is, in addition to the density of mussels, influenced by the general environmental status of the ecosystem. To ensure re-establishment of *Z. marina* in fjords, where nutrient loading has been reduced, reductions of phytoplankton biomass alone may not be enough. The density of mussels and the effect they have on sediment biogeochemistry must be considered as well.

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