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Epifaunal and infaunal responses to submarine mine tailings in a Norwegian fjord

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ABSTRACT

Disposal of mine tailings in marine shallow water ecosystems represents an environmental challenge, and the present paper reports results from a field study in Frønfjorden, Norway, which is subject to such disposal. Structural and functional responses of benthic infauna and epifauna were investigated along a gradient from heavy tailings deposition to reference conditions. The tailings clearly impacted the faunal composition, with lowered species number close to the outfall. Total abundance of infauna increased in the most impacted area due to dominance of opportunistic species, whereas the epifauna was reduced and represented by a few scattered specimens only. In the most impacted area functional responses included an increase in mobile carnivores/omnivores and species utilizing symbionts. Sessile and tube-living taxa, and deposit and suspension feeders decreased, probably due to smothering in combination with tailings-associated changes of the substrate. Functional diversity decreased for both infauna and epifauna, but less than the structural diversity.

1. Introduction

Disposal of mine tailings represents an environmental challenge due to their vast volumes and potential toxicity (Cornwall, 2013; Ramirez-Llodra et al., 2015). Tailings are composed by the fine particle waste remaining after the mineral product has been extracted from the ore-bearing rock. The tailings mainly consist of crushed rock, but may in addition contain potentially harmful substances such as heavy metals and remnants of process chemicals (Koski, 2012; Ramirez-Llodra et al., 2015). Traditionally, tailings have been stored in land dams, but sea disposal has been favored in regions where land availability, climatological conditions or geological instability may result in dam failure or leakages (Arnesen et al., 1997; Kvassnes and Iversen, 2013; Ramirez-Llodra et al., 2015). In submarine tailing disposal (STD), tailings are discharged through a submerged pipeline at relatively shallow depths (< 100 m), aiming at creating a gravity flow that deposits the tailings on the seafloor below the mixing-zone (Ramirez-Llodra et al., 2015). Up to several million tons of fine-grained mineral particles may be disposed annually by a single mine. The safe and environmentally-acceptable storage of these fine-fraction wastes is one of the most important environmental issues faced by the mining industry (Cornwall, 2013).

Marine life may be affected by STD through smothering, toxic effects from process chemicals, oxidative release of trace metals, altered sediment composition and physical damage from sharp tailing particles (Olsgard and Hasle, 1993; Ellis et al., 1995; Dold, 2014; Ramirez-Llodra et al., 2015; Morello et al., 2016). The fauna living in the sediments, the macroinfauna, is particularly used for monitoring of effects because it can be sampled quantitatively, hence allowing for assessing the degree of influence based on numerical measures of the species assemblages. Within the European Water Framework Directive, macroinfauna is one of the indicators of environmental status, together with other biological and supporting quality elements of the water bodies (see e.g. Van Hoey et al., 2010).

On the other hand, the benthic epifauna that lives on or just above the sediment surface is far less studied. The epifauna includes a variety of living strategies from sessile suspension feeders, slowly moving detritus feeders, to slowly or actively moving predators. In general, there is much less information on epifaunal responses to disturbances, both with regard to composition and functional attributes (Pearson and Rosenberg, 1978), but this is sparsely studied (Fields et al., 2019). As several epifaunal species feed directly on sinking material or the most recently deposited organic matter on the sediment surface, they may be more sensitive indicators of recent chemical contamination, and perhaps respond in different manners and on faster time scales than

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infauna (Jørgensen et al., 2011). Moreover, with regard to mine tailings, they may be particularly vulnerable as they live where the tailings chronically disturb the sediment surface and builds up.

Most studies addressing the environmental effects of waste effluents and other anthropogenic stress factors have focused on changes in species diversity and community structure. In recent years, there has also been a growing concern about which species attributes are affected, and what the consequences are for the ecosystem functioning (Beauchard et al., 2017). Application of techniques such as Biological Traits Analysis (BTA) has proved useful to improve the understanding of the actual ecological significance of environmental variables and human activities (Bremner, 2008; Oug et al., 2012; Beauchard et al., 2017). In BTA, species traits are expressed as variables describing species performances. The traits may also be correlated to environmental variables like natural gradients or gradients in disturbances and contaminants (Oug et al., 2012; Fleddum et al., 2013; Rand et al., 2018), which is one of the reasons why trait-based approaches now have become useful tools in environmental monitoring and management. Partly originating from the concept of BTA, is analysis of functional diversity. Functional diversity is the diversity of all the species traits in a community and is commonly assumed to be a better predictor of ecosystem productivity and vulnerability than species diversity (Petchey and Gaston, 2002, 2006).

Norway is the country with most STDs (past and active) globally (Ramirez-Llodra et al., 2015). One of these is located in Frønfjorden, western Norway, arising from production of liquid marble. The discharges commenced in 1982, and regular monitoring has been conducted since 1989, focusing on water quality (turbidity, oxygen, temperature and salinity), sediment quality and macrofaunal communities. Main results from the monitoring of macroinfauna for the period 1993 to 2010 have been presented by Brooks et al. (2015). The study showed clear spatial and temporal changes in the species communities that were correlated with distance from the outfall and proportion of fine tailings in the sediments.

In the last years, more comprehensive environmental studies have been carried out in the fjord. Field and experimental setups have been used to assess ecological effects on macroinfauna (Trannum et al., 2018; submitted) and bioavailability and toxic effects of process chemicals (Brooks et al., 2018; Brooks et al., 2019). Also sediment conditions have been investigated using sediment profile imagery (SPI) (Schaanning et al., 2009).

The present study reports results from faunal investigations in Frønfjorden in the period 2013–2016. The main focus is on the sampling in 2016 when epifauna was sampled in parallel with macroinfauna. Moreover, both structural and functional changes are assessed. The results are interpreted in relation to sediment conditions and tailings disposal, and with regard to new knowledge from recent experimental studies.

2. Materials and methods

2.1. Study area and discharge information

Frønfjord is a fjord in Møre and Romsdal county in the west coast of Norway. It is 12 km long with a maximum depth of around 70 m. The tidal range is about 2 m and the fjord is well flushed. In general, the oxygen-levels have been good and indicate that the fjord is not eutrophic (DNV GL, 2016). The water masses are relatively homogeneous from autumn to early spring. The formation of the pycnocline typically occurs in spring due to increased freshwater runoff and increased temperature, especially in the surface layer (DNV GL, 2016).

The fjord has received mine tailings from Omya Hustadmarmor AS in Elnesvågen since 1982. The production plant receives marble from adjacent mines, where it is grounded, washed and sieved. The final product is liquid marble, which is used in paper production as filler or coating (Ramirez-Llodra et al., 2015). The tailings are discharged at 20 m depth through a submerged pipe, and deposited in a fjord basin at

40–70 m depth (Skei, 2014). Since 2007, the discharges have been 300–500 k tons per year (Ramirez-Llodra et al., 2015). The tailings are very fine-grained; 30% < 4 µm, 60% < 20 µm and 80% < 63 µm. They do not contain elevated levels of metals (Farkas et al., 2017), but both flocculation chemicals (anionic polyacrylamide) and cationic flotation chemicals are used in the process. In October 2014, the previously used flotation chemical (Lilafлот) was substituted with FLOT2015. FLOT2015 is formed mainly by a mixture of unsaturated fatty acids (mono-, di- and/or triesterified) with a methyl triethanol ammonium (MTA) moiety that belongs to the family of the esterquats tensioactives (Brooks et al., 2018). In contact with water, the product undergoes hydrolytic reactions that break the ester linkage and yield a mixture of fatty acids and MTA. The MTA compound is measurable and provides an indication of the presence of the flotation chemical in the sediments (Escudero-Oñate, pers. comm.).

Prior to discharge, freshwater used in the processing is removed and seawater is added to reduce spreading of the particles and maximize transport downwards to the seabed. In areas near the outfall almost all of the top few meters of sediment can be made up of fine (< 63 µm) CaCO₃ particles. As a result of the discharge, the depth of the fjord in certain areas has gradually decreased by up to 25 m from that recorded prior to the deposit. As sediment has accumulated around the outfall pipes, they have been relocated and extended within the deposit area (DNV GL, 2016).

2.2. Field work

Infauna and epifauna were sampled at six and five stations, respectively, located along a transect from the inner part of the fjord across the vicinity of the outfall to the fjord mouth (Fig. 1). Station NY1 was situated in an inner fjord basin separated from the main fjord basin by islets and sills. NY0, NY3 and NY4 were located in the deposit area, NY5 in the mixing zone and NY6 just outside the mixing zone. Infauna and epifauna were sampled at adjacent positions, but epifauna sampling had to be omitted at the fjord mouth (NY7) due to coarse bottom conditions with large stones.

The infauna was sampled in 2013, 2015 and 2016 (Table 1) using a 0.1 m² van Veen grab. The sediment catch was washed through a 1 mm sieve with circular holes and the retained material preserved in 10% buffered formaldehyde solution in seawater. In 2013 and 2015 three replicate samples were taken, whereas in 2016 two replicates were taken. Sediment samples for analysis of grain size (upper 5 cm layer), total organic carbon (TOC) (0–5 cm in 2013 and 2015; 0–1 cm in 2016) and nitrogen (TN) (0–1 cm, 2016 only) were taken from a separate grab. The sampling was conducted according to ISO 16665:2014.

The epifauna was sampled in 2016 using a 1.8 m wide Agassiz trawl equipped with a coarse outer net and 25 mm mesh inner net. A coarse mesh was chosen to avoid clogging of the net from sediment tailings when sampling in the deposit area. Three replicate hauls were taken at each station. Trawl lengths were standardized to a sailing distance of 200 m giving a bottom dredging length of about 160 m. Samples were washed on 5 mm sieves for cleaning of mud and tailings. Larger organisms were counted and weighed in the field and discarded, whereas smaller organisms were preserved in 96% ethanol.

2.3. Laboratory analyses

For grain size analysis, the samples were initially split by wet sieving at 63 µm, then dried, and the coarse fractions passed through 2 mm, 1 mm, 500 µm, 250 µm, 125 µm sieves, according to Wentworth scale (Wentworth, 1922). The percent of each fraction, including the fine fraction (< 63 µm), was determined by weighing. Total organic carbon (TOC) and total nitrogen (TN) were determined using a Carlo Erba element analyzer based on chromatographic detection of CO₂ and N₂ gases, respectively, after oxygen had been removed in a combustion at 1800 °C.

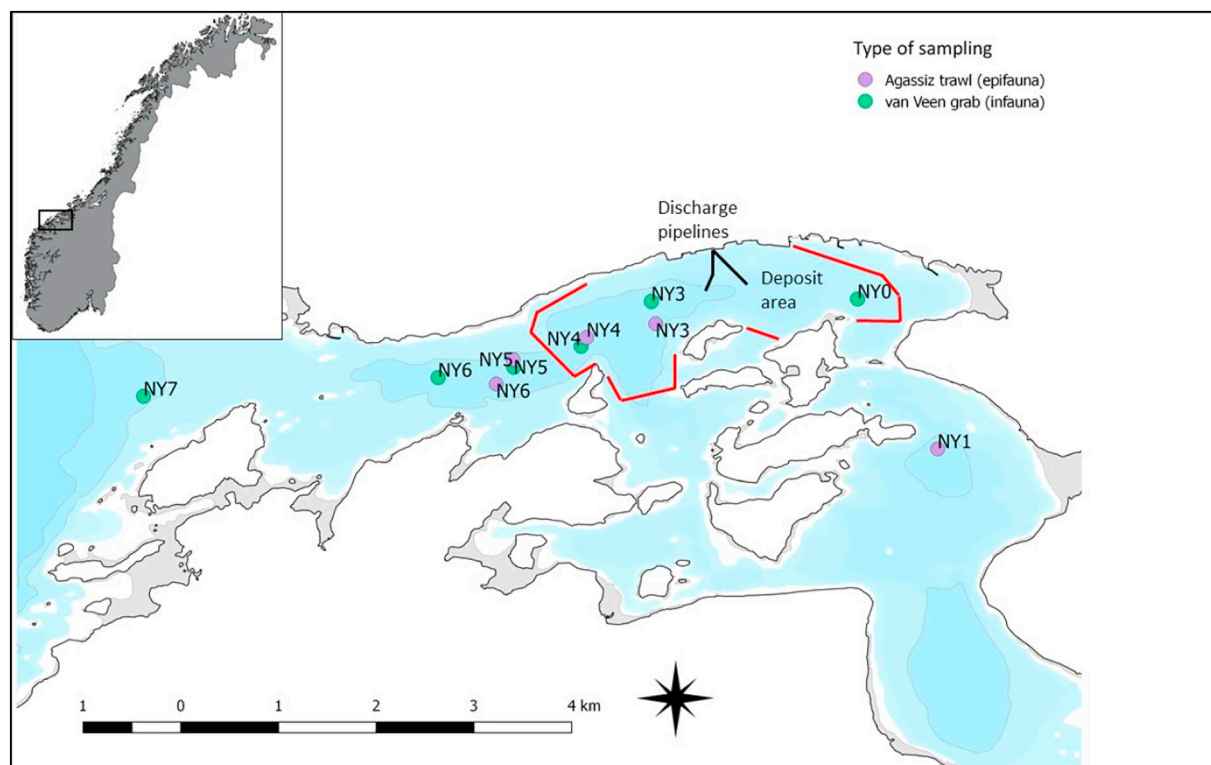


Fig. 1. Map of sampling stations in Frønfjorden, 2013–2016. The discharge pipes are indicated by the black lines. The deposit area is situated within the red lines. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

All organisms were sorted into main taxonomic groups and then transferred to ethanol (> 80%). They were identified to species or lowest possible taxon following standardized methods (ISO 16665:2014). The species names were checked according to World Register of Marine Species (<http://www.marinespecies.org/>). Biomass determination was performed for the 2016 samples. For infauna this was measured as bulk weight for the main taxonomic groups. Free-living and errant annelids were weighed separately, and also large organisms like sea urchins and large bivalves. A separate category was applied for unidentified organic material like animal parts and segments, constituting maximum 0.07% of the total biomass. The samples were placed on a filter paper, and the individuals were blotted dry for a few seconds, and then placed on a new filter paper and weighed. The weight was read immediately to avoid evaporation of the ethanol; in gram with a resolution of four decimals. For epifauna each species was weighed separately. This was done on board the ship for the organisms that were identified in the field, and in the laboratory for the remaining organisms. In the field, the resolution was lower (one decimal), but the methodology was otherwise the same as for the infauna.

Table 1

Position and depth of epifaunal and infaunal sampling, Frønfjorden (mean positions). Sampling year is also presented. Latitude and longitude in decimal degrees (WGS 84).

	Station	Equipment	Latitude	Longitude	Depth (m)	Year
Infauna	NY0	van Veen grab	62.84583	7.14600	37–38	2013, 2015
	NY3	van Veen grab	62.84330	7.10495	64	2015, 2016
	NY4	van Veen grab	62.83842	7.09203	66–70	2013, 2015, 2016
	NY5	van Veen grab	62.83575	7.07913	54–56	2013, 2015, 2016
	NY6	van Veen grab	62.83395	7.06432	54–62	2013, 2015
	NY7	van Veen grab	62.82890	7.00610	52–64	2013, 2015, 2016
	Epifauna	NY1	Agassiz trawl	62.83304	7.16564	50–55
NY3		Agassiz trawl	62.84131	7.10636	50–67	2016
NY4		Agassiz trawl	62.83927	7.09291	68–70	2016
NY5		Agassiz trawl	62.83644	7.07881	54–56	2016
NY6		Agassiz trawl	62.83401	7.07606	57–58	2016

Table 2
Overview of traits and categories from the traits database used in the present study.

	Trait		Categories			
Activity	Adult life habit	AH	1 Non-tube sessile			
			2 Permanent tube			
			3 Semi-permanent tube			
			4 Burrower			
			5 Surface crawler			
6 Swimmer						
Degree of attachment	DA	1 None				
		2 Temporary				
		3 Permanent				
Adult mobility (relative)	AM	1 None				
		2 Low				
		3 Medium				
		4 High				
Sediment dwelling depth	SD	1 0 cm (surface)				
		2 0–1 cm				
		3 1–5 cm				
		4 5–15 cm				
		5 > 15 cm				
Size and shape	Body form	BF	1 Short cylindrical			
			2 Flattened dorsally			
3 Flattened laterally						
4 Ball-shaped						
5 Long, thin threadlike						
6 Irregular						
Normal adult size	NS	1 < 0.5 cm				
		2 0.5–1 cm				
		3 1–3 cm				
		4 3–6 cm				
		5 6–10 cm				
		6 > 10 cm				
Life history	Feeding	FH	1 Suspension/filter feeder			
			3 Surface deposit feeder			
			5 Dissolved matter/symbionts			
			6 Sandlicker/large detritus			
			7 Scavenger			
			8 Carnivore/omnivore			
			9 Parasite/commensal			
			10 Subsurface deposit feeder			
			11 Deep deposit feeder			
			Reproduction	Larval type	LT	1 Planktotrophic
						2 Lecithotrophic
Bioturbation potential	Sediment reworking	SR	1 Epifauna			
			2 Surficial modifier			
			3 Upward conveyor			
			4 Downward conveyor			
			5 Biodiffusor			
			6 Regenerator			

For infauna, species lists from 2013, 2015 and 2016 were checked for variability in species names or identification level and harmonized. All calculated abundances were square-root transformed prior to analyses to balance between abundant and rare species. The analyses were performed with PRIMER package version 6.1.13.

2.4.3. Functional analyses

Functional analyses encompass methods to express the importance and variety of life strategies and organism performances in species assemblages. In this study, functional community composition and diversity were calculated separately for the infauna and epifauna, and assessed in relation to the STD. The life strategies and performances (species traits) used in the analyses are shown in Table 2. Each trait is divided in several categories which express different states of the trait. Each species is scored for their affinities to the categories. The scores are assigned according to the “fuzzy coding” procedure (Chevenet et al., 1994) with values ranging from 0 (= no affinity) to 3 (= dominant).

Traits data for the species were extracted from a database held at Norwegian Institute for Water Research where information has been

compiled based on literature and by consulting experts (Oug et al., 2012). For the trait “sediment reworking” (SR), that expresses the bioturbation activity of the species, the information is based on the approach by Queiros et al. (2013). Several taxa in the present dataset lacked traits information. In these cases, rare taxa were omitted from the analyses (infauna), whereas frequently occurring and abundant taxa were assigned traits. Information was acquired from scientific literature or open databases; BIOTIC (MarLIN, 2006) and Polytraits (<http://polytraits.lifewatchgreece.eu>; Faulwetter et al., 2014) or by entering information from taxonomically related taxa. Some traits in the database, mainly related to reproduction and life duration, lacked information for many species (see Oug et al., 2012). Traits with missing information for more than one third of the species in the present study were omitted.

Functional diversity was described using the Quadratic entropy index (FD_{RAO}) (Rao, 1982; Leps et al., 2006). Index values were calculated separately for each trait and averaged for each station following procedures described by Leps et al. (2006) and Darr et al. (2014). Functional redundancy was used to examine the relationship between taxonomic and functional diversity, and was calculated as the ratio between FD_{RAO} and the Shannon–Wiener diversity index; i.e. FD_{RAO}/H' (van der Linden et al., 2012). Functional redundancy expresses to which degree different species represent the same ecosystem functions, i.e. a low ratio value indicates that a few attributes are shared among the present species. The calculations were performed using Canoco 5 software.

Functional community composition was described using Biological Traits Analysis (BTA) for species assemblages. In the analysis, species abundance data are combined with traits data for each species to calculate community weighted means (CWMs or “trait profiles”) expressing the functional composition of the species assemblages (Bremner et al., 2003, 2006; Oug et al., 2012; Beauchard et al., 2017). For the macroinfauna, the CWMs were subject to multivariate ordination using metric multidimensional scaling (MDS) based on Euclidean distance (dissimilarity) measure (=Principal coordinate analysis). Infaunal species data were calculated as mean abundances per stations (0.1 m^2), and square-root transformed to reduce the influence of the numerically dominant species. The calculations were carried out using PRIMER package version 6.1.13. The epifauna results were rather sparse (see Section 3), and were here deemed insufficient for a reliable multivariate ordination of functional features. Thus, for epifauna, selected traits along the station transect were plotted separately. Mean abundances for each station (per 10 m^2) were calculated and square-root transformed.

3. Results

3.1. Environmental data

Inside the fjord, the sediment was very fine grained, with a clear gradient of increasing fine fraction towards the discharge (Table 3). The fine fraction was as high as 99.4% at the closest station (NY3) and only 14.4% at the outermost station (NY7). The sediment was classified as pelite (i.e. composed of mud and silt) at the three stations inside the fjord and as fine sand at NY7. All sediments were poorly sorted based on the inclusive graphic standard deviation (IGSD), with the lowest value at NY3 and the highest at NY5. The skewness was zero at NY3 (i.e. a symmetrical grain size distribution), and showed the highest value at NY7. The kurtosis was 0.73–0.75 at the stations within the fjord and 1.74 at NY7.

The amount of organic carbon (TOC) was lowest at station NY7, outside the sill (3.5 mg/g), and highest at station NY4 within the deposit (13.6 mg/g) (Table 3). Total nitrogen was below the detection limit of 1 mg/g at all stations.

3.2. Infauna

3.2.1. Species richness, diversity and biomass

A total of 272 taxa and 14,869 individuals were recorded. The annelids constituted the most abundant group with 53 taxa, followed by

Table 3

Sediment grain size characteristics, sediment classification, and content of total organic carbon (TOC) and total nitrogen (TN), Frænfjorden (NY0/NY6 from 2015, otherwise from 2016). Grain size parameters from 0 to 5 cm, TOC/TN from 0 to 1 cm.

	% < 63 μm	Median D ₅₀ (φ)	Sorting (φ)	Skewness (φ)	Kurtosis (φ)	Classification	TOC (mg/g)	TN (mg/g)
NY0	99.4	–	–	–	–	Pelite	3.7 ^a	–
NY3	99.4	5.966	1.225	0.000	0.738	Pelite	8.1	< 1.0
NY4	94.9	5.870	1.295	– 0.010	0.752	Pelite	13.6	< 1.0
NY5	72.0	5.204	1.642	0.030	0.728	Pelite	12.8	< 1.0
NY6	20.1	–	–	–	–	Fine sand/gravel	10.4 ^a	–
NY7	15.4	3.225	1.393	– 0.039	1.737	Fine sand	3.5	< 1.0

^a 0–5 cm.

molluscs (36 taxa), crustaceans (34 taxa) and echinoderms (10 taxa). In 2016, 142 taxa were recorded at the four sampled stations. In 2013 and 2015, 178 taxa (five stations) and 206 taxa (six stations), respectively, were found. Complete species data are presented as supplementary information (Supplementary material 1).

The composition of the infaunal species communities changed distinctly from outside of the fjord (station NY7) through the mixing zone (station NY5) to the deposit area (stations NY4, NY3) (Fig. 2; data from 2016). The number of species generally decreased, whereas abundances increased quite markedly. The total biomass was highest outside the fjord and decreased with about 25% in the deposit area. The changes for species groups were, however, remarkable. Tube-building annelids were reduced from being biomass dominants outside the fjord to almost zero biomass in the inner deposit area. Starfish and brittle stars

(Asteroidea/Ophiuroidea) were clearly reduced in the deposit area, whereas bivalves and free-living annelids increased and dominated the biomass (Fig. 2).

Corresponding with the changes in species richness and abundances, the species diversity (*H'*, *ES*₁₀₀) decreased from outside of the fjord and the mixing zone inwards to the STD (Fig. 3). The results for 2016 were generally similar to the previous monitoring in 2013 and 2015, but the decrease was lesser in 2016 close to the discharge outlet (NY3).

3.2.2. Community analysis

In the community analysis the sampling stations were arranged along a distance gradient in the fjord with the outer station (NY7) to the left and the inner stations (NY3, NY0) most close to the outfall to the

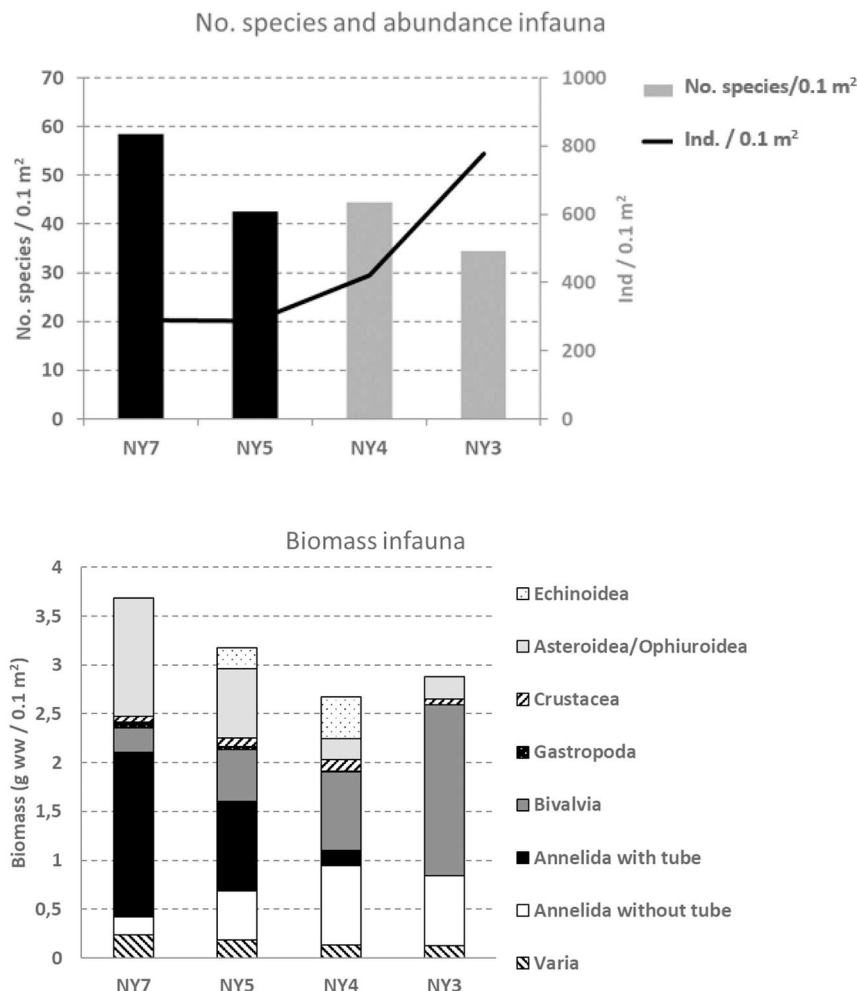


Fig. 2. Number of species, abundance and biomass of infauna in Frænfjorden, 2016 (mean values pr).

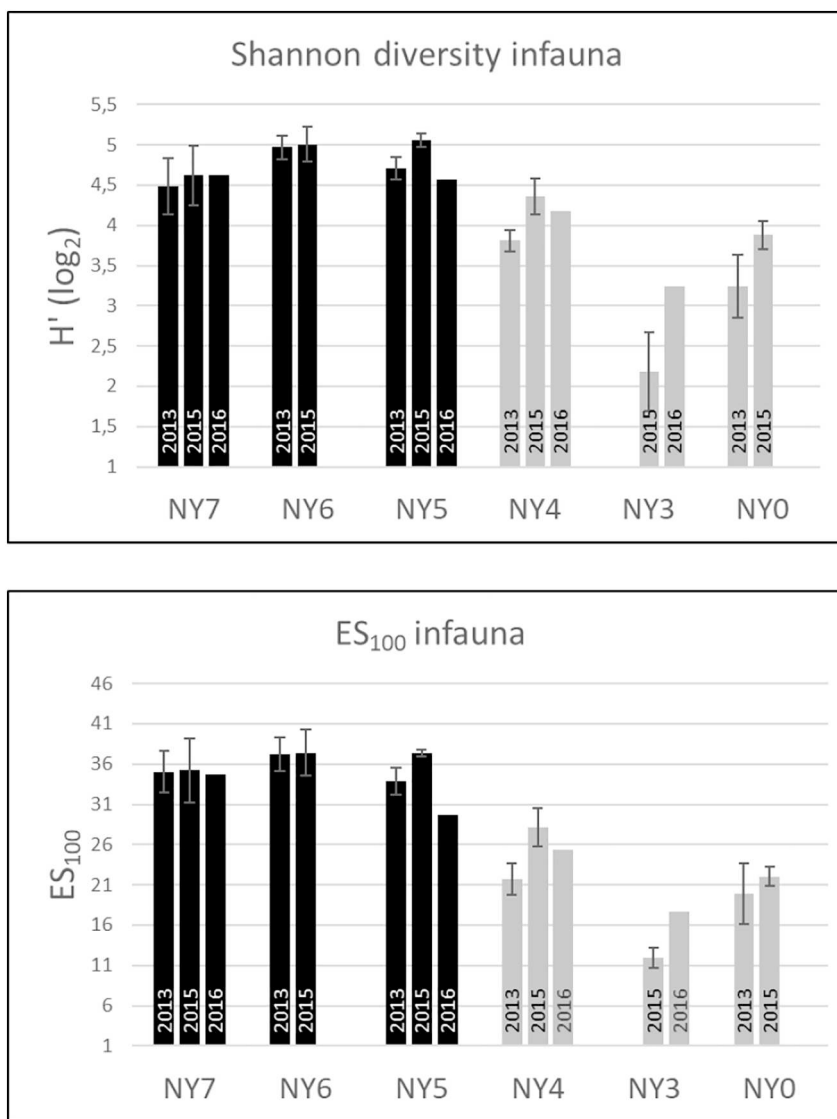


Fig. 3. Shannon-Wiener ($H' \log_2$) and Hurlbert's index (ES_{100}) of infauna in Frænfjorden, 2013–2016 (mean values pr. 0.1 m²). Stations in black are outside the deposit area and stations in grey inside.

right (Fig. 4). The pattern was quite parallel for the three years, but there was a difference between the years indicated by the 2013-samples being placed in the upper part and the 2016-samples in the lower part of the diagram. The distance gradient clearly showed that the species composition changed gradually with distance from the outfall, i.e. in relation to the deposition of tailings.

3.2.3. Functional diversity and community functioning

There were traits information for 203 taxa in the present study. All taxa that had insufficient or no information and were removed had total abundances < 10 individuals, except one taxon with 11 individuals (Hydroidolina indet), one with 12 individuals (*Amphiura* sp.) and one with 21 individuals (Golfingiidae indet).

The functional diversity (FD_{Rao}) varied less than the species diversity along the fjord gradient (Fig. 5). In 2016, FD_{Rao} was more or less similar at all stations, whereas in 2013 and 2015, FD_{Rao} largely paralleled the variation in species diversity. The functional redundancy was largely stable, both along the fjord gradient and temporally, except for a high value in 2015 (NY3) co-occurring with low species diversity. The high value indicates that there were relatively many different functions (low redundancy) represented among the species present.

The analysis of the community functioning (trait profiles = CWMs)

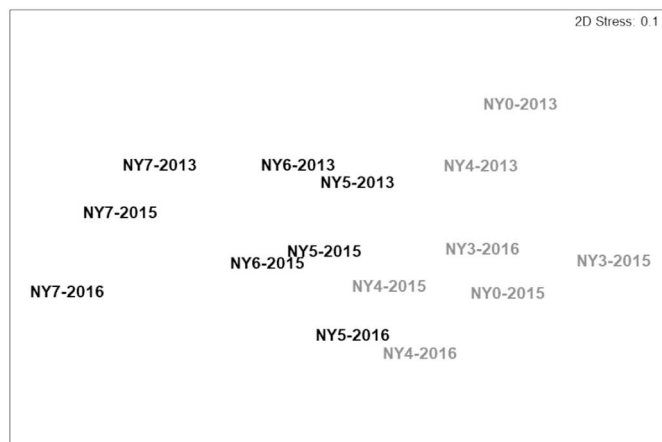


Fig. 4. Plot of nMDS-ordination of infauna in Frænfjorden, 2013–2016 (square-root transformed data). Stations in black are outside the deposit area and stations in grey inside. Stations NY6 and NY0 were not sampled in 2016 and NY3 not in 2013.

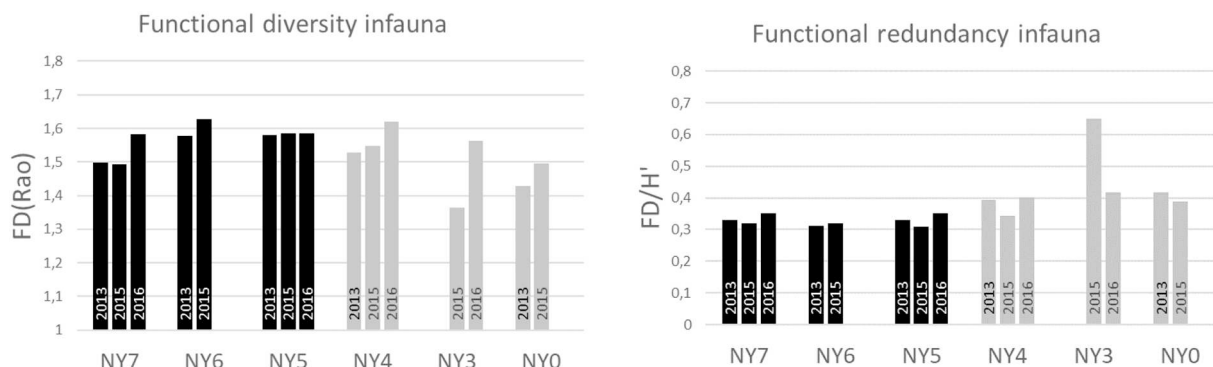


Fig. 5. Functional diversity (FD(Rao)) and functional redundancy (FD/H') of infauna in Frænfjorden, 2013–2016 (mean values pr. 0.1 m²). High FD/H' ratio is indicative of low functional redundancy. Stations in black are outside the deposit area and stations in grey inside.

illustrated that rather many traits changed along a gradient from the outer fjord (NY7) across the mixing zone (NY6, NY5) and into the STD (NY4, NY3, NY0) (Fig. 6: horizontal axis). Trait features (categories) that were prevalent at the outer part of the fjord and decreased in the STD, included semi-permanent or permanent tube, permanent and temporary attachment, no mobility and surface deposit feeding. In contrast, features that characterized the STD included no attachment, high mobility, surface crawling, carnivorous feeding and to some extent feeding on dissolved matter (symbionts). There were smaller differences between the sampling years (vertical axis), mainly indicated by larger proportions of long thin surface deposit feeders in the outer part of the fjord in 2013 and 2015, and increased proportions of laterally flattened dissolved matter feeders in the deposit area in 2015 and 2016. The functional changes reflect the faunal changes from a community dominated by echinoderms and tube-living annelids outside the fjord to dominance of bivalves and free-living annelids within the STD (Fig. 2).

The traits sediment dwelling depth and larval type appeared to be less clearly related to the main gradients (axis correlations < 0.7 for all categories; not entered in Fig. 6). This indicates, for instance, that in the

most impacted deposit area both shallow and deep digging organisms were found, despite the large change in sediment structure and the prevalence of unattached burrowing and mobile organisms.

3.3. Epifauna

3.3.1. Species richness, diversity and biomass

A total of 61 species taxa were identified in the samples. The Agassiz dredge collected some infaunal species and scattered specimens of small species that would pass through the meshes of the collecting net. Deleting the infaunal and small species as well as three species without traits, 36 epifaunal species were included in the present analyses. The echinoderms constituted the most abundant group with 17 taxa, followed by annelids (8 taxa) and crustaceans (7 taxa). Complete species data are presented as supplementary information (Supplementary material 2).

Species numbers, abundances and diversity (H') changed markedly from outside of the deposit area to within the STD (Fig. 7). At the two stations within the deposit area (NY4 and NY3) only five and three species, respectively, were found. Also, the abundances were strongly

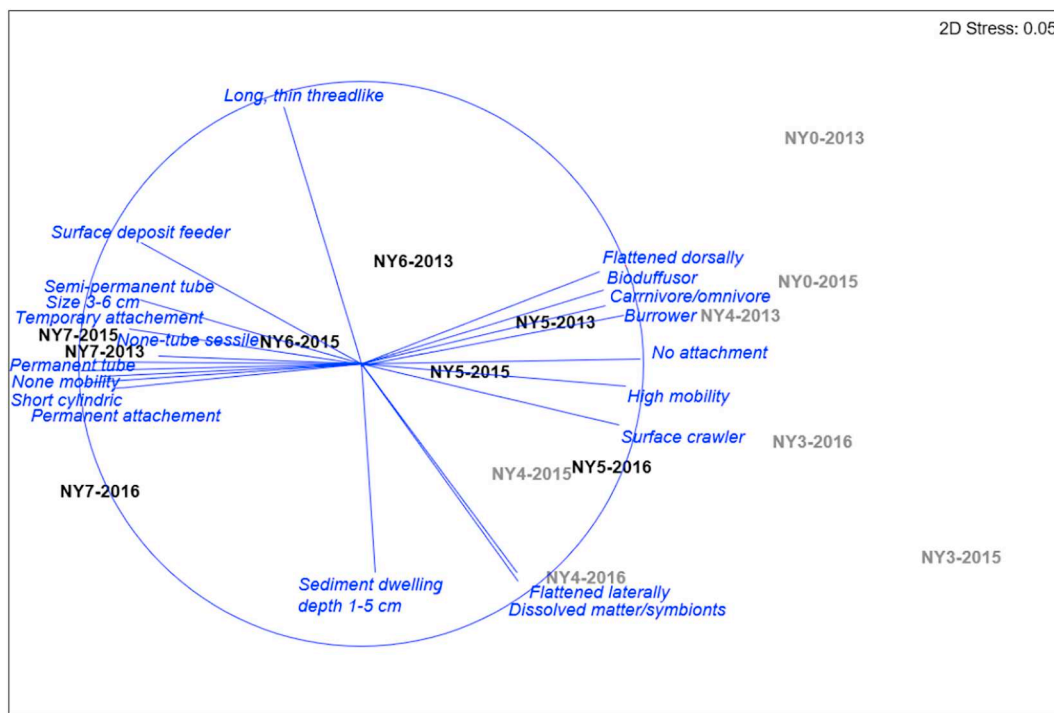


Fig. 6. Principal coordinate analysis (PCoA) of species traits of Frænfjorden infauna (2013–2016); plot of station centroids with trait categories added according to correlations. Trait categories are illustrated as vectors pointing in the direction of maximum increase, long vectors indicate strong trends. For clarity, only traits with high correlation to the axes (Pearson correlation coefficient > 0.7) are shown. Stations in black are outside the deposit area and stations in grey inside.

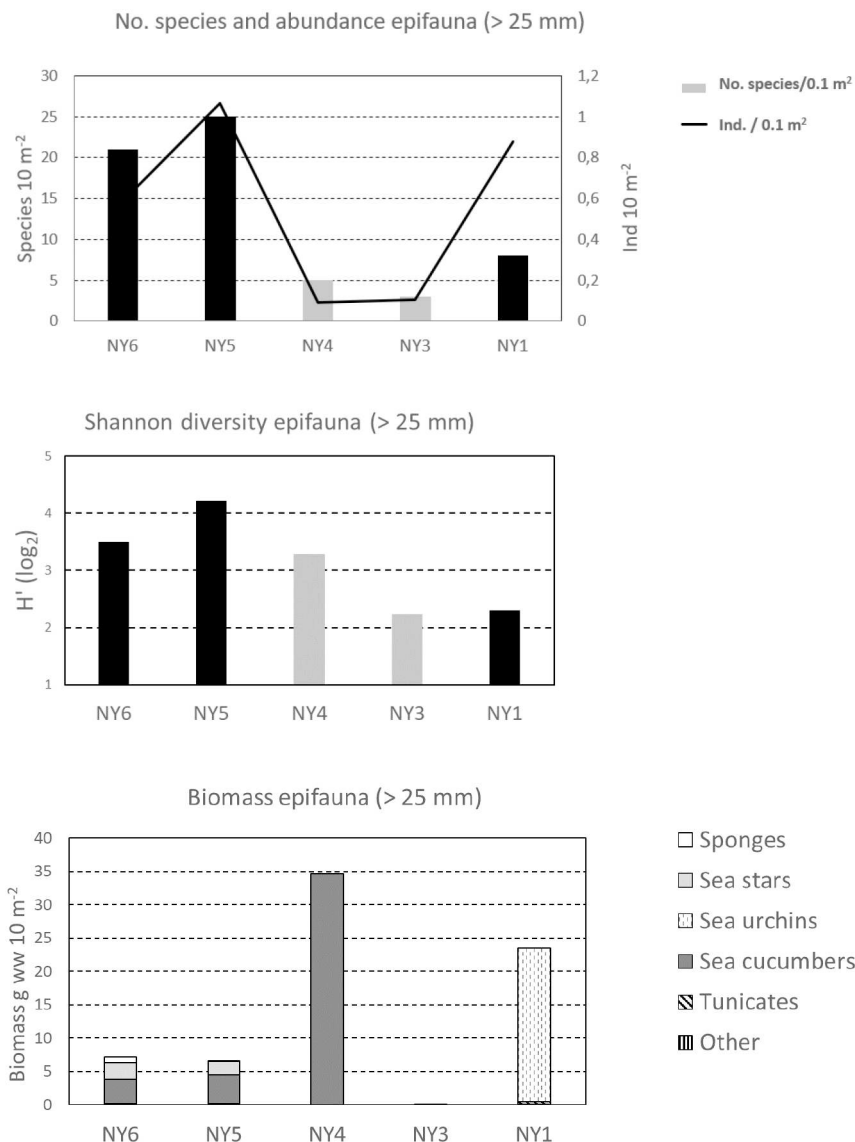


Fig. 7. Faunal characteristics of epifauna in Frænfjorden 2016. Number of species is total number in three replicate samples per station. Abundances and biomass (g wet weight) are calculated as mean values per 10 m² dredged area. Upper figures; stations in black are outside the deposit area and stations in grey inside.

reduced. In the inner fjord basin (NY1), species number, abundances and diversity (H') increased, but species number and H' were lower than at the outer part of the fjord. The biomass was generally low, but showed a high value at NY4 due to a few very large sea cucumbers (*Parastichopus tremulus*). The inner fjord station (NY1) was dominated by sea urchins (*Brissopsis lyrifera*), which contributed to the major part of the biomass.

3.3.2. Community analysis

In the MDS-ordination of species composition the samples from the deposit area (stations NY4, NY3) were clearly separated from the mixing zone (NY6, NY5) and the inner fjord basin (NY1) (Fig. 8). Within both station groups, there was a geographical pattern as the outer stations were placed to the left and the inner stations to the right. This pattern indicates that the species composition changed gradually and that the fauna in the STD was substantially affected. The replicate samples were generally grouped, but it appeared that differences among replicates were rather large and almost of the same magnitude as differences between stations.

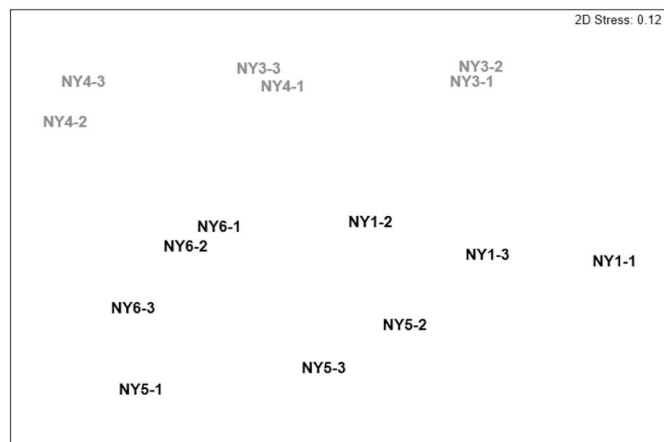


Fig. 8. Plot of nMDS-ordination of epifauna in Frænfjorden, 2016, three replicates per station (square-root transformed data). Stations in black outside the deposit area and stations in grey inside.

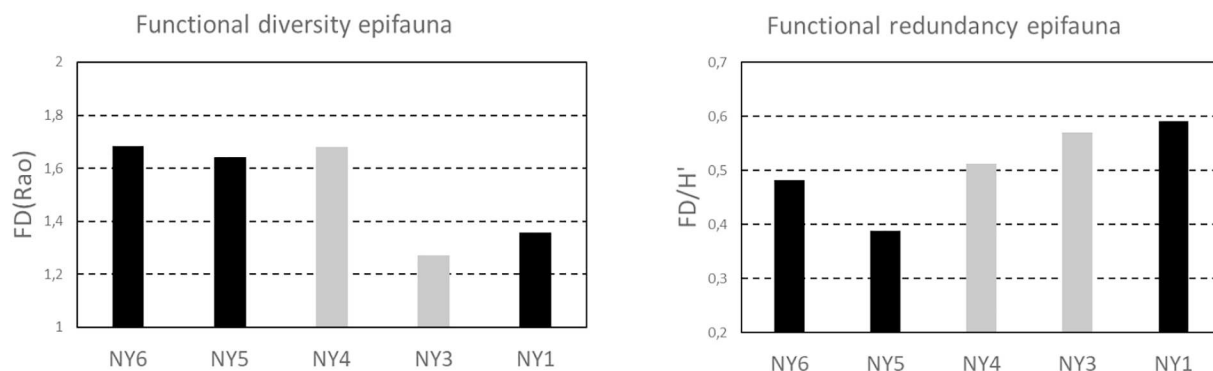


Fig. 9. Functional diversity (FD_{Rao}) and functional redundancy of epifauna in Frøenfjorden, 2016 (sum of three hauls); stations in black outside the deposit area and stations in grey inside.

3.3.3. Functional patterns

All 36 epifauna species had traits information. The functional diversity (FD_{Rao}) of epifauna was lowest close to the effluent outfall (station NY3), but was also rather low in the inner fjord basin (NY1) (Fig. 9). The functional redundancy (FD_{Rao}/H' ratio) varied rather much. The high values at the stations in the deposit area indicate that there were relatively many functions represented (low redundancy), but it should be considered that the number of species at these stations was very low making comparisons with more species-rich stations unreliable.

Functional attributes of the epifauna species assemblages showed several responses from outside of the deposit area to within the area (Fig. 10). For the main life style (“adult life habit”) it appears that sessile living forms were missing in the deposit area, whereas surface crawling increased somewhat. For feeding there was a marked disappearance of suspension feeding, and closest to the outfall (station NY3) also of subsurface deposit feeding, while the proportion of scavengers and carnivores increased. The size distribution of organisms was similar at most stations, but close to the outfall (NY3) the

organisms were smaller, and in the inner basin (NY1) one size group (moderate size) dominated, obviously due to the abundance of sea urchins. With regard to sediment reworking, the attribute of biodiffusion, i.e. faunal activity causing random and local sediment mixing increased in the STD, whereas “epifauna”, i.e. faunal activity limited to surficial sediment only, decreased.

4. Discussion

4.1. Tailings gradient

The sediments in the deposit area were heavily influenced by the discharges of the fine limestone tailings. During sampling, the sediments were observed to have a yellow-grey, fine-grained, sticky, and highly compact structure. The grain-size analyses verified that these sediments consisted almost exclusively of silt and clay (pelite) with only very small fractions of coarser material (low values for sorting and kurtosis) (Table 3). Generally, low particle heterogeneity is a

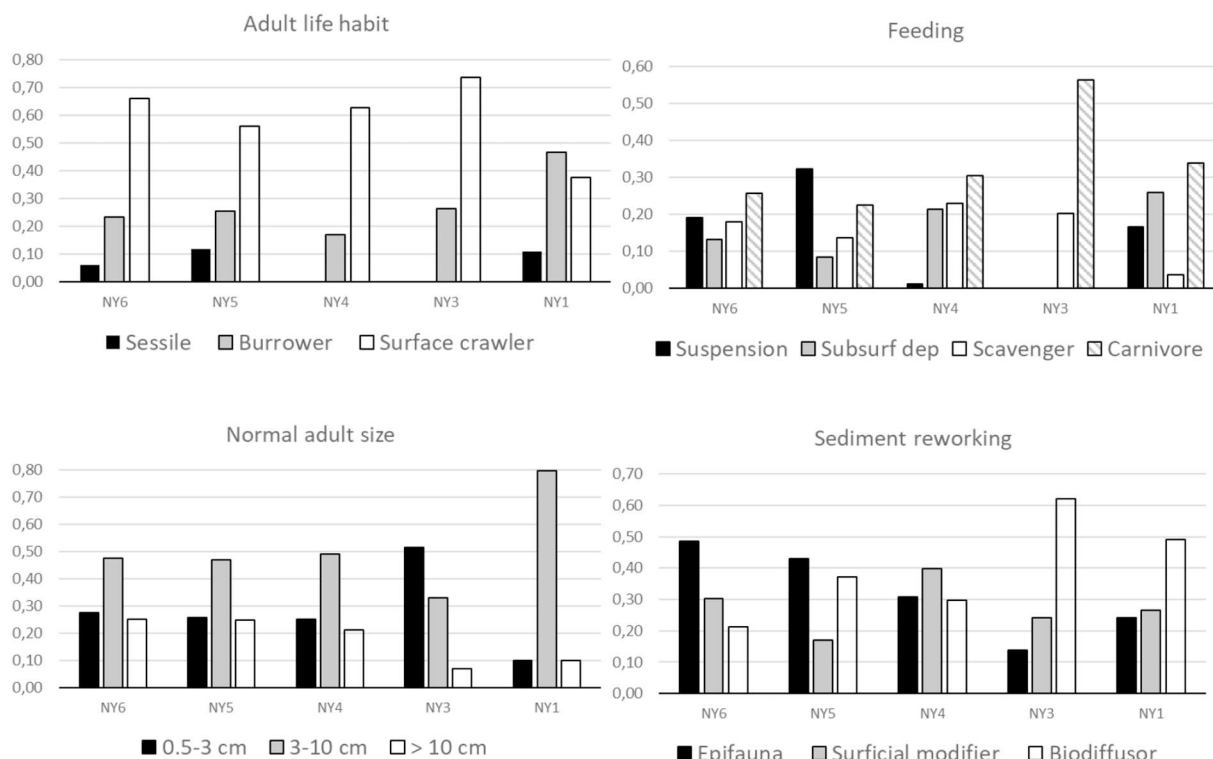


Fig. 10. Relative importance of functional features (modalities) for the traits “adult life habit”, “feeding”, “normal adult size” and “sediment reworking” for epifauna at each station 2016 (the most dominant functional features within each trait are presented). Data from replicate samples have been summed for each station.

characteristic of mine tailings (Morello et al., 2016). In the mixing zone and outer part of the fjord, the sediments were more normal with significant amounts of sand and coarser particle fractions.

With regard to the organic matter, total organic carbon (TOC) was lowest at the station outside the sill (NY7), and highest at the second most impacted station (NY4) (Table 3). However, as the tailings were composed of limestone it cannot be excluded that the TOC-analysis may have incorporated some inorganic carbon, which was assumed to be the case in an analysis of the Hustadmarmor-tailings (Trannum et al., 2018). Thus the pattern in TOC should not be given too much weight. Further, it is also difficult to draw any conclusion from the content of total nitrogen (TN), which was below detection limit at all stations. Coarser sediments naturally have lower content of organic matter, which can explain the low TN-content outside the fjord sill. Inside the fjord, on the other hand, larger concentrations were expected, i.e. in an adjacent fjord the TN-content ranges from 1.8 to 4.4 mg/g (Borgersen et al., 2017). Tailings are in general assumed to have a low organic matter content (Shimmield et al., 2010), and in an analysis of tailings from Hustadmarmor as well as two other mines, total nitrogen was below detection limit in all tailings in contrast to a natural fjord sediment (Trannum et al., 2018; Trannum pers. com.).

4.2. Faunal gradient

Not unexpectedly, species composition and diversity of the benthic fauna changed markedly along the tailings gradient from the outer to the inner fjord. The species richness decreased for both faunal groups, in particular for epifauna where only a few species were recorded close to the outfall. In contrast, the infauna showed an increase in total abundances due to dominance of small bivalves and annelids. The increase may represent a change in the fauna with prevalence of opportunistic species that are particularly adapted to hostile or physically stressed environments. In studies of tailings-impacted fjords conducted elsewhere, similar gradients in infauna with dominance of opportunistic species have been recorded (Ellis and Hoover, 1990; Trannum and Vögele, 2001; Josefson et al., 2008; Berge et al., 2011; Schaanning et al., 2019).

The different responses of epifauna and infauna may be related to their living modes. Whereas the epifauna may be particularly vulnerable to disposal of particles because they live where the tailing deposit builds up and the sediment surface is constantly disturbed, the infauna may be protected by living buried in the sediment. Studies by Atkinson et al. (2011) and Fleddum et al. (2013) have shown that epifaunal assemblages responded more pronounced to impacts of heavy trawling than infaunal assemblages. Of course, the disturbance factor differs, but similarly to tailings disposal, trawling makes the sediment surface unstable and reduce habitat heterogeneity (Thrush and Dayton, 2002; Puig et al., 2012). Moreover, Jørgensen et al. (2011) proposed that as many epifaunal species feed directly on sinking material or the most recently deposited organic matter on the sediment surface, they may be more sensitive to chemical contamination, and perhaps respond in difference manners and on faster time scales than infauna. Such difference in the response seems to be supported by the present work.

The present finding agrees well with previous studies of infauna in the fjord. Brooks et al. (2015) documented clear changes in the community structure that were related to distance from the outfall and amounts of fine tailings in the sediments. In particular, species diversity (H') decreased in relation to percentage of fine tailings in surface sediments, whereas the proportion of disturbance-tolerant species increased. Brooks et al. (2015) further showed that species diversity was gradually reduced over time, and that the proportion of tolerant species increased most close to the outfall.

4.3. Functional pattern

Corresponding with the changes in species composition along the tailings gradient, there were also profound changes in functional

features. For infauna, especially sessile, tube-living and surface deposit feeders decreased, whereas mobile surface crawling and burrowing carnivores and omnivores increased. For epifauna several similar changes were observed, especially in that sessile permanently attached forms decreased and surface-living forms remained. Moreover, epifaunal suspension feeders were missing in the most impacted area. The functional diversity decreased for both faunal groups, but relatively less than the structural diversity. The functional redundancy essentially did not change much along the gradient.

The reduction of sessile tube-building species is probably the most obvious consequence of the tailings disposal. Tube-builders are generally dependent on finding the appropriate material for constructing their tubes, and therefore exhibit more sediment-specific preferences during settlement (Gray, 1971; Pinedo et al., 2000; Duchêne, 2010). In the present study, the outermost station (NY7) was situated outside the fjord in a more open current-influenced fjord environment that is generally advantageous to sessile tube-building species. The most abundant species at this station was for instance the annelid *Owenia borealis*, which builds solid tubes of rather coarse sand particles and shell fragments. The difference in the abundance of this species from outside to inside of the fjord may have contributed to the strength of the gradient in tube-building, but nevertheless, the reduction inside the deposit area was distinct. Similar functional changes were recorded in the previous monitoring studies (Brooks et al., 2015), as well in other fjords subject to mine tailings disposal, for instance Jøssingfjord (southern Norway) receiving tailings from titanium extraction (Olsgard and Hasle, 1993). Moreover, in a recolonization experiment conducted with mine tailings from the present plant, tube-building annelids were particularly affected (Trannum, pers. com.).

At the innermost station (NY3), almost all deposit feeders were absent, evidenced for both infauna and epifauna. Deposit feeders are generally considered relatively tolerant to pollution (Pearson and Rosenberg, 1978; Gaston et al., 1998; Roth and Wilson, 1998; Nunes et al., 2008). However, these assessments have mostly been based on organic enrichment, or pollutants in combination with enrichment. In the case of mine tailings, the available nutrient material is essentially diluted, also at deeper sediment layers. In the present study, also sub-surface and deep deposit feeders decreased along the tailings gradient, but less strongly than surface deposit feeders (correlation < 0.7 ; not entered in Fig. 6). These results accord well with a previously conducted mesocosm-experiment where all deposit feeders (surface, sub-surface and deep deposit feeders) were strongly affected by tailings (Trannum et al., 2018).

Concurrent with the reduction of sessile forms, there was an increase of mobile species towards the deposit area. Mobile species are by their nature able to move in and out of disturbed patches and may search actively for available resources such as newly settled material, and to escape if the conditions are too hostile. Typically, many highly mobile species are carnivores or omnivores, as also appears from the correlation of these features in the present analyses. It may be assumed that their feeding can be performed without being directly exposed to the tailings. Several forms are also scavengers that may feed on dead organic matter arising from pelagic production in the water column. The main species groups involved in the pattern comprise annelids, crustaceans and some echinoderms. One of the species was the cumacean *Diastylis lucifera*, which was found at high densities in the deposit area (stations NY3, NY4). *D. lucifera* has been categorized in ecological group III (tolerant) in AMBI (AMBI library updated 2017). In a polluted area not far from Frænfjorden, *D. lucifera* was recorded together with other highly tolerant species (Trannum et al., 2011). Furthermore, members of the genus *Diastylis* have been among the first colonizers of defaunated sediments (Brunswig et al., 1976), which concurs well with the present case where the substrate can be more or less barren. Mobile carnivores and omnivores are also known to increase in response to other disturbance factors, e.g. bottom trawling (Kaiser and Spencer, 1994; Demestre et al., 2000; Fleddum et al., 2013) and smelter effluent contaminants (Oug, 1998). This feeding group was the only group

which was not significantly affected by tailings in the mesocosm experiment by Trannum et al. (2018).

Feeding on dissolved matter or with symbionts tended to increase in the deposit area. This can mainly be attributed to a change of species within the bivalve genus *Thyasira*. Most close to the effluent outfall *Thyasira sarsi* was found (station NY3), whereas *Thyasira equalis* was relatively abundant more distant in the deposit area (station NY4). Both species use symbiotic sulfur-oxidising bacteria to exploit sulfides in reducing sediments (Dando et al., 2004; Keuning et al., 2011), but while *T. sarsi* is highly pollution-tolerant and often associated with organic-rich sediments, *T. equalis* thrives in sediment with lower organic content, and little or no H₂S (Keuning et al., 2011). Indeed, H₂S-smell was recorded at one of the most tailings-impacted stations, in line with the dominance of *T. sarsi*.

Both infauna and epifauna were characterized by small species closest to the discharge. For infauna, there was a relative increase in the body size of 3–6 cm further away from the discharge and an increase in the size of 1–3 cm towards the discharge (correlation < 0.7; not entered in Fig. 6). For epifauna, the size 0.5–3 cm dominated at the most impacted station (NY3). Reduced size is a general characteristic of polluted areas (Pearson and Rosenberg, 1978) including physically disturbed sediments (Kaiser et al., 2000; Fleddum et al., 2013). Small size of the species may be a consequence of selective pressures for morphological simplicity and rapid maturation (Ryu et al., 2011). Dominance of small species has been recorded in other fjords influenced by sedimentation (Holte and Gulliksen, 1998), including mine tailings (Olsgard and Hasle, 1993).

Sediment reworking is a key mediator of several important geochemical processes in marine systems (Queiros et al., 2013). This trait is therefore both reflecting species performances as well as activities that modifies the environment (“effect trait”). Processes of sediment reworking, particularly the transport of particles up or down, are important for distribution of tailings, and may also provide a mechanism for incorporating pollutants into the substrate (Bradshaw et al., 2006). In the present study, the category of biodiffusers, which incorporates species performing random and local mixing over short distances, tended to increase in the deposit area. The category also incorporates “gallery biodiffusers”, which refers to species constructing a branched tube system where both local mixing (generally upper part of tube system) and non-local transport (from bottom of tube system) take place (François et al., 2002). In the present study both these groups were represented, e.g. by nephtyid and phyllocid annelids that live in the uppermost sediment, and “gallery biodiffusers” like glycerid annelids that make deep tubes. These species are also carnivores, and may thus be protected from the most adverse impacts both by their feeding and by hiding into the sediment.

The functional diversity (FD) decreased along the tailings gradient, but not as much as the species diversity (*H'*). This suggests that rather many functions were still present in the impacted and species-poor areas, which particularly was the case for the second most impacted station (NY4). This was also evidenced by the lower functional redundancy for epifauna than for infauna, although comparisons may be unreliable because of the low species numbers. Species assemblages with low functional redundancy are generally interpreted to be sensitive to species loss (functions get lost with loss of species) (Liu et al., 2019), whereas high redundancy would indicate that the ecosystem functions are robust to changes in diversity (Micheli and Halpern, 2005).

4.4. Faunal impact factors

The tailings deposition initiates a distance gradient in several sediment characteristics, which all can contribute to the gradual changes in faunal composition. In addition, the natural fjord gradient will also structure the fauna. The outermost station is situated outside the sill, and the fauna may vary from the fauna inside the fjord due to different

hydrographical and sediment conditions. Nevertheless, the faunal changes evidenced in the mixing zone and deposit area clearly point to an effect of the tailings.

Based on the previous monitoring data, Brooks et al. (2015) illustrated that infaunal diversity and several functional features such as permanent tube, adult size and life duration were correlated to the percentage of fine tailings in the sediments. In general, sediment composition is one of the most important structuring variables for the fauna also along natural gradients (Gray and Elliott, 2009). In the present case with very fine tailings, it is also likely that the particles may lead to suffocation and clogging of feeding and respiratory organs (Trannum et al., 2018; Brooks et al., 2019), which may explain the decrease of deposit feeders and epifaunal suspension feeders. Such clogging could possibly also intensify effects of chemical toxicity and particle sharpness. Edged triangular and rectangular shapes were evident observed in a SEM-EDX-analysis of microstructure of tailings from this plant (Trannum et al., 2018). Sharp tailings may create additional risk for the fauna (Olsgard and Hasle, 1993).

Smothering and the more or less constant disturbance of the sediment surface represent a chronic stressor for the fauna. Olsgard and Hasle (1993) concluded that sedimentation of mine tailings of 3–4 cm y⁻¹ clearly impacted the macrofauna, while sedimentation of 1 mm y⁻¹ did not result in any observable effects. It is also likely that infauna and epifauna may have different tolerance to this stressor in particular, which may underlie some of the difference in sensitivity between these two groups in the present study. At least in the context of dredging, studies have shown that infaunal species may be able to escape > 10 cm of burial (Jackson and James, 1979; Maurer et al., 1982; Bellchambers and Richardson, 1995), whereas epibenthic species are often unable to escape > 1 cm (Kranz, 1974).

Tailings deposition may also reduce sediment heterogeneity (Morello et al., 2016), which was confirmed by the grain size distribution in the present study (Table 3). Reduced sediment heterogeneity can restrict the niche availability and hence faunal diversity (Gray, 1974; Gray and Elliott, 2009). The same factor may reduce sediment permeability, i.e. water movement through the sediment, which can reduce the oxygen content and alter sediment chemistry (Gray and Elliott, 2009; Ramirez-Llodra et al., 2015). In an experimental setup with addition of crushed marble, Näslund et al. (2012) found that oxygen penetration was reduced in the marble-treatments compared to the coarser control-sediment, which supports such explanation. In the SPI-survey of Frænfjorden, the oxygen penetration depth was reduced towards the effluent outfall (Schaanning et al., 2009), and as mentioned above H₂S was recorded in the sediments during field sampling.

Chemical toxicity may also have contributed to the impoverished fauna towards the discharge. The added flocculation chemicals are generally not considered highly toxic, but there has been more concern regarding the flotation chemicals (Brooks et al., 2018; Trannum et al., 2018). The flotation chemical in the present case (FLOT2015) is composed of fatty acids and esterquats. Methyl triethanol ammonium (MTA), the chemical marker for FLOT2015, was recorded in mussels placed in cages at stations NY4 and NY5. Significant biomarker responses were recorded, with most intensity at NY4 (Brooks et al., 2018). The chemical has also been traced in sediment and pore water, with increased concentration at the most impacted station (NY3) (Escudero-Oñate & Ferrando-Climent, pers. com.). Moreover, a toxic response was observed in a sediment-contact assay with the amphipod *Corophium* sp. (Brooks et al., 2019). Thus, the experimental studies indicate a toxic effect of the effluent discharges, which likely can be linked to the benthic community responses as well. It should be noted that the chemical FLOT2015 was introduced in 2014 to replace the previously used chemical (Lilafлот), which was considered less degradable and more toxic. This may perhaps explain why the species diversity (*H'*, ES₁₀₀), as well as the functional diversity, increased from 2015 to 2016 at the stations within the STD. In spring 2015, it may have been too early for the fauna to improve. Also, the degradation of the flotation chemical can have contributed to the

oxygen depletion described above. The tailings themselves are mainly inorganic, and although total organic carbon (TOC) was not very informative in the present case, the finding with total nitrogen (TN) below 1 mg/g at all stations indicates that the nutrient-value for the benthos was low. Thus the indications of degradation of organic matter are mainly interpreted to be caused by the added chemical. The present and previous studies suggest that the ecological effects in the fjord are not only caused by smothering and altered sediment composition, but that also chemical toxicity effects may be involved.

In several aspects, the infaunal responses resemble the gradient described by Pearson and Rosenberg (1978) with regard to organic enrichment; the total abundance increased close to the discharge due to a peak of opportunistic species. The biomass was also slightly higher at the most impacted station (NY3) compared to the second most impacted station (NY4). Moreover, for the stations inside the fjord, the number of species was slightly higher at NY4 than the station in the mixing zone (NY5), although this should not be given too much weight. Thus, this finding proposes that the SAB-curve in Pearson and Rosenberg (1978) can be valid also with regard to mine tailings. This similarity was also proposed by Olgard and Hasle (1993). In addition, there were similarities regarding the functional responses, particularly with the relative increase in mobile surface crawling and carnivore/omnivore species towards the outfall. Pearson and Rosenberg (1978) also described a progressive simplification of trophic variety in response to increasing enrichment, which seems to be valid with regard to mine tailings indicated by the gradient in functional diversity. However, there are some notable differences; while deposit feeders increase with increasing disturbance in the classical model, they were reduced in the present case, probably due to a reduction of available organic matter in the sediment in contrast to sediments subject to organic enrichment. Also the reduction of tube-builders towards the outfall contrasts with the classical model, presumably due to the change in the sediment composition and physical disturbance. These differences again points to the added value traits analyses can represent in a pollution assessment.

4.5. Implications for environmental monitoring of STD-impacted fjords

In standard environmental monitoring, assessment of the ecological status is based on various diversity indices calculated from an infaunal species list. The present study demonstrates that such assessment may underestimate effects, and that epifauna can represent a more sensitive tool. Already in 1978, Pearson and Rosenberg stated that detailed studies of epifauna in the context of pollution were strongly needed (Pearson and Rosenberg, 1978), but still this ecosystem compartment is far less studied than the infauna. At the same time, neither epifauna nor infauna alone is considered a sufficient surrogate for the community as a whole. Jørgensen et al. (2011) suggested that benthic monitoring ideally should be carried out by combining both techniques, which is supported by the present work. A monitoring programme including the most vulnerable ecosystem compartment is also according to the precautionary principle.

Another improvement would be to include biological traits analyses in regular environmental monitoring, which is also recommended in other studies (Elliott and Quintino, 2007; Rand et al., 2018; Liu et al., 2019). Indeed, increasingly more studies emphasize the functional importance of individual species, rather than species diversity, in mediating processes that are needed to maintain efficient and productive ecosystems (Diaz et al., 2006; Karel et al., 2008; Oug et al., 2012; Fleddum et al., 2013; Gagic et al., 2015; Beauchard et al., 2017). In the present case, the traits-analysis identified which ecological functions were most sensitive to mine tailings, and notably these were not necessarily the same functions as for other forms for disturbances. As long as traits information is available from accessible databases, the analyses are not highly time-consuming, and such supplement could be performed without increasing monitoring budgets significantly.

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