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Predation of intertidal infauna on juveniles of the bivalve *Macoma balthica*

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Abstract

Juveniles of the bivalve *Macoma balthica* live on tidal flats in the Wadden Sea. This study examined the interaction of *Macoma* with the infaunal polychaetes *Arenicola marina* and *Nereis diversicolor* and the gastropod *Retusa obtusa*. The distribution of *M. balthica* spat on the flats, shortly after settlement in April, showed a positive correlation with the *Arenicola* distribution and a negative correlation with *Nereis* distribution. There were no locations where *Macoma* spat and *Retusa* occurred together. In August, *Macoma* spat had grown too large for predation by intertidal infauna. Small individuals of *Macoma* spat were found in stomachs of *Arenicola* (0.14 worm^{-1}) and *Nereis* (0.05 worm^{-1}). Laboratory experiments showed that *Nereis* and *Retusa* could reduce *Macoma* spat abundance, both in the absence and presence of sediment and alternative prey. *Arenicola* reduced the abundance of small *Macoma* ($<1 \text{ mm}$) in sediment without, but not with, alternative prey. In field experiments, we manipulated the density of *Arenicola* in $0.25\text{--}1 \text{ m}^2$ plots and of *Nereis* in 0.03 m^2 cages and examined the effect on *Macoma* density several weeks later. We found a significant negative relation between densities of polychaetes and *Macoma* spat for both polychaete species in these experimental plots. Peculiarly, we found a significant positive relation between manipulated *Nereis* density and adult *Macoma* density in the cages; we cannot explain this. Consumption rates, calculated both from stomach contents and from field experiments, were 45 to $102 \text{ Macoma m}^{-2} \text{ d}^{-1}$ for *Arenicola* and 5 to $116 \text{ Macoma m}^{-2} \text{ d}^{-1}$ for *Nereis*. These values are higher than recorded consumption rates by epibenthic predators in the same area. Nevertheless, between-year differences in year-class strength could not be explained by differential abundance of these polychaetes. In conclusion, *Arenicola* and *Nereis* had a negative effect on the abundance of *Macoma* $<1.5 \text{ mm}$, which was at least partly caused by direct consumption. *Retusa obtusa* can eat juvenile *Macoma*, but probably did not so in the study area, because there were no locations where *Retusa* and *Macoma* spat occurred together in the period that *Macoma* was $<2 \text{ mm}$. © 2002 Elsevier Science B.V. All rights reserved.

Keywords: *Macoma balthica*; *Nereis diversicolor*; *Arenicola marina*; Spat; Predation; Enclosure; Tidal flat; Wadden Sea; The Netherlands

1. Introduction

Juvenile shellfish such as *Macoma balthica*, *Cerastoderma edule* and *Mya arenaria* are smaller than $300 \mu\text{m}$ at settlement after a pelagic larval stage.

These young macrobenthic animals thus belong to the meiobenthos and have to cope with other conditions than the older and larger stages. The effect of infaunal polychaetes on meiofaunal and juvenile macrofaunal abundance has been studied in many locations. Infaunal polychaetes had little effect in some studies (Reise, 1979; Kennedy, 1993) and a large impact in others (Commuto, 1982; Ambrose, 1984a,b;

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Commito and Shrader, 1985; Ronn et al., 1988; Desroy et al., 1998; Tita et al., 2000). Negative effects of polychaetes can be divided into direct effects due to predation and indirect effects due to disturbance. Deposit-feeding infauna usually ingest, move and disturb large amounts of sediment. The number of meiofaunal organisms that is killed by such disturbance can be much higher than the number that is actually eaten (Tita et al., 2000).

Macoma balthica is one of the most common and widespread bivalves in the intertidal and subtidal Wadden Sea. *Macoma balthica* spawns in early spring (late March and April) (Honkoop and Van der Meer, 1997). The eggs and larvae are pelagic for a short period and then, at a length of 190 μm to 300 μm , settle mainly on the low tidal flats (Günther, 1991; Armonies and Hellwig-Armonies, 1992). They can reach a length of 4–7 mm in September of their first year (Armonies, 1996).

Two major groups of predators on *Macoma* are generally acknowledged: wading birds (Hulscher, 1982; Zwarts and Blomert, 1992) and epibenthic organisms such as shrimps, crabs and fish (Reise, 1978; Van der Veer et al., 1998; Hiddink et al., 2002). There are, however, also many indications of predation and/or disturbance by infaunal invertebrates on juvenile bivalves (Sarvala, 1971; Reise, 1979; Ratcliffe et al., 1981; Berry, 1988; Flach, 1992; Thiel and Reise, 1993; Olafsson et al., 1994; Cummings et al., 1996). So far, effects of infaunal predation and disturbance on *Macoma* in the Wadden Sea have not been examined quantitatively.

Infauna is generally very numerous. For example, the large deposit-feeding polychaete *Arenicola marina* may reach densities of over 45 m^{-2} (Flach and De Bruin, 1993) and the omnivorous scavenger *Nereis diversicolor* 400 m^{-2} (Essink et al., 1998a). In comparison, densities of the most abundant epibenthic predator, the shrimp *Crangon crangon*, are generally < 100 m^{-2} (Beukema, 1992; Van der Veer et al., 1998), and average densities of waders are several orders of magnitude lower at about one per 10 000 m^2 (Zwarts and Wanink, 1984). Since many species of infaunal animals are so numerous, even a low consumption per predator may heavily affect the abundance of *Macoma* spat, and thus the later year-class strength of adults.

The question we want to answer in this study is whether infauna affects densities of juvenile and adult

Macoma. The study was primarily aimed at the effects of predation.

We examined the interaction of juvenile *Macoma* with the lugworm *Arenicola marina*, the ragworm *N. diversicolor* and the gastropod *Retusa obtusa*. *Arenicola* is a large deposit-feeding polychaete that lives in U-shaped burrows in intertidal areas. On the head side of the burrow, surface sediment descends through a funnel and is ingested (Retraubun et al., 1996). A few hours after digestion, the sediment is deposited as a cast at the surface through the tail shaft. The worm ingests vast quantities of sediment every day. It is estimated that a large *Arenicola* may ingest 80 cm^3 of sand per day (Cadée, 1976). *Arenicola* is known as a bioturbator, disturbing other infauna such as the amphipod *Corophium volutator* and small bivalves (Flach, 1992). Flach and Tamaki (2001) found a negative correlation between adult *Arenicola* and juvenile *Macoma* density on tidal flats of the Wadden Sea. This correlation was strongest from July to September, suggesting that post-settlement processes cause this correlation.

Nereis is an omnivorous scavenger. It lives in a burrow in the sediment, where it can use three different food-searching strategies (Tita et al., 2000). While deposit feeding, the worm swallows surface sediment unselectively, and the larger part of the gut is filled with inorganic sediment. *Nereis* can also filter feed, by pumping water through a mucous web. This net-bag is swallowed with the trapped particles afterwards (Riisgard et al., 1992; Vedel et al., 1994). The third feeding mode is active hunting on meiofauna from the burrow. *Nereis* is known to prey on spat of the cockle *Cerastoderma edule* and *Macoma* both in the field and in the laboratory (Reise, 1979, 1985; Ronn et al., 1988).

The small predatory gastropod *Retusa* (up to 6 mm shell length) lives in the top layer of the sediment. Its main foods are the gastropod *Hydrobia ulvae* and Foraminifera. Prey is ingested whole (Stamm, 1995). Mean shell length of ingested *Hydrobia* was 1.37 mm with a maximum of 1.9 mm in a study by Berry (1988). *Retusa* can eat *Macoma* in laboratory experiments (Ratcliffe et al., 1981).

As burying depth is positively related with *Macoma* size, adults are probably less vulnerable to sediment disturbance by infauna than juveniles. Because infaunal predators are relatively small, it seems unlikely that the older *Macoma*, which usually reach a shell length

of 5 mm within one year, are vulnerable to infaunal predation.

The intertidal distribution of *Macoma* was compared with the distribution and density of the predators to examine which species showed overlapping distributions. The effect of these species on both 0-group and older *Macoma* was studied with stomach content analysis and in laboratory and field experiments. From stomach content analysis and predator density manipulations predation/disturbance rates in the field were estimated. From these studies, we quantified the effect of these predators on the density and distribution of *Macoma*.

2. Methods

2.1. Study area

Our study was carried out at the Groninger Wad, which is part of the Lauwers basin, in the eastern Dutch Wadden Sea (6°31'E, 53°26'N) (Fig. 1). The average tidal range is 2.4 m. The tidal flat area ranges from -1.4 to +1.0 m NAP (Dutch ordinance level); saltmarshes occur at the higher levels. Average high water (MHW) is at +1.0 m, mean low water (MLW) at -1.4 m NAP. On the salt marsh side, brushwood groynes of former landreclamation works enclose the tidal flat area above +0.3 m NAP. Sediments range from fine sand to fine silt. The area studied (7.9 km²) was enclosed by salt marsh on the south side, the shallow shipping gully to Noordpolderzijl on the eastern side, the deep (10 m) tidal channel Zuidoost-Lauwers on the northern side and partly by another shallow tidal channel on the western side.

Abundant species in the study area are the bivalves *Macoma balthica*, *Cerastoderma edule*, *Mya arenaria* and *Scrobicularia plana*, the gastropod *Hydrobia ulvae*, the polychaetes *Nereis diversicolor*, *Arenicola marina*, *Tharyx marioni*, *Heteromastus filiformis*, *Scoloplos armiger* and *Eteone longa* and the crustaceans *Crangon crangon* and *Carcinus maenas* (Tydeman, 2000).

2.2. Collection of experimental animals

All animals for field and laboratory experiments were collected at the Groninger Wad. *Macoma* spat

was retrieved from the top layer of the sediment, on locations where spat was abundant. The samples were sieved over 1000, 500, 300 and 125 µm and the fractions were sorted under a binocular microscope. *Macoma* was stored in sediment in a dish with a shallow layer of aerated seawater. The size range studied was 0.3 to 2 mm for 0-group and 5–20 mm for 1+ group *Macoma*. *Nereis* and *Arenicola* were excavated with a spade and intact worms were picked out of the sediment by hand. The polychaetes were stored in the laboratory in buckets with a 10 cm layer of sediment from the field, placed in an aquarium with running seawater (S = 30). *Retusa obtusa* was collected by sieving the upper sediment layer in the field over 1 mm. They were stored in the laboratory in small containers with a 2 cm layer of sediment from the field.

2.3. Field distribution

On 10–14 April and again on 23–25 August 2000 the densities of *Macoma* spat, 1+ group *Macoma*, *Nereis*, *Arenicola* and *Retusa* were measured at 57 intertidal stations at the Groninger Wad (Fig. 1). *Macoma* spat was sampled in April by coring 5 times with a 4.6 cm² corer to a depth of 2 cm. Samples were sieved through 1000, 500, 300 and 125 µm mesh. The fractions were sorted at 6–25 × magnification under a binocular dissecting microscope. *Nereis*, *Retusa* and 1+ *Macoma* were sampled by coring 5 times with an 83 cm² corer to a depth of 15 cm in April and once with an 83 cm² corer to a depth of 15 cm in August. Samples were sieved over 1 mm and sorted in the laboratory. Per species, the number of individuals was counted. As *Nereis* individuals were often broken, the number of heads per sample was counted. In August, the number of *Macoma* spat was also counted in these samples (1 × 83 cm²). All samples were stored at -25 °C until sorting. The lugworm density was estimated from the number of casts on the sediment in a 0.5 × 0.5 m grid in four locations within 10 m of the station. According to Flach and Beukema (1994) (in March and August) and Farke et al. (1979) (in July), the number of casts underestimates the density of lugworms by on average 6%.

2.4. Stomach content analysis

The guts of 87 *Arenicola* (84 ± 26 mm, 2.7 ± 1.7 g wet weight, WW) and of 120 *Nereis* (55 ± 13 mm)

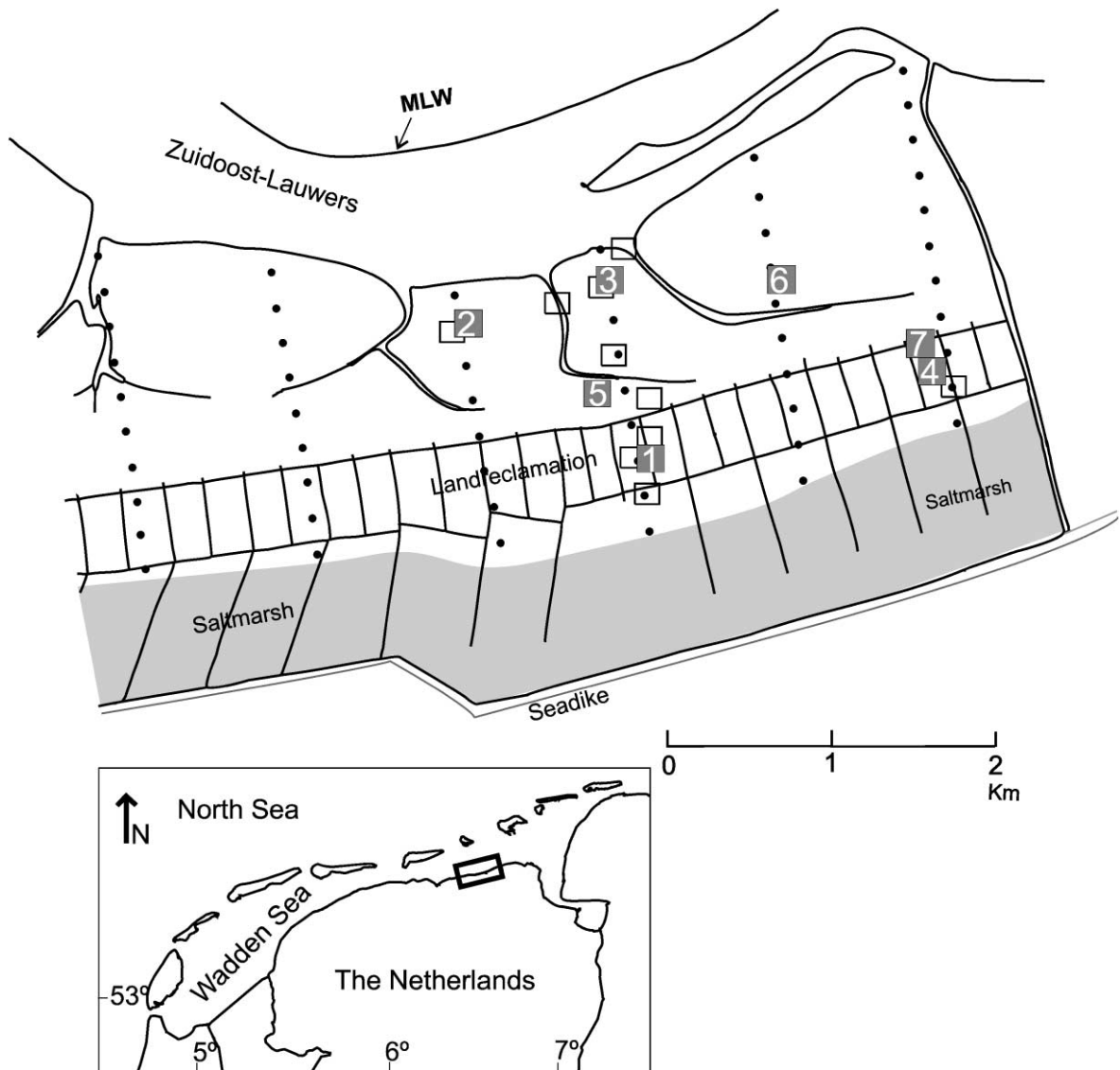


Fig. 1. Map of the study area at the Groninger Wad, in the eastern Dutch Wadden Sea. Locations of sampling stations (●), locations where polychaetes for stomach content analyses were collected (□) and enclosure experiments with location number (■) (see Table 2). MLW = mean low water.

were examined for the presence of *Macoma*. The polychaetes were collected on 15 June 1999, 23 June 1999, 8 July 1999, 26 April 2000, 4 May 2000, 16 May 2000, 25 May 2000 and 4 April 2001 in locations where juvenile *Macoma* was present (Fig. 1). In these locations, the densities of the polychaetes and the bivalve were measured as described in the section 'field distribution'. Polychaetes were stored in 4%

formalin in seawater until analysis in the laboratory. The polychaetes were cut open over their whole length and the gut content was rinsed into a petri dish with water. The whole gut content was analysed under a binocular microscope at 10–40 × magnification. Only *Macoma* that were presumed to be alive at the moment of ingestion were counted. While old shells are white, fresh ones are usually translucent with the

two valves still attached, sometimes with flesh still inside. All *Macoma* were measured with a calibrated micrometer eyepiece.

2.4.1. Gut throughput time

The stomachs of *Nereis* and *Arenicola* are filled and emptied with sediment several times a day. To calculate daily predation rates from stomach contents, we need to know the time it takes prey (and sediment) to pass from the mouth to the anus (the gut throughput time, GTT) (Plante and Mayer, 1996). Cadée (1976) studied the faeces production of *Arenicola* in the Wadden Sea under temperature conditions similar to those on the Groninger Wad. An average *Arenicola* produces 11.0 to 12.8 cm³ of faeces d⁻¹, or 0.45 to 0.53 cm³ h⁻¹ (egestion rate ER). If the volume of sediment in the gut of the worm is known, the throughput time of the sediment can be calculated. The ingestion and egestion rates in lugworms are almost equal, as only a small fraction of the ingested material is digested (Plante and Mayer, 1996).

Arenicola were collected on the tidal flats of the Groninger Wad; we used animals from the same locations and dates as for stomach content analysis. Before processing, the length and WW (after blotting on paper for 10 s) of the animals were measured (79 ± 25 mm, 2.22 ± 1.32 g WW, n=42). They were cut open over their whole length and gut contents were rinsed out with water into a 10 cm³ cylinder with 0.1 cm³ scaling. The sample was left to set for 30 s. The amount of sediment (volume in cm³) was read from the scaling. GTT was calculated from the estimated average volume and literature values of ER (Cadée, 1976) as: GTT (h) = gut capacity (cm³)/ER (cm³ h⁻¹).

We estimated the GTT of deposit-feeding *Nereis diversicolor* in an experiment where we fed the worm sand that was coloured with methylene blue and enriched with dead microalgae (Instant Algae, Reed Mariculture). Sixteen containers were filled with sediment on the tidal flats. *Nereis* was collected and three of them were allowed to bury themselves in each of the containers directly after collection (61 ± 16 mm, n=58). Containers were transported to the laboratory and stored in an aquarium with running seawater in a climate room at 10 or 15 °C. They were left to acclimate for one week. The sediment from the field (with all natural food sources) provided food for the worms in this period.

At the start of the experiment, a 1 mm layer of the enriched blue sediment was spread over the sediment of all containers. For the first 8 h, we sampled one container each hour. After 8 h, we sampled one container every 2 h. The last samples were taken after 24 h. When a container was sampled, the *Nereis* was sieved out over 1 mm and killed by brief immersion in 70% ethanol. Using this method, the worms stopped moving within a few seconds without ejection of gut contents. The length of the worm was measured to the nearest mm and it was then cut into 2 mm pieces, starting at the tail. We measured at what distance from the head (in mm) coloured sediment was found in the guts and calculated this distance as a percentage of the total length. The experiment was carried out at 10 (n=34) and 15 °C (n=25) (normal water and sediment temperatures in April and June) to examine the effect of temperature on the turnover rate. The filling of *Nereis* was calculated as the average of all samples in percentages per h, excluding both non-feeding and thus empty animals and animals that had already filled completely with blue sediment.

2.5. Laboratory experiments

All laboratory experiments were executed in a climate room with a 16/8 h day/night cycle at 10 °C and S=30.

In a pilot experiment, five *Nereis* were added to a 15 (l) × 10 (w) × 13 (h) cm container with a layer of sand and 100 specimens of 0-group *Macoma* for 24 h. Four *Arenicola* were put in a bucket (201 cm²) with a 10 cm thick layer of sediment for 68 h. After this period the stomachs were examined for the presence of 0-group *Macoma* as described above.

Other laboratory experiments with *Nereis* and *Retusa* were executed in 9 cm diameter 1 cm high petri dishes. Experiments with *Arenicola* were performed in plastic containers with dimensions 15 (l) × 10 (w) × 13 (h) cm that were filled with a 7 cm layer of sediment and submerged in an aquarium with running seawater. In all experiments, we used 0-group *Macoma* that were retained on a 300 µm sieve and passed through a 1 mm sieve at the start of the experiment (shell length between 400 and 1400 µm).

We performed three types of laboratory experiments. Table 1 provides details on the number of replicates and duration of the laboratory experiments.

Table 1

Laboratory experiments on the effects of predation by polychaetes and *Retusa* on juvenile *Macoma balthica*. In experiments where sediment from the field was used, an unknown number of *Macoma* were present in this sediment and no *Macoma* were added

| Predator | Experiment type | Sediment | # Predators | Container | <i>Macoma</i> start | Duration (days) | n predator | n control |
|----------------------------|------------------|----------|-------------|-----------------|---------------------|-----------------|------------|-----------|
| <i>Arenicola marina</i> | Azoic sediment | Azoic | 1 | 15 × 10 × 13 cm | 100 | 20 | 6 | 9 |
| <i>Arenicola marina</i> | Alternative prey | Field | 1 | 15 × 10 × 13 cm | Unknown | 13 | 4 | 4 |
| <i>Arenicola marina</i> | Alternative prey | Field | 2 | 15 × 10 × 13 cm | Unknown | 14 | 6 | 4 |
| <i>Arenicola marina</i> | Alternative prey | Field | 2 | 15 × 10 × 13 cm | Unknown | 14 | 6 | 4 |
| <i>Nereis diversicolor</i> | No sediment | None | 2 | Petridish 9 cm | 20 | 3 | 10 | 10 |
| <i>Nereis diversicolor</i> | Azoic sediment | Azoic | 2 | Petridish 9 cm | 20 | 3 | 10 | 10 |
| <i>Nereis diversicolor</i> | Alternative prey | Field | 2 | Petridish 9 cm | Unknown | 5 | 5 | 5 |
| <i>Nereis diversicolor</i> | Alternative prey | Field | 2 | Petridish 9 cm | Unknown | 7 | 5 | 5 |
| <i>Nereis diversicolor</i> | Alternative prey | Field | 2 | Petridish 9 cm | Unknown | 6 | 8 | 8 |
| <i>Retusa obtusa</i> | No sediment | None | 5 | Petridish 9 cm | 20 | 3 | 10 | 10 |
| <i>Retusa obtusa</i> | Azoic sediment | Azoic | 5 | Petridish 9 cm | 20 | 3 | 10 | 10 |
| <i>Retusa obtusa</i> | Alternative prey | Field | 5 | Petridish 9 cm | Unknown | 5 | 5 | 5 |

Generally, the duration of experiments with *Arenicola* was longer than for *Nereis* and *Retusa*, because the experimental units with *Arenicola* contained much more sediment (since lugworms need a thick layer to show their natural burrowing behaviour and cannot forage otherwise). As the amount of sediment was much larger, we expected a smaller effect of *Arenicola* and therefore the duration of experiments was longer. For the same reason, the experimental densities were higher for *Arenicola* than for *Nereis* and *Retusa* experiments (Table 1).

The 'no-sediment' experiments indicate whether the predators were able and willing to eat or kill *Macoma*. 20 *Macoma* and 2 *Nereis* or 5 *Retusa* were put in a petri dish with seawater but without sediment. In controls, no predator was added. This experiment could not be performed with *Arenicola* because this polychaete cannot show its natural foraging behaviour (ingesting sediment) without sediment. The number of remaining live *Macoma* was counted after three days.

In the 'azoic sediment' experiment, we examined whether the predators killed *Macoma* if both prey and predator were buried in the sediment. Experiments with 2 *Nereis* or 5 *Retusa* were done in a 0.5 cm layer of azoic sediment (< 125 µm, without organic material, incinerated at 550 °C for 2 h) with seawater. In controls, no predator was added. 20 *Macoma* were added to each petri dish at the start of the experiment and the number of remaining live *Macoma* was counted after three days. 'Azoic sediment' experiments with *Arenicola* were performed in plastic con-

tainers, which were filled with 7 cm of azoic (all fauna killed by heating to 100 °C for 3 h) sediment, which was enriched with dead microalgae (Instant Algae, Reed Mariculture) to stimulate foraging. 100 *Macoma* were added to each container. The containers were submerged in an aquarium with running seawater. After 20 days, the whole content of each container was sieved over 1000 and 300 µm and the number of live *Macoma* per fraction was counted under the binocular microscope at 10 × magnification.

In an 'alternative prey' experiment, we examined whether the predators killed *Macoma* in sediment that contained abundant alternative prey, such as diatoms, nematodes and foraminiferans. The containers were filled with sediment from the field, from locations where *Macoma* was abundant (25 cm³ of sediment for *Nereis* and *Retusa* (4 mm thick), a 7 cm thick layer for *Arenicola*). This sediment already contained *Macoma* spat (in the size range of 300 to 1000 µm) and alternative prey; no *Macoma* was added to the sediment afterwards. For *Nereis* and *Arenicola*, three of these experiments were conducted, for *Retusa* one (Table 1). As the locations where and the time when the sediment was collected varied between experiments, there was a large variation in the number of *Macoma* present at the start of the experiments. The duration of these experiments varied from 5 to 14 days and was longer for *Arenicola* because the sediment volume was larger in their experimental units. Samples were treated as described for the other experiments.

Arenicola often died or came out of the sediment during the experiments; these treatments were discarded and caused the unbalanced design of some experiments in Table 1.

2.6. Field experiments

In the field experiments, we tried to estimate the impact of *Nereis* and *Arenicola* on the density of 0-group and older *Macoma*. Therefore, we manipulated the density of *Arenicola* and *Nereis* by adding animals to experimental plots on the tidal flats or killing animals in these plots. We examined the effect on the number of *Macoma* after two weeks to two months. We assumed that the number of *Macoma* at the start of the experiment was similar in the plots. Table 2 gives an overview of the locations (see also Fig. 1), period, duration and the applied manipulation (number of polychaetes added) per experiment. We did three consecutive experiments for both *Arenicola* and *Nereis*, but we sampled the third *Arenicola* experiment twice, once for juvenile and once for adult *Macoma*. The second *Arenicola* experiment (locations 2 and 3) was executed in two locations simultaneously and analysed as one experiment.

2.6.1. *Arenicola*

If a lugworm is dug from the sediment and laid down in another location (to experimentally increase densities), it buries itself there. Flach (1992) showed that increased densities of lugworms in experimental plots slowly decrease towards the natural densities, but that differences in densities between treatments remain apparent for months. Densities of *Arenicola* could be reduced by stabbing with a spade on the presumed location of the worm in the sediment, between the cast and the funnel. We tried to do this without severely disturbing the ambient sediment. Plot sizes in the experiments were 0.25 (0.5 × 0.5 m), 0.5 (0.7 × 0.7 m) and 1 (1 × 1) m². In general, we added between –5 (removal) and 50 lugworms per plot, all treatments in duplicate, aiming at experimental densities between 0 and 60 m⁻². Table 2 gives the number of worms added per plot. During the experiment, the number of casts in each plot was counted at least twice and the average was used as the lugworm abundance. Only casts of adult *Arenicola* (casts at least 2 cm wide) were counted. The experiments lasted between 13 and 58 days. At the end of the experiment, in two experiments the density of 0-group *Macoma* was determined (locations 1 and 4), in one

Table 2

Field density manipulation experiments. The densities *Arenicola marina* and *Nereis diversicolor* were manipulated, all treatments in duplicate. The number of removed *Arenicola* (stabbed to death) was not known exactly and indicated as negative numbers added. For location numbers see Fig. 1

| Location | 1 | 2 and 3 | 4 ^a | 4 ^a | 5 | 6 | 7 |
|--|--------------------------------------|---------------------|----------------|----------------|----------------------------|---------------------------------|---------------------------------|
| Species | <i>Arenicola marina</i> | | | | <i>Nereis diversicolor</i> | | |
| Type | Plot | Plot | Plot | Plot | Cage | Cage | Cage |
| Date Start | 22-Jun-00 | 04-Apr-01 | 01-May-01 | 01-May-01 | 26-May-00 | 19-Apr-01 | 03-May-01 |
| Date End | 28-Jul-00 | 17-Apr-01 | 30-May-01 | 28-Jun-01 | 08-Jun-00 | 03-May-01 | 14-Jun-01 |
| Duration (days) | 36 | 13 | 29 | 58 | 13 | 14 | 42 |
| Plot size (m ²) | 0.25 | 1 | 0.5 | 0.5 | 0.03 | 0.03 | 0.03 |
| <i>Macoma</i> age sampled | 0-grp | 0-grp and adults | 0-grp | Adults | 0-grp and adults | 0-grp and adults | 0-grp and adults |
| Natural # of worms in plot | 5 | 15 | 5 | 5 | 4 | 7 | 8 |
| N | 8 | 10 | 11 | 11 | 8 | 12 | 12 |
| Manipulation (# added, treatments in duplicate) | a –2 b 0 c 5 d 10 e f | –15 | –5 | –5 | 0 10 20 50 | 0 10 20 30 40 50 | 0 10 25 40 55 70 |

^a One experiment sampled twice.

experiment only the density of older *Macoma* was estimated (location 4) and in one experiment, the density of both adults and juveniles was determined (locations 2 and 3, Table 2). In the first experiment, the whole top layer (3 cm, 0.25 m²) of the sediment was sieved over 1 mm. In the other two experiments, 0-group *Macoma* was sampled by coring 10 and 16 times with a 4.5 cm² corer, scattered over the whole plot. Samples were sieved through 1000, 500, 300 and 125 µm mesh. The fractions were sorted at 6–25× magnification under a binocular dissecting microscope. Adult *Macoma* were sampled in the second and fourth experiment, by coring 5 and 9 times with an 83 cm² corer and sieving over 1 mm. All samples were stored at –25 °C until sorting.

2.6.2. *Nereis*

Nereis is a motile worm that can and does leave its burrow. The worms were, therefore, enclosed in a cage in density manipulation experiments. The cage was a 19 cm diameter 50 cm long diameter PVC tube and had five 8 cm diameter holes in the sides, which were covered with 1 mm mesh nylon (Fig. 2). The top of the cage, which was difficult to reach for *Nereis*, was covered with 2 mm nylon mesh. The cages were pushed into the sediment until the lower side of the side holes was at the sediment level. No *Nereis* was killed in the experimental plots, as this was not possible without severe sediment disturbance. We used 8 to 12 cages per experiment, to which between 0 to 70 *Nereis* were added, each treatment in duplicate (see Table 2).

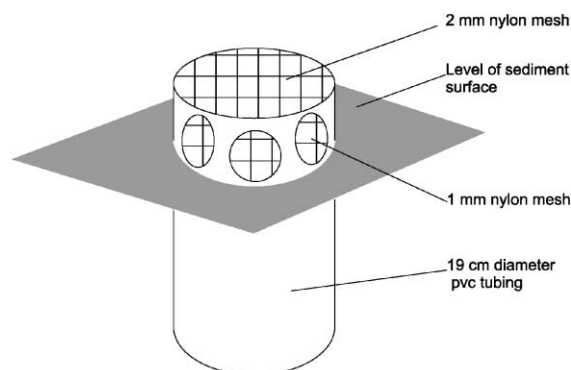


Fig. 2. Construction of the enclosure cage used in the *Nereis diversicolor* density manipulation experiments.

At the end of the experiment, the top layer of the sediment was removed to 2 cm depth and collected in a plastic bag to count juvenile *Macoma*. The rest of the sediment was removed to a depth of 20 cm and sieved over 1 mm to count *Nereis* and adult *Macoma*. We took care to add up the numbers of *Nereis* and *Macoma* from the two different samples. Samples were treated as in the lugworm experiment.

2.7. Statistical analysis

The distribution of *Macoma* was compared with the distribution of the polychaetes and *Retusa* with a correlation of the densities at the 57 sampled stations. For normally distributed data, we used the Pearson's correlation. If the densities were not normally distributed, we used a Spearman's rank correlation.

The laboratory experiments were analysed with an ANOVA or two factor-ANOVA (Type III), with the factors predator presence and sediment presence or experiment number (only if applicable) and their interaction.

We analysed the density manipulation experiments with a General Linear Model (GLM) (type III) (Crawley, 1993) in which experiment number was used as a categorical factor and polychaete density as a quantitative factor. In the GLM we only give the interaction term in the final model if it was significant.

3. Results

3.1. Field distribution

Fig. 3 presents the spatial distribution and densities of 0-group and 1+ *Macoma balthica*, *Arenicola marina*, *Nereis diversicolor*, and *Retusa obtusa* in April and August 2000. In April, *Macoma* spat locally reached densities of over 10 000 m⁻². Nearly all 0-group *Macoma* were found in the 125 µm fraction and thus had passed through the 300 µm sieve. They were concentrated in the low sandy area close to the main tidal channel. Their distribution overlapped with that of *Arenicola* (density 0–30 m⁻² at locations where 0-group *Macoma* occurred, Pearson's correlation coefficient $r = +0.404$, $p = 0.002$) and to a lesser extent with the that of *Nereis* (0–700 m⁻² at locations where *Macoma* occurred, although their densities

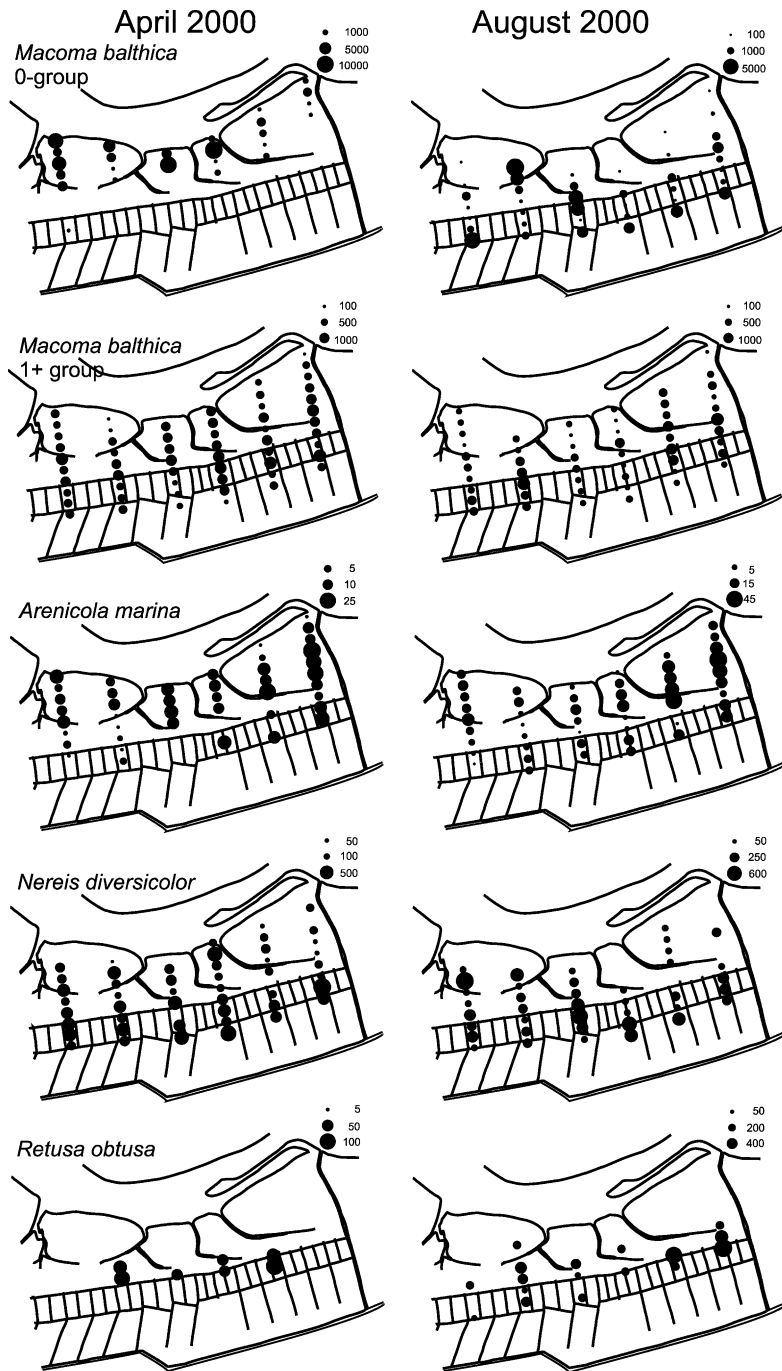


Fig. 3. Distribution patterns of 0-group and 1+ *Macoma balthica*, *Arenicola marina*, *Nereis diversicolor* and *Retusa obtusa* (ind m⁻²) in April and August 2000 at the Groninger Wad. See Fig. 1 for the locations of the sampling stations.

correlated negatively, Pearson's correlation coefficient $r = -0.331$, $p = 0.012$). *Retusa* was rare in April ($0-120 \text{ m}^{-2}$) and concentrated at the border of the former land reclamation works. There were no stations where both *Retusa* and *Macoma* were present (Spearman's rank correlation coefficient $r = -0.313$, $p = 0.018$).

In August, most 0-group *Macoma* were present at stations adjoining the saltmarsh and land reclamation works. The highest *Macoma* density observed was 6506 ind m^{-2} . They had a mean length of $4.2 \pm 1.3 \text{ mm}$. There was a negative correlation between the numbers of *Macoma* and *Arenicola* (Pearson's correlation coefficient $r = -0.277$, $p = 0.037$), but still there were many stations where both *Macoma* and *Arenicola* were numerous. There was a significant positive correlation between the number of *Macoma* and *Nereis* in August (Pearson's correlation coefficient $r = +0.384$, $p = 0.003$). *Retusa* density increased to maximally 1084 ind m^{-2} in August. There were many stations with *Macoma* and without *Retusa* and there was no significant correlation between the densities of *Retusa* and 0-group *Macoma* (Pearson's correlation coefficient $r = +0.080$, $p = 0.55$).

There was a significant positive correlation between 1+ *Macoma* and *Retusa* densities in April 2000 (Pearson's correlation coefficient $r = +0.360$, $p = 0.006$) and a significant positive relation with *Retusa* density in August 2000 (Pearson's correlation coefficient $r = +0.389$, $p = 0.003$). Other correlations were non-significant.

3.2. Stomach content analysis

Fig. 4 shows the length-frequency distribution of *Macoma* found in polychaete stomachs from the field. We found $0.14 \pm 0.51(\text{SD})$ *Macoma* per *Arenicola* stomach (12 *Macoma* found in 87 stomachs). These *Macoma* were between 0.19 and 0.92 mm long with an average of 0.41 mm. In the stomach of *Nereis*, 0.05 ± 0.26 *Macoma* were found (7 *Macoma* found in 120 stomachs). These *Macoma* were between 0.21 and 1.5 mm long with an average of 0.59 mm. Preliminary experiments showed that 43% of *Macoma* between 0.5 and 1 mm died when submerged in fluids from the intestine of lugworms.

The average size of *Arenicola* examined for the volume of the stomach contents was $79 \pm 25 \text{ mm}$ length and $2.22 \pm 1.32 \text{ g WW}$. Cadée (1976) used

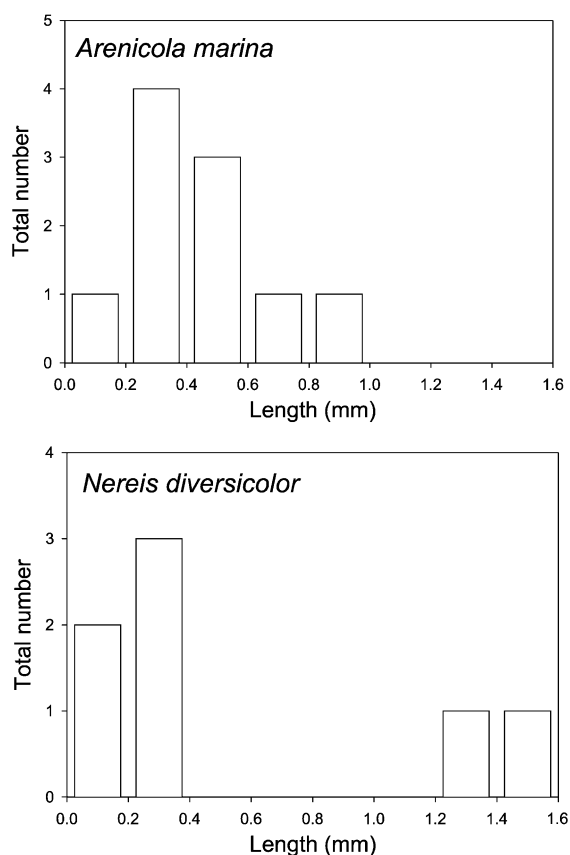


Fig. 4. Length-frequency distributions of *Macoma balthica*, found in the stomachs of (top) *Arenicola marina* and (bottom) *Nereis diversicolor*.

lugworms with wet weights between 1 and 9 g and found that the faeces production only doubled in this weight range. The mean volume of sediment in the intestines of the lugworms was $0.57 \pm 0.54 \text{ cm}^3$. Cadée (1976) found that an average *Arenicola* produced 11.0 to 12.8 cm^3 of faeces d^{-1} . Thus, at a faeces production of $12 \text{ cm}^3 \text{ d}^{-1}$ this means that the complete stomach content is replaced 21.6 times d^{-1} and that the gut throughput time (GTT) is 66 min.

Nereis filled its stomach at a rate of $15\% (\pm 14) \text{ h}^{-1}$. There was no significant difference between the filling rate at 10 and $15 \text{ }^\circ\text{C}$ (t-test, $p = 0.48$). The 15% filling h^{-1} of the stomach equals 3.6 fillings d^{-1} and a GTT time of 6.7 h.

Table 3 gives an order-of-magnitude calculation of the effect of *Arenicola* and *Nereis* predation on the

Table 3

Numbers of *Macoma balthica* ($\pm 95\%$ confidence intervals) in the stomach contents of *Arenicola marina* and *Nereis diversicolor*. From stomach contents, gut throughput times (GTT) and density in the field, mortality rates of *Macoma* were calculated. All values are averages from April to June

| Species | <i>Arenicola marina</i> | <i>Nereis diversicolor</i> |
|---|-------------------------|----------------------------|
| N | 87 | 120 |
| # <i>Macoma balthica</i> worm ⁻¹ | 0.14 \pm 0.11 | 0.05 \pm 0.05 |
| GTT (h) | 1.1 | 6.7 |
| Consumption worm ⁻¹ d ⁻¹ | 3.0 \pm 2.3 | 0.2 \pm 1.0 |
| Density worm (n m ⁻²) | 24 \pm 6.2 | 272 \pm 116.5 |
| Consumption m ⁻² d ⁻¹ | 71 \pm 73 | 53 \pm 408 |
| Density <i>Macoma balthica</i> (n m ⁻²) | 17,064 \pm 11,225 | 18,458 \pm 11,780 |
| % <i>Macoma balthica</i> consumed d ⁻¹ | 0.4% \pm 0.2% | 0.3% \pm 1.3% |

Macoma spat mortality in the field, at the locations where the polychaetes were collected. Although the consumption per worm thus estimated was much lower for *Nereis* (0.2 *Macoma* d⁻¹) than for *Arenicola* (3.0 *Macoma* d⁻¹), the mortality caused by the two species was approximately equal (0.3–0.4% d⁻¹) because *Nereis* densities in the field were much higher.

3.3. Laboratory experiments

In 25 examined *Nereis* from the pilot experiments, we found 8 *Macoma*, which measured 0.97 \pm 0.35 mm with a range of 0.60–1.50 mm. Out of 4 examined *Arenicola* from the pilot experiments, one contained one *Macoma* of 0.57 mm.

3.3.1. *Nereis diversicolor*

3.3.1.1. No sediment and azoic sediment. The number of surviving *Macoma* was significantly lower in treatments where *Nereis* was present than in no-predator treatments (Fig. 5). There was no difference in the number of surviving *Macoma* with and without sediment in the absence of *Nereis*, but in the presence of *Nereis* the number of surviving *Macoma* was significantly higher in the presence of sediment (Table 4A).

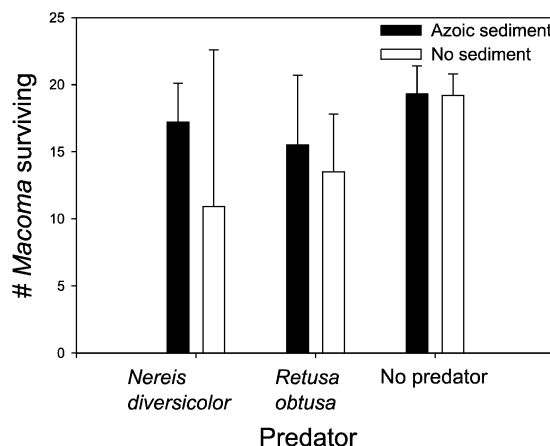


Fig. 5. Numbers of surviving 0-group *Macoma balthica* (+SD) in laboratory predation experiments with *Nereis diversicolor* and *Retusa obtusa*, with and without sediment. All experiments started with 20 *Macoma*.

3.3.1.2. Alternative prey. Experiments with sediment from the field, which contained *Macoma* and many other meiofaunal organisms, showed that the number of surviving *Macoma* was significantly lower in the presence of *Nereis* than without these polychaetes (Table 5A, Fig. 6).

3.3.2. *Retusa obtusa*

3.3.2.1. No sediment and azoic sediment. The number of surviving *Macoma* was significantly lower in

Table 4

Two factor-ANOVA table. Effect of *Nereis diversicolor* and *Retusa obtusa* on the number of surviving *Macoma balthica*, with and without sediment in the laboratory experiment

| Source | Df | MS | F-ratio | p-value |
|--------------------------------------|----|-------|---------|---------|
| A. <i>Nereis diversicolor</i> | | | | |
| <i>Nereis</i> presence | 1 | 371.6 | 19.91 | 0.0001 |
| Sediment presence | 1 | 113.2 | 6.07 | 0.0186 |
| <i>Nereis</i> \times Sediment | 1 | 223.5 | 11.97 | 0.0014 |
| Residual | 37 | 18.7 | | |
| Total | 40 | | | |
| B. <i>Retusa obtusa</i> | | | | |
| <i>Retusa</i> presence | 1 | 153.3 | 10.15 | 0.0029 |
| Sediment presence | 1 | 0.48 | 0.03 | 0.8589 |
| <i>Retusa</i> \times Sediment | 1 | 13.0 | 0.86 | 0.3586 |
| Residual | 37 | 15.1 | | |
| Total | 40 | | | |

Table 5

Two factor-ANOVA table. Effect of *Nereis diversicolor* and *Arenicola marina* on the number of surviving *Macoma balthica* in sediment from the field (with alternative prey) in the laboratory experiment

| Source | Df | MS | F-ratio | p-value |
|---|----|----------|---------|---------|
| <i>A: Nereis diversicolor</i> | | | | |
| Experiment number | 2 | 64,677.0 | 120.06 | 0.0000 |
| <i>Nereis</i> presence | 1 | 4120.6 | 7.65 | 0.0098 |
| <i>Nereis</i> presence \times experiment | 2 | 321.5 | 0.60 | 0.5572 |
| Residual | 29 | 539.7 | | |
| Total | 34 | | | |
| <i>B: Arenicola marina</i> | | | | |
| Experiment number | 2 | 27,468 | 30.07 | 0.0000 |
| <i>Arenicola</i> presence | 1 | 3281 | 3.59 | 0.0713 |
| <i>Arenicola</i> presence \times experiment | 2 | 6992 | 7.65 | 0.0030 |
| Residual | 22 | 913 | | |
| Total | 27 | | | |

treatments where *Retusa* was present than in no-predator treatments (Fig. 5). There was no effect of sediment presence on the number of surviving *Macoma* in the presence of *Retusa* (Table 4B).

3.3.2.2. Alternative prey. *Retusa* ate *Macoma* in the presence of alternative prey (Fig. 7). The number of surviving *Macoma* at the end of the experiment was significantly lower in the presence of *Retusa* (ANOVA, $df=9$, F-ratio = 5.79, $p=0.043$).

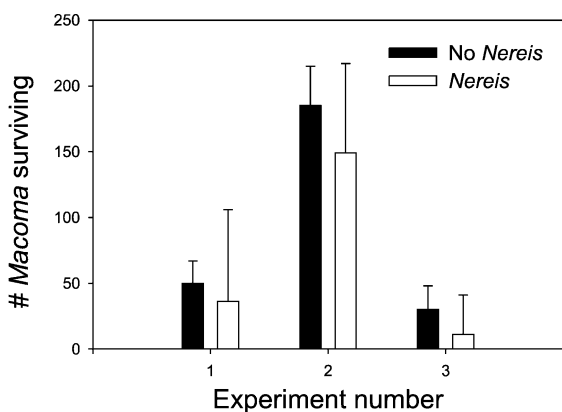


Fig. 6. Numbers of surviving 0-group *Macoma balthica* (+SD) in three laboratory predation experiments with and without *Nereis diversicolor*, in natural sediment (with alternative prey present). The initial numbers of *Macoma* in the sediment varied between experiments and were unknown.

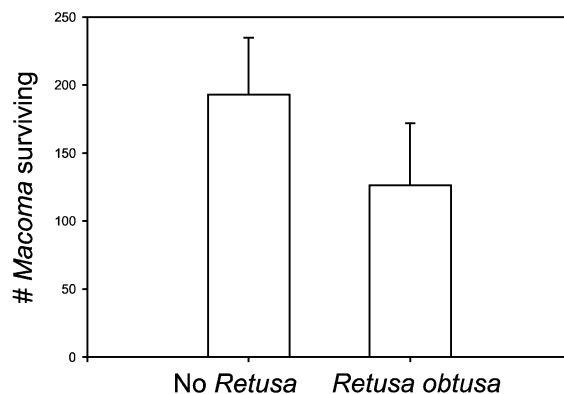


Fig. 7. Numbers of surviving 0-group *Macoma balthica* (+SD) in laboratory predation experiments with and without *Retusa obtusa*, in natural sediment (with alternative prey present). The initial numbers of *Macoma* in the sediment varied between experiments and were unknown.

3.3.3. Arenicola marina

In the absence of *Arenicola*, an anoxic layer of sediment was usually present below the sediment surface, while this layer was never present with *Arenicola*.

3.3.3.1. Azoic sediment. *Arenicola* decreased the number of juvenile *Macoma balthica* in laboratory experiments in the presence of azoic sediment without other prey (ANOVA, $df=14$, F-ratio = 12.11, $p=0.004$) (Fig. 8). The number of juvenile *Macoma* retained on the 1 mm sieve (at the start of the experiment all *Macoma* could pass through this sieve),

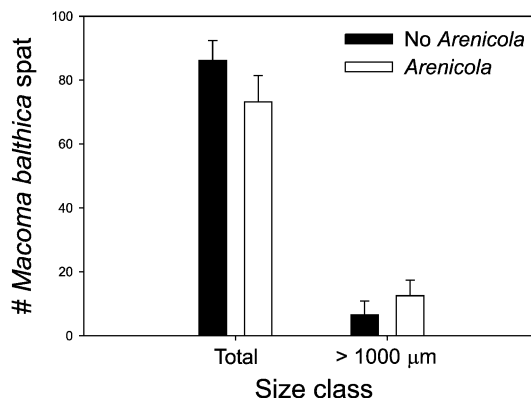


Fig. 8. Number of surviving 0-group *Macoma balthica* (+SD) in laboratory predation experiments with and without *Arenicola marina* in azoic sediment. The experiment started with 100 *Macoma*.

however, was significantly higher in the presence of *Arenicola* (ANOVA, $df = 14$, F-ratio = 6.27, $p = 0.026$).

3.3.3.2. Alternative prey. *Arenicola* did not have a significant effect on 0-group *Macoma* abundance in experiments with sediment from the field containing alternative prey (Fig. 9, Table 5B). In these experiments, there was no effect of lugworm presence on the size of *Macoma*. As the number of *Macoma* at the start of the experiment was unknown and probably different in each container, large variations in these numbers may have affected the outcome of this experiment.

3.4. Field experiments

In the *Arenicola* density manipulation experiments, many *Nereis* (in ambient densities) were present in the plots, while in the *Nereis* density manipulation experiments no adult and only few juvenile *Arenicola* were present in the cages.

3.4.1. *Arenicola marina*

Manipulated lugworm densities ranged from 2 to 54 m^{-2} . There was a significant negative effect of *Arenicola* abundance on the density of 0-group *Macoma* (Table 6, Fig. 10), whereas there was no effect on the number of 1+ *Macoma* (Table 6, Fig. 11). It is remarkable, however, that there was a (just)

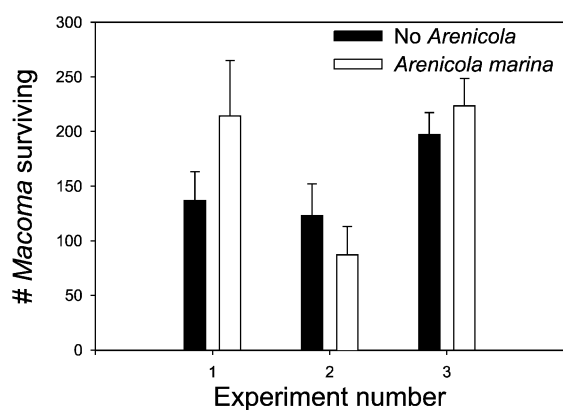


Fig. 9. Number of surviving 0-group *Macoma balthica* (+SD) in three laboratory predation experiments with and without *Arenicola marina*, in natural sediment (with alternative prey present). The initial numbers of *Macoma* in the sediment varied between experiments and were unknown.

Table 6

GLM table. Density manipulation experiments with *Arenicola marina* in the field, effect on the number of surviving 0-group and adult *Macoma balthica*

| Source | Df | MS | F-ratio | p-value |
|---|----|---------|---------|---------|
| <i>0-group Macoma balthica</i> , $R^2 = 0.80$ | | | | |
| Experiment | 2 | 1.64E8 | 35.93 | 0.0000 |
| <i>Arenicola</i> density | 1 | 0.47E8 | 10.44 | 0.0034 |
| Residual | 25 | 0.04E8 | | |
| Total | 28 | | | |
| <i>Adult Macoma balthica</i> , $R^2 = 0.76$ | | | | |
| Experiment | 1 | 907,163 | 58.93 | 0.0000 |
| <i>Arenicola</i> density | 1 | 1388 | 0.09 | 0.7674 |
| Residual | 18 | 15,393 | | |
| Total | 20 | | | |

significant positive relation between the numbers of lugworms and adult *Macoma* in the first experiment (linear regression, $p = 0.044$, $R^2 = 0.37$, numbers of *Macoma* = 5.99 numbers of *Arenicola* + 265).

3.4.2. *Nereis diversicolor*

Densities of *Nereis* were manipulated successfully. Densities in the cages ranged from 100 to 2200 m^{-2} . In the third experiment, at location 7, four out of twelve cages accidentally enclosed crabs with a carapace width of more than 15 mm. In this experiment, both *Nereis* and *Carcinus* had an effect on the 0-group *Macoma* densities, according to the formula # *Ma-*

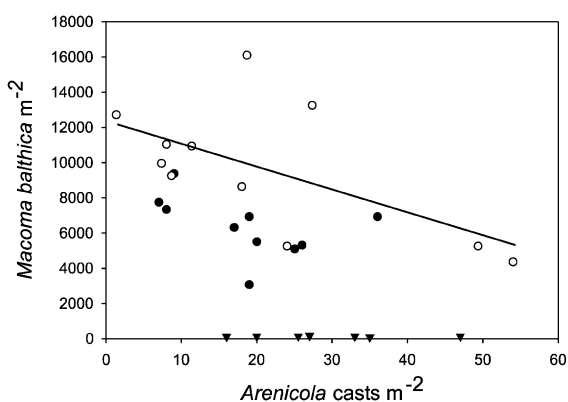


Fig. 10. Relations between the numbers of *Arenicola marina* and 0-group *Macoma balthica* in three density manipulation experiments in the field. ∇ : location 1, \bullet : locations 2 and 3, \circ : location 4. Only significant regression lines are drawn.

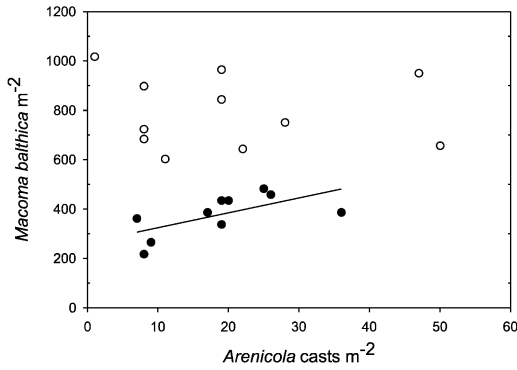


Fig. 11. Relations between the number of *Arenicola marina* and 1+group *Macoma balthica* in two density manipulation experiments in the field. ●: locations 2 and 3, ○: location 4. Only significant regression lines are drawn.

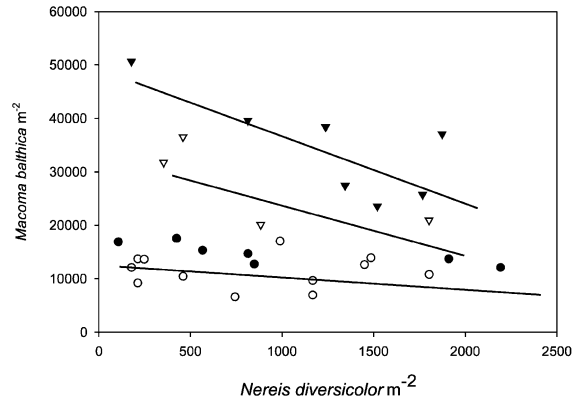


Fig. 12. Relations between the numbers of *Nereis diversicolor* and 0-group *Macoma balthica* in three density manipulation experiments in the field. ●: location 5, ○: location 6, ▼: location 7 without crabs, ▽: location 7 with crabs. Only significant regression lines are drawn.

$coma = 48,705 - 11 \times \# Nereis - 326 \times \# Carcinus$ (Multiple regression, $p = 0.0167$, $R^2 = 0.64$). Thus, during the 42 day study period, one crab ate the same amount of *Macoma* as $(326/11) = 28$ *Nereis*. There was a significant negative effect of *Nereis* abundance on the density of 0-group *Macoma balthica* (Table 7, Fig. 12). In this analysis, the second experiment (at location 7) is divided into treatments with and without crabs and thereby analysed separately. There was a significant positive effect of *Nereis* density on the number of 1+ *Macoma* (Fig. 13, Table 7), but there was no significant effect of experiment number and crab presence.

Table 7
GLM table. Density manipulation experiments with *Nereis diversicolor* in the field, effect on the number of surviving juvenile and adult *Macoma balthica*

| Source | Df | MS | F-ratio | p-value |
|---|----|---------|---------|---------|
| <i>0-group Macoma balthica</i> , $R^2 = 0.87$ | | | | |
| Experiment | 3 | 1.27E6 | 56.28 | 0.000 |
| <i>Nereis</i> density | 1 | 116,596 | 5.15 | 0.032 |
| Residual | 25 | 22,642 | | |
| Total | 29 | | | |
| <i>Adult Macoma balthica</i> $R^2 = 0.36$ | | | | |
| Experiment | 3 | 38.8008 | 1.80 | 0.1737 |
| <i>Nereis</i> density | 1 | 149.011 | 6.90 | 0.0145 |
| Residual | 25 | 21.6034 | | |
| Total | 29 | | | |

By dividing the slope of the regression line (relation number of worms with number of *Macoma*) by the duration of the experiment for the significant experiments, it is possible to estimate the consumption worm⁻¹ d⁻¹. This is necessary to be able to estimate the consumption/disturbance rates, because the duration of the experiments was not equal. Table 8 shows that one *Arenicola* killed 4.24 *Macoma* d⁻¹ in the third experiment. One *Nereis* consumed 0.02 *Macoma*

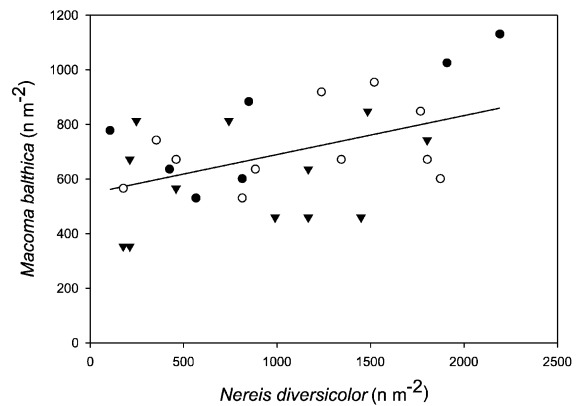


Fig. 13. Relations between the numbers of *Nereis diversicolor* and 1+group *Macoma balthica* in three density manipulation experiments in the field. ●: location 5, ○: location 6, ▼: location 7. Only significant regression lines are drawn.

Table 8

Relation between the numbers of polychaetes and *Macoma balthica* spat in the density manipulation experiments in the field with *Arenicola marina* and *Nereis diversicolor*. ‘Slope’ is the slope of the regression line that described the relation between the numbers of *Macoma* and the numbers of polychaetes. By dividing this slope by the duration of the experiment, the number of *Macoma* that was eaten (or disturbed) per polychaete per day was calculated (only for the significant relations)

| Location | Species | p-value | R ² | Slope | <i>Macoma</i> predator ⁻¹ day ⁻¹ |
|----------|---------------------------------------|---------|----------------|--------|--|
| 1 | <i>Arenicola marina</i> | 0.65 | 0.00 | – 0.92 | |
| 2 and 3 | <i>Arenicola marina</i> | 0.07 | 0.36 | – 117 | |
| 4 | <i>Arenicola marina</i> | 0.03 | 0.41 | – 123 | 4.24 |
| 5 | <i>Nereis diversicolor</i> | 0.03 | 0.64 | – 0.23 | 0.02 |
| 6 | <i>Nereis diversicolor</i> | 0.95 | 0.00 | – 0.10 | |
| 7 | <i>Nereis diversicolor</i> | 0.04 | 0.59 | – 12.6 | 0.30 |
| 7 | <i>Nereis diversicolor</i> with crabs | 0.24 | 0.57 | – 9.3 | |

d⁻¹ in the first and 0.30 d⁻¹ in the third experiment, in treatments without crabs.

4. Discussion

In the period that *Macoma balthica* were smaller than 1.5 mm, a negative effect of *Nereis diversicolor* presence on the abundance of 0-group *Macoma* was observed in field and laboratory experiments. A negative effect of *Arenicola marina* was found in the field, but not in all laboratory experiments.

As small *Macoma* were found in the stomach contents of both polychaetes in the field and laboratory, polychaete predation on *Macoma* is a mechanism that can at least partly explain these density reductions. Above 1.5 mm shell length, the bivalves reached a size refuge. In gut contents of *Nereis* in both laboratory and field, we never found a *Macoma* larger than 1.5 mm. The largest *Macoma* found in *Arenicola* was 0.92 mm. *Arenicola* even had a positive effect on numbers of 0-group *Macoma* larger than 1 mm in one of the laboratory experiments. This suggests that the presence of *Arenicola* increases the growth rate of 0-group *Macoma*, through a so far unknown mechanism.

Arenicola is probably not a real predator on *Macoma*, as predation presupposes an active and selective searching for prey. *Arenicola* feeds on sediment in bulk and inadvertently ingests *Macoma* spat, which have the same size as the sand grains. It does not matter whether *Macoma* dies after active selection or after bulk feeding on sediment, it dies anyway.

Retusa ate *Macoma* spat in laboratory experiments, both with and without sediment, but the number of

Macoma eaten by one *Retusa* was low. As there was no overlap in the distribution of *Retusa* and *Macoma* spat in April, *Retusa* cannot have been a predator on *Macoma* spat in the field. In August, there was overlap in distribution and *Retusa* densities had increased. Since *Retusa* swallows its prey whole (Stamm, 1995) and the largest *Hydrobia ulvae* that have been recorded in *Retusa* stomachs measured 1.9 mm (Berry, 1988), we expect no predation on *Macoma* larger than 2 mm. Therefore, in August *Macoma* was too large (4.2 mm) for *Retusa* predation.

The estimates of the magnitude of consumption were comparable in field experiments and stomach content analysis, both in *Arenicola* and *Nereis*. For *Arenicola*, from stomach content analysis, we expect a consumption of 71 *Macoma* m⁻² d⁻¹ at the average density of 24 *Arenicola* m⁻² (Table 3). From field experiments, we expect a consumption of 102 *Macoma* m⁻² d⁻¹ at this lugworm density. For *Nereis*, from stomach content analysis, we expect a consumption of 53 *Macoma* m⁻² d⁻¹ at the average density of 272 *Nereis* m⁻² (Table 3). From field experiments, we expect a consumption of 5 to 116 *Macoma* m⁻² d⁻¹ at this ragworm density (Table 8).

In some of the experiments (especially in the field), the effects of predation/ingestion and disturbance could not be viewed separately from each other. The sediment reworking activities of some infaunal invertebrates, such as the lugworm (Flach, 1992), may cause disturbance or death to *Macoma* and other small benthos (Hunt and Scheibling, 1997). As *Macoma* is very mobile during its early life history (Armonies, 1994), disturbance may stimulate the small benthos to migrate away from locations with many polychaetes (Flach and De Bruin, 1994). This increased migration

can possibly explain the reduced number of *Macoma* in the experiments with *Arenicola*, a very active bioturbator, as *Macoma* was not enclosed in cages in these experiments. However, as calculated above, the stomach contents of *Arenicola* can explain 70% of the disappearance of *Macoma* in field experiments and therefore killing by ingestion seems to be the major cause of mortality by lugworms.

As *Macoma* spat can stand burial in anoxic sediment for 10 d without mortality (Elmgren et al., 1986), we do not expect that burial under *Arenicola* casts had any adverse effects on *Macoma* spat.

Other authors have also described adverse effects of polychaetes on juvenile bivalves. Flach (1992) manipulated *Arenicola* densities in the field and found a negative correlation between densities of bivalve spat (*Cerastoderma edule*, *Macoma balthica*, *Angulus tenuis*, *Mya arenaria*, *Ensis spec.*) and lugworm densities, although the numerical effects were much smaller than in the current study. Flach assumed that disturbance (especially by the funnels) was the mechanism explaining reduced densities. The priapulid *Halicryptus spinulosus* reduced the number of settling *Macoma* postlarvae in the Baltic. As no *Macoma* was found in the guts of *Halicryptus*, this effect was probably due to disturbance (Aarnio et al., 1998).

We found an unexpected significant positive relation between *Nereis* and 1+group *Macoma* abundance in field experiments. As these experiments were conducted in cages that did not allow migration of adults in and out of the experimental plots (meshes of maximally 2×2 mm), this effect cannot be explained by selective im- or emigration. Therefore, the lower densities of large *Macoma* at low experimental *Nereis* densities and vice versa must be explained by a higher mortality at low *Nereis* abundance, for which we know no mechanism. The two highest points in Fig. 13 play a major role in causing the positive relation between *Macoma* and *Nereis* densities. Without these two data points, no relation between *Nereis* and adult *Macoma* density exists. Therefore, we believe this relation to be a Type I error (though with a very low probability, $p=0.0087$).

From field experiments, it also became clear that polychaetes were not the only factor affecting *Macoma* spat densities. For example, in the third *Nereis*-enclosure experiment, *Nereis* and crabs inside the cages consumed substantial numbers of *Macoma*. Since the

density outside the cages ($319\text{--}391\text{ m}^{-2}$) where all predators were present was lower than inside the cages where crabs and *Nereis* were present ($593\text{--}1433\text{ m}^{-2}$), there were other factors, besides crabs and *Nereis*, that caused a reduction in spat densities that we did not cover in this experiment.

In many other studies, crustacean predation is considered very important for bivalve spat abundance (Beukema et al., 1998; Van der Veer et al., 1998; Strasser and Günther, 2001; Hiddink et al., 2002). Combined, the shore crab *Carcinus maenas* and the brown shrimp *Crangon crangon* consumed $7.3\text{ Macoma m}^{-2}\text{ d}^{-1}$ on the low tidal flats and $2.7\text{ Macoma m}^{-2}\text{ d}^{-1}$ on the high tidal flats at the Groninger Wad (Hiddink et al., 2002), while polychaete consumption can be higher than $100\text{ m}^{-2}\text{ d}^{-1}$ (Table 3). A further indication of the importance of polychaete predation comes from the field experiments: in the enclosure experiment one small crab consumed the same amount of bivalves as 28 *Nereis*. The ratio of the densities of these two species was, however, more like 1/175, showing the numerical importance of *Nereis* predation. Thus, the number of *Macoma* killed by polychaetes seems to be higher than the number eaten by epibenthic crustaceans, especially in the period that the spat is smaller than 1 mm (Strasser et al., 2001).

Year-class strength of *Macoma* is determined somewhere between the egg stage in early spring and the post-settlement stage next August (Honkoop et al., 1998). Egg production and shrimp predation can partly explain the year-to-year variability in *Macoma* recruitment (Beukema et al., 1998). Based on the present study, it can be hypothesised that polychaetes play a larger role in determining the year-class strength of *Macoma* than the epibenthos, because they consume large amounts of bivalves shortly after settlement.

Average densities per year from the long term-monitoring of the macrobenthos on five stations on the Groninger Wad by RIKZ (Essink, 1978; Essink et al., 1998b) were used to examine the effect of *Nereis* (1969 to 1999) and *Arenicola* (1976 to 1999) on the abundance of *Macoma* spat. Our experiments showed that polychaetes affect *Macoma* spat in spring, but in the monitoring study spat was not present in the spring samples. Therefore, the density of *Macoma* spat in summer and/or autumn (August–November)

was related to polychaete-density in spring of the same year (between February and May). A prerequisite for a between-year correlation between the densities of *Macoma* and the polychaetes is that there should be variation in the densities of both *Macoma* and the polychaetes. As the densities and standard deviations of the average densities per year were 622 ± 667 for *Macoma* spat, 278 ± 165 for *Nereis* and 33 ± 35 for *Arenicola*, there was a large enough variation in densities between years. Nevertheless, we found no relation between the densities of *Macoma* and *Arenicola* or *Nereis* (Spearman's rank correlations, *Macoma-Arenicola* $r = +0.083$, $p = 0.706$, $n = 24$, *Macoma-Nereis* $r = +0.001$, $p = 0.994$, $n = 31$). Therefore, although the polychaetes consume large quantities of *Macoma* spat, polychaete density does not seem to be a major factor determining spat density in autumn.

In the calculation of predation rates from the stomach contents of *Arenicola* and *Nereis*, the values of GTT used were important. For *Arenicola* the GTT of 66 min that we calculated was longer than GTT of 15 and 63 min as calculated by Kermack (1955) and Plante and Mayer (1994), but very similar to values of 1 to 1.5 h calculated by Plante and Mayer (1996). Thus, an underestimation of predation pressure is more likely than an overestimation. The GTT of 6.7 h that we used in our calculations for *Nereis* is slightly shorter than the value found by Masson et al. (1995) (75% of the gut of emptied after 8 h) and may have led to an overestimation of predation pressure.

We can draw the general conclusions that both *Arenicola* and *Nereis* caused a reduction in *Macoma* spat abundance. Effects on densities in the field were expected from stomach content analysis and were also shown in enclosure experiments. Nevertheless, in the area studied the polychaetes were not an important factor that determined the year-class strength of *Macoma*. *Retusa* was no significant predator, because in early spring there is no overlap in distribution with *Macoma* spat, while later in the year *Macoma* probably outgrew *Retusa* mouth size.

Similar effects of polychaetes can be expected in other bivalve species with small benthic spat, such as *Mya arenaria* and *Cerastoderma edule*. We did not find these bivalves in the stomachs of the polychaetes, but that was probably because settlement of these species occurred later in the season. *Retusa* impact on

these species may be larger because, due to the later settlement, periods of spat and *Retusa* abundance matched better than for *Macoma*.

The importance of polychaetes for *Macoma* spat abundance probably depends on the scale examined. In the intertidal, polychaetes seem to be important predators on *Macoma* spat. However, *Arenicola* and *Nereis* are rare or absent in the subtidal Wadden Sea and coastal North Sea (Holtmann et al., 1998; Dekker and De Bruin, 1998) while *Macoma* spat is also very rare at these locations (Hiddink and Wolf, in press). Therefore, we hypothesise that *Macoma* spat lives in the intertidal despite the presence of polychaete predators. The impact of worms in the intertidal is probably much lower than the potential predation pressure of epibenthic crabs and shrimps in the subtidal.

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