Marine Biology
June 2008, Volume 154, Number 4 : Pages 671-682
http://dx.doi.org/10.1007/s00227-008-0960-9

Bacterivory of a mudflat nematode community under different environmental conditions

Pierre-Yves Pascal[1, *], Christine Dupuy[1], Pierre Richard[1], Jadwiga Rzeznik-Orignac[2], Nathalie Niquil[1]

[1] Littoral, Environnement et Sociétés (LIENSS) UMR 6250 CNRS-Université de La Rochelle, 2 Rue Olympe de Gouges, 17042 La Rochelle cedex, France
[2] Biologie des organismes marins et écosystèmes (BOME) UMR-CNRS 5178–USM 0401– MNHN, 61 Rue Buffon, 75231 Paris, France

*: Corresponding author : Pascal P. Y., email address : pypascal@univ-lr.fr

Abstract:

The fate of the benthic bacterial biomass is a topic of major importance in understanding how soft-bottom environments function. Because of their high abundance, production and nutritional value, benthic bacteria may constitute an important food resource for benthic fauna. The trophic role of bacteria for a nematode community on the Brouage mudflat (Marennes-Ole´ron-France), dominated by three species: Chromadora macrolaima (64% of the abundance), Daptonema oxycerca (15%) and Ptycholaimellus jacobi (8%), was determined in grazing experiments using ^{15}N pre-enriched bacteria. On intertidal flats, seasonal, tidal and circadian cycles induce strong variations in environmental conditions. Grazing experiments were performed in order to measure the effects of abiotic (temperature, salinity and luminosity) and biotic (bacterial and algal abundances) factors on assimilation rates of bacteria by nematodes. In order to assess simultaneously bacteria and algal assimilation rates, algal abundances were modified adding ^{13}C pre-enriched Navicula phyllepta. Assimilation rate was significantly lower at 5°C; moreover, general trend shows a prominent temperature effect with an optimum around 30°C. Assimilation at salinity 18 was not significantly different from the assimilation at salinity 31. Assimilation was higher under light conditions than in the dark. Above 10^9 bacteria ml^{-1}, assimilation of bacteria remained unaffected by bacterial abundance. However, assimilation of algae increased with the algal concentration. Nematode kept feeding under conditions of stress, which are typical of the surficial sediment habitat and they appeared to be principally dependent on the algal resource.

Keywords: Nematode, bacteria, grazing, environmental factor, mudflat

Introduction

Bacteria play a major role in cycling organic matter in marine sediments through the remineralization of nutrients and organic matter and production of particulate and dissolved carbon (e.g. Legendre and Rassoulzadegan 1996; Rivkin et al. 1996). Although bacterial abundance remains stable, around 109 cells ml-1 (review in Schmidt et al. 1998), bacterial production rates vary greatly (Sander and Kalff 1993). This discrepancy between abundance and production may be induced by bottom-up, biochemical and top-down control (van Oevelen et al. 2006a). In the top-down control situation, bacterial carbon is assumed to be regulated by higher trophic levels of benthic food webs. The majority of animals feeding on sedimentary deposits are more dependent on attached bacteria than on nonliving organic debris (e. g. Fenchel 1972). Models of benthic ecosystems emphasize the role of bacteria and their immediate grazers as a major route through which organic material is processed (e. g. Kuipers et al. 1981). Consequently, quantitative data dealing with the link between bacteria and benthic fauna are essential for understanding the extent to which this trophic link structures energy and material fluxes in the communities. Results concerning the impact of meiofaunal grazing on bacteria are conflicting (Kemp 1990). Montagna (1984b) suggested that meiofaunal grazing pressure (principally through

polychaetes) represents a significant stimulatory effect on the microbial community and may be important in sandy sediments (Montagna and Bauer 1988). However, quantitative studies on meiobenthos bacterivory are sparse.

Although they are small and inconspicuous, nematodes are consistently the most abundant meiobenthic taxon in mudflat sediments. Their average densities of 10^6 ind m^{-2} represents a biomass of roughly 0.2 to 2 gC m^{-2} and some authors have suggested that their ecological significance is great in terms of food-web relationships (review in Platt and Warwick 1980; Heip et al. 1985).

Benthic bacteria can constitute a significant food source for nematodes (Lopez et al. 1979; Tietjen 1980; Montagna 1984b; Montagna 1995; Moens et al. 1999b). According to Wieser (1960), there are four different feeding groups of nematodes: selective deposit feeders (1A), non-selective deposit feeders (1B), epigrowth feeders (2A) and omnivore-predators (2B). Nematodes of each feeding groups are potential bacterivores, even predacious may benefit directly from bacterial carbon (Moens et al. 1999b). Deposit and epigrowth feeders feed on bacteria and unicellular eukaryotes in different ways. Deposit-feeding species have no teeth and generally swallow the food whole and undamaged. They feed predominantly on bacteria associated with detritus. The epigrowth feeding species puncture the cell membrane with their teeth and ingest only the cell contents (juice feeders). Diatoms and other benthic microalgae are known to be important trophic sources for many epigrowth feeders but the importance of bacteria as a food source remains poorly documented (Moens and Vincx 1997). The aim of this study was to experimentally assess rates of bacteria uptake by a nematode community from an intertidal mudflat using ^{15}N enriched bacteria as tracers. The intertidal habitat studied is subject to a wide range of environmental varying factors. Three relevant time scales drive these environmental variations: long-term (seasonal cycle), medium-term (lunar cycle) and short-term (solar and tidal cycles) (Guarini et al. 1997). Variations concern both biotic (i.e.

temperature, salinity and luminosity) and abiotic factors (i.e. bacterial and algal abundances). As those variations may influence the feeding behavior of nematodes, one aim of the present study is to determine if nematodes bacterivory is constant in the mudflat or influenced by environmental factors. Other aim is to describe feeding behavior of nematodes when an alternative algal resource is available.

For this purpose, a mudflat nematode community from surficial sediment was put in microcosms, in contact with labeled preys: a bacterial community and one algal species. Grazing experiments were performed in order to evaluate effects of abiotic (temperature, salinity and luminosity) and biotic (bacterial and algal abundances) factors on rates of prey uptake. We focused on the surficial mudflat sediment nematode community because the surficial sediment (i) has high bacterial production, (ii) contains the highest nematodes densities and (iii) undergoes faster and more wide-ranging changes in environmental factors than do the deeper layers.

Material and methods

Study site

The Brouage intertidal mudflat is located in the eastern part of the Marennes-Oléron Bay (Atlantic coast of France). Meteorological conditions exhibit a strong seasonality typical of a temperate climate. Temperature and salinity of emerged sediments are more extreme during summer tidal cycles (Guarini et al. 1997). Minimum and maximum mud temperatures are 5°C and 34°C respectively. The maximum daily range of mud temperature due to emersion and immersion cycle reaches 18°C (Guarini et al. 1997). Salinity of overlaying water is controlled by the river Charente freshwater input, ranging from 25 to 35 over the year (Héral et al. 1982). Salinity of the upper layers of sediment may also decrease with rainfall. The sediment surface irradiance shifts from dark during submersion and night emersions to high levels of incident light during daytime emersions. This irradiance can reach 2000 µM of photons m^{-2} s^{-1}

(Underwood and Kromkamp 2000). Details of numerous benthic organisms and processes are available concerning this intertidal zone (gathered in Leguerrier et al. 2003; Leguerrier et al. 2004; Degré et al. 2006).

^{15}N enriched bacteria and ^{13}C enriched algae as tracer

The method used was described in Pascal et al. (2008). This method is based on the assumption that grazers ingest unselectively enriched and natural bacteria. Briefly, one centimeter-depth of surficial sediment was sampled during ebb tide in the Brouage mudflat (45°55N, 1°06W) (Fig. 1). Bacteria from surficial sediment were cultured in a liquid bacterial culture medium containing $^{15}NH_4Cl$ 1 g l^{-1} (99% ^{15}N-enriched NH_4Cl CortecNet), rinsed by centrifugation and frozen until the grazing experiments. An axenic clone of the *Navicula phyllepta* diatom (CCY 9804, Netherlands Institute of Ecology NIOO-KNAW, Yerseke, The Netherlands) was cultured in a liquid medium containing $NaH^{13}CO_3$, then rinsed and freeze-dried until the grazing experiments. Isotopic composition of enriched prey was assessed using mass spectrometer. For these experiments, labeled preys were mixed with sediment from the Brouage mudflat that had been previously sieved through a 50 µm mesh. The abundance of bacteria and algae in the slurry was estimated in order to determine the ratio between enriched and unenriched prey.

Grazing experiments

The top centimeter of sediment was collected on March 13, 2006. At the time of sampling, sediment presented a temperature of 7°C and a salinity of 29. Sediment sampled was first sieved through a 500 µm mesh to remove macrofauna, then through a 50 µm mesh to extract meiofauna. One ml of the sediment remaining in the mesh was put into each microcosm (Pyrex beakers, ø = 4.5 cm) and the fraction which passed through the mesh was mixed with ^{15}N enriched bacteria. This slurry contained 10.5×10^8 bacteria ml^{-1}, with the ^{15}N enriched bacteria being twice as abundant as unenriched ones. Four ml of this slurry were put into each

microcosm. Each experiment was carried out in triplicate. Control samples were frozen (-80°C) in order to kill any nematodes.

A kinetic study was performed to validate its linear or hyperbolic shape in order to calculate the grazing rate. Incubations for this kinetic study were run for 1 to 12 hours, including the 3 hours run that was used for all other experiments. Incubations were made under the following standardized conditions that were close to the year-round mean values recorded on the study site: temperature (20°C), salinity (31), luminosity (darkness), bacterial abundance (10.5×10^8 bacterial cell ml^{-1}) and algal abundance (15 µgChla g^{-1}).

For each other experiment one environmental incubation factor was modified. In order to decrease salinity, cultured bacteria were rinsed with 0.2 µm filtered-sea-water diluted with 0.2 µm filtered water (final salinity of 18). The light effect was tested with a light intensity of 83 µM photons m^{-2} s^{-1}. Bacterial abundances (total enriched and non-enriched) tested were 4, 7 and 17 cells ml wt sed-1 with respectively the following ratio between abundance of total and enriched bacteria: 6.1, 2.0 and 1.3. Algal abundance was modified by adding various quantities of cultured *N. phyllepta* enriched in ^{13}C while bacterial abundances (total enriched and non-enriched) were kept constant at 10×10^8 cells ml^{-1}. Using two isotopes for labeling bacterial (^{15}N) and algal food (^{13}C) offers the opportunity to assess bacterial and algal ingestion rates simultaneously. Algal abundance (total enriched and non-enriched) were 26, 64 and 114 µgChla g dry sed^{-1} with respectively the following ratio between abundance of total and enriched algae: 2.4, 1.3 and 1.2.

Incubations were halted by freezing the microcosms at -80°C. Samples were thawed and nematodes were extracted from the sediment using Ludox TM (Heip et al. 1985). For each sample, at least 700 nematode specimens were picked up randomly and individually with Pasteur pipette, rinsed twice in Milli-Q water to remove adhering particle and transferred in aluminium cup.

149 *Isotope analysis and calculations*

150 $\delta^{15}N$ and $\delta^{13}C$ of prey (bacteria and algae) and grazers (nematodes) were measured using
151 an EA-IRMS (Isoprime, Micromass, UK). Nitrogen isotope composition is expressed in the
152 delta notation ($\delta^{15}N$) relative to air N_2: $\delta^{15}N = [(^{15}N/^{14}N)_{sample} / (^{15}N/^{14}N)_{reference})-1] \times 1000$.
153 Carbon isotope composition is expressed in the delta notation ($\delta^{13}C$) relative to Vienna Pee
154 Dee Belemnite (VPDB): $\delta^{13}C = [(^{13}C/^{12}C)_{sample} / (^{13}C/^{12}C)_{reference})-1] \times 1000$.

155 The ^{15}N incorporated was defined as excess (above the background level) ^{15}N and is
156 expressed in terms of specific uptake (*I*). *I* was calculated as the product of excess ^{15}N (*E*) and
157 the biomass of N per grazer. *E* is the difference between the background ($F_{background}$) and the
158 sample (F_{sample}) ^{15}N fraction: $E = F_{sample} - F_{background}$, with $F = {}^{15}N / (^{15}N + {}^{14}N) = R / (R + 2)$
159 and *R* = the nitrogen isotope ratio. For the $F_{background}$, we used highest control values ($\Delta^{15}N$ =
160 11.78 and $\Delta^{13}C$ = -16.34) measured using killed (frozen) grazers. *R* was derived from the
161 measured $\delta^{15}N$ values as: $R = ((\delta^{15}N/1000)+1) \times R\text{air}N_2$ where $R\text{air}N_2 = 7.35293\ 10^{-3}$
162 (Mariotti 1982). The uptake of bacterial carbon was calculated as Uptake = ($I \times$ (% $C_{enriched\ bacteria}$ /
163 % $N_{enriched\ bacteria}$) / ($F_{enriched\ bacteria} \times$ incubation time). This uptake was multiplied by
164 the ratio between the abundance of total and enriched bacteria determined by DAPI counts.

165 Incorporation of ^{13}C was calculated analogously, with $F = {}^{13}C / (^{13}C + {}^{12}C) = R / (R + 1)$,
166 $R\text{air}N_2$ is replaced by $R_{VPDB} = 0.0112372$ and Uptake = I / ($F_{enriched\ bacteria} \times$ incubation time).
167 The uptake measured was multiplied by the ratio between the abundance of total and enriched
168 diatom, determined from fluorometrical measurements.

169 Enriched *N. phyllepta*-produced carbon consisted of 22.95 ± 0.54% ^{13}C. The C/N ratio of
170 enriched bacteria was 3.49 and bacterial nitrogen consisted of 2.88 ± 0.03% ^{15}N. The average
171 weight of nematodes was 0.33 ± 0.18 µg DW and each nematode was composed on average
172 of 0.11 ± 0.05 µgC and 22.28 ± 5.82 ngN (N = 72 samples of at least 700 specimens each).

Uptake expressed as $gC_{bacteria}\ gC_{nematode}^{-1}\ h^{-1}$ was obtained by dividing the uptake of bacteria ($gC\ ind^{-1}\ h^{-1}$) by the mean nematode weight ($gC\ ind^{-1}$).

Variations of assimilation rates with respect to the salinity and luminosity were tested using two-tailed test. One-way analysis of variance (ANOVA) was used in order to test the impact of temperature and algal and bacterial abundance on the uptake rates of bacteria and algae. The Tukey test was used for post-hoc comparisons.

Nematode community composition

Nematode communities used in the grazing experiments were extracted from sediment with Ludox TM (Heip et al. 1985). In order to determine the taxonomic composition of the community studied, 303 nematodes were collected at random, determined to species or generic level under the microscope and sorted by feeding group as indicated by Wieser (1953; 1960).

Results

Composition of the nematode community

In the sample collected for the taxonomy of the nematode community, 19 species belonging to 18 genera were observed (Tab. 1). Three species made up more than 87 % of the community: *Chromadora macrolaima* (64%), *Daptonema oxycerca* (15%) and *Ptycholaimellus jacobi* (8%). The other species were much less abundant, 11 representing less than 1 %.

The community was dominated by epigrowth feeders 2A (75%) due to high abundances of *C. macrolaima* and *P. jacobi*. Non-selective deposit feeders 1B (21%) were the second most abundant trophic group due to high abundance of *D. oxycerca*. Selective deposit feeders 1A (2%) and omnivores/predators 2B (1%) exhibited marginal abundances in the community studied.

Uptake of microbes by nematodes

198 Nematodes isotopic compositions and rates of bacterial ad algal uptakes are presented in
199 Table 2.

200 The kinetic experiment showed that bacterial uptake by the nematode community
201 increased linearly during the twelve hours of incubation (Fig. 2). The linear regression slope
202 suggested an assimilation rate of bacteria of 32 pgC ind^{-1} h^{-1} equivalent to 25×10^{-5} $gC_{bacteria}$
203 $gC_{nematode}^{-1}$ h^{-1} ($r^2 = 0.98$).

204 Temperature had a significant effect on the assimilation rate of bacteria ($F = 7.5$, $p<0.005$).
205 Temperature tested fluctuated between 5 and 40°C and were in the range of those found in the
206 study area (Guarini et al. 1997). Uptake of bacteria was reduced at 5°C, then increased with
207 temperature to reach an optimum at around 30°C and then decreased (Fig. 3). This rate
208 increased from 3 pgC ind^{-1} h^{-1} to 25 pgC ind^{-1} h^{-1} when the temperature rose from 5°C to
209 30°C and then decreased, reaching 18 pgC ind^{-1} h^{-1} at 40°C. Overall trend is showing a
210 prominent temperature effect but assimilation rates observed at 10, 20, 30 and 40°C were
211 nevertheless not significantly different from each other but were significantly different from
212 those observed at 5°C.

213 The assimilation rate at a salinity of 31 (19 ± 2 pgC ind^{-1} h^{-1}) was similar (two-tailed test, p
214 $= 0.72$) to that found for a salinity of 18 (18 ± 4 pgC ind^{-1} h^{-1}) (data not shown).

215 Light significantly affects the feeding activity of nematodes (two-tailed test, $p<0.05$).
216 Assimilation rates were more than twice as high (41 ± 11 pgC ind^{-1} h^{-1}) under light (83 µM of
217 photons m^{-2} s^{-1}) than in darkness (19 ± 2 pgC ind^{-1} h^{-1}) (data not shown).

218 Assimilation rates were significantly linked with bacterial abundance ($F = 52$, $p<0.001$)
219 (Fig. 4). Its value was null for the lowest tested abundance (4×10^8 cells ml^{-1}), was amplified
220 more than fourfold when abundance increased from 7×10^8 cells ml^{-1} to 10×10^8 cells ml^{-1}
221 and remained stable between this last value and 17×10^8 cells ml^{-1}.

Using two isotopes to label bacterial (^{15}N) and algal food (^{13}C) offers the opportunity to assess bacterial and algal uptake rates simultaneously. Assimilation of algae increased linearly when algal abundance increased ($r^2 = 0.99$, $p<0.05$). Bacteria represented 25, 16 and 8 % of algal plus bacterial intake when algal concentrations were respectively 26, 64 and 114 μgChla g^{-1}, with bacterial abundance remaining constant (Fig. 6). Plotted data indicated that assimilation of bacteria declines when the algal concentration is high (114 μgChla g^{-1}) and the bacterial concentration remains constant (Fig. 5). Nevertheless, these differences were not significant and assimilation of bacteria was unaffected by algal abundance ($F = 3.24$, $p = 0.11$).

Discussion

Nematode community

Rzeznik-Orignac *et al.* (2003) studied the Brouage mudflat nematode community and showed that in the lower part of the mudflat and over four seasons (2000-2001), six dominant species represented 45% of the community: *Metachromadoroides remanei* (11.5%), *Terschellingia longicaudata* (11.2%), *P. jacobi* (8.6%), *C. macrolaima* (8.6%), *Sabatiera pulchra* (5.2%) and *D. oxycerca* (0.7%). In our study, performed in March 2006, only three species, although all mentioned in the Rzeznik-Orignac *et al.* study, represented 87% of the community (Tab. 1). As the sampling site was the same, difference in nematode community composition may be due to inter-seasons or inter-years fluctuations. More probably, major difference has to be put into relation with the sampling method used in each of these studies. In the present one, only the top centimeter of sediment was sampled, whereas the top 5 cm were sampled in the Rzeznik-Orignac *et al.* (2003) study. As nematodes exhibit a strong vertical distribution of species in sediment (e. g. Platt 1977; Steyaert et al. 2001), the community in the present study is not representative of the total Brouage mudflat nematode community. Moreover, the three major species of the community under study belong to the

247 *Daptonema* and *Ptycholaimellus* genera that were mainly found in the 5 mm surficial layer of
248 sediment (Steyaert et al. 2003) and the third one, belonging to the family Chromadoridae is a
249 typical surface-dwelling epigrowth feeder (Platt 1977).

250 Feeding habits on diatoms of some genera close to *Chromadara* are known from culture
251 experiments: *Chromadorita tenuis* (Jensen 1987) and *Chromadora macrolaimoides* (Tietjen
252 and Lee 1973). *Chromadora macrolaima* and *Ptycholaimellus jacobi* break or pierce the
253 frustule of diatoms to suck out their contents whereas *Daptonema oxycerca* swallow the
254 whole diatom cell (Wieser 1953; Wieser 1960). Our nematode community is mainly
255 composed by three dominant species known or suspected to feed predominantly on diatoms.
256 The entire nematode community presents however higher abundances of bacterial grazers
257 (Rzeznik-Orignac et al. 2003). As a result, extrapolation of the present grazing results to the
258 rest of the community or to other communities must be realized with caution.

259 *Grazing experiments*

260 Like all various methods previously developed and applied to measure bacterivory, the
261 method used in the present study presents methodological shortcomings that make
262 interpretation of the resulting problematic. For instance, sieving the sediment changes the
263 bacterial availability, bacteria being not attached to particle as in natural situation. Indeed,
264 disruption of microbial-meiofaunal spatial relationships is known to affect grazing rate with
265 slurry method (Carman et al. 1989). Nematode grazing can be highly dependent on their
266 bacterial prey's activity, size and species (Tietjen et al. 1970; Tietjen and Lee 1973; Romeyn
267 and Bouwman 1983; Grewal and Wright 1992; Moens 1999; Moens et al. 1999a). One major
268 hypothesis in grazing experiments is that grazers did not select for or against the added
269 labeled bacteria over the bacteria present in the sediments. Consequently, the cultured bacteria
270 community must present characteristics that are roughly similar to the natural one. Despite the
271 fact that our culturing process modified the specific composition of the natural bacterial

community, the size, activity and diversity of the bacterial consortium used in the present study would be more representative of the natural community than in most previous grazing experiments (Pascal et al. 2008).

Bacterial and algal uptake

^{15}N accumulation in nematodes was linear over the 12-hour incubation period (Fig. 2). The constant accumulation of labeled food in nematodes had already been observed with comparable incubation times (Herman and Vranken 1988; Moens et al. 1999c). Since the nematode defecation intervals are very short (e. g. Thomas 1989) and since the gut is emptied completely with each defecation (Duncan et al. 1974), the gut contents would thus be renewed every few minutes. For these reasons, it has been suggested that during long incubation times, the linear accumulation of the label indicates assimilation rather than ingestion (Schiemer 1987; Moens et al. 1999c). The slope of this linear curve gives an assimilation rate of 32 pgC ind^{-1} h^{-1}. Assuming an assimilation rate of 25% (Herman and Vranken 1988; Somerfield et al. 2005), the ingestion rate would be four times higher, at 128 pgC ind^{-1} h^{-1}. As all other grazing experiments were run for 3 hours, they provided the assimilation rate of food.

Abiotic factors

Behavioral responses of plant-parasites and terrestrial nematodes to several stimuli such as electrical, mechanical and chemical stimuli and physical factors such as temperature and light have been well described (review in Croll 1970). However, similar studies concerning the effect of environmental conditions on the feeding behavior of marine or brackish-water nematodes are scarce and limited in scope.

Temperature has received attention as a factor influencing the growth and reproduction of estuarine and marine nematodes (review in Heip et al. 1985). The influence of temperature on feeding has only been studied in the predacious nematode Enoploides (Moens et al. 2000), the epigrowth feeder *Chromadora macrolaimoides* (Tietjen and Lee 1973) and in two brackish-

297 water bacterivorous species: *Pellioditis marina* and *Diplolaimelloides meyli* (Moens et al.
298 1996; Moens and Vincx 2000). In the present study, nematodes exhibited a classical response
299 which was more or less related to a bell-shaped function also observed by Moens and Vincx
300 (2000). The assimilation rate of bacteria rose when the temperature increased and reached its
301 upper value at 30°C before declining. As assimilation rates recorded for the tested
302 temperatures were statistically different at 5°C, it may be suggested that the nematode grazing
303 rate is lower in winter when the temperature drops to under 5°C. However, nematodes may
304 adjust their optimum temperature conditions along seasons (Gee 1985). Studied nematodes
305 were probably adapted to low temperature as sediment temperature was 7°C at the time of the
306 sampling. Montagna (1984b) did not observe different grazing rates between winter and
307 summer, but the temperature range in his study area was small (18 and 23°C).
308 Salinity plays a major role in determining the spatial structure of the nematode community
309 along permanent gradients (Soetaert et al. 1995) or in its seasonal variations (Chatterji et al.
310 1995). However nematodes can also be affected by short-term variations in salinity (Forster
311 1998) and intertidal habitats are subject to major changes in interstitial salinity over short time
312 periods. During low tide, raindrops implode into fine sediments, disturbing and mixing the
313 surface sediment of mudflats. Moreover, freshwater run-off passively diffuses into the
314 interstitial pores of the sediment. In their review of marine nematode ecology, Heip et al.
315 (1985) compiled an extensive list of marine and estuarine species with their salinity
316 tolerances. *C. macrolaima*, the most abundant species in the present study, is found in areas
317 with salinities ranging from 35 to 24 whereas *D. oxycerca*, the second most abundant species,
318 is found in area ranging from 35 to 0.9. This species seemed to be able to tolerate gradual
319 changes in salinity but not rapid ones. After exposure to salinity of 3.33 for 10 min and 48 h,
320 Forster (1998) recorded adult mortalities of 10-35% and 70% respectively. This species from
321 the lower level of the intertidal zone is able to osmoregulate but unable to sustain water

322 regulation over long periods (Forster 1998). In the present study, assimilation at salinity 18
323 was not significantly different from the assimilation at salinity 31. Since the nematodes were
324 not acclimated until the grazing experiment, they seemed to be able to cope with rapid, but
325 limited, osmotic stress. Our conclusions concerning the minor role of salinity in bacterial
326 assimilation by nematodes are in accordance with Moens and Vincx's (2000) results. Among
327 the three factors they investigated (temperature, salinity and food abundance) affecting *P.*
328 *marina* and *D. meyli* food intake, salinity played a minor role in bacterial assimilation by
329 nematodes.

330 When exposed to air at low tide, benthic organisms at the sediment's surface are subject to
331 the highest solar and UVB radiation that can be experienced by marine organisms. There are
332 few investigations assessing the effects of luminosity on nematodes. The predacious
333 nematode Enoploides caught approximately twice as many prey nematodes in the dark as in
334 light (Moens et al. 2000). On the other hand, Sundbäck et al. (1996a) concluded that ambient
335 UVBs did not exert any strong selective pressure on the meiofaunal community of a muddy
336 microtidal area. Nozais et al. (1999) observed a deleterious effect of UVBs on the nauplia
337 stages of harpacticoid copepods from a tidal mudflat. However, they did not observe any
338 effect on nematode abundance. In our study, light may not present damaging effect as
339 nematodes fed actively, moreover bacterial assimilation was enhanced when light increased.
340 Uptake rates of bacteria by foraminifera (Pascal et al. In press) and the gastropoda *Hydrobia*
341 *ulvae* (Pascal et al., in prep) that were obtained by grazing experiments performed under the
342 same conditions as in present study did not demonstrate any effect of light. Consequently, the
343 response we observed should not be due to experimental bias and seems to be specific to
344 nematodes. Montagna et al. (1995) observed a significant correlation between algal
345 production and the grazing rate of a nematode community from the Brouage mudflat. In their
346 experience, algal production was increased by increasing the light intensity above

microcosms. They concluded that nematode's ingestion rate increases with algal production. In intertidal mudflats, benthic diatoms migrate in the surface sediment along the diurnal cycle (e. g. Serôdio et al. 1997). The nematode community under study was dominated by epigrowth feeders (Tab. 1), with a diet mainly composed of benthic diatoms. Those nematodes may graze when luminosity is high during low tide in order to graze in algal biofilm and maximize their algal intake. This feeding behavior has not been documented for nematodes, but harpaticoid copepods were shown to graze at a higher rate just after the mudflat became exposed (Decho 1988). Buffan-Dubau and Carman (2000) also observed a midday feeding peak by ostracods and harpaticoid copepods. This result suggests that nematodes would have a feeding behavior principally controlled by algae and that bacteria may be taken up accidentally with algae.

Biotic factors

For nematodes, the effects of bacterial concentration on growth, fecundity, population development and feeding rates have been studied (Nicholas et al. 1973; Schiemer et al. 1980; Schiemer 1982a; Schiemer 1982b; Schiemer 1983; Vranken et al. 1988; Moens and Vincx 2000). Nematode ingestion rates are generally proportional to food availability (Nicholas et al. 1973; Schiemer et al. 1980; Moens et al. 1996). However, the relationship between assimilation rate and food concentration is not linear and have been described by a Michaelis-Menten function (Schiemer 1982b). This function is consistent with Holling's prey-dependent type II functional response (Holling 1959). Assimilation rates depend on the amount of ingested food and efficiency with which the material is assimilated. The assimilation efficiency have been found to decrease when food concentration increases: at a high food concentration *Plectus palustris* presented a low assimilation efficiency (12%) (Duncan et al. 1974) whereas at a food concentration one order of magnitude lower, the assimilation efficiency reached 57% (Schiemer et al. 1980). A similar conclusion was drawn by Moens et

al. (2006) on the marine bacterivore *Pellioditis marina*. Low efficiencies at high concentrations appear to result from short gut retention times, with the gut transit being too fast for effective digestion (Taghon et al. 1978). In the present study, it is not possible to determine if assimilation rates fluctuations are due to variations of ingestion rates or assimilation efficiencies. The number of bacterial concentrations tested was limited, however the response observed could be described by a sigmoid function. This function is consistent with Holling's prey-dependent type III functional response (Holling 1959). The threshold value for significant assimilation appeared to fall between 4 and 7×10^8 cells ml^{-1} and the threshold value for constant assimilation appeared to be between 7 and 10×10^8 cells ml^{-1}. Those values are in the range of data provided by literature for fast growing opportunistic bacterivores nematodes typical of plant material or root systems: an optimal grazing rate was obtained for *Caenorhabditis briggsae* (Nicholas et al. 1973) and *P. marina* and *D. meyli* (Moens and Vincx 2000) at a bacterial concentration of 5, 25 and 5×10^8 cells ml^{-1} respectively. Our study, dealing with tidal flat microalgal grazers community, suggests that the optimal level of bacterial foraging is also reached at high bacterial density, when bacterial concentrations attain 1×10^9 cells ml^{-1}. As bacterial concentrations are rarely lower in marine sediments, (Hondeveld et al. 1992; Schmidt et al. 1998; Hamels et al. 2004), bacterial assimilation rates would seldom be lower than 18 pgC ind^{-1} h^{-1}.

The relationship between the assimilation rate of algae and the algal concentration is poorly documented. Montagna et al. (1995) observed a proportional rise in the algal grazing rate with increasing algal concentration. In the present study, algal assimilation increased linearly without reaching a plateau. This function may be consistent with Holling's prey-dependent type I functional response (Holling 1959). Thus, the algal concentration threshold was not reached and maximal algal assimilation may be higher than those we obtained. Under natural conditions, the chlorophyll a content of the first centimeter of sediment varies between

0 and 50 µgChla g^{-1} (review in MacIntyre et al. 1996). However, through vertical migration, benthic microalgae were concentrated near the surface during diurnal low tides and produced a biofilm with an average thickness of 50 µm (Herlory et al. 2004). In this thin layer of algal mat, the concentration of chlorophyll a can reach 150 µgChla g^{-1} (Serôdio et al. 1997) and even 300 µgChla g^{-1} (Kelly et al. 2001). Thus, the higher algae abundance used in the present study (114 µgChla g^{-1}) is not representative of conditions occurring in the natural environment and the nematode community feeding on the algal biofilm could thus present a higher assimilation rate of algae.

In our experiment, algal assimilation by nematodes always represented more than three times the bacterial assimilation. Nematodes are able to select potential food items. *Chromadora macrolaimoides* have a preference for diatoms and chlorophytes whereas they assimilate fewer bacteria (Tietjen and Lee 1973). *D. oxycerca* is able to swallow particles of different sizes and large frustules of diatoms (Boucher 1974). The three dominant species of the community studied, i.e. *C. macrolaima*, *D. oxycerca* and *P. jacobi*, were found to dominate the Brouage nematode community during spring diatom blooms (Rzeznik-Orignac et al. 2003) indicating that they are highly dependent on algal resources. The use of natural stable isotopic analyses led to a similar conclusion elsewhere, that the microphytobenthos constitutes the main food source for the nematode community dwelling in the surficial centimeter of mud in the study area (Riera et al. 1996). Montagna et al. (1984b), looking at the grazing rates of a nematode community from a saltmarsh, found that diatoms are selected 14 more times than bacteria. Algal carbon accounted on average for more than 90% of carbon grazed by a nematode community from a microtidal sandy sediment (Sundbäck et al. 1996b). In the present study, nematodes also ingested algae at a higher rate than bacteria. Depending on the algal concentration, the nematode community assimilates 3 to 11 times more diatoms than bacteria.

Nematode production can be estimated on the basis of the P/B ratio, bearing in mind that the choice of this ratio may be inaccurate (Heip et al. 1982). With an individual biomass of 0.11 µgC ind^{-1}, the estimation of production yielded 0.11 ngC ind^{-1} h^{-1}, given a P/B ratio of 9 as often advocated (Gerlach 1971; Warwick and Price 1979; Bouvy 1988). Using a 10% factor for energy conversion efficiency (Bouvy 1988), a nematode carbon demand of 1.13 ngC ind^{-1} h^{-1} can be calculated. In the present study, the maximum ingestion rate of bacteria measured would represent 4% of this energy demand. On the other hand, the maximum algal ingestion measured would correspond to 15% of this demand. To get 100% of their energy needs, nematodes may graze at a higher rate on the algal compartment, as discussed above. Nematodes may also be dependent on other food sources such as detritus, protozoa, oligochaetes or nematodes (Moens and Vincx 1997). The role of Dissolved Organic Matter in nematode nutrition also remains elusive, although it is likely to be highly relevant (Lopez et al. 1979; Meyer-Reil and Faubel 1980; Montagna 1984a; Jensen 1987). Bacterial carbon was found to constitute 6% of the total carbon requirement of a mudflat nematode community (van Oevelen et al. 2006b). On the other hand, using inverse modeling, Van Oevelen et al. (2006c) suggested that mudflat community of nematodes relied for 50 % on algae and 39 % on bacteria. Variations between studies can be due to differences in methodology and to nematode community composition.

In the brouage mudflat, nematode present a mean densities of 2112 ind 10 cm^{-2} (Rzeznik-Orignac et al. 2003) whereas biomasses of benthic bacteria represent 0.846 gC m^{-2} (Degré et al. 2006). If nematodes grazing rates measured in the present study are representative of those of total community, it would imply that 0.02 % of bacterial biomass is assimilated by nematodes each day. This extrapolation is debatable, as studied nematodes feed predominantly on diatoms whereas total community may be more dependent on bacterial resource, consequently grazing of bacteria is probably underestimated.

Conclusion

Caution must be taken in interpreting our results, since the impact of each environmental factor on the feeding behavior of the nematode community was studied separately, whereas in natural environment all these factors covary greatly. The combination of temperature and salinity factors was found to have a higher impact than each factor taken alone (Tietjen and Lee 1972; Tietjen and Lee 1977). Moreover, nematodes may respond to environmental changes at a seasonal scale, by physiological adjustment and shifting of their optimum conditions (Gee 1985). However, the nematode community studied appeared to have adapted to the highly variable environment constituted by the surficial sediment of intertidal mudflats, except at low temperatures (5°C), and their feeding activity is only slightly decreased by temperature, salinity or light stress. Due to high abundance of bacteria in the marine sediment, nematodes may never be food limited with bacteria. Nematodes kept feeding under conditions of stress which were typical of the surficial sediment habitat, moreover they appeared to be principally dependent on the algal resource. Consequently, the community of nematodes dwelling in the top centimeter of the Brouage mudflat may also have a feeding strategy which is strongly linked to the formation of algal biofilm during the diurnal ebb.

Acknowledgements

We thank Prescilla Perrichon and Stéphanie Bollard for their help in sample preparation, Gaël Guillou for isotopic analyses, Utte Wollenzien from NIOO-CEMO for giving us a *N. phyllepta* strain and Janet Heard Carnot for English corrections. This work was financially supported by the “Conseil Général de Charente Maritime”, the ANR-VASIREMI and ECCO and “PNEC Chantier Littoral Atlantique”.

Experiments of the present study comply with current laws of French country.

References

Boucher G (1974) Premières données écologiques sur les nématodes libres marins d'une station de vase côtière de Banyuls. Vie Milieu 23: 69-100

Bouvy M (1988) Contribution of the bacterial and microphytobenthic microflora in the energetic demand of the meiobenthos in an intertidal muddy sediment (Kerguelen Archipelago). P. S. N. Z. I: Mar Ecol 9: 109-122

Buffan-Dubau E, Carman KR (2000) Diel feeding behavior of meiofauna and their relationships with microalgal resources. Limnol Oceanogr 45: 381-395

Carman KR, Dobbs FC, Guckert JB (1989) Comparison of three techniques for administering radiolabeled substrates for trophic studies: uptakes of label by harpaticoid copepods. Mar Biol 102: 119-125

Chatterji A, Ansari ZA, Mishra JK, Parulekar AH (1995) Seasonality in meiofaunal distribution on a tropical beach at Balramgari, northwest coast of India. Indian J Mar Sci 24: 49-55

Croll NA (1970) The behaviour of nematodes: their activity, senses and responses. Edward Arnold, London

Decho AW (1988) How do harpaticoides grazing rates differ over a tidal cycle? Field verification using chlorophyll-pigment analyses. Mar Ecol Prog Ser 45: 263-270

Degré D, Leguerrier D, Armynot du Chatelet E, Rzeznik-Orignac J, Auguet J-C, Dupuy C, Marquis E, Fichet D, Struski C, Joyeux E, Sauriau P-G, Niquil N (2006) Comparative analysis of the food webs of two intertidal mudflats during two seasons using inverse modelling: Aiguillon Cove and Brouage Mudflat, France. Est Coast Shelf Sci 69: 107-124

Duncan A, Schiemer F, Klekowski RZ (1974) A preliminary study of feeding rates on bacterial food by adult femals of a benthic nematode, *Plectus palustris* de Man 1880. Pol Arch Hydrobiol 21: 249-258

Fenchel T (1972) Aspects of decomposer food chains in marine benthos. Verh Dtsch Zool Ges 65: 14-22

Forster SJ (1998) Osmotic stress tolerance and osmoregulation of intertidal and subtidal nematodes. J Exp Mar Biol Ecol 224: 109-125

Gee JM (1985) Seasonal aspects of the relationship between temperature and respiration rate in four species of intertidal harpaticoid copepod. J Exp Mar Biol Ecol 93: 147-156

Gerlach SA (1971) On the importance of marine meiofauna for benthos communities. Oecologia 6: 176-190

Grewal PS, Wright RT (1992) Migration of *Caenorhabditis elegans* (Nematod: Rhaabditidae) larvae towards bacteria and the nature of bacterial stimulus. Fund Appl Nemat 15: 159-166

507 Guarini JM, Blanchard GF, Gros P, Harrison SJ (1997) Modeling the mud surface
508 temperature on intertidal flats to investigate the spatio-temporal dynamics of the
509 benthic microalgal photosynthetic capacity. Mar Ecol Prog Ser 153: 25-36

510 Hamels I, Sabbe K, Muylaert K, Vyverman W (2004) Quantitative importance, composition,
511 and seasonal dynamics of protozoan communities in polyhaline versus freshwater
512 intertidal sediments Microb Ecol 47: 18-29

513 Heip CHR, Herman P, Coomans A (1982) The productivity of marine meiobenthos.
514 Academia Analecta 44: 1-20

515 Heip CHR, Vincx M, Vranken G (1985) The ecology of marine nematodes. Oceanogr Mar
516 Biol Annu Rev 23: 399-489

517 Héral M, Razet D, Deslous-Paoli JM, Bertome JP, Garnier J (1982) Caractéristiques
518 saisonnières de l'hydrologies du complexe estuarien de Marennes-Oléron (France).
519 Revue des travaux de l'institut des pêches maritimes 46: 97-119

520 Herlory O, Guarini JM, Richard P, Blanchard GF (2004) Microstructure of
521 microphytobenthos biofilm and its spatio-temporal dynamics in an intertidal mudflat
522 (Aiguillon Bay, France). Mar Ecol Prog Ser 282: 33-44

523 Herman PMJ, Vranken G (1988) Studies of the life history and energetics of marine and
524 brackish water nematodes. II. Production, respiration and food-uptake by *Monhystera*
525 *disjuncta*. Oecologia 77: 457-463

526 Holling CS (1959) Some characteristics of simple types of predation and parasistism.
527 Canadian Entomologist 91: 385-398

528 Hondeveld BMJ, Bak RPM, van Duyl FC (1992) Bacterivory by heterotrophic nanoflagellates
529 in marine sediments measured by uptake of fluorescently labeled bacteria. Mar Ecol
530 Prog Ser 89: 63-71

531 Jensen KT (1987) Feeding ecology of free-living aquatic nematodes. Mar Ecol Prog Ser 35:
532 187-196

533 Kelly JA, Honeywill C, Paterson DM (2001) Microscale analysis of chlorophyll-a in cohesive
534 intertidal sediments: the implications of microphytobenthos distribution. J Mar Biol
535 Ass U K 81: 151-162

536 Kemp PF (1990) The fate of benthic bacterial production. Rev Aquat Sci 2: 109-124

537 Kuipers BR, de Wilde PAWJ, Creutzberg F (1981) Energy flow in a tidal flat ecosystem. Mar
538 Ecol Prog Ser 5: 215-221

539 Legendre L, Rassoulzadegan F (1996) Food-web mediated export of biogenic carbon in
540 oceans: hydrodynamic control. Mar Ecol Prog Ser 145: 179-193

541 Leguerrier D, Niquil N, Boileau N, Rzeznik J, Sauriau PG, Le Moine O, Bacher C (2003)
542 Numerical analysis of the food web of an intertidal mudflat ecosystem on the Atlantic
543 coast of France. Mar Ecol Prog Ser 246: 17-37

544 Leguerrier D, Niquil N, Petiau A, Bodoy A (2004) Modeling the impact of oyster culture on a
545 mudflat food web in Marennes-Oléron Bay (France). Mar Ecol Prog Ser 273: 147-162

546 Lopez G, Riemann F, Schrage M (1979) Feeding biology of the brackish-water Oncholaimid
547 nematod *Adoncholaimus thalassophygas.* Mar Biol 54: 311-318

548 MacIntyre HL, Geider RJ, Miller DC (1996) Microphytobenthos: the ecological role of the
549 "secret garden" of unvegetated, shallow-water marine habitats. I. Distribution,
550 abundance and primary production. Estuaries 19: 186-201

551 Mariotti A (1982) Apports de la géochimie isotopique à la connaissance du cycle de l'azote.
552 PhD Thesis, Paris, pp 476

553 Meyer-Reil LA, Faubel A (1980) Uptake of organic matter by meiofauna organisms and
554 interrelationships with bacteria. Mar Ecol Prog Ser 3: 251-256

555 Moens T (1999) Feeding ecology of free-living estuarine nematodes. An experimental
556 approach. PhD Thesis302

557 Moens T, Bergtold M, Traunspurger W (2006) Feeding ecology of free-living benthic
558 nematodes. In: Eyulaem A, Andrassy I, Traunspurger W (eds) Freshwater nematodes:
559 ecology and taxonomy. CAB International publishing, Cambridge, pp 105-131

560 Moens T, Herman P, Verbeeck L, M. S, Vincx M (2000) Predation rates and prey selectivity
561 in two predacious estuarine nematode species. Mar Ecol Prog Ser 205: 185-193

562 Moens T, Verbeeck L, de Maeyer A, Swings J, Vincx M (1999a) Selective attraction of
563 marine bacterivorous nematodes to their bacterial food. Mar Ecol Prog Ser 176: 165-
564 178

565 Moens T, Verbeeck L, Vincx M (1999b) Feeding biology of a predatory and a facultatively
566 predatory nematode (*Enoploides longispiculosus* and *Adoncholaimus fuscus*). Mar
567 Biol 134: 585-593

568 Moens T, Verbeeck L, Vincx M (1999c) Preservation and incubation time-induced bias in
569 tracer-aided grazing studies on meiofauna. Mar Biol 133: 69-77

570 Moens T, Vierstraete A, Vincx M (1996) Life strategies in two bacterivorous marine
571 nematods: preliminary results. P. S. N. Z. I: Mar Ecol 17: 509-518

572 Moens T, Vincx M (1997) Observations on the feeding ecology of estuarine nematodes. J
573 Mar Biol Ass U K 77: 211-227

574 Moens T, Vincx M (2000) Temperature, salinity and food thresholds in two brackishwater
575 bacterivorous nematode species: assessing niches from food absorption and respiration
576 experiments. J Exp Mar Biol Ecol 243 137–154

577 Montagna PA (1984a) Competition for dissolved glucose between meiobenthos and sediment
578 microbes. J Exp Mar Biol Ecol 76: 177-190

579 Montagna PA (1984b) *In situ* measurement of meiobenthic grazing rates on sediment bacteria
580 and edaphic diatoms. Mar Ecol Prog Ser 18: 119-130

581 Montagna PA (1995) Rates of metazoan meiofaunal microbivory: a review. Vie Milieu 45: 1-
582 9

583 Montagna PA, Bauer JE (1988) Partitioning radiolabeled thymidine uptake by bacteria and
584 meiofauna using metabolic blocks and poisons in benthic feeding studies. Mar Biol
585 98: 101-110

586 Montagna PA, Blanchard GF, Dinet A (1995) Effect of production and biomass of intertidal
587 microphytobenthos on meiofaunal grazing rates. J Exp Mar Biol Ecol 185: 149-165

588 Nicholas WL, Grassia A, Viswanathan S (1973) The efficiency with which *Caenorhabditis*
589 *briggsae* (Rhabditinae) feeds on the bacterium *Escherichia coli*. Nematologica 19:
590 411-420

591 Nozais C, Desrosiers G, Gosselin M, Belzile C, Demers S (1999) Effect of ambient UVB
592 radiation in a meiobenthic community of a tidal. Mar Ecol Prog Ser 189: 149-158

593 Pascal PY, Dupuy C, Mallet C, Richard P, Niquil N (2008) Bacterivory by benthic organism
594 in sediment: quantification using-^{15}N enriched bacteria. J Exp Mar Biol Ecol 355: 18-
595 26

596 Pascal PY, Dupuy C, Richard P, Niquil N (In press) Bacterivory in the common foraminifer
597 *Ammonia tepida*: isotope tracer experiment and the controlling factors. J Exp Mar Biol
598 Ecol

599 Platt HM (1977) Vertical and horizontal distribution of free-living marine nematodes from
600 Strangford Lough, Northern Ireland. Cah Biol Mar 18: 261-273

601 Platt HM, Warwick RM (1980) The significance of free-living nematodes to the littoral
602 ecosystem. In: Price JH, Irvine DEG, Farnham WF (eds) The shore environment, Vol
603 2. Academis Press, London and New-York, pp 729-759

604 Riera P, Richard P, Grémare A, Blanchard GF (1996) Food source of intertidal nematods in
605 the Bay of Marennes-Oléron (France), as determined by dual stable isotope analysis.
606 Mar Ecol Prog Ser 142: 303-309

607 Rivkin RB, Legendre L, Deibel D, Tremblay JE, Klein B, Crocker K, Roy S, Silverberg N,
608 Lovejoy C, Mesplé F, Romero N, Anderson MR, Matthews P, Savenkoff C, Vézina
609 AF, Therriault JC, Wesson J, Bérubé C, Ingram RG (1996) Vertical flux of biogenic
610 carbon in the ocean: Is there food web control? Science 272: 1163-1166

611 Romeyn K, Bouwman L (1983) Food selection and consumption by estuarine nematodes.
612 Hydrobiol Bull 17: 103-109

613 Rzeznik-Orignac J, Fichet D, Boucher G (2003) Spatio-temporal structure of the nematode
614 assemblages of the Brouage mudflat (Marennes Oléron, France). Est Coast Shelf Sci
615 58: 77-88

616 Sander BC, Kalff J (1993) Factors controlling bacterial production in marine and freshwater
617 sediments. Microb Ecol 26: 79-99

618 Schiemer F (1982a) Food dependence and energetics of freeliving nematodes. I. Respiration,
619 growth and reproduction of *Caenorhabditis briggsae* (Nematoda) at different levels of
620 food supply. Oecologia 54: 108-121

621 Schiemer F (1982b) Food dependence and energetics of freeliving nematodes. II. life history
622 parameters of *Caenorhabditis briggsae* (Nematoda) at different levels of food supply.
623 Oecologia 54: 122-128

624 Schiemer F (1983) Comparative aspects of food dependance and energetics of free living
625 nematodes. Oikos 41: 32-42

626 Schiemer F (1987) Nematoda. In: Pandian TJ, Vernberg FJ (eds) Animal energetic. Academic
627 Press, New York, pp 185-211

628 Schiemer F, Duncan A, Klekowski RZ (1980) A bioenergetic study of a benthic nematod,
629 *Plectus palustris* de Man 1880, throughout its life style. II Growth, fecundity and
630 energy budget at different levels of bacterial food and general ecological
631 considerations. Oecologia 44: 205-212

632 Schmidt JL, Deming JW, Jumars PA, Keil RG (1998) Constancy of bacterial abundance in
633 surficial marine sediments. Limnol Oceanogr 43: 976-982

634 Serôdio J, da Silva JM, Catarino F (1997) Nondestructive tracing of migratory rhythms of
635 intertidal benthic microalgae using in vivo chlorophyll a fluorescence. J Phycol 33:
636 542-553

637 Soetaert K, Vincx M, Wittoeck J, Tulkens M (1995) Meiobenthic distribution and nematode
638 community structure in five European estuaries. Hydrobiologia 311: 185-206

639 Somerfield PJ, Warwick RM, Moens T (2005) Methods for the study of marine benthos. In:
640 Eleftheriou A, McIntyre AD (eds) Meiofauna techniques. Blackwell Science, Oxford,
641 pp 229-272

642 Steyaert M, Herman PMJ, Moens T, Widdows J, Vincx M (2001) Tidal migration of
643 nematodes of an estuarine tidal flat (the Molenplaat, Schelde Estuary, SW
644 Netherlands). Mar Ecol Prog Ser 224: 299-304

645 Steyaert M, Vanaverbeke J, Vanreusel A, Barranguet C, Lucas C, Vincx M (2003) The
646 importance of fine-scale, vertical profiles in characterizing nematode community
647 structure. Est Coast Shelf Sci 58: 353-366

648 Sundbäck K, Nilsson C, Odmark S, Wulff A (1996a) Does ambient UV-B radiation influence
649 marine diatom-dominated microbial mats? A case study. Aquat Microb Ecol 11: 151-
650 159

651 Sundbäck K, Nilsson P, Nilsson C, Jonsson B (1996b) Balance between autotrophic and
652 heterotrophic components and processes in microbenthic communities of sandy
653 sediments: a field study. Est Coast Shelf Sci 43: 689-706

654 Taghon GL, Self RLF, Jumars PA (1978) Predicting particle selection by deposit feeders: a
655 model an its implications. Limnol Oceanogr 23: 752-759

656 Thomas JH (1989) Genetic analysis of defecation in *Caenorhabditis elegans*. Genetics 124:
657 855-872

658 Tietjen JH (1980) Microbial-meiofaunal interrelationships: a review. Microbiology 110: 335-
659 338

660 Tietjen JH, Lee JJ (1972) Life cycles of marine nematodes. Influence of temperature and
661 salinity on the development of *Monhystera denticulata* Timm. Oecologia 10: 167-176

662 Tietjen JH, Lee JJ (1973) Life history and feeding habits of the marine nematode
663 *Chromadora macrolaimoides* Steiner. Oecologia 12: 303-314

664 Tietjen JH, Lee JJ (1977) Life histories of marine nematodes. Influence of temperature and
665 salinity on the reproductive potential of *Chromadorine germanica* Bütschli.
666 Mikrofauna Meeresboden 61: 263-270

667 Tietjen JH, Lee JJ, Rullman J, Greengart A, Trompeter J (1970) Gnotobiotic culture and
668 culture physiological ecology of the marine nematode *Rhabditis marina* Bastian.
669 Limnol Oceanogr 15: 535-543

670 Underwood GJC, Kromkamp JC (2000) Primary production by phytoplancton and
671 microphytoplancton in estuaries. In: Nedwell DB, Raffaelli DG (eds) Estuaries,
672 Advances in Ecological Research. Academic Press, pp 93-153

673 van Oevelen D, Middelburg JJ, Soetaert K, Moodley L (2006a) The fate of bacterial carbon in
674 sediments: modeling an in situ isotope tracer experiment. Limnol Oceanogr 51: 1302-
675 1314

676 van Oevelen D, Moodley L, Soetaert K, Middelburg JJ (2006b) The trophic significance of
677 bacterial carbon in a marine intertidal sediment: Results of an *in situ* stable isotope
678 labeling study. Limnol Oceanogr 51: 2349-2359

679 van Oevelen D, Soetaert K, Middelburg JJ, Herman PMJ, Moodley L, Hamels I, Moens T,
680 Heip CHR (2006c) Carbon flows through a benthic food web: integrating biomass,
681 isotope and tracer data. J Mar Res 64: 453-482

682 Vranken G, Herman PMJ, Heip CHR (1988) Studies of the life history and energetics of
683 marine and brackish-water nematodes I. Demography of *Monhystera disjuncta* at
684 different temperature and feeding conditions Oecologia 77: 296-301

685 Warwick RM, Price JH (1979) Ecological and metabolic studies on freeliving nematodes
686 from an estuarine mudflat. Est Coast Mar Sci 9: 257-271

687 Wieser W (1953) Die Beziehungen zwischen Mundhölengestalt, Ernährungsweise und
688 Vorkommen bei freilebenden marinen nematoden. Ein ökologisch-morphhlogische
689 studie. Arkiv fur Zoologi 4: 439-484

690 Wieser W (1960) Benthic studies in Buzzards Bay. II. The meiofauna. Limnol Oceanogr 5:
691 121-137

692

693

694 ## Table and Figures captions
695

696 **Table 1.** List of species ranked by dominance (feeding types according to Wieser (1953;
697 1960)). 1A: selective deposit feeders; 1B: non-selective deposit feeders; 2A: epigrowth
698 feeders; 2B: omnivores/predators.

699 **Table 2.** Nematodes isotopic composition ($\Delta^{15}N$ and $\Delta^{13}C$ mean ± SD, N = 3) and bacterial
700 and algal uptake rates calculated.

701 **Figure 1**. Map of the study site.

702 **Figure 2.** Assimilation of bacterial carbon (mean ± SD, N =3) as function of incubation time
703 (h).

704 **Figure 3.** Assimilation rate of bacterial carbon (mean ± SD, N =3) as function of temperature
705 (°C). Different letters above bars indicate significant differences between incubation
706 conditions (ANOVA; Tukey test).

707 **Figure 4.** Assimilation rate of bacterial carbon (mean ± SD) as function of bacterial
708 abundance (10^8 cell.ml^{-1}). Different letters above bars indicate significant differences between
709 incubation conditions (ANOVA; Tukey test).

710 **Figure 5.** Assimilation rate of bacterial carbon (mean ± SD) as function of algal abundance
711 (µgChla.g^{-1}).

712 **Figure 6.** Uptake rate of algal carbon ○ (mean ± SD) and bacterial carbon ● (mean ± SD) as
713 function of algal abundance (µgChla g^{-1}). Bacterial abundance was constant (10.5×10^8 cells
714 ml^{-1}).

Table

Table 1

Genera species	Feeding type	Abundance relative (%)
Chromadora macrolaima	2A	64.2
Daptonema oxycerca	1B	15.2
Ptycholaimellus jacobi	2A	7.9
Sabatieria pulchra	1B	2.6
Axonolaimus paraspinosus	1B	1.7
Praeacanthonchus punctatus	2A	1.7
Halalaimus sp.	1A	1.0
Aegialoalaimus sp.	1A	1.0
Sphaerolaimus gracilis	2B	0.7
Spilophorella sp.	2A	0.7
Metachromadora sp.	2A	0.7
Theristus sp.	1B	0.3
Parodontophora marina	1B	0.3
Tripyloides marinus	1B	0.3
Eleutherolaimus sp.	1B	0.3
Desmolaimus zeelandicus	1B	0.3
Daptonema hirsutum	1B	0.3
Terschellingia sp.	1A	0.3
Viscosia sp.	2B	0.3

Table 2

	$\Delta^{15}N$		Bacteria uptake	$\Delta^{13}C$		Algae uptake
	Control	Normal	(pg C ind^{-1} h^{-1})	Control	Normal	(pg C ind^{-1} h^{-1})
Kinetics (hours)						
1		12.01 ± 0.25	3.67 ± 4.06			
2		14.24 ± 0.86	20.00 ± 7.03			
3	11.78 ± 0.23	15.20 ± 0.38	18.51 ± 2.05	-15.4 ± 1.32		
5		20.63 ± 1.49	28.80 ± 4.84			
8		27.72 ± 1.55	32.41 ± 3.15			
12	11.56 ± 0.87	32.66 ± 4.91	28.31 ± 6.65	-15.8 ± 0.77		
Temperature (°C)						
5		12.41 ± 0.53	3.38 ± 2.87			
10		14.60 ± 0.50	15.26 ± 2.70			
30		16.38 ± 0.79	24.92 ± 4.27			
40		15.18 ± 1.73	18.42 ± 9.38			
Irradiance						
Light		19.42 ± 2.01	41.43 ± 10.88			
Salinity						
18		15.03 ± 0.66	17.62 ± 3.57			
Bacterial abundance (10^8 cells ml wt sed^{-1})						
4.2		11.75 ± 0.20	-0.65 ± 4.39			
7.0		12.25 ± 0.40	3.34 ± 2.93			
17.4		15.86 ± 0.10	18.34 ± 0.45			
Algal abundance (µg Chla g dry sed^{-1})						
25.6		15.39 ± 0.59	19.58 ± 3.19		-4.44 ± 6.53	58.94 ± 32.35
64.3		15.58 ± 0.51	20.59 ± 2.78		25.60 ± 4.79	107.11 ± 12.24
113.7	11.50 ± 0.17	14.34 ± 0.61	13.87 ± 3.33	-16.34 ± 0.53	52.14 ± 33.29	161.20 ± 78.30

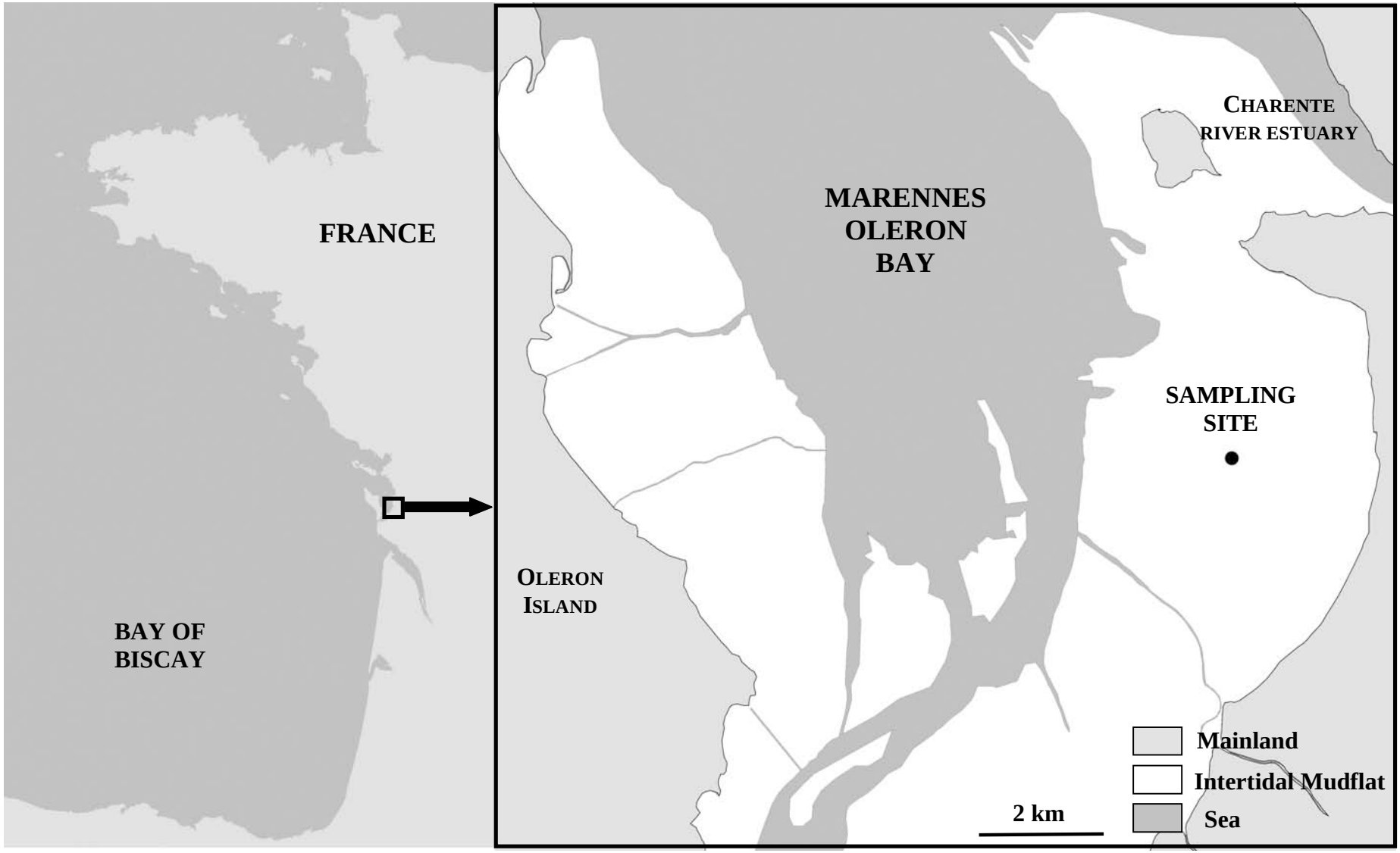
FRANCE
BAY OF
BISCAY
MARENNES
OLERON
BAY
CHARENTE
RIVER ESTUARY
SAMPLING
SITE
OLERON
ISLAND
Mainland
Intertidal Mudflat
Sea
2 km

Fig. 1

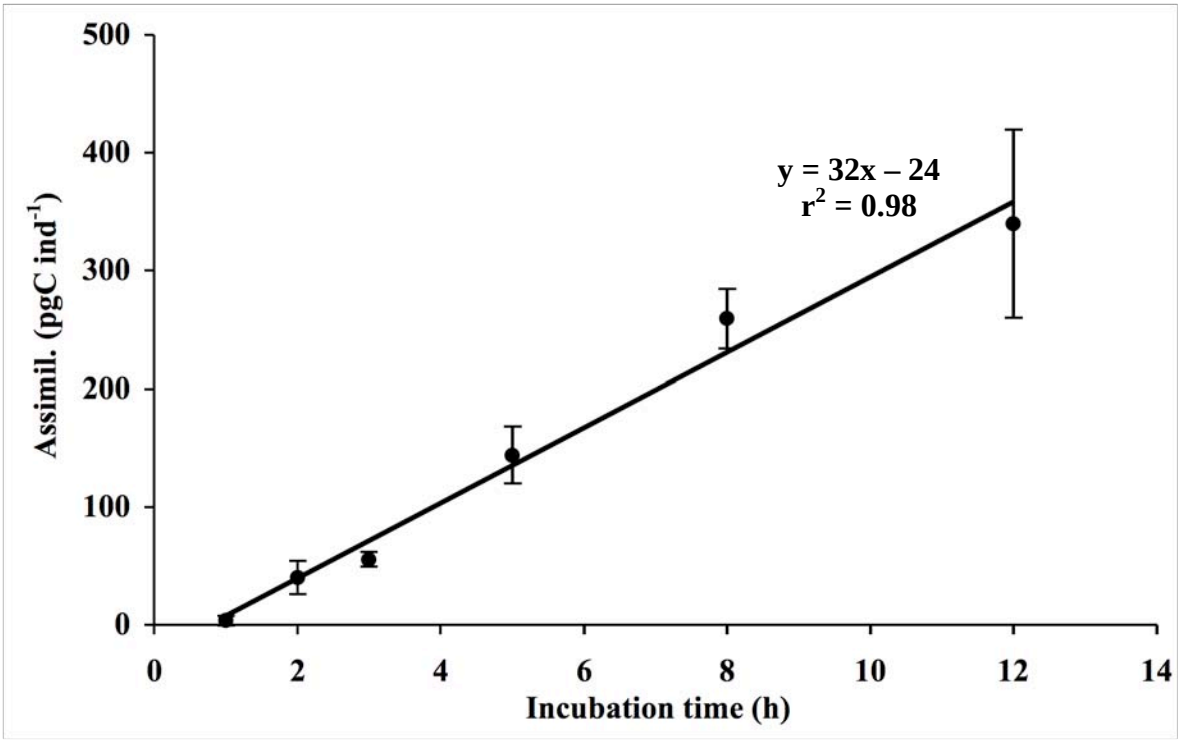
y = 32x – 24
r2 = 0.98
Assimil. (pgC ind-1)
Incubation time (h)
0
100
200
300
400
500
0
2
4
6
8
10
12
14

Fig. 2

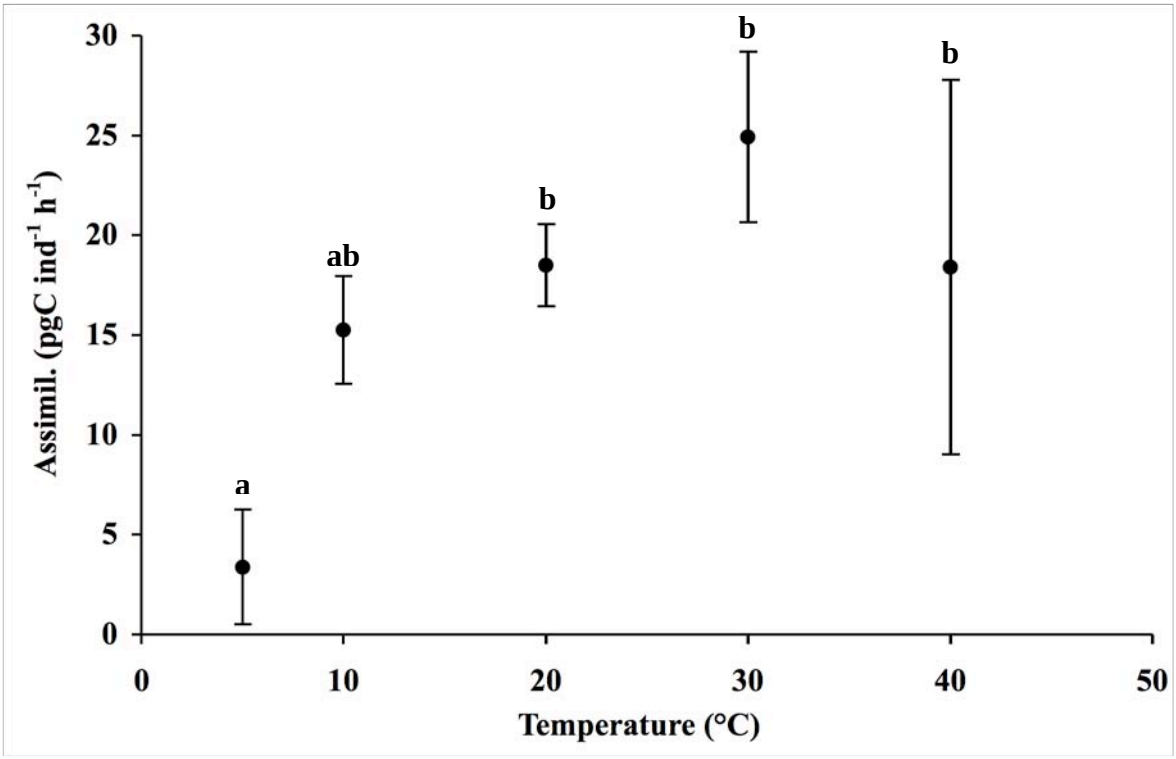
a
ab
b
b
b
Assimil. (pgC ind-1 h-1)
Temperature (°C)
0
5
10
15
20
25
30
0
10
20
30
40
50

Fig. 3

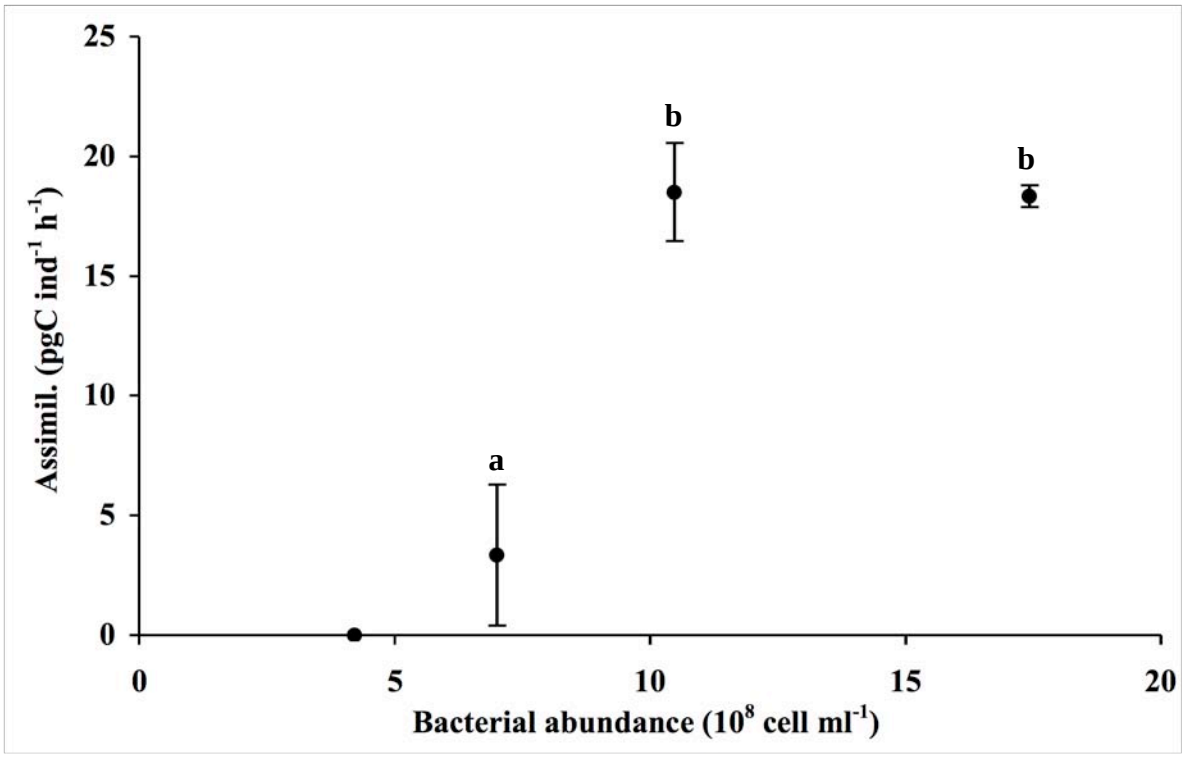
25
20
15
10
5
0
Assimil. (pgC ind-1 h-1)
b
b
a
0
5
10
15
20
Bacterial abundance (10^8 cell ml-1)

Fig. 4

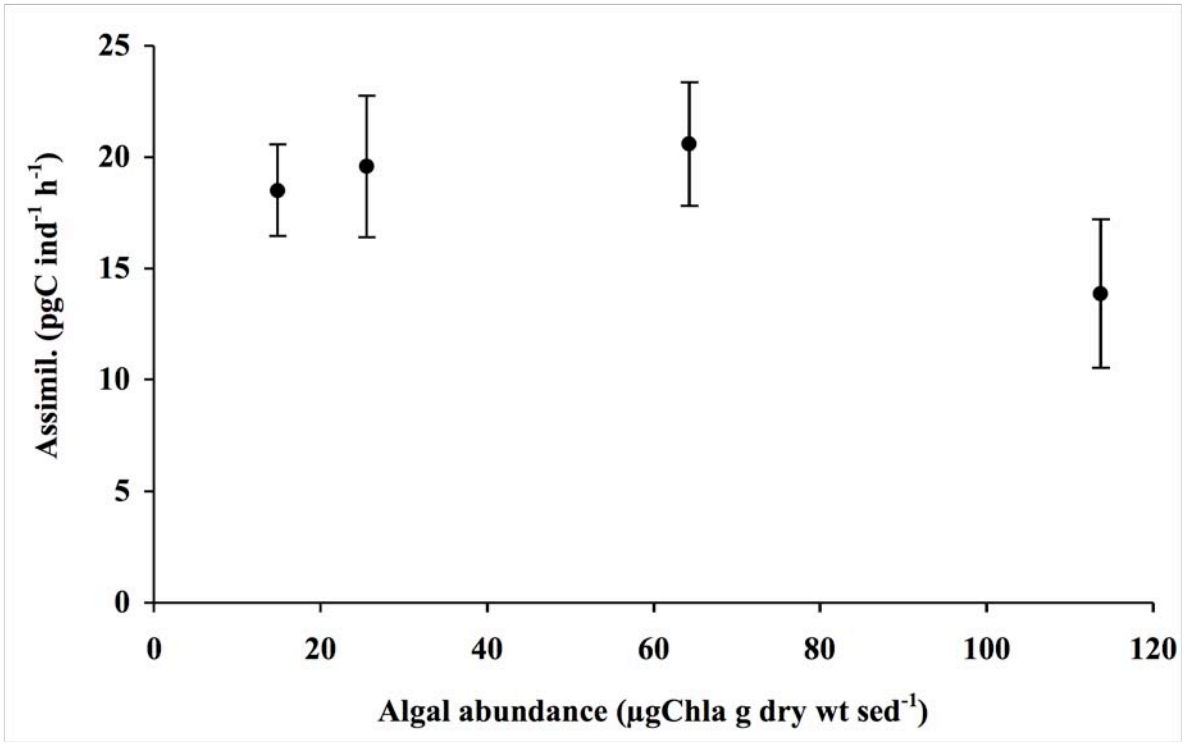
25
20
15
10
5
0
Assimil. (pgC ind-1 h-1)
0
20
40
60
80
100
120
Algal abundance (µgChla g dry wt sed-1)

Fig. 5

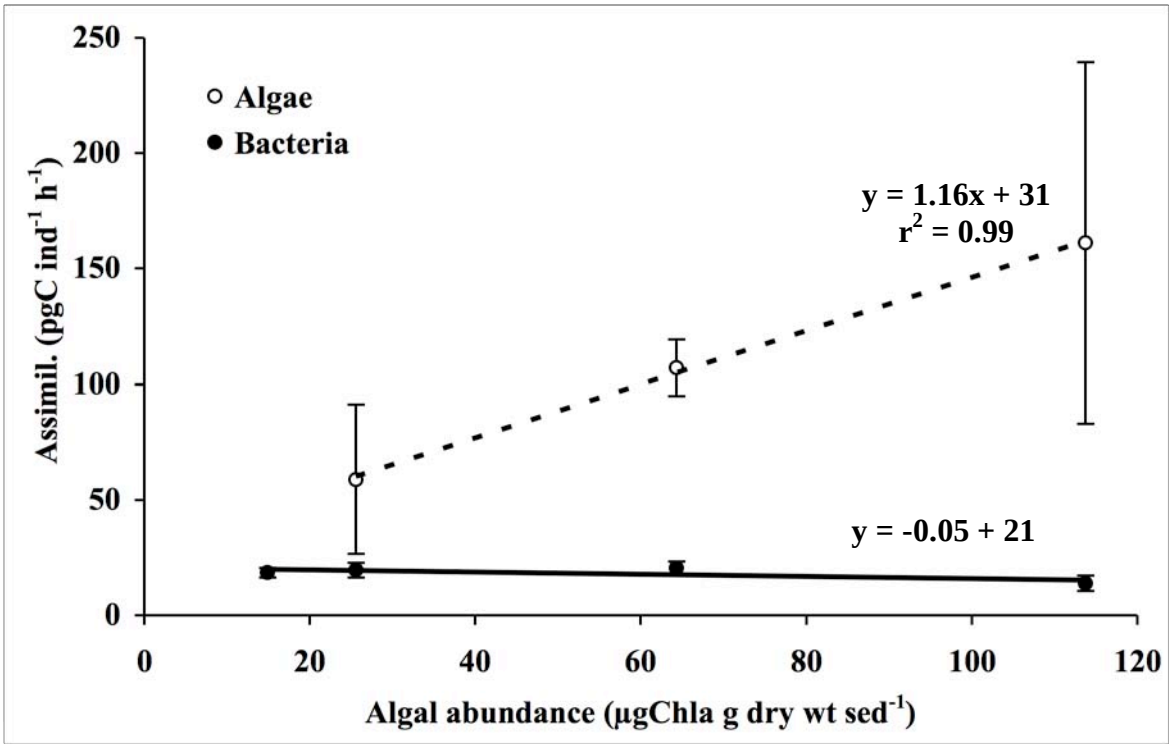
Algae
Bacteria
y = 1.16x + 31
r2 = 0.99
y = -0.05 + 21
Assimil. (pgC ind-1 h-1)
Algal abundance (µgChla g dry wt sed-1)
0
50
100
150
200
250
0
20
40
60
80
100
120

Fig. 6