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Turnover of dead jellyfish: stimulation and retardation of microbial activity

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ABSTRACT: Little is known about the fate of jellyfish biomass after their populations crash. We measured turnover rates of dead *Periphylla periphylla* in a pelagic *in situ* experiment in Raunefjorden, Norway. Decay was exponential, with decay-coefficients of 0.67 to 1.12 d^{-1}; smaller jellyfish turned over faster than larger ones. The rapid turnover suggests that large amounts of carbon and nitrogen are transferred to the pelagic and/or benthic foodwebs at the collapse of jellyfish blooms. The interactions between bacteria and dead jellyfish were examined in a shipboard experiment in Lurefjorden, Norway, using natural bacterial assemblages. Growth and morphology of the bacteria in the incubations suggested that only certain morphotypes proliferated in the vicinity of jellyfish tissue. Subsequent experiments with homogenized *P. periphylla* tissue and bacterial isolates from various phylogenetic groups confirmed that some bacteria could use jellyfish as substrate, while others were inhibited. Tests with sensitive isolates revealed that inhibition of bacterial growth depended on the body part, size and concentration of *P. periphylla* used. Because only some bacteria were able to use *P. periphylla* as a substrate, we suggest that the role of dead jellyfish as trophic links in specific pelagic foodwebs depends partly on bacterial community composition.

KEY WORDS: *Periphylla periphylla* · Degradation · Bacteria · Inhibition · Growth

INTRODUCTION

Although many fish, turtles, birds and invertebrates feed on jellyfish, top-down trophic control of jellyfish populations is often lacking, and populations may therefore grow large (Arai 2005). This pattern, combined with the reputation of jellyfish as voracious predators of zooplankton (Purcell & Arai 2001, Purcell & Decker 2005), other jellyfish (Purcell 1997), fish eggs and larvae (Purcell & Arai 2001), has led to the traditional view of jellyfish representing trophic dead ends or sinks in pelagic foodwebs (Hansson & Norrman 1995, Arai 2005). Although challenged, some reports suggest that jellyfish biomass has increased in the past decades (Mills 2001), and that jellyfish biomass may be linked to climatic variability and change (Brodeur et al. 1999, Lynam et al. 2004, Purcell 2005, Purcell & Decker 2005). In any case, mass-occurrences of jellyfish are common, often seasonal, features of many coastal systems (Schneider 1989, Hansson 1997, Mills 2001) and may contribute substantially to the carbon standing stock of planktonic foodwebs (e.g. Schneider 1989, Graham et al. 2001). Yet the fate of the carbon and nutrients bound in these populations after their crashes is unknown.

Bacteria thrive in the dissolved organic carbon (DOC) released by live jellyfish, both in the laboratory (Hansson & Norrman 1995) and the field (Riemann et al. 2006, this volume). We therefore expect that dead jellyfish return energy and nutrients to the foodweb via rapid degradation and remineralization processes in the water column and on the seafloor by components of the microbial loop.

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Other gelatinous material, such as abandoned appendicularian houses and other marine snow particles provide both food and habitats for microbes and larger animals (e.g. Alldredge 1972, Kiørboe 2000). Small structures, such as houses from the appendicularian *Oikopleura dioica*, may be completely disintegrated in deep water columns (Davoll & Silver 1986), or sink to the bottom at speeds exceeding 100 m d^{-1} in shallower environments (e.g. Hansen et al. 1996). Stemman et al. (2004) estimated that 90% of the particles <1 cm are remineralized within the euphotic zone in oceanic environments. In contrast, in the Gulf of Oman, dead jellyfish (*Crambionella orsini*) have been observed by video to roll down the continental slope and form meter sized benthic patches at 3300 m depth (Billet et al. 2006). Direct measurements of degradation rates of larger, dead gelatinous zooplankton are scarce, and little is known about the contribution of dead jellyfish to the pelagic nutrient pool. Larger live medusae accumulate damage through the season and are gradually broken down in the water column (Mills 1993). Mesocosm experiments with the moon jellyfish *Aurelia aurita* suggest pelagic turnover rates of ~4 to 5 days (Hansson 1997). This result implies that turnover rates may be sufficiently high to make jellyfish quantitatively important as nutrient sources during breakdown of blooms.

In Lurefjorden, western Norway, the coronate scyphomedusa *Periphylla periphylla* abounds year round and performs daily vertical migrations between the surface and several hundred meters (Fosså 1992, Youngbluth & Båmstedt 2001, Sørnes 2005). This makes *P. periphylla* well suited for experiments where large quantities of similarly sized animals are needed. Here, we used *P. periphylla* to study the degradation of dead gelatinous zooplankton. We quantified turnover rates of dead medusae through time in a pelagic *in situ* experiment and conducted a shipboard experiment that targeted the microbial activity associated with decaying jellyfish. A final series of laboratory incubations using bacterial isolates was executed to further assess positive and negative effects on specific bacterial species. In a companion paper, we examined the distribution of bacterial activity and microbial community composition relative to jellyfish biomass in Lurefjorden (Riemann et al. 2006).

MATERIALS AND METHODS

We carried out a series of experiments aimed at quantifying decomposition rates of various sizes of jellyfish in an *in situ* pelagic setting, as well as a series of shipboard and laboratory experiments investigating the interactions between dead jellyfish and the microbial community.

In situ* degradation experiment.** ***Experimental set-up: *Periphylla periphylla* were collected with a MIK net (2 m mouth diameter, 1 to 3 mm graded mesh) between 300 m and the surface in the deep basin of Lurefjorden, on 8 October 2003. On board the ship the jellyfish were emptied into clean, water filled buckets and stored at 5°C in darkness during transport back to the laboratory. *P. periphylla* were weighed individually (wet weight, to nearest 0.1 g) and placed individually in tagged nylon mesh bags (80% polyamide, 20% elastane). The bags stretched with their contents and had a mesh size of ~5 to 10 mm. This large mesh size allowed for colonization of both microbes and zooplankters, while ensuring transport of solutes and materials to the surrounding water. The bags were horizontally spaced at approximately 30 cm intervals and suspended individually at 1 or 8 m depth with fishing lines from the mesocosm platform in Raunefjorden, off Espegrend Marine Biological Station. These depths represented 2 different temperatures; mean temperature (±SD) was 10.1 ± 0.5°C at 1 m and 12.5 ± 0.1°C at 8 m. A small lead weight anchored the bag in the water. A total of 250 jellyfish, equally divided between 4 size categories (42.6 ± 8.14, 121 ± 12.3, 223 ± 12.3 and 300 ± 36.1 g, mean ± SD), were suspended, with the 3 smallest categories at the shallower depth. The size groups were chosen such that their initial weights did not overlap. We estimated that this number of jellyfish would allow for daily sampling for several weeks if needed, while allowing for randomization of samples. Because we needed to process many individuals, the time from capture until the last jellyfish had been suspended was approximately 12 h. Temperature and salinity were monitored throughout the study period.

At each daily sampling, 3 jellyfish of each size category were gently pulled to the surface and weighed in their bags, but without the line, weight and label. Wet weights were subsequently corrected for bag weight (mean ± SD, 14.8 ± 0.62 g in seawater, n = 7 bags), and the rate of change in wet weight was used as a proxy for degradation rates. The largest jellyfish were homogenized and frozen for subsequent determination of carbon and nitrogen content. Sampled jellyfish were discarded.

Carbon and nitrogen analysis: Known volumes of jellyfish homogenate were collected on precombusted (450°C for 4 h) Whatman GF/F filters. The filters were analyzed with a Carlo Erba model 1108 high-temperature combustion elemental analyzer, using standard procedures and a combustion temperature of 1030°C. Acetanilide was used for standardization and results were corrected for carbon content of blank filters. The first sample was taken after 6 h.

Shipboard experiments. We conducted 2 shipboard experiments in Lurefjorden to investigate the de-

velopment of bacteria around dead *Periphylla periphylla*.

Experimental set-up: In the first experiment we incubated 6 large dead *Periphylla periphylla* (101 ± 13.6 g, mean ± SD) individually in 5 l containers and monitored the development of the bacterial community. We also included 3 controls without *P. periphylla*. In the second experiment, we monitored bacterial abundance in 2 l containers with individual small (31.0 ± 1.0 g, mean ± SD, n = 3) or medium (63.8 ± 6.0 g, n = 4) dead *P. periphylla* and in 5 controls without *P. periphylla*. The setups for both experiments were similar, and the experiments were conducted in parallel in Lurefjorden on 23 to 25 April 2004. All materials were acid washed in 1 M HCl, and gloves were used at all times.

Sterile seawater for the incubation experiments was obtained by filtering seawater from ~100 m depth in Lurefjorden, obtained with a Rosette sampler, through a polypropylene capsule filter (5 to 10 µm nominal pore size, Advantec MFS) followed by a 0.22 µm capsule filter (Millipak-40, Millipore). Prior to filtration, the virgin capsule filters were flushed with ~5 l Milli-Q. The containers were inoculated with a 10% volume of surface (5 m) seawater, which had been 1.0 µm filtered (polycarbonate, MSI) to remove flagellates. The total volume was 4820 ml for the large and 1650 ml for the small containers.

Small and medium sized *Periphylla periphylla* were caught in MIK nets between 300 m and the surface. Larger specimens were scooped up directly from the surface water with buckets at night. We selected healthy looking intact animals and suffocated these in air for half an hour before the start of the experiment. At this time the jellyfish were also weighed and measured. The jellyfish were rinsed in 3 successive baths of 0.2 µm filtered seawater and subsequently added to the pre-filled containers. We denoted this t = 0. The experimental chambers were gently aerated to ensure oxic conditions and covered at all times to prevent light-generated breakdown of the porphyrin of the *P. periphylla*. The containers were partly submerged in a flowing surface-water-bath with a temperature of 10 to 11.5°C. The temperature at the inflow and outflow of the water bath was always the same. The ambient water temperature was ~8°C at the surface (Riemann et al. 2006).

We monitored the experiments for ~50 h and sampled approximately every 8 h (i.e. t = 2, 10, 18, 26, 33, 42 and 50 h). Samples were taken for determination of bacterial abundance in both the small and large containers (see below). The large containers were also sampled for total organic carbon (TOC), nutrients and bacterial production (see below). The required water volumes for the latter analyses made these unfeasible in the small containers.

Bacterial abundance: Aliquots of 25 (small containers) or 50 ml (large containers) were fixed with 0.2 µm filtered formalin and stored at 4°C for one month before bacterial enumeration. Bacteria in samples from the control incubations without added jellyfish were stained with SYTO 13 (Molecular Probes) and counted on a Becton Dickinson FACSCalibur flow cytometer (Gasol & del Giorgio 2000) using fluorescent beads (True counts, Becton Dickinson) as standards. In incubations with jellyfish, interference from released jellyfish cells prevented enumeration by flow cytometry. Instead, bacteria were stained with SYBR Green I (Molecular probes), filtered onto 0.22 µm black polycarbonate filters (Osmonics) and mounted in glycerol (Noble & Fuhrman 1998). More than 200 bacteria or >15 fields filter^{-1} were counted at 1250× using epifluorescence microscopy (Axioplan, Zeiss). Pictures were taken using an Olympus DP50 camera. To estimate variability among filters, duplicate filters were counted on 3 occasions. The average SD among filters was 6%.

Bacterial production: Bacterial production was measured by [^{3}H]-thymidine incorporation (Fuhrman & Azam 1982). For each sample, triplicate aliquots (10 ml) and a fixed blank were incubated with [methyl-^{3}H]-thymidine (10 nM final, Amersham Pharmacia Biotech) in polyethylene vials in a water bath with flowing surface seawater for ca. 1 h. Samples with 5% trichloroacetic acid added prior to the addition of [^{3}H]-thymidine served as blanks. Samples were filtered onto 0.2 µm mixed cellulose ester filters (Advantec MFS), rinsed carefully with ice-cold 5% trichloroacetic acid and counted by liquid scintillation spectrometry. Bacterial carbon production was calculated using 1.1×10^{18} cells mole^{-1} thymidine incorporated (Riemann et al. 1987) and a carbon-to-cell ratio of 20 fg C bacterium^{-1} (Lee & Fuhrman 1987).

TOC: Duplicate 12 ml samples were frozen in 15 ml polypropylene tubes and analyzed with a Shimadzu TOC-5000 high temperature catalytic oxidation instrument. Samples were acidified and sparged prior to analysis. Calculation of carbon concentrations was made using potassium hydrogen phthalate as a standard.

Nutrients: Three 12 ml samples were filtered through glass fiber filters (GF/F, Whatman) into 15 ml polypropylene tubes and frozen for subsequent analysis of ammonia, phosphate and nitrate/nitrite on a Bran & Luebbe TRAACS 800 autoanalyzer (Grasshoff et al. 1983).

Growth of bacterial isolates in response to *Periphylla periphylla*. To complement the shipboard experiments, we examined effects of *P. periphylla* on growth of marine bacterial isolates from various phylogenetic groups (Tables 1 & 2) in a laboratory setting. Generally, overnight bacterial cultures were diluted,

Table 1. Bacterial isolates used in incubation experiments with *Periphylla periphylla* homogenate

Name	GenBank accession number[c]	Phylogeny
BAL3[a]	U63935	α-Proteobacteria
BAL11[a]	U63939	α-Proteobacteria
BAL18[a]	U63944	γ-Proteobacteria
BAL203[b]	AY962019	γ-Proteobacteria
BAL37[a]	U63953	β-Proteobacteria
BAL208[b]	AY962020	Actinobacteria
BAL209[b]	AY962021	Actinobacteria
BAL22[a]	U63946	Bacteroidetes
BAL17[a]	U63943	Bacteroidetes

[a]Isolated by Pinhassi & Hagström (2000)
[b]Isolated from the Baltic Sea Proper on ZoBell plates 17 May 2004
[c]Nucleotide sequences and information can be accessed via www.ncbi.nlm.nih.gov/Entrez/

exposed to *P. periphylla* homogenate (see below), and optical density (OD) was monitored over time. Since the inhibition of bacterial growth observed in the shipboard experiments was suspected to be caused by a proteinaceous toxin (B. Johnston, pers. comm.), we included a control consisting of autoclaved *P. periphylla* homogenate in our set-up. Further, we included a negative control in which the added *P. periphylla* volume was substituted by sterile seawater (salinity = 33‰), to control for added salt from the *P. periphylla* tissue.

Periphylla periphylla homogenate: In all experiments, we used homogenate made immediately prior to each experiment. *P. periphylla* were caught in Lurefjorden 12 January 2005. Individual live animals of various sizes and individual body parts were blended with a food processor and immediately frozen in 50 ml Falcon tubes. Immediately prior to each experiment, *P. periphylla* samples were thawed and further homogenized with an Ultra-Turrax T25 (Janke & Kundel) at 24 000 rpm for several minutes. The homogenates were subsequently centrifuged (10 000 × *g*, 10 min, 4°C) to remove particles, which would otherwise interfere with OD measurements. Half of the supernatant was used untreated and the other half was autoclaved and used as control. Thawed samples and homogenates were stored on ice in darkness during set-up of experiments. The concentration of homogenate was quantified volumetrically, i.e. as ml added homogenate per ml ready solution, in all experiments. The homogenates were not characterized further.

The use of diluted, non-concentrated homogenates implies that the concentration in the experiments by definition can never exceed concentrations in a real, dead jellyfish. In the experiments with isolates, the homogenate was diluted ~8 to 100 times in the bacterial growth medium depending on the experiment. In nature, concentrations of solutes will build up in the boundary layer around the dead *Periphylla periphylla* and decrease with distance away from the jellyfish. The concentration will also depend on the amount, size and abundance of dead *P. periphylla*, molecular diffusion and ambient water flow.

General experimental set-up: The general set-up was similar for all experiments with bacterial isolates. While details pertaining only to specific experiments are described in their respective sections below, the general set-up was as follows. Bacterial isolates grown overnight in ZoBell medium (5 g peptone, 1 g yeast extract, 800 ml filtered seawater, 200 ml Milli-Q water, autoclaved 121°C, 20 min) were diluted with fresh ZoBell medium to an OD of 0.04 to 0.06 and aliquoted into autoclaved sodium glass-tubes (VWR 109986-17). *Periphylla periphylla* homogenate, autoclaved *P. periphylla* homogenate or sterile seawater (negative control) was added to a final volume of 4 ml. Bacterial growth, measured as OD directly in the tubes, was monitored over time using a Biowave CO8000 cell density meter (600 nm, VWR). Each measurement was corrected for background OD with a blank sample without bacteria. Separate blanks were used for each treatment (*P. periphylla* homogenate, autoclaved *P. periphylla* homogenate and seawater [salinity =

Table 2. Summary of experiments with bacterial isolates and *Periphylla periphylla* homogenate

Experiment	Isolates	Final *P. periphylla* concentration (ml ml^{-1})	Contents of homogenate (weight of animals used)
Screening	BAL3, BAL11, BAL18, BAL203, BAL208, BAL209, BAL37, BAL22, BAL17	0.04	Whole animal (92 g)
Concentration	BAL18, BAL208, BAL3	0.0125, 0.025, 0.05, 0.10, 0.15	Whole animal (~600 g)
Body parts	BAL18, BAL208	0.05	Tentacles from 5 animals (no data), tops from 5 animals (no data), bodies from 5 animals (no data), whole animal (83 g)
Size	BAL18, BAL208	0.05	Whole animals (5.9–900 g)

33‰], as well as for different concentrations of additions when applicable) as optical properties of these 3 treatments differ. All experiments were carried out in triplicate in darkness at room temperature (18 to 19°C) on a shaking table. Size and parts of *P. periphylla* specimens, concentration of homogenate and bacteria used in the various experiments are summarized in Table 2.

Screening: In the first experiment, we monitored the growth of 9 different bacterial isolates of various phyla, when exposed to *Periphylla periphylla* homogenate at a concentration of 0.04 ml *P. periphylla* homogenate ml^{-1} culture (Table 2). The volumetric concentrations of homogenate used were chosen to approximate concentrations in the shipboard experiment where whole jellyfish were added to 2 to 5 l volumes, corresponding to enrichment of ~0.02 to 0.05 g *P. periphylla* ml^{-1} seawater. An assumed density of *P. periphylla* of ~1 g cm^{-3} was applied in the conversion. The size of the *P. periphylla* used (Table 2) was approximately that of an average *P. periphylla* at the time of capture in Lurefjorden (Riemann et al. 2006). From this experiment, isolates were selected for further experimentation.

Response to increasing concentration of Periphylla periphylla homogenate: We estimated growth of 3 bacterial strains as a function of increasing volumetric concentrations of *P. periphylla* homogenate. 0, 50, 100, 200, 400, or 600 µl of homogenate was added to 3.4 ml medium with bacteria (Table 2). Sterile seawater was added to a final volume of 4 ml to ensure similar salt content in all treatments. The *P. periphylla* used here was larger than in the first experiment in order to ensure that all treatments got homogenate from the same animal, i.e. a comparable exposure. OD measurements were conducted using separate blanks without bacteria for each concentration and treatment (*P. periphylla* homogenate, autoclaved *P. periphylla* homogenate and seawater).

Impact of Periphylla periphylla size: Differently sized *P. periphylla* may possibly contain different compounds or amounts of growth suppressants per unit volume. We therefore examined the effect of *P. periphylla* size on bacterial growth by exposing 2 bacterial strains to equal volumetric amounts of homogenate, made from each of 10 whole animals ranging in size from 5.9 to 900 g wet weight (Table 2). In this experiment, separate *P. periphylla* size-specific and treatment-specific blanks without bacteria were used to ensure that the bacterial growth signal, measured as OD, was independent of potential differences in optical properties between homogenates.

Impact of Periphylla periphylla body parts: While larger metazoans are generally sensitive to nematocyst toxin, we hypothesized that microbes may react to components present in the body tissue of the jellyfish, rather than to the nematocyst toxins per se. We examined this by exposing 2 bacterial isolates to equal volumetric amounts of homogenate made from either tentacles, top dome (i.e. the ectoderm and mesoglea above the coronal furrow in the exumbrella), body (i.e. all other tissue except the tentacles), tentacles, and whole animals (Table 2). As the compounds affecting bacterial growth were unknown, no corrections were made for potential differences in tissue structure, chemical composition or density. Tissue specific blanks were used to ensure that the bacterial growth signal, measured as OD, was independent of any differences in optical properties between homogenates from different tissues.

Statistical analyses. Statistical analyses were done using the software R (R development Core Team 2005) and the package *nlme* (Pinheiro et al. 2005), unless otherwise noted. Details pertaining to statistical analyses are incorporated in the results.

RESULTS

In situ degradation experiment

We analyzed the degradation data by fitting exponential decay models to the wet weights as a function of time for each size group. The overall fits, as well as the coefficients were statistically significant for all sizes, with *b*-coefficients of 0.67 to 1.12 d^{-1} (Fig. 1A). We tested for differences between size groups in the exponentially decreasing part of the degradation curves with a homogeneity-of-slopes-model on the natural logarithms of wet weights. This analysis included the first 2 time-points for the small size group and the first 5 time-points for the other size groups. The slopes of the size groups were different as indicated by the interaction between initial size and time ($p < 0.000001$). A Tukey Honest Significant Difference (Tukey HSD) test revealed that all groups differed from one another (all p-values ≤0.0008). For the 3 size groups placed at a common temperature, turnover scaled with initial weight to a power of −0.43 ± 0.09, which is similar to the hypothesized 1/3 from surface to volume relationships (Fig. 1B), i.e. for similarly shaped objects with a linear dimension *r*:

$$\frac{surface}{volume} \propto \frac{r^2}{r^3} = \frac{1}{r} \propto \frac{1}{weight^{1/3}}$$

The C:N ratio of the decaying jellyfish increased in the beginning of the experiment and then decreased (Fig. 1C,D). The fit (not shown) of a second order polynomial confirmed this pattern; it gave a parabola with a maximum C:N at 3.4 d and a significant negative coefficient of the quadratic term ($p = 0.0055$).

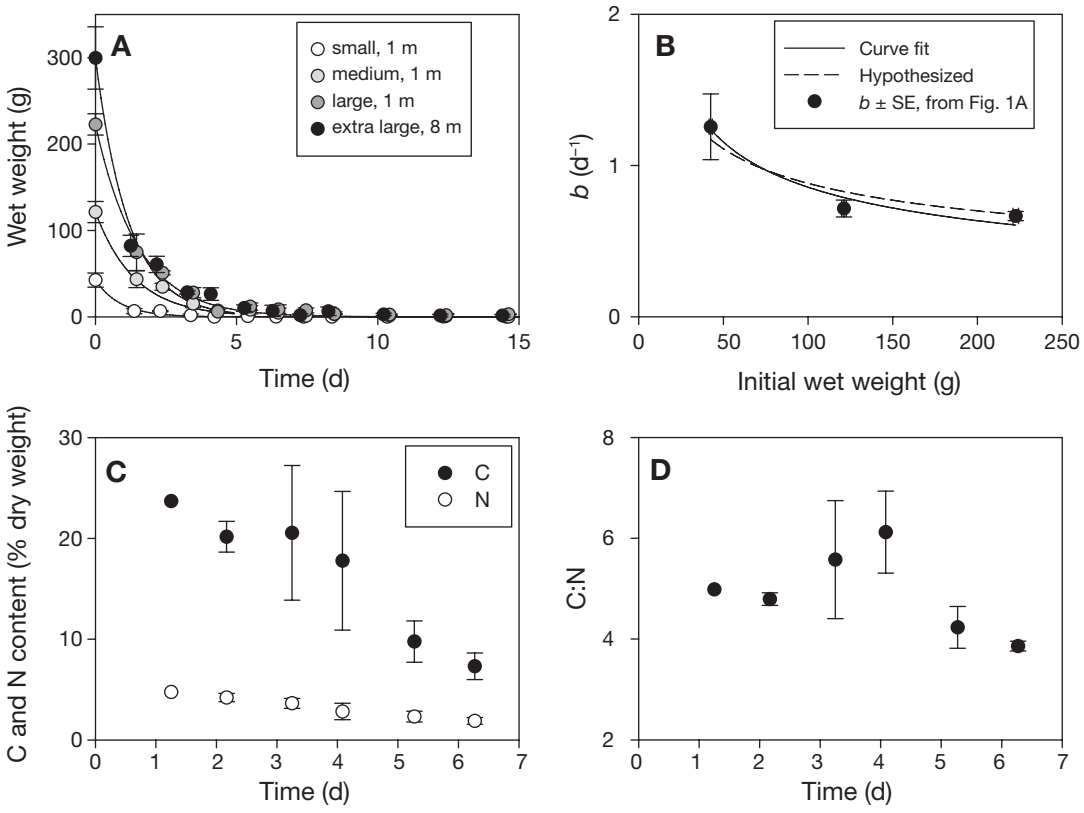

Fig. 1. *Periphylla periphylla. In situ* degradation experiment. (A) *P. periphylla* degradation. Data are mean ± SD. The curve fits are exponential decay models to the original non-averaged data; $y = ae^{-bx}$, where y is wet weight (g) and x is time. Curve fits with SE of coefficients in parentheses for the different size categories are: small $y = 42.08(\pm 0.91)e^{-1.12(\pm 0.22)x}$, $R^2 = 0.93$, n = 82; medium $y = 121.2(\pm 1.4)e^{-0.716(\pm 0.056)x}$, $R^2 = 0.98$, n = 84; large $y = 222.6(\pm 1.6)e^{-0.666(\pm 0.031)x}$, $R^2 = 0.99$, n = 80; extra large $y = 299.3(\pm 4.1)e^{-0.844(\pm 0.031)x}$, $R^2 = 0.97$, n = 84. All coefficients and curve fits are statistically significant with p-values < 0.05. Temperature during the first week was 10.1 ± 0.5°C at 1 m and 12.5 ± 0.1°C at 8 m. (B) Rate coefficients as a function of initial *P. periphylla* wet weight for the 3 size groups kept at the shallow depth. The dashed line is a power function with a forced slope of –1/3, which is that hypothesized from surface to volume ratio (see 'Results'), while the fitted solid line has a power of ~0.4 ± 0.09. (C) Carbon and nitrogen contents as a function of time for the extra large category. Data are only shown for the first week because 95% of the initial wet weight was degraded during the first 5 d. (D) C:N ratios from individual data points reported in (C)

Shipboard experiments: development of microbes around dead *Periphylla periphylla*

We hypothesized that the rapid breakdown observed in the *in situ* experiment in Raunefjorden was due mainly to microbial colonization and a large release of dissolved organic matter. This was further examined in the shipboard experiment where we monitored bacterial growth, carbon and nutrient release around dead *Periphylla periphylla*.

TOC and nutrients in the *Periphylla periphylla* enriched containers increased with time (Fig. 2C,D). We modeled the TOC data by linear regression with individual components in the slope, allowing for different variations in the 2 groups. The fixed effects of the slope differed between the enriched containers and the non-enriched controls ($p < 0.0001$). A Mann-Whitney test confirmed that PO_4^{3-} ($p = 0.020$) and NH_4^+ ($p = 0.020$) concentrations were higher in the enriched containers than in the controls, while $NO_2^- + NO_3^-$ concentrations were not ($p = 0.12$).

Despite the tremendous amount of released carbon and nutrients, bacteria showed strong bimodality in their response to the jellyfish enrichment. Growth was high in some experimental chambers, while negative in others (Fig. 2A,B). Growth rates varied between –0.2 and 7.0 d^{-1} during the first 40 h. The pattern was repeated in both experiments, and the controls were generally stable in all parameters measured, indicating a true experimental signal (Fig. 2). To statistically confirm this visual impression, we regressed the logarithms of the bacterial abundances on the time for each individual container separately. A Siegel-Tukey test (Lehman 1975) applied the individual slopes revealed a difference in the spreads between the *Periphylla periphylla* enriched group and the controls for both small ($p = 0.005$) and large ($p = 0.024$) containers (Fig. 2A,B).

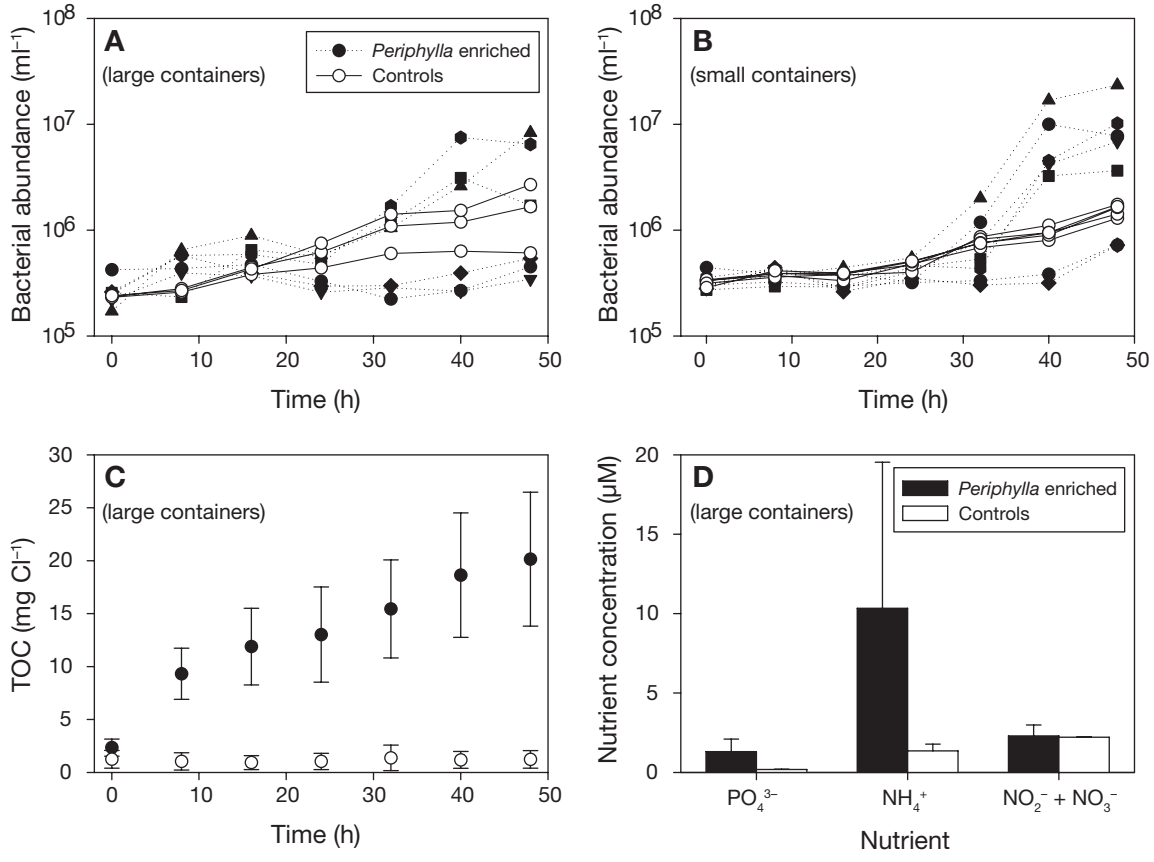

Fig. 2. Shipboard incubation experiment with natural bacterial assemblages from Lurefjorden. Development of bacterial concentrations in the incubation experiments in (A) large and (B) small containers. Symbol types represent individual containers, while fill represents treatment; black: containers with added *Periphylla periphylla* substrate; white: control containers without added substrate. (C) Mean (± SD) total organic carbon (TOC) content as a function of time in the large containers. Fills as in (A,B). (D) Mean (+ SD) nutrient concentrations in the large containers at the termination of the experiment

We examined bacterial morphology in the samples (Fig. 3). The bacterial assemblage used as inoculum in the incubations, the communities growing in the control incubations, as well as the bacterial communities in incubations where a decrease in bacterial abundance was seen, consisted of a mix of different bacterial morphotypes (e.g. Fig. 3A). In contrast, incubations with pronounced bacterial growth towards the end of the experiment were dominated (>90% of all cells) by single morphotypes (Fig. 3B–E). These monoculture-like bacteria extensively colonized cnidocytes released from *Periphylla periphylla* (Fig. 3H–J). Although these morphotypes varied between incubations, they generally consisted of large rod-shaped cells. In one incubation, only small coccoid cells were seen (Fig. 3B). No flagellates were observed, indicating that flagellate grazing during incubations was insignificant.

When overlaying the data in Fig. 2A,B with the bacterial morphology information (Fig. 3) and correcting for *Periphylla periphylla* sizes and water volumes used, an interaction between cell type and *P. periphylla* enrichment appears (Fig. 4A). Linear regressions revealed a biomass increase of the monoculture-like cells ($p = 0.0065$) and a decrease of the other cell types ($p = 0.012$) with increasing enrichment (Fig. 4A). The slopes of these lines differed ($p = 0.0065$). In this regression analysis we allowed for heteroscedasity when using the function gls of *nlme* (Pinheiro et al. 2005). The bacterial production measured in the large containers partially mirrored this picture (Fig. 4B). However, the large individual variation with time prevented adequate statistical analysis.

Growth of bacterial isolates in response to *Periphylla periphylla*

Screening

We hypothesized that inhibiting compounds in *Periphylla periphylla* would be destroyed by autoclavation. Thus, a lower OD in *P. periphylla* than in autoclaved

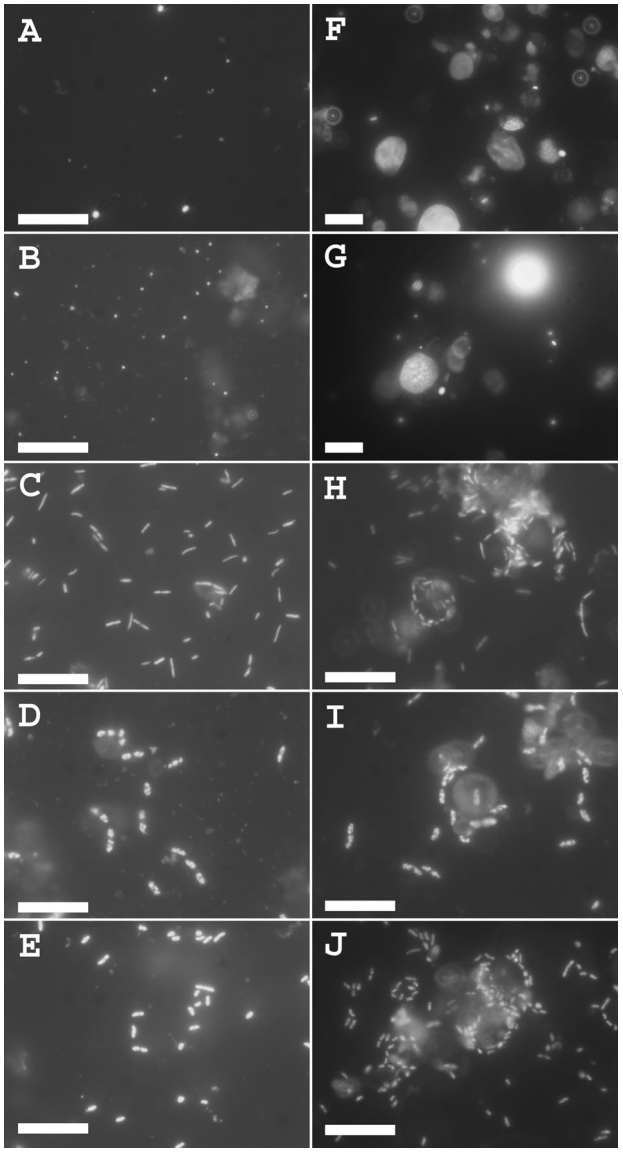

Fig. 3. Bacterial development in the shipboard incubation experiment. Scale bars = 10 µm. (A) Inoculum showing different cell morphologies at t = 0 h. (B–E) The different morphotypes, with monoculture-like appearance contributing >90% dominance at t = 40 h. (F–J) *Periphylla periphylla* cnidocytes and bacterial colonization of these. (F–G) Cells from *P. periphylla* at t = 8 h. (H–J) Colonization by bacterial morphotypes shown in (C–E) at t = 40 h

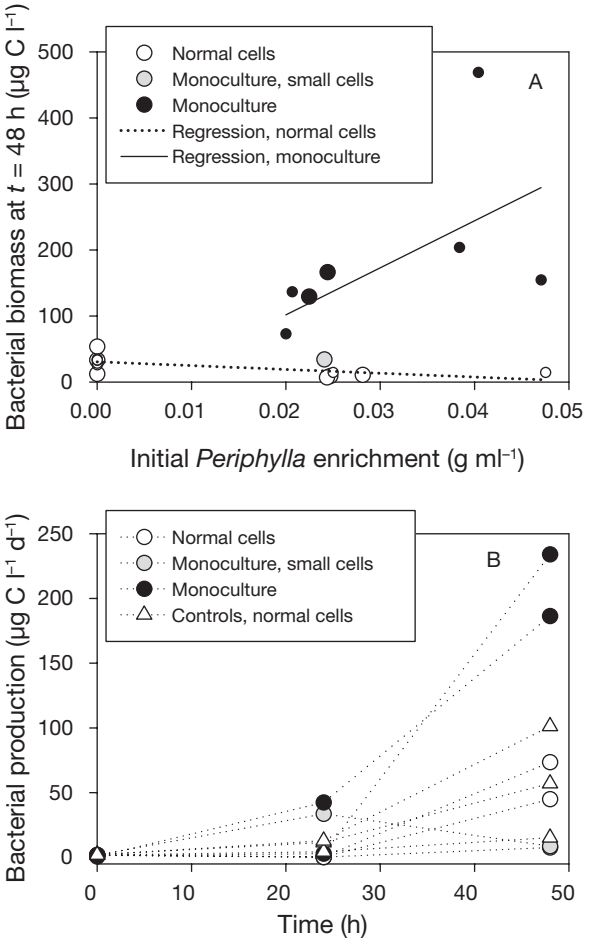

Fig. 4. Bacterial (A) biomass and (B) production in the shipboard incubation experiment. The cell morphotypes dominating in the microscopical counts (cf. Fig. 3, see also 'Results') are indicated by fills. 'Normal' cells imply a variety of cell types similar to that in Fig. 3A, and 'monoculture', implies cell morphologies similar to that in Fig. 3B–E. (A) Bacterial biomass as a function of initial enrichment. Large circles indicate the large container experiment (cf. Fig. 2A) and small circles the small container experiment (cf. Fig. 2B). Biomass at t = 48 h was calculated by assuming 20 fg C bacterium^{-1} (Lee & Fuhrman 1987). (B) Bacterial production as a function of time in the large container incubations as measured by thymidine incorporation. Dotted lines connect individual containers

P. periphylla treatments would suggest a net inhibitory effect of the *P. periphylla*, while a higher OD in autoclaved or raw *P. periphylla* than in the seawater controls would suggest a net enrichment. The screening experiment confirmed that different bacteria respond differently to *P. periphylla* (Fig. 5). We analyzed the OD curves as a function of time, accounting for the longitudinal data of the individual test tubes:

$$y_{ij} = (\beta_0 + b_{0i}) + (\beta_1 + \gamma_1 P_i + \delta_1 C_i + b_{1i}) t_{ij} + (\beta_2 + \gamma_2 P_i + \delta_2 C_i) t_{ij}^2 + \varepsilon_{ij} \quad (1)$$

Here, y_{ij} denotes the OD of test tube i at the j'th timepoint t_{ij}. Greek letters denote fixed effects, lower case Roman letters denote tube specific random effects and ε_{ij} denotes the homoscedastic normal errors. P_i and C_i are binary variables taking the value 1 if test tube i contains raw *Periphylla periphylla* (for P_i) or seawater (for C_i), and 0 otherwise. Thus, β_1 and β_2 are inter-

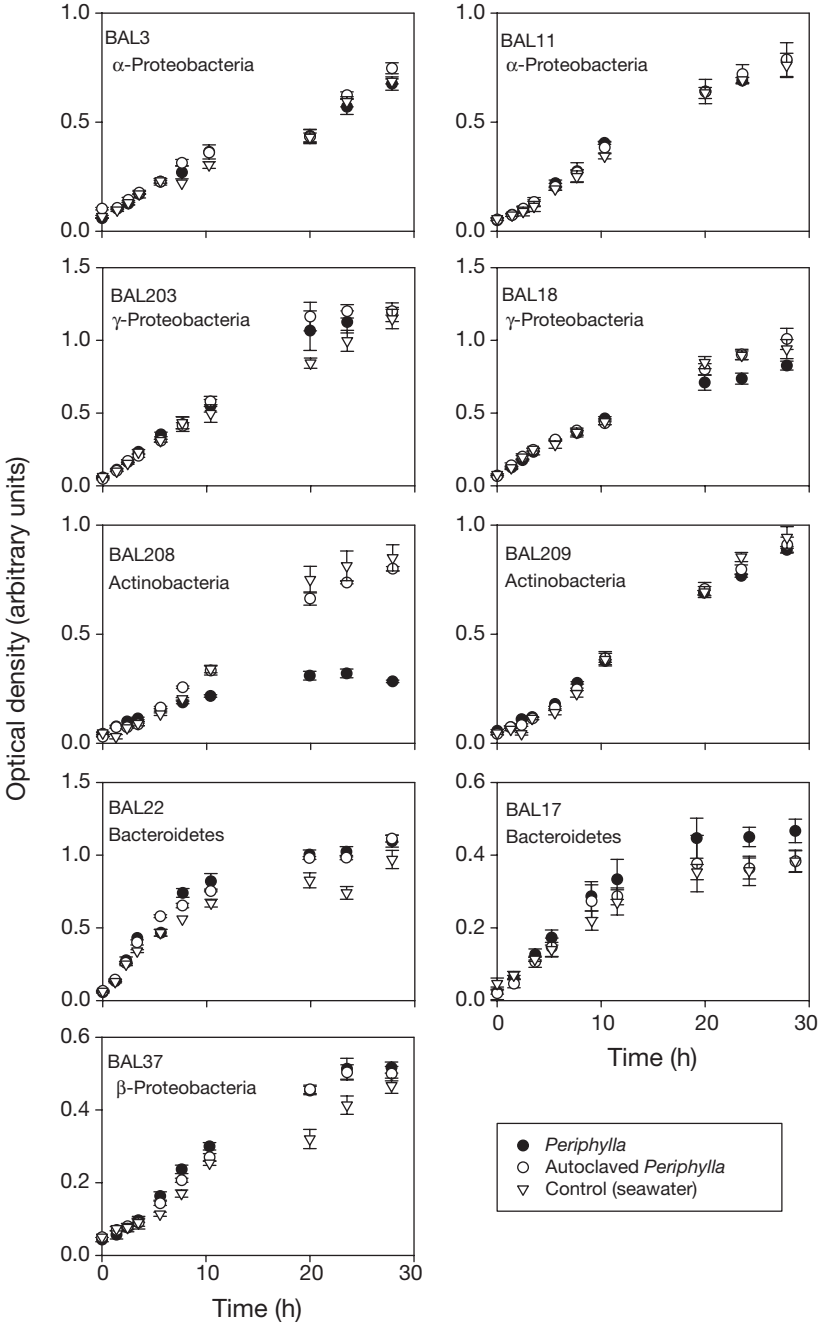

Fig. 5. Screening experiment with bacterial isolates. Optical density as a function of time for cultures with various bacterial isolates with added *Periphylla periphylla* homogenate, autoclaved *P. periphylla* homogenate or sterile seawater at equal volumetric concentrations. Name and phylogeny of isolates are indicated in the panels. Data are mean ± SD

preted as the average contribution to the linear and quadratic terms, respectively, for autoclaved *P. periphylla* and the corresponding γ's and δ's show how much *P. periphylla* and seawater deviate from the autoclaved *P. periphylla*. The differences ($\delta_1 - \gamma_1$) and ($\delta_2 - \gamma_2$) show how much larger the seawater controls are than the *P. periphylla* treatments. The linear and quadratic terms represent the initial growth and the slowing down of the growth for larger time values, respectively.

Periphylla periphylla had no effect on the 2 α-Proteobacteria (BAL3 and BAL11) (Fig. 5, Table 3). The β-Proteobacterium (BAL37) grew better on raw and autoclaved *P. periphylla* relative to the seawater controls, while there were no differences between raw and autoclaved *P. periphylla* (Fig. 5, Table 3). Similarly, BAL17 (Bacteroidetes) grew slower in the seawater treatment than in the *P. periphylla* treatments, but growth did not slow down as fast as in the *P. periphylla* treatments (Fig. 5, Table 3). The other Bacteroidetes isolate (BAL22) also grew better in the *P. periphylla* treatment than in the seawater control, while there were no differences between the raw and autoclaved *P. periphylla* treatments (Fig. 5, Table 3). *P. periphylla* affected the Actinobacteria (BAL208, BAL209) negatively. For BAL208, the initial growth was lower in the *P. periphylla* treatment than in the other treatments and growth also leveled off most rapidly in the *P. periphylla* treatment (Fig 5, Table 3). For BAL209 the growth curves were so linear that the quadratic term could be dropped. Doing so revealed that initial (linear) growth was lowest in the *P. periphylla* treatment (Fig. 5, Table 3). For BAL18 (γ-Proteobacteria) initial growth did not differ between treatments; however, the *P. periphylla* treatment slowed down most rapidly (Fig. 5, Table 3). BAL3, BAL18, and BAL208 were selected for further experiments.

Concentration experiment

While BAL208 responded negatively to increasing *Periphylla periphylla* concentrations, the signal was less obvious for BAL3 and BAL18 (Fig. 6). To quantify the signal as a function of homogenate addition we adjusted Eq. 1 to let coefficients depend linearly on *P. periphylla* concentration (*x*, dimensionless):

Table 3. Results from statistical analysis (Eq. 1) of screening experiment (Fig. 5). Estimated coefficients and their respective individual p-value in parentheses. **Bold** indicates statistical significance (i.e. $p < 0.05$). For BAL209 the growth curves were so linear that the quadratic term could be dropped. The lower of the 2 BAL209 lines reports the analysis without the quadratic term

Isolate	γ_1 *Periphylla* vs. Autoclaved	δ_1 Seawater vs. Autoclaved	$\delta_1 - \gamma_1$ Seawater vs. *Periphylla*	γ_2 *Periphylla* vs. Autoclaved	δ_2 Seawater vs. Autoclaved	$\delta_2 - \gamma_2$ Seawater vs. *Periphylla*
BAL3	0.0050	−0.0023	−0.0073	−0.0023	0.000053	0.00028
(α-Proteobacteria)	(p = 0.27)	(p = 0.63)	(p = 0.12)	(p = 0.15)	(p = 0.75)	(p = 0.87)
BAL11	0.0021	−0.0029	−0.0050	−0.00099	0.00081	0.00018
(α-Proteobacteria)	(p = 0.53	(p = 0.34)	(p = 0.14)	(p = 0.39)	(p = 0.43)	(p = 0.12)
BAL37	0.0039	**−0.0107**	**−0.015**	−0.00013	**0.00027**	**0.00040**
(β-Proteobacteria)	(p = 0.14)	**(p = 0.0001)**	**(p = 0.0001)**	(p = 0.17)	**(p = 0.0043)**	**(p = 0.00001)**
BAL18	0.0044	0.0056	0.00115	**−0.00040**	−0.00023	0.00017
(γ-Proteobacteria)	(p = 0.25)	(p = 0.15)	(p = 0.76)	**(p = 0.0037)**	(p = 0.091)	(p = 0.205)
BAL203	−0.0076	**−0.023**	**−0.015**	0.00017	**0.00055**	0.00037
(γ-Proteobacteria)	(p = 0.26)	**(p = 0.0006)**	**(p = 0.0190)**	(p = 0.42)	**(p = 0.0135)**	(p = 0.088)
BAL17	0.0032	**−0.0077**	**−0.011**	−0.000008	**0.00024**	**0.00025**
(Bacteroidetes)	(p = 0.24)	**(p = 0.0058)**	**(p = 0.0002)**	(p = 0.92)	**(p = 0.0076)**	**(p = 0.0058)**
BAL22	0.0089	−0.015	**−0.0242**	−0.0030	0.00032	**0.00062**
(Bacteroidetes)	(p = 0.25)	(p = 0.051)	**(p = 0.0025)**	(p = 0.28)	(p = 0.25)	**(p = 0.027)**
BAL208	**−0.012**	**−0.0016**	**−0.013**	**−0.00031**	0.000072	**0.00038**
(Actinobacteria)	**(p = 0.0061)**	**(p = 0.070)**	**(p = 0.0020)**	**(p = 0.042)**	(p = 0.63)	**(p = 0.0127)**
BAL209	−0.0011	−0.0021	−0.0010	−0.000023	0.00015	0.00017
(Actinobacteria)	(p = 0.77)	(p = 0.56)	(p = 0.78)	(p = 0.86)	(p = 0.26)	(p = 0.19)
BAL209	**−0.0017**	**0.0019**	**0.0036**			
(Actinobacteria)	**(p = 0.048)**	**(p = 0.024)**	**(p = 0.001)**			

$$y_{ij} = (\beta_0 + b_{0i}) + [\beta_1 + (\delta_1 + \gamma_1 P_i) x_{ij} + b_{1i}] t_{ij} + [\beta_2 + (\delta_2 + \gamma_2 P_i) x_{ij}] t_{ij}^2 + \varepsilon_{ij} \quad (2)$$

Eq. 2 is the model that gave the lowest value for the BIC criterion among different similar choices (see Pinheiro et al. 2005). Here, the variables have the same meaning as in Eq. 1, and x_{ij} denotes the *Periphylla periphylla* concentration at the j'th time point t_{ij}. The coefficients γ_1 and γ_2 represent the average difference for the concentration dependence of the coefficients between the *P. periphylla* and the autoclaved *P. periphylla* treatments.

For BAL3 the δ_1 and δ_2 do not differ from 0 (Table 4) implying that OD for the autoclaved *Periphylla periphylla* treatment was independent of concentration. There was, however, an effect of the raw *P. periphylla* treatment as $\gamma_1 > 0$ and $\gamma_2 < 0$ (Table 4). Thus, for small time values the *P. periphylla* treatment grew faster than the autoclaved *P. periphylla* treatment. As time increases, the quadratic term becomes more important, and the *P. periphylla* curve slows down faster than the autoclaved *P. periphylla* curve. This effect increases slightly with increasing concentration (Fig. 6, Table 4).

For BAL18, neither γ_1 nor γ_2 differ from 0 indicating that there were no differences between the 2 treatments (Table 4). However, because $\delta_1 > 0$ and $\delta_2 < 0$ (Table 4) small concentrations of both raw and autoclaved *Periphylla periphylla* enhance OD, while large concentrations suppress OD.

For BAL208, $\delta_1 > 0$, while δ_2 does not differ from 0 (Table 4). This implies an increase in OD with increasing concentrations for the autoclaved *Periphylla periphylla* treatment. While γ_1 does not differ from 0, $\gamma_2 < 0$ (Table 4). This means that the OD-curves start out equal, but for larger time values the *P. periphylla* curve turns downwards away from the autoclaved *P. periphylla* curve. This effect increases with increasing concentration (Table 4). We note that although the qualitative effect is similar in the *P. periphylla* treatment for BAL3 and BAL208, this effect is quantitatively much stronger on BAL208 than on BAL3; i.e. $\gamma_{2(BAL208)} / \gamma_{2(BAL3)} \approx 5$ (Table 4).

Impact of *Periphylla periphylla* size

We examined if *Periphylla periphylla* of various sizes suppress bacterial growth differently by exposing bacteria to the same volumetric concentration of homogenates made from *P. periphylla* of widely different sizes (x_i) (Fig. 7). Optical density (y_i) was measured after ~19 h. We fitted the non-linear model

$$y_i = \beta_0 + \beta_1 \exp(-x_i/\theta) + \varepsilon_i \quad (3)$$

separately for the raw and autoclaved *P. periphylla* treatments by the package *nls* of R. The errors ε_i are assumed to be independently normally distributed variables with the same variance.

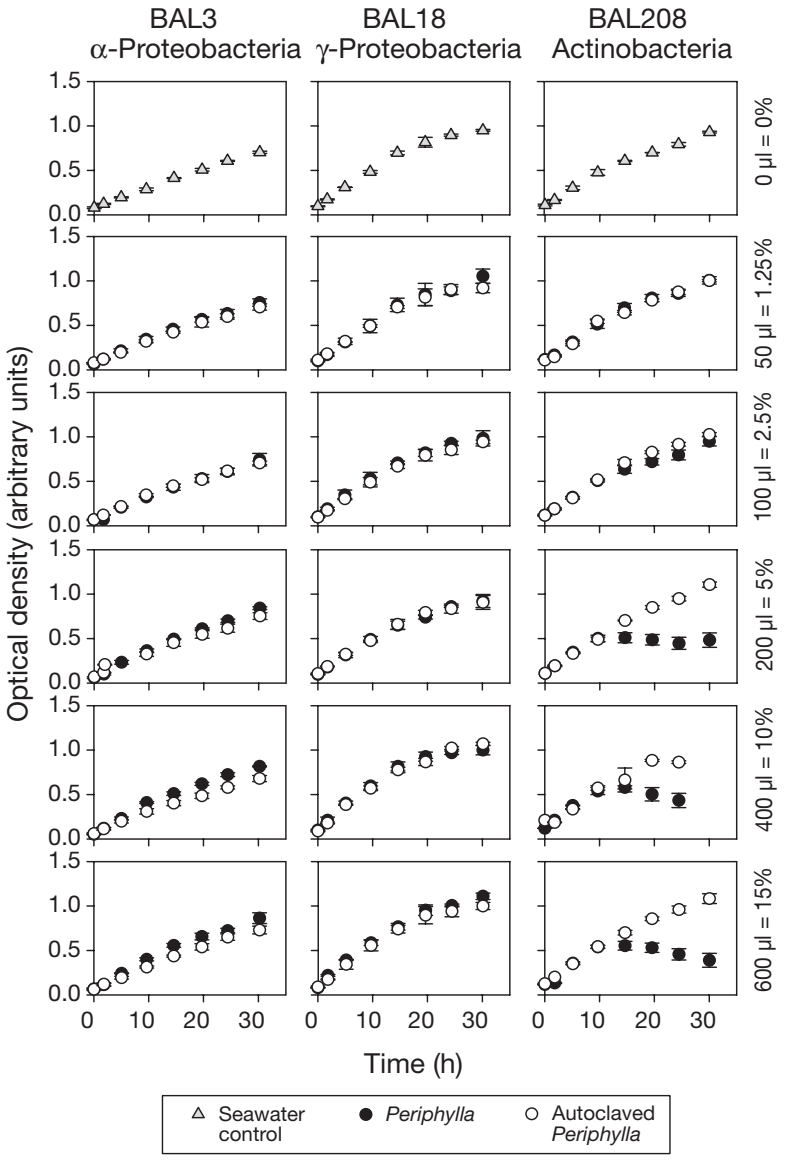

Fig. 6. Bacterial growth as a function of *Periphylla periphylla* homogenate concentration for 3 bacterial isolates. Each panel represents data of bacterial abundance (optical density) as a function of time for a given isolate and *P. periphylla* addition. The isolates are indicated by the column names and the volume of *P. periphylla* homogenate used at the right of each panel row. Data are mean ± SD

For the *Periphylla periphylla* treatment in BAL208 $\beta_1 > 0$ ($p < 0.0001$), indicating a decreased bacterial growth when exposed to homogenized *P. periphylla* of increasing size. In contrast, for the *P. periphylla* treatment in BAL18 $\beta_1 < 0$ ($p = 0.031$), indicating a weak increase in OD with increasing *P. periphylla* size. For autoclaved *P. periphylla* $\beta_1 > 0$ for both BAL18 ($p = 0.003$) and BAL208. In the latter case we are unable to report a p-value due to problems with convergence in the *nls* package. However, fitting with the *optim* package in R also gives a positive value, but it does not provide a p-value. In accordance with the visual impression of the figures, the asymptotic levels of the 2 treatments, that is the values for large *P. periphylla,* differ for BAL208 ($p < 0.000001$) while not for BAL18 ($p = 0.58$).

Impact of *Periphylla periphylla* body parts

Different tissues of *Periphylla periphylla* yielded different responses (Fig. 8). For BAL18, a 2-way ANOVA revealed an effect of body part ($p < 0.00001$), no effect of *P. periphylla* treatment ($p = 0.49$) and no interaction between treatment and body part ($p = 0.78$). A closer examination employing Tukey's HSD method (*TukeyHSD* in R) and including the control group 'seawater' showed that the 'top dome' falls below and the other body parts above the seawater control, but only the 'top dome' stands out from the others (Table 5).

For BAL208, the 2-way ANOVA showed effects of body part, treatment

Table 4. Results from statistical analysis (Eq. 2) of concentration experiment (Fig. 6). Estimated coefficients and their respective individual p-values in parentheses. **Bold** indicates statistical significance (i.e. $p < 0.05$)

Isolate	β_1	δ_1	γ_1	β_2	δ_2	γ_2
BAL3	**0.027**	0.0000022	**0.000017**	**−0.00019**	−0.00000006	**−0.00000031**
(α-Proteobacteria)	**(p < 0.0001)**	(p = 0.28)	**(p < 0.0001)**	**(p < 0.0001)**	(p = 0.35)	**(p = 0.0001)**
BAL18	**0.050**	**0.000014**	**0.0000073**	**−0.00071**	**−0.00000032**	−0.00000018
(γ-Proteobacteria)	**(p < 0.0001)**	**(p = 0.0001)**	**(p = 0.056)**	**(p < 0.0001)**	**(p = 0.0024)**	(p = 0.12)
BAL208	**0.041**	**0.000017**	−0.00000050	**−0.00047**	−0.00000024	**−0.0000016**
(Actinobacteria)	**(p < 0.0001)**	**(p < 0.0001)**	(p = 0.90)	**(p = 0.0001)**	(p = 0.075)	**(p < 0.0001)**

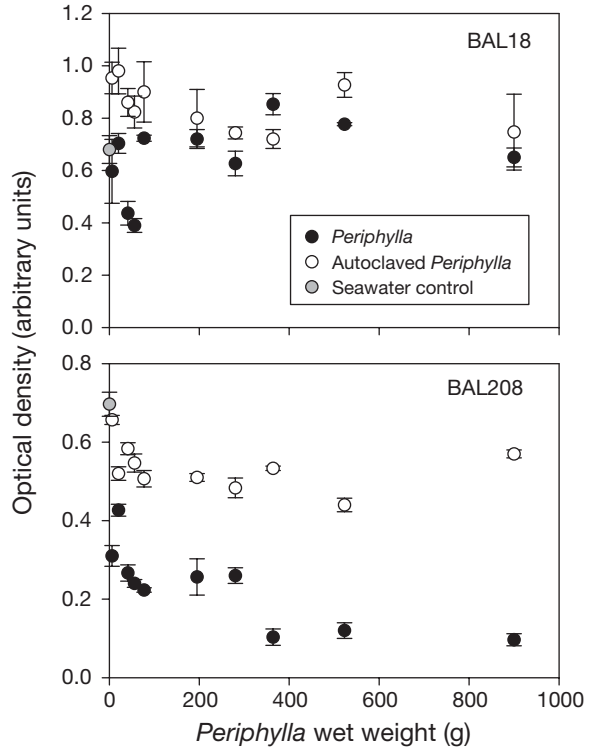

Fig. 7. Response of 2 isolates to homogenate made from differently sized *Periphylla periphylla*. Data are reported as optical density at t = 19 h and are mean ± SD. The isolate names are indicated in the panels

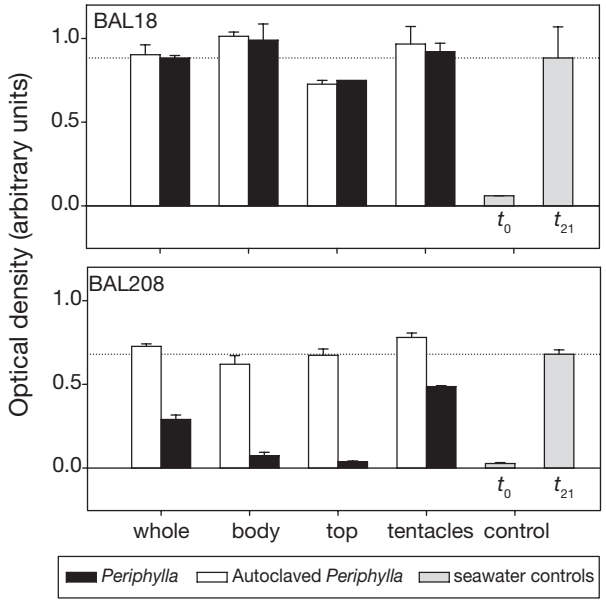

Fig. 8. Optical density at t = 21 h for 2 bacterial isolates exposed to homogenate made from different *Periphylla periphylla* tissues. The seawater control is given also for t = 0 and equals the start concentration for all treatments. The isolate names are indicated in the panels. BAL18 was also observed at t = 30 h with similar results (not shown)

and an interaction between body part and treatment (all p-values <0.000001). Tukey's HSD method generally revealed that the autoclaved treatments did not differ from the seawater control, while OD in the *Periphylla periphylla* treatments were lower than in the controls (Table 6). The *P. periphylla* treatments 'body' and 'top dome' did not differ from one another (Fig. 8). Apparently, the growth suppressing compounds are strongest or more concentrated in the top dome and weakest in the tentacles (Fig. 8, Table 6). Most of this effect is destroyed in the autoclavation.

DISCUSSION

Degradation of jellyfish

The shipboard and laboratory experiments suggested that only some bacterial species thrive around dead *Periphylla periphylla,* while others are inhibited by this jellyfish (Figs. 2 to 5). Despite this, pelagic turnover of jellyfish is rapid (Fig. 1). If correcting for temperature differences, assuming a Q_{10} of 2.3 (Bidle et al. 2002), the decay times of 4.1 to 7 d (10 to 12°C) observed here (Fig. 1) are faster than those measured on *Aurelia aurita* immobilized in nets in large tanks with Swedish fjord water (4.9 d, 15 to 17°C, initial weight 149 to 428 g, Hansson 1997). Assuming extensive microbial colonization and degradation of *P. periphylla* tissue, nitrogen-rich compounds would presumably be hydrolyzed faster than carbon-rich pools such as polysaccharides, analogous to degradation of marine snow (Smith et al. 1992). In accordance, the C:N ratio increased during the first 3 to 4 days (Fig. 1D) of degradation. We did not quantify bacterial production or colonizers in the *in situ* experiment. When pulling up the jellyfish, clouds of motile organisms, including oncaid and harpacticoid copepods, as well as particulate matter originating from the jellyfish were visible (J. Titelman, T. Sørnes, pers. obs.). Presumably, colonizers consist of organisms such as bacteria, flagellates, ciliates and copepods, similar to those observed on marine snow (Kiørboe 2000). Ultimately, the residence time in suspension determines the contribution of dead jellyfish as a substrate for pelagic organisms. Settling rates are a function of diameter and density, both of which may be affected by differential remineralization (Berelson 2002). Inhibitory effects on microbial activity by *P. periphylla* are apparently size and tissue dependent (Figs. 7 & 8), perhaps affecting residence time in the water column.

Some jellyfish regulate their buoyancy to become neutral or even positively buoyant, while others are negatively buoyant (Mills 1981, Graham et al. 2001). Live *Periphylla periphylla* in Lurefjorden hang motion-

Table 5. Results from Tukey HSD test of body part experiment for BAL18. Values are Bonferroni adjusted p-values. **Bold** indicates significant differences. The tested groups correspond to those depicted in Fig. 8. Treatments were not considered in this analysis as the 2-way ANOVA showed no effect of treatment for this isolate

	Top	Body	Tentacles	Whole	Seawater control
Top	–				
Body	**0.00005**	–			
Tentacles	**0.0012**	0.69	–		
Whole	**0.016**	0.14	0.79	–	
Seawater	0.093	0.23	0.80	0.99	–

less for up to 2 h at mid depths (Youngbluth & Båmstedt 2001), while they sink slowly if not pulsing when residing in the less saline surface water (Sørnes 2005). Similarly, live hydromedusae sink at rates from 0 to ~1700 m d^{-1} in the laboratory, depending on species and condition (Mills 1981). Unfortunately, there are few published sinking rates available, and dead and live jellyfish likely sink at different rates. For comparative purposes only, abandoned houses (mean length 68 ± 24 cm) of the large appendicularian *Bathochordeaus* sink at ~820 m d^{-1} (Hamner & Robison 1992), while houses (mean diameter = 1.13 mm) of the smaller *Oikopleura dioica* sink at ~120 m d^{-1} (Hansen et al. 1986), and dead *Aurelia aurita* medusae are often found drifting at the surface (pers. obs.).

Live jellyfish release DOC at a rate of ~0.012 mg C g wet $weight^{-1}$ d^{-1} (*Aurelia aurita,* in Hansson & Norrman 1995) and may contribute substantially to the pool of TOC in jellyfish rich environments (*Periphylla periphylla*, in Riemann et al. 2006). In comparison dead jellyfish leak DOC to the surrounding water at a rate of ~0.36 mg C g wet $weight^{-1}$ d^{-1} (calculated from Fig. 2), suggesting that dead jellyfish may provide hotspots of substrate in pelagic (and benthic) environments. Rapid turnover of dead jellyfish by microbes and zooplankters (Fig. 1) may be quantitatively important for returning energy to the pelagic foodweb at times of high jellyfish abundance. However, the extent of degradation mediated by bacteria varies with jellyfish species, size and body part, as well as with the composition of the local bacterial assemblage.

Periphylla periphylla as a substrate

In Lurefjorden, the density gradients are weak below ~50 m (Eiane et al. 1999, Riemann et al. 2006). The density of *Periphylla periphylla* from Lurefjorden is crudely estimated at ~1.05 ± 0.02 g cm^{-3} (mean ± SD, from wet weights and morphometrics of 15 animals), suggesting that dead *P. periphylla* would sink slowly out of the water column. Given decay coefficients of 0.67 to 1.12 d^{-1} (Fig. 1) a significant portion of dead jellyfish in such systems remain subject to some pelagic degradation, and the weight of dead jellyfish would be substantially reduced, of course depending on the depth at which the final travel started (cf. Fig. 1).

Table 6. Tukey HSD test of *Periphylla* body part experiment for BAL208. Values are Bonferroni adjusted p-values resulting from testing the groups in the vertical list against the groups in the horizontal header (all against all). **Bold** indicates significant differences. The tested groups correspond to those depicted in Fig. 8. Treatments are indicated in parentheses

	Top (*Periphylla*)	Top (autoclaved)	Body (*Periphylla*)	Body (autoclaved)	Tentacles (*Periphylla*)	Tentacles (autoclaved)	Whole (*Periphylla*)	Whole (autoclaved)	Seawater control
Top (*Periphylla*)	–								
Top (autoclaved)	**<0.00001**	–							
Body (*Periphylla*)	0.786	**<0.00001**	–						
Body (autoclaved)	**<0.00001**	0.366	**<0.00001**	–					
Tentacles (*Periphylla*)	**<0.00001**	**<0.00001**	**<0.00001**	**0.00039**	–				
Tentacles (autoclaved)	**<0.00001**	**0.00441**	**<0.00001**	**0.00004**	**<0.00001**	–			
Whole (*Periphylla*)	**<0.00001**	**<0.00001**	**<0.00001**	**<0.00001**	**<0.00001**	**<0.00001**	–		
Whole (autoclaved)	**<0.00001**	0.366	**<0.00001**	**0.00440**	**<0.00001**	0.366	**<0.00001**	–	
Seawater control	**<0.00001**	0.999	**<0.00001**	0.236	**<0.00001**	**0.00809**	**<0.00001**	0.529	–

Youngbluth & Båmstedt (2001) suggested that *P. periphylla* in Lurefjorden die a natural death in absence of predators. Video observations with the ROV 'Aglantha' at the bottom of Lurefjorden and neighboring fjords (e.g. Sognefjorden, ~1200 m) from 1997 to 2004 have usually revealed few dead jellyfish (J. Titelman, T. Sørnes & U. Båmstedt pers. obs.), suggesting low mortality or that *P. periphylla* are degraded or consumed before reaching the bottom. Also, occasional observations from shallow-water bottoms (5 m) suggest that dead *P. periphylla* may rapidly attract benthic scavengers.

To examine the degradation of *Periphylla periphylla,* we conducted a shipboard experiment using natural bacterial assemblages from Lurefjorden. Despite tremendous carbon and nutrient release from the dead jellyfish (Fig. 2), only some bacterial morphotypes were apparently able to use this substrate while others were inhibited (Figs. 2 to 4). The positive response was observed as extensive growth and a monoculture-like appearance in some incubations, and as pronounced colonization of *P. periphylla* cells. In corroboration, our laboratory experiments using 9 bacterial isolates, demonstrated that homogenized *P. periphylla* tissue selectively inhibited several bacteria, one γ-Proteobacterium (BAL18) and 2 Actinobacteria (BAL208 and BAL209) (Fig. 5). Hence, some, but not all, bacterial species can actively degrade *P. periphylla* tissue. Therefore, the bacterial community composition may affect the role of jellyfish as trophic links in specific pelagic foodwebs.

We chose bacterial isolates from various phylogenetic groups to get an indication of whether certain groups were inhibited by *Periphylla periphylla*. Our finding that a γ-Proteobacterium (BAL18) and the Actinobacteria (BAL208 and BAL209) are sensitive to *P. periphylla* contrast with those of Long & Azam (2001) and Grossart et al. (2004), who showed that these phylogenetic groups were the most prolific producers of antagonistic molecules and also the most resilient to them, while members of Bacteroidetes were the most sensitive to antagonistic compounds. Here, members of Bacteroidetes rather benefited from the *P. periphylla* homogenate in the laboratory experiments (Fig. 5). Bacteroidetes also dominated in Lurefjorden, thriving at depths with high *P. periphylla* biomass (Riemann et al. 2006). The likely functional variability of bacterial species within these broad phylogenetic groups and the unknown resemblance of inhibitory compounds produced by *P. periphylla* (this study) and by bacteria (Long & Azam 2001, Grossart et al. 2004) make direct comparisons between these studies speculative. Still, our findings demonstrate that *P. periphylla* specifically inhibits certain bacteria. Similarly, methanolic extracts of the jellyfish *Cassiopeia* sp. inhibited some strains of *Bacillus* and *Pseudomonas* (Bhosale et al. 2002).

Inhibition of bacterial growth

Bacteria that responded negatively to fresh *Periphylla periphylla* generally grew well on autoclaved extract. This suggests that at least one of the compounds inhibiting bacterial growth is of proteinaceous nature and denatured by autoclavation. However, some inhibition occurred also in the autoclaved treatments, albeit with signals of a lower magnitude. For example, the scaling to *P. periphylla* size and body tissue resembled those in the fresh treatments (Figs. 7 & 8). Thus, several compounds, probably of different nature, likely interact to cause the inhibition. Interestingly, the non-pigmented top dome generated the strongest inhibition (Fig. 8). This tissue is generally low in protein and contains only ~1/9 the amount of protein per unit dry weight relative to the pigmented muscular tissue and tentacles (U. Båmstedt et al. unpubl.). The reddish pigment in *P. periphylla* is porphyrin (Jarms et al. 2002), which becomes toxic to the jellyfish itself and also to many Gram-positive bacteria (but not Gram-negative bacteria) when photoactivated (Malik et al. 1990). Apart from the Actinobacteria (BAL208 and BAL209), all isolates were Gram-negative, among them BAL18 that responded negatively to *P. periphylla* (Figs. 5 & 7). The top dome does not contain pigments. Also, our experiments were conducted in darkness to avoid photoactivation. An alternative inhibitor may be nematocyst toxin. Cnidarian nematocyst toxins are generally cytolytic, hemolytic and neurotoxic (Bailey et al. 2003). Analyses of the sodium channel neurotoxin from nematocysts of another cnidarian (a sea anemone, *Actinia equina*) demonstrated a conserved stretch of residues that were similar to precursors of antimicrobial peptides from frogs (Anderluh et al. 2000). However, in the present study the response to tentacles, where nematocyst density is highest, was less dramatic than to other tissues (Fig. 8). Thus, neither porphyrin nor nematocyst toxin solely explain the inhibitory effects of *P. periphylla* on bacterial growth. Antimicrobial properties of different tissues may also depend on their enervation (Kasahara & Bosch 2003). In the freshwater cnidarian *Hydra*, tissue lacking neurons induced a stronger inhibitory response of strains of *Escherichia coli* and *Bacillus subtilis* than did tissue with neurons (Kasahara & Bosch 2003). Such an explanation may contribute to the strong signal observed for the dome tissue (Fig. 8).

The concentration experiment suggested that the inhibitory compounds generally decreased the maximum sustainable bacterial concentration for BAL208 (Actinobacteria) (Fig. 6, Table 4). The negative effects of increasing *Periphylla periphylla* concentration exist also for the α-Proteobacterium (BAL3), despite its growth being generally higher in the *P. periphylla*

treatment than in the autoclaved treatment (Fig. 6, Table 4). This indicates that higher concentration of jellyfish compounds (or more dead jellyfish) may negatively affect bacteria that are seemingly unaffected at lower concentrations. The isolates' different growth dynamics were also evident in the 'body part' experiment. The variable response of BAL18 between experiments may stem from variation in *P. periphylla* sizes used (cf. Fig. 7, Table 2).

While both BAL18 and BAL208 responded negatively to *Periphylla periphylla* homogenate in general, the response of BAL18 was more pronounced for small *P. periphylla* than for large, while for BAL208, the opposite was the case (Fig. 7). Given that bacterial growth depended on *P. periphylla* size (Fig. 7), it is possible that additional isolates would have reacted negatively to the *P. periphylla* homogenate had we used a differently sized animal to make homogenate for the initial screening (Fig. 5). *P. periphylla* size varies over several orders of magnitude (0.1 to ~5000 g wet weight) (Youngbluth & Båmstedt 2001, Sørnes 2005, Riemann et al. 2006). In addition to being subject to potential differential degradation of various tissues, a dead large *P. periphylla* would thus potentially increase both the power and amount of inhibitory compounds available to sensitive bacteria. Given the variation in bacterial inhibition to various *P. periphylla* sizes, concentrations and tissues, it seems likely that *P. periphylla* contain several compounds probably both of non-nematocystic (cf. Zhang et al. 2003) and nematocystic origin to which bacteria respond.

CONCLUSION

Dead jellyfish, and the chemical plumes (cf. Kiørboe & Jackson 2001) around them, constitute habitats and food for both microorganisms and larger invertebrates. The fate of dead jellyfish and the role of jellyfish as sinks or links in pelagic foodwebs are determined by a combination of (1) physical and chemical constraints preventing or facilitating break-up, degradation and sinking and (2) species-specific suitability as a substrate for microbes and the specific microbial community composition (cf. Riemann et al. 2006). In addition, other studies (Bullard & Hay 2002) have revealed that (3) species-specific palatability to scavenging animals is also of importance for the fate of dead gelatinous matter.

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