

RESEARCH ARTICLE

Niche overlap between a cold-water coral and an associated sponge for isotopically-enriched particulate food sources

Dick van Oevelen^{1*}, Christina E. Mueller¹, Tomas Lundälv², Fleur C. van Duyl³, Jasper M. de Goeij⁴, Jack J. Middelburg⁵

1 Department of Estuarine and Delta Systems, Royal Netherlands Institute for Sea Research (NIOZ-Yerseke) and Utrecht University, Yerseke, The Netherlands, **2** Sven Lovén Centre for Marine Sciences, Tjärnö, University of Gothenburg, Strömstad, Sweden, **3** Department of Marine Microbiology and Biogeochemistry, Royal Netherlands Institute for Sea Research (NIOZ- Texel) and Utrecht University, Texel, The Netherlands, **4** Department of Freshwater and Marine Ecology, Institute for Biodiversity and Ecosystem Dynamics, University of Amsterdam, The Netherlands, **5** Utrecht University, Department of Earth Sciences, Utrecht, The Netherlands

* Dick.van.Oevelen@nioz.nl



OPEN ACCESS

Citation: van Oevelen D, Mueller CE, Lundälv T, van Duyl FC, de Goeij JM, Middelburg JJ (2018) Niche overlap between a cold-water coral and an associated sponge for isotopically-enriched particulate food sources. PLoS ONE 13(3): e0194659. <https://doi.org/10.1371/journal.pone.0194659>

Editor: Kay C. Vopel, Auckland University of Technology, NEW ZEALAND

Received: October 27, 2017

Accepted: March 7, 2018

Published: March 26, 2018

Copyright: © 2018 van Oevelen et al. This is an open access article distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Data Availability Statement: All relevant data are available at <https://doi.org/10.5281/zenodo.1198189> (DOI: [10.5281/zenodo.1198189](https://doi.org/10.5281/zenodo.1198189)).

Funding: This research was supported by the CALMARO project (FP7/2007-2013) within the European Community's Seventh Framework Program (FP7/2007-2013), the Netherlands Organisation for Scientific Research (VIDI grant no. 864.13.007 to DvO) and has received funding from the European Union's Horizon 2020 research and

Abstract

The cold-water coral *Lophelia pertusa* is an ecosystem engineer that builds reef structures on the seafloor. The interaction of the reef topography with hydrodynamics is known to enhance the supply of suspended food sources to the reef communities. However, the reef framework is also a substrate for other organisms that may compete for the very same suspended food sources. Here, we used the passive suspension feeder *Lophelia pertusa* and the active suspension feeding sponge *Hymedesmia coriacea* as model organisms to study niche overlap using isotopically-enriched algae and bacteria as suspended food sources. The coral and the sponge were fed with a combination of ¹³C-enriched bacteria/¹⁵N-enriched algae or ¹⁵N-enriched bacteria/¹³C-enriched algae, which was subsequently traced into bulk tissue, coral skeleton and dissolved inorganic carbon (i.e. respiration). Both the coral and the sponge assimilated and respired the suspended bacteria and algae, indicating niche overlap between these species. The assimilation rates of C and N into bulk tissue of specimens incubated separately were not significantly different from assimilation rates during incubations with co-occurring corals and sponges. Hence, no evidence for exploitative resource competition was found, but this is likely due to the saturating experimental food concentration that was used. We do not rule out that exploitative competition occurs in nature during periods of low food concentrations. Food assimilation and respiration rates of the sponge were almost an order of magnitude higher than those of the cold-water coral. We hypothesize that the active suspension feeding mode of the sponge explains the observed differences in resource uptake as opposed to the passive suspension feeding mode of the cold-water coral. These feeding mode differences may set constraints on suitable habitats for cold-water corals and sponges in their natural habitats.

innovation programme under grant agreements No. 678760 (ATLAS) and No. 679849 (SponGES). This output reflects only the author's view and the European Union cannot be held responsible for any use that may be made of the information contained therein. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing interests: The authors have declared that no competing interests exist.

Introduction

Sessile suspension feeders extract organic resources from the water column and contribute significantly to the cycling of organic matter and inorganic nutrients in many marine ecosystems [1–4]. Being non-mobile, the growth of sessile suspension feeders is mostly limited by availability of space and food in the ambient water column [5]. Some suspension feeders, such as calcifying corals, significantly modify the topography of the seafloor by forming large reef structures. The interaction of the reef topography with hydrodynamics is known to enhance the supply of limiting resources to the reef communities making them archetypical examples of ecosystem engineers [6]. Yet, these structures may also be used as habitat by other suspension feeders that in turn may compete with the ecosystem engineer for the very same suspended resources.

Cold-water corals are passive suspension feeders that grow in the deep sea, where organic resources are scarce due to degradation of surface-produced organic matter during the downward transit to the seafloor [7]. *Lophelia pertusa* (Linnaeus, 1758) is the dominant reef-building cold-water coral species in the North Atlantic Ocean [8] that forms reef structures of several meters to hundreds of meters in height [9–11]. These structures interact with the (tidal) hydrodynamics [12, 13] in a way that the flux of limiting organic resources to the reef communities is temporarily enhanced [14–16]. Moreover, field and laboratory observations indicate that *L. pertusa* feeds on a broad range of different food sources ranging from dissolved to particulate suspended organic matter [17–20], which further increases its capacity to utilize resources under limiting conditions.

The carbonate reef framework is also an important site for settlement, refuge and feeding ground for many associated organisms [21]. A dominant taxonomic group of associated organisms in cold-water coral reef communities are sponges [22, 23]. Sponges are active suspension feeders with a high filtering capacity that allows them to extract food particles from the water column [24]. The food spectrum of deep-water sponges is broad [25–27] and thereby overlaps, at least partially, with that of cold-water corals. Habitat formation provided by the cold-water coral reef framework may therefore lead to exploitative competition for limiting suspended resources between cold-water corals and associated sponges if they share suspended resources.

Hymedesmia coriacea (Fristedt, 1885) is a sponge species that is frequently found on Atlantic cold-water coral reefs [28], where it often lives in close contact with living *L. pertusa* and sometimes overgrows the coral [29]. We used *L. pertusa* and *H. coriacea* as coral–sponge couple to study resource competition using isotopically-labelled algae and bacteria as suspended food sources. The coral *L. pertusa* and the sponge *H. coriacea* were fed with a combination of ^{13}C -enriched bacteria/ ^{15}N -enriched algae or ^{15}N -enriched bacteria/ ^{13}C -enriched algae, both separately and placed together. Food assimilation into tissue and coral skeleton, and respiration was used to investigate resource uptake and metabolic activity of the deep-water coral and sponge. A comparison of food processing rates of separate coral and sponge specimens versus the coral-sponge couple allowed identifying whether niche overlap occurred.

Materials and methods

The experiments described below follow a comparable approach as in Mueller et al. [30], Middelburg et al. [31] and in particular Van Oevelen et al. [32]. The data of the ‘coral only’ experiments in the present manuscript are identical to those described in Van Oevelen et al. [32], but are here presented in different units to facilitate comparison with the associated sponge.

Sampling location and maintenance

Corals and sponge specimens were collected from the Tisler Reef under permission granted by the Norwegian Directorate of Fishery. The Tisler Reef is located east of the Tisler islands in the sill area of the Koster fjord that connects to the Skagerrak at the border between Norway and Sweden [10]. The living parts of the reef cover a large part of the sill and extend over an area of approximately $1,200 \times 200$ m at a water depth of 70 to 155 m [10]. The flow direction over the sill fluctuates irregularly between NW and SE with current velocities that are generally between 0 to 50 cm s^{-1} but can reach peaks in excess of 70 cm s^{-1} [33]. The water temperature generally varies between 6 to 9°C throughout the year [10], but occasionally higher temperatures (exceeding 12°C) have been observed [34]. The particulate organic carbon (POC) concentration in the benthic boundary layer varies between 43.5 to $106.3 \mu\text{g POC L}^{-1}$ and depositional fluxes of POC ranges from 18 – $1485 \text{ mg POC m}^{-2} \text{ d}^{-1}$ with an annual average of $459 \text{ mg POC m}^{-2} \text{ d}^{-1}$ [33].

Both species were collected from a water depth of approximately 110 m using the remotely operated vehicle Sperre Subfighter 7500 DC. Specimens were placed in cooling boxes filled beforehand with cold seawater (7 – 8°C) and transported within a few hours to the laboratory at the Sven Lovén Centre in Tjärnö (Sweden). After arrival, coral samples were clipped to approximately the same size and living coral polyps from the reef branches with encrusting sponge were removed to prepare sponge specimens. All specimens were maintained in aquaria (~ 20 L) placed in a dark thermo-constant room (7°C) for up to 6 wk before used in the experiment as acclimation time. Sand-filtered (1 – 2 mm particle size) bottom water from 45 m depth out of the adjacent Koster fjord (salinity 31) was continuously flushed through the aquaria ($\sim 1 \text{ L min}^{-1}$). From experience at the station, it is known that the sand-filtered water still contains sufficient organic particles so that no extra food had to be provided during the acclimation period.

Preparation of labeled food substrates

The labelled food sources used in this experiment are identical to those described in Van Oevelen et al. [32]. Bacteria ($\sim 1 \mu\text{m}$) were cultured by adding a few mL of natural seawater from the Oosterschelde estuary (Netherlands) to M63 culture medium adjusted after Miller [35]. In the medium either 50% of glucose (3 g L^{-1}) or 50% of NH_4Cl (1.125 g L^{-1}) was replaced by its heavy isotope equivalent (Cambridge Isotopes, 99% ^{13}C , 99% ^{15}N) to obtain ^{13}C or ^{15}N isotopically-enriched bacteria. After 3 d of culturing in the dark, bacteria were concentrated by centrifugation ($14500 \times g$), after which the concentrate was rinsed 3 times with $0.2\text{-}\mu\text{m}$ filtered sea water to remove residual label and stored frozen (-20°C) until further use.

Single (^{13}C or ^{15}N) isotopically-enriched diatoms were cultured axenically in F/2 culture medium adjusted after Guillard [36]. In this medium, either 80% of the NaHCO_3 or 70% of the NaNO_3 was replaced by its heavy isotope equivalent (Cambridge Isotopes, 99% ^{13}C , 99% ^{15}N). The medium was injected with a sterile culture inoculum of the diatom *Thalassiosira pseudonana* (Hasle & Heimdal, 1970) ($\sim 5 \mu\text{m}$). After 3 wk of culturing with a 12 h light–dark cycle (at a cell density of around 3 – $4 \times 10^6 \text{ cells mL}^{-1}$), diatoms were concentrated by centrifugation at $450 \times g$. The concentrate was rinsed three times with $0.2\text{-}\mu\text{m}$ filtered seawater to remove residual label and kept at -20°C until further use.

Experimental set up and procedure

Prior to the start of the experiment, circular Plexiglas incubation chambers (10 L) were filled with $5\text{-}\mu\text{m}$ filtered Koster fjord bottom water and placed in a thermo-controlled room (7°C) at the Tjärnö Marine Laboratory. A coral specimen, i.e. a sponge-free *Lophelia pertusa*

fragment (10.35 ± 1.31 g DW fragment⁻¹, 16.83 ± 2.79 polyps fragment⁻¹, mean \pm sd, $n = 6$) and a sponge specimen, i.e. dead coral branches overgrown by the encrusting sponge *H. coriacea* (0.03 ± 0.01 g DW sponge⁻¹, mean \pm sd, $n = 12$), and were placed separately or together in the incubation chamber (10 L). Subsequent analysis (see below) showed that the coral carbon biomass ($10,128 \pm 3,459$ $\mu\text{mol C}$, mean \pm sd, $n = 12$) in the incubations was substantially higher than the sponge carbon biomass (451 ± 146 $\mu\text{mol C}$, mean \pm sd, $n = 12$). Coral fragments were kept in an upright position by inserting them into elastic silicone tubes that were mounted on an acrylic plate, which was fixed to the chamber base. A steady water circulation maintained by a motor-driven paddle in the upper part of the incubation chamber (speed: 2 rpm).

The corals and sponges in the incubation chambers were exposed to a 10-day feeding period (12 h feeding day⁻¹, 12 h flushing day⁻¹). A total food pulse of 108 $\mu\text{mol C L}^{-1}$ was given at the beginning of each daily feeding period consisting of ¹³C bacteria/ ¹⁵N algae or ¹³C algae/ ¹⁵N bacteria in a diatom:bacteria ratio of 1:3 (Table 1), resulting in a total food pulse of 2,708 $\mu\text{mol C}$ algae and 8,125 $\mu\text{mol C}$ bacteria per chamber. After each feeding period, chambers were flushed with 5- μm filtered Koster fjord bottom water (140 mL min⁻¹) for 12 h (flushing period) to avoid accumulation of waste products such as NH₄⁺ and CO₂, to remove remaining food particles and to renew the O₂ supply.

After the last flushing period, the incubation chambers were closed for 48 h to measure the production of ¹³C dissolved inorganic carbon (¹³C-DIC) as a proxy for respiration. Pilot experiments (C. E. Mueller unpubl. data), prior work [17, 30–32] and literature reports indicate that changes of pH (standard deviation of 0.04 around a mean pH of 7.8 NBS), dissolved oxygen and ammonium concentration [37] in these 48 h are not sufficient to negatively influence coral or sponge physiology. Samples for DIC analysis were taken before (control) and after the respiration incubation. Water was filtered (GF/F) in a 20-mL headspace vial, which was subsequently poisoned with 10 μL HgCl₂ and closed with an aluminium cap fitted with a rubber septum and stored upside down for further analysis. After the 48-hours closed incubation, the coral and sponge specimens were stored frozen (-20 °C) for later analysis of the incorporation of ¹³C and ¹⁵N in their tissues and ¹³C incorporation in the calcium carbonate skeleton of the coral.

Corals and sponges for background isotope measurements (controls) were incubated in parallel under ‘acclimatization’ conditions: i.e. without food addition to the sand-filtered seawater (1–2 mm particle size).

Sample treatment and analysis

Sponge tissue was scraped from the coral surface, homogenized by pestle and weighed. Coral samples were weighed and homogenized by grinding with a ball Mill for 20 s (MM 2000, Retsch, Haan, Germany). 30 mg of ground coral material, 2–3 mg ground sponge material or a subsample of the POM-filter was transferred to pre-combusted silver boats and decalcified by

Table 1. Experimental design.

Treatment	¹³ C-Alg + ¹⁵ N-Bac	¹⁵ N-Alg + ¹³ C-Bac
Coral	$n = 3$	$n = 3$
Sponge	$n = 3$	$n = 3$
Coral + Sponge	$n = 3$	$n = 3$

Experimental design and replicates (n) used in this study. The coral *L. pertusa* and the sponge *H. coriacea* were exposed separately and together to a food mixture consisting of ¹³C-labelled algae + ¹⁵N-labelled bacteria and to a food mixture composed of ¹⁵N-labelled algae + ¹³C-labelled bacteria.

<https://doi.org/10.1371/journal.pone.0194659.t001>

acidification [38] to analyze the organic coral tissue, sponge tissue and POM, respectively. Subsequently, each sample was simultaneously measured for ^{13}C and ^{15}N using a thermo Electron Flash EA 1112 analyzer (EA) coupled to a Delta V isotope ratio mass spectrometer (IRMS).

A headspace of 3 mL was created in each DIC sample vial by injecting N_2 gas through the vial septum. Samples were then acidified with 20 μL of concentrated H_3PO_4 to transform DIC into CO_2 . When CO_2 had exchanged with the vial headspace, 10 μL sample of the headspace gas was injected into an elemental analyzer isotope-ratio mass spectrometer (EA-IRMS). Calculations for ^{13}C respiration followed the description for ^{13}C tissue assimilation.

The incorporation of metabolic-derived ^{13}C in coral skeleton was measured following Mueller et al. [30] and Tanaka et al. [39]; 30 mg of each coral sample (i.e. the organic fraction and inorganic skeleton) was transferred to a silver boat and measured on the EA-IRMS for total ^{13}C content. The incorporation into the ^{13}C in the inorganic skeleton was obtained by subtracting ^{13}C assimilation in the organic fraction from the ^{13}C incorporation in the total (organic fraction and inorganic skeleton). Calcification based on metabolic-derived inorganic carbon may only be a small amount of the total calcification [~8%, 40], but is used here as proxy for changes in total calcification [30].

The uptake rates of tracer C or N from the enriched substrates have all been normalized to organic C or N biomass (e.g., $\mu\text{mol C}_{\text{tracer}} \text{mmol C}^{-1} \text{d}^{-1}$) to allow direct comparison of the uptake rates between species. These rates are calculated as from the delta notations obtained from the IRMS: $\delta X (\text{‰}) = (R_{\text{sample}}/R_{\text{ref}} - 1) \times 1000$, in which X is the element (C or N), R_{sample} is the heavy: light isotope ratio in the sample and R_{ref} is the heavy: light isotope ratio in the reference material ($R_{\text{ref}} = 0.0111797$ for C and $R_{\text{ref}} = 0.0036765$ for N). The atomic fraction of the heavy isotope (F) in a sample is calculated as $F = R_{\text{sample}}/(R_{\text{sample}} + 1)$. The excess (above background) atomic fraction is the difference between the F in an experimental sample and the atomic fraction in a control sample: $E = F_{\text{sample}} - F_{\text{control}}$. The excess incorporation of ^{13}C or ^{15}N was divided by the atomic enrichment of each specific food source to convert to total C_{tracer} or N_{tracer} incorporation, respectively, and divided by the incubation time to convert to daily rates. Calcification rates are calculated similarly using the $\delta^{13}\text{C}$ (‰) increase in the CaCO_3 pool.

Respiration rates are inferred from the excess (E) of ^{13}C in the DIC pool during the 48-h incubation as: $E = F_{\text{end}} - F_{\text{start}}$. The excess DIC values are multiplied with the DIC pool (i.e., $\mu\text{mol DIC L}^{-1}$) and chamber volume (10 L), and divided by the atomic fraction enrichment of the food source and the incubation time. Respiration are normalized to the total organic C in the incubation chamber (i.e. depending on the incubation; 'coral organic carbon tissue', 'sponge organic carbon tissue' or 'coral + sponge organic carbon tissue').

Data analysis

A net growth efficiency (NGE) for the coral and the sponge was calculated from the 'single species' experiments from the C incorporation rate into tissue and respiration rates as: $\text{NGE} = \text{Tissue incorporation} / (\text{Tissue incorporation} + \text{Respiration})$. Only 'single species' experimental data were for this calculation, because total respiration in mixed assemblages cannot be attributed to single species. As discussed below, this NGE calculation ignores other potential losses including cell shedding or dissolved organic carbon release.

Differences in the C and N incorporation rates between the coral and sponge were tested on \log_{10} -transformed (to meet the normal distribution assumption) data from the 'single species' experiments in 2-way ANOVAs, with C or N uptake as dependent variable and species (i.e. coral or sponge) and food source (i.e. bacteria or algae) as dependent factors. To test whether the incorporation of the coral or sponge was affected by the presence of the other

species, incorporation rates by the coral and sponge were tested on \log_{10} -transformed data in 3-way ANOVAs, with C or N uptake as dependent variable and treatment (i.e. coral or sponge 'only' versus coral and sponge together), food source (i.e. bacteria or algae) and element (i.e. C or N) as dependent factors. Significant differences in calcification rates were tested for in an ANOVA on \log_{10} -transformed data with treatment (i.e. coral only and coral with sponge) and food source as dependent factors. Significant differences in respiration rates and the net growth efficiency were tested for with \log_{10} -transformed data in an ANOVA with treatment (i.e. coral only and sponge only) and food source as dependent factors. Data are presented as mean \pm standard deviation. All raw data presented in this paper are made freely downloadable from doi:[10.5281/zenodo.1198189](https://doi.org/10.5281/zenodo.1198189).

Results

Carbon and nitrogen uptake and stoichiometry

Both the cold-water coral *Lophelia pertusa* and the sponge *Hymedesmia coriacea* incorporated algae and bacteria in all treatments (Fig 1). Mean tracer carbon (i.e. carbon from the isotopically-enriched food sources) incorporation rates by *L. pertusa* ranged from 0.08–0.17 $\mu\text{mol C}_{\text{tracer}} \text{mmol C}_{\text{coral}}^{-1} \text{d}^{-1}$ (Fig 1A) and tracer N incorporation rates ranged from 0.06–0.1 $\mu\text{mol N}_{\text{tracer}} \text{mmol N}_{\text{coral}}^{-1} \text{d}^{-1}$ (Fig 1D). For *H. coriacea*, tracer C incorporation ranged from 1.9–6.2 $\mu\text{mol C}_{\text{tracer}} \text{mmol C}_{\text{sponge}}^{-1} \text{d}^{-1}$ (Fig 1B) and tracer N incorporation rates ranged from 0.54–5.3 $\mu\text{mol N}_{\text{tracer}} \text{mmol N}_{\text{sponge}}^{-1} \text{d}^{-1}$ (Fig 1D). The tracer incorporation rates of the coral and the sponge were not significantly different between treatments (coral: $F_{1,19} = 2.87$, $P = 0.11$, sponge: $F_{1,19} = 1.53$, $P = 0.23$), food sources (coral: $F_{1,19} = 0.35$, $P = 0.56$, sponge: $F_{1,19} = 3.19$, $P = 0.09$) and elements (coral: $F_{1,19} = 2.55$, $P = 0.13$, sponge: $F_{1,19} = 0.57$, $P = 0.46$) nor was the interaction between treatment and food source (coral: $F_{1,19} = 2.83$, $P = 0.11$, sponge: $F_{1,19} = 0.52$, $P = 0.48$). However, the incorporation rates of the sponge were significantly higher than those of the coral in the 'single species' experiments (C: $F_{1,9} = 101$, $P < 0.001$, N: $F_{1,9} = 106$, $P < 0.001$). The C_{tracer} incorporation rates did not differ between food sources ($F_{1,9} = 3.14$, $P = 0.11$), while the N_{tracer} incorporation rates did differ significantly between food sources ($F_{1,9} = 12.7$, $P < 0.01$).

It is not straightforward to evaluate the stoichiometry of resource uptake, as the uptake of C and N from algae (and bacteria) was measured in different incubations and thereby on different coral and sponge specimens (Table 1). Therefore, we calculated the stoichiometric uptake based on average C- and N-incorporation rates of both food sources and consider that only broad-scale differences will become apparent using this approach. The stoichiometric uptake of the coral *L. pertusa* is broadly in agreement with the C:N ratio of its tissue and the food sources, although there seems to be a preferential uptake of bacterial C in the coral only treatment (Fig 2A and 2C). The stoichiometric incorporation of bacteria by the sponge was broadly in agreement with the sponge tissue and the resource (Fig 2B). However, the sponge seemed to preferentially incorporate C from the algae in the 'sponge only' and 'coral + sponge' treatments (Fig 2D).

Coral calcification

The ^{13}C tracer from algal and bacterial food sources was also traced back in the calcium carbonate skeleton of the coral (Fig 3). Calcification rates of the two food sources and treatments were in the same order of magnitude and ranged from 2.1–9.7 $\text{nmol C}_{\text{tracer}} \text{mmol C}_{\text{skeleton}}^{-1} \text{d}^{-1}$ (note the nmol scale for tracer). Calcification rates did not differ significantly between treatments ($F_{1,8} = 3.0$, $P = 0.12$) or food source ($F_{1,8} = 2.7$, $P = 0.14$).

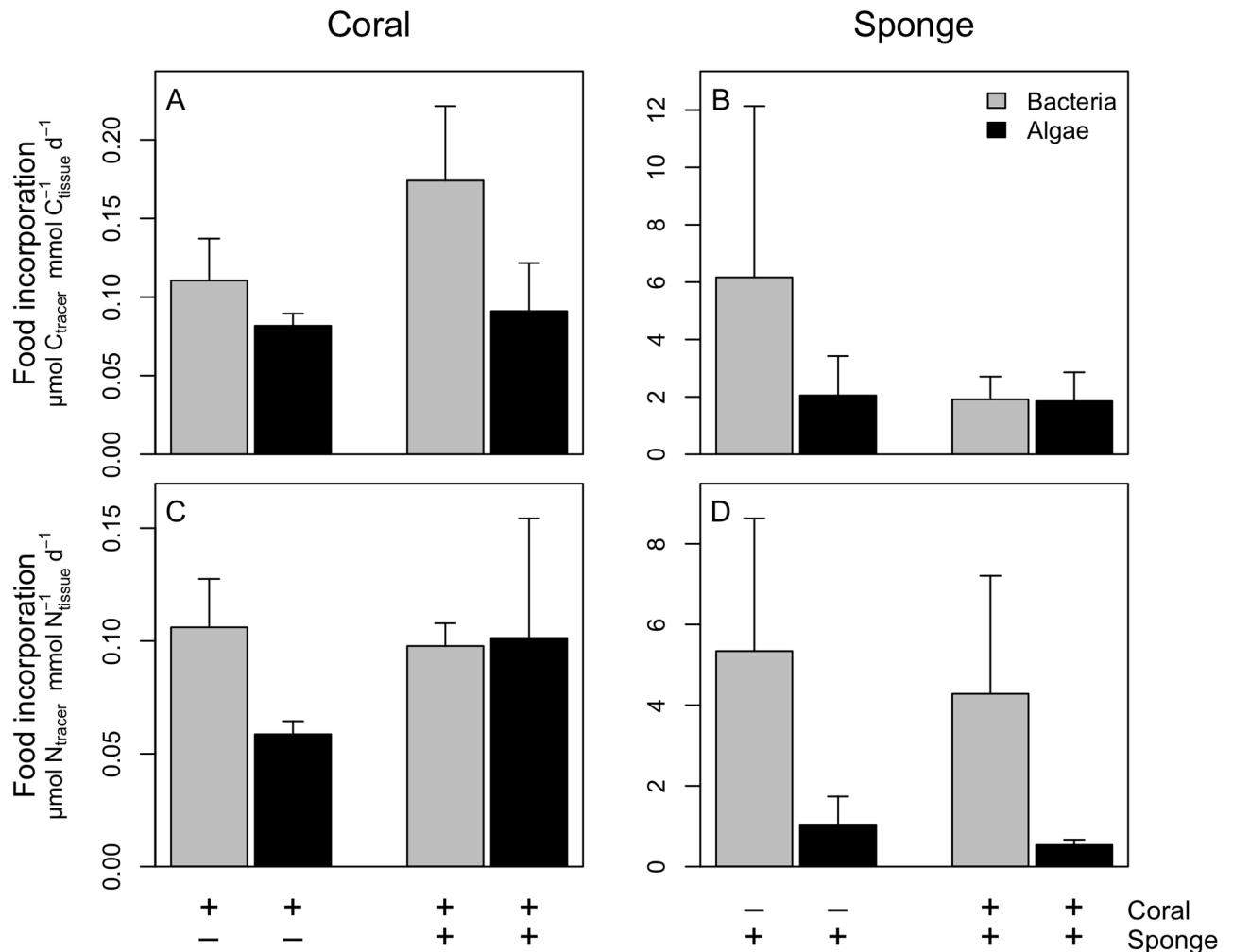


Fig 1. Food incorporation by the coral and sponge. Food incorporation rates, expressed in $\mu\text{mol } C_{\text{tracer}} \text{ mmol } C_{\text{tissue}}^{-1} \text{ d}^{-1}$ of algal and bacterial carbon (A, B) and nitrogen (C, D) in the tissue of the cold-water coral *Lophelia pertusa* (A, C) and the sponge *Hymedesmia coriacea* (B, D). The presence (+) or absence (-) of a species is indicated below the figures. Note scale differences between the subpanels.

<https://doi.org/10.1371/journal.pone.0194659.g001>

Respiration

The closed incubation after the 10-d feeding period showed respiration of the offered food sources by the coral and sponge in all treatments (Fig 4). Coral respiration rates of the isotope-enriched food ranged from 0.84–2.1 $\mu\text{mol } C_{\text{tracer}} \text{ mmol } C_{\text{coral}}^{-1} \text{ d}^{-1}$ whereas sponge respiration rates ranged from 54–66 $\mu\text{mol } C_{\text{tracer}} \text{ mmol } C_{\text{sponge}}^{-1} \text{ d}^{-1}$. The respiration rates in the treatment with corals and sponges cannot be attributed to a single taxon, but the biomass-specific respiration of the coral-sponge ‘consortium’ was between the coral and sponge rates and ranged from 20–40 $\mu\text{mol } C_{\text{tracer}} \text{ mmol } C_{\text{coral+sponge}}^{-1} \text{ d}^{-1}$. Respiration rates differed significantly between the coral and sponge ($F_{1,9} = 78.2, P < 0.001$), but did not differ significantly between food sources ($F_{1,9} = 2.3, P = 0.16$).

Carbon budget of the experiment

To estimate the fate of the added food sources during the experimental period, a carbon budget was calculated for the coral and sponge (based on the ‘single species only’ treatment data)

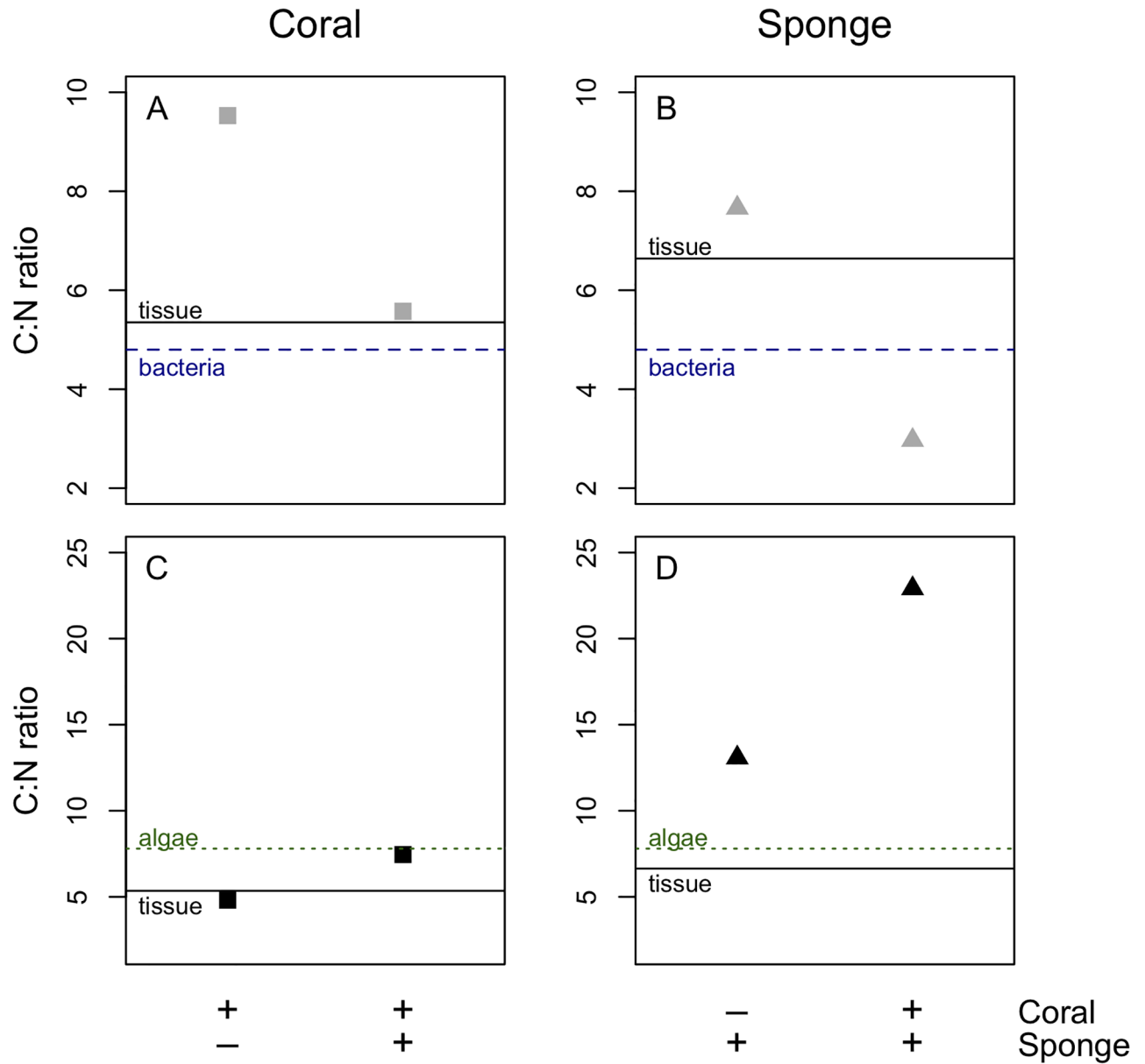


Fig 2. Stoichiometry of food incorporation. A) Mean C:N ratio of bulk coral tissue (solid line), bacterial food source (dashed line) and assimilation of bacteria into coral tissue (grey squares), B) mean C:N ratio of bulk sponge tissue (solid line), bacterial food source (dashed line) and assimilation of bacteria into sponge tissue (grey triangles), C) mean C:N ratio of bulk coral tissue (solid line), algal food source (dashed line) and assimilation of algae into the coral tissue (black squares) and D) mean C:N ratio of bulk sponge tissue (solid line), algal food source (dashed line) and assimilation of algae into sponge tissue (black triangles). The presence ('+') or absence ('-') of a species is indicated below the figures. Note scale differences between subpanels A and B versus C and D.

<https://doi.org/10.1371/journal.pone.0194659.g002>

results for incorporation, calcification and respiration under the assumption that these rates were constant over the 10-day feeding period. Note that the contribution of respiration in the budget was only determined at the end of the 10-day feeding period, while the incorporation rates were averaged over the whole period and that other loss processes are ignored (see Discussion below).

The total amount of carbon processed by the coral during the 10-d experiment (i.e. sum of incorporation, respiration and calcification) in the separate food treatments amounted to

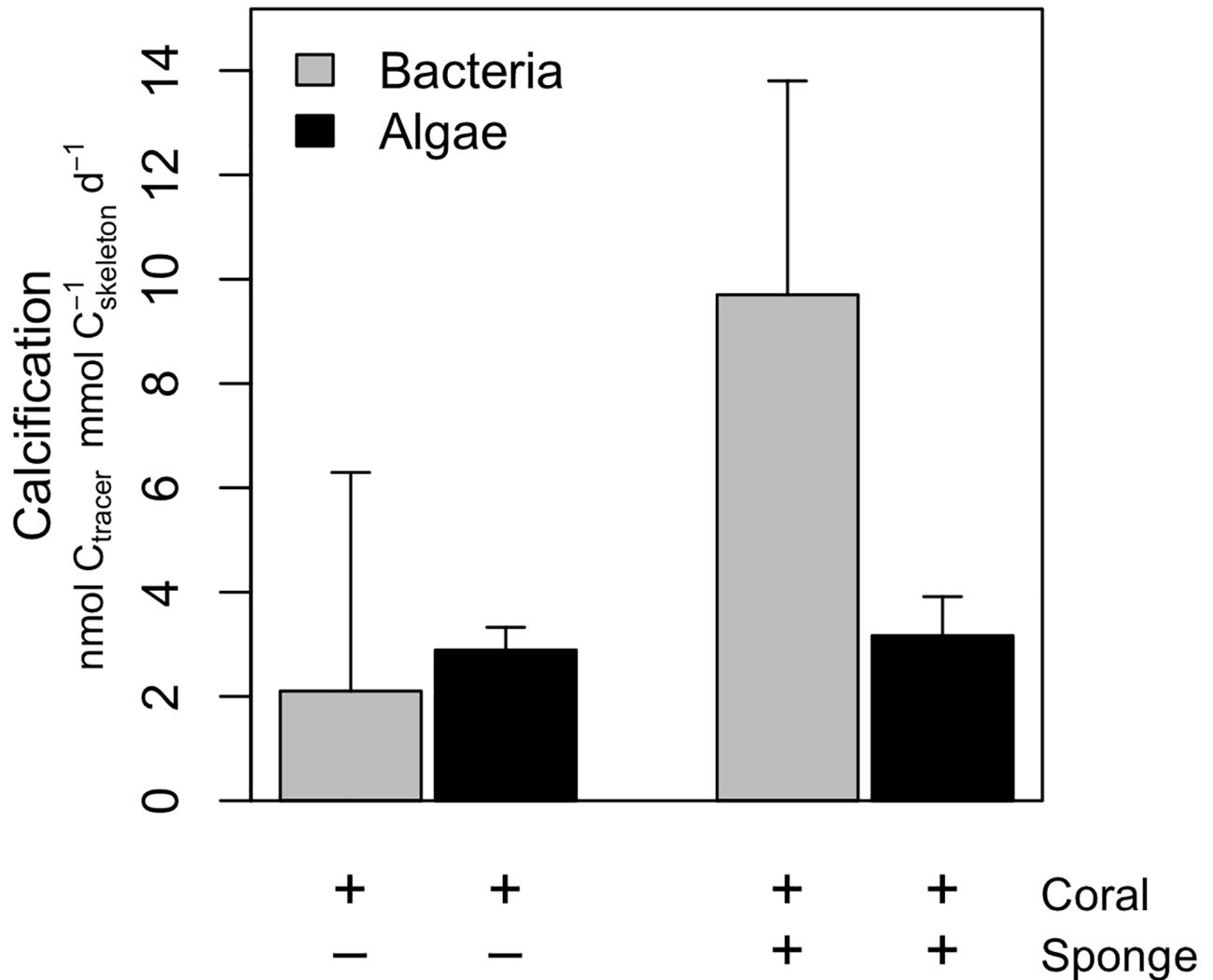


Fig 3. Coral calcification. Calcification rates expressed as $\text{nmol } C_{\text{tracer}} \text{ mmol } C_{\text{skeleton}}^{-1} \text{ d}^{-1}$ of the cold-water coral *Lophelia pertusa* when fed ¹³C-enriched bacteria or ¹³C-enriched algae in the presence (+) or absence (-) of sponges as indicated below the figure.

<https://doi.org/10.1371/journal.pone.0194659.g003>

273 μmol bacterial-C and 111 μmol algal-C, and for the sponge it was 345 μmol bacterial-C and 219 μmol algal-C (Table 2). For both taxa, the dominant fate was respiration. Of the total carbon added (2,708 μmol algal-C and 8,125 μmol bacterial-C), between 10 and 13% of the algal-C and between 0.014 and 0.027% of the added bacterial-C could be accounted for, while the remainder was not processed or could not be accounted for. The comparatively small differences in total processing rates between the two taxa in the experiment (Table 2) contrast with the clear differences in the processing rates presented in Figs 1 and 3, which is explained by the 29 ± 9 times (mean \pm standard deviation) higher coral biomass in the incubations as compared to the sponge biomass.

Net growth efficiency

The net growth efficiency ranged from 5.8 to 11.0% (Fig 5). The net growth efficiency did not differ significantly between species ($F_{1,9} = 0.34, P = 0.57$) and food source ($F_{1,9} = 0.010, P = 0.92$).

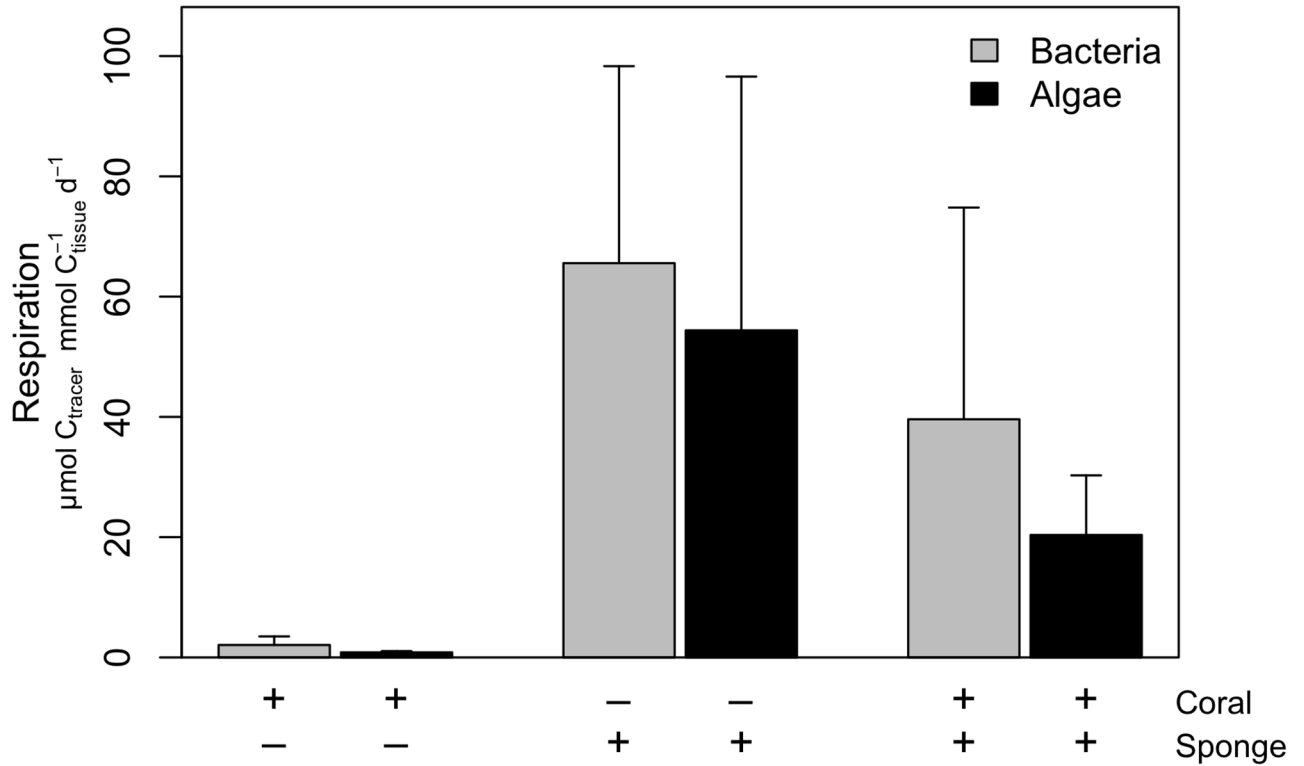


Fig 4. Respiration rates. Respiration rates expressed as $\mu\text{mol C}_{\text{tracer}} \text{mmol C}_{\text{tissue}}^{-1} \text{d}^{-1}$ of the incubations fed with ^{13}C -enriched bacteria and ^{13}C -enriched algae with only corals, with only sponges and with both taxa. The presence ('+') or absence ('-') of a species is indicated below the figure.

<https://doi.org/10.1371/journal.pone.0194659.g004>

Discussion

Niche overlap between a coral and its associated sponge

The coral and the sponge both assimilated and respired the suspended bacteria and algae, indicating niche overlap between these suspension feeders. The incorporation rates were, however, not significantly different for the two food sources for both taxa, although bacteria were added in a threefold higher quantity as compared to the algae. While it was surprising to see that the mean incorporation and calcification rates of the coral when fed with bacteria were higher in the 'coral + sponge' treatment as opposed to the 'coral only', this difference was not significant.

Table 2. Carbon budget during the 10-d experiment. Total carbon budget ($\mu\text{mol C}$, mean \pm sd) of the coral *Lophelia pertusa* and the sponge *Hymedesmia coriacea* during the 10-day experiment. The amount 'Unaccounted' was calculated as the added food minus the other terms.

		Coral	Sponge
Bacteria	Tissue incorporation	14.8 \pm 1.7	37.2 \pm 43.7
	Respiration	256.1 \pm 101.8	308.2 \pm 82.8
	Calcification	2.3 \pm 4.5	
	Unaccounted	7,852 \pm 105	7,777 \pm 75
Algae	Tissue incorporation	9.4 \pm 2.3	8.0 \pm 4.8
	Respiration	98.5 \pm 40.7	210.7 \pm 157.0
	Calcification	3.1 \pm 0.53	
	Unaccounted	2,597 \pm 43	2,489 \pm 161

<https://doi.org/10.1371/journal.pone.0194659.t002>

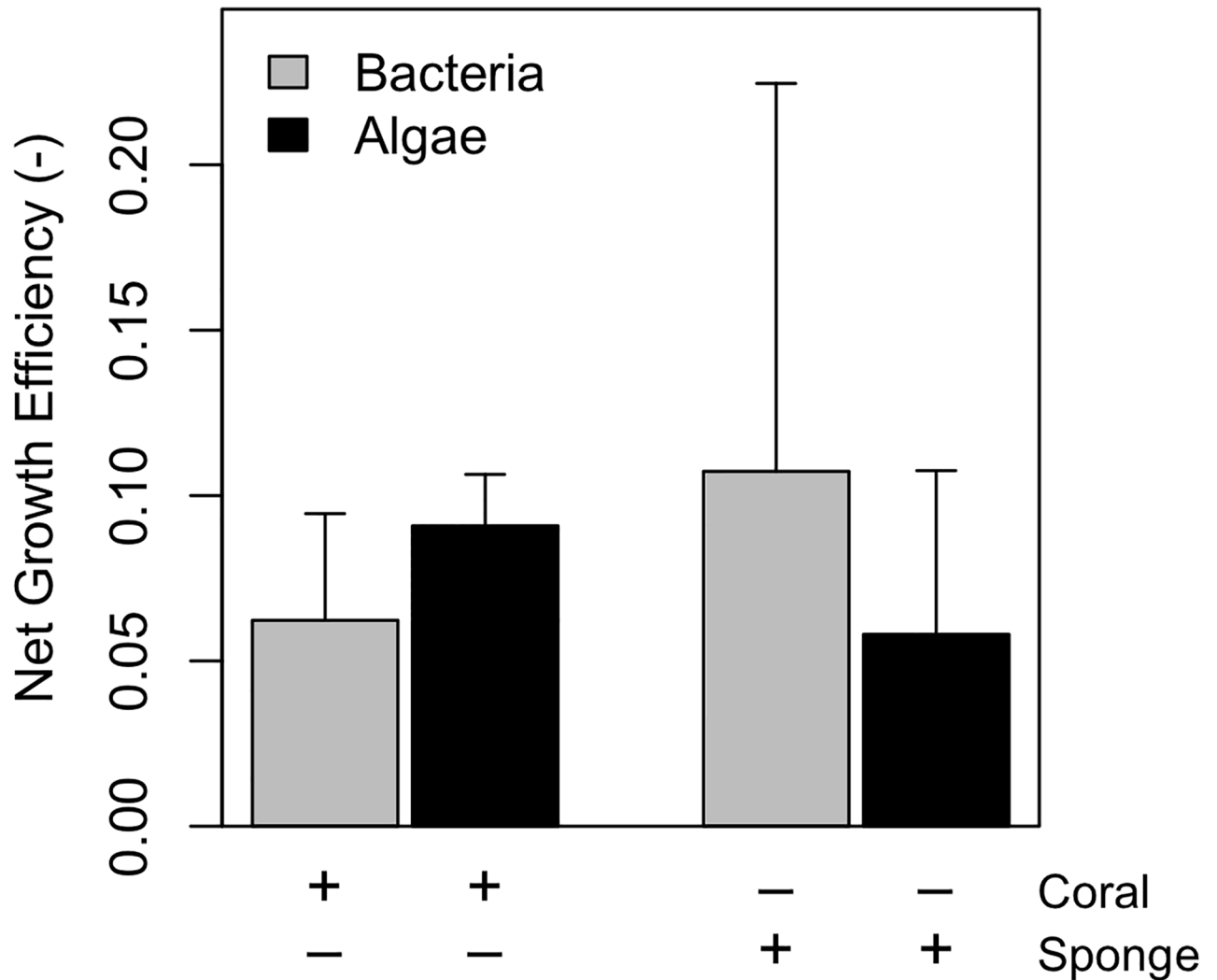


Fig 5. Net growth efficiency. The net growth efficiency (NGE) of the incubations fed with ¹³C-enriched bacteria and ¹³C-enriched algae with corals or sponges. The presence ('+') or absence ('-') of a species is indicated below the figure.

<https://doi.org/10.1371/journal.pone.0194659.g005>

The coral *L. pertusa* is a known opportunistic suspension feeder [29], capable of feeding of dissolved organic matter [17, 18] and variously sized suspended particulates [17, 41–43].

The sponge *Hymedesmia coriacea* did not show a clear preferential feeding on bacteria as reported for other sponges [24, 44, 45]. Similar uptake of pico- (equaling bacteria) and nano-plankton (equaling algae)-derived carbon under comparable conditions has been observed for the temperate sponge *Mycale lingua* (Pile et al. 1996), which is found on CWC reefs [46]. Smaller-sized phytoplankton may therefore be an important diet contribution of CWC reef sponges. The algal cells are retained in the aquiferous canal system of the sponge and while particles <4 μm are preferentially retained in the filter cell, i.e. choanocytes, of the sponge, larger particles are captured by pinacocytes, epithelial cells [47]. Hence, a modulation of selective cell types might explain the differences in assimilation rates among species and allows niche segregation between suspension feeders on the CWC coral reef. Besides uptake of suspended particles, uptake of dissolved organic matter and dissolved inorganic carbon (i.e. chemototrophy by symbiotic microbes) have been observed for CWC reef sponges [48, 49].

Niche overlap between taxa may also result in exploitative competition for suspended resources. We did not find experimental evidence for exploitative competition as the co-occurrence of a coral and its associated sponge did not significantly reduce incorporation rates. However, exploitative competition is only expected when resource concentrations are limiting. The provided food concentrations during the incubations ($108 \mu\text{mol C L}^{-1}$) were substantially higher as compared to natural conditions where organic carbon concentrations of $5.8 \mu\text{mol C L}^{-1}$ [Røst reef, 50], 3.6 to $8.9 \mu\text{mol C L}^{-1}$ [Tisler reef, 33] and 0.75 to $10.8 \mu\text{mol C L}^{-1}$ (several North Atlantic reef locations [several Atlantic reefs, 51] have been measured. Excess feeding was decided for to ensure uptake of measurable food quantities by the investigated species. As a result, only a small fraction of the resources that were fed during this study could be traced back in tissue, skeleton (only for coral) or in the dissolved inorganic carbon pool following respiration. Although this budget ignores release of $^{13}\text{C}/^{15}\text{N}$ tracer by corals in the form of secondary metabolites or dissolved organic matter [50], and the release of cellular debris and undigested food [49], it is evident that most labelled food was not consumed during the daily feeding period. Food availability at cold-water coral reefs is however strongly variable throughout the year [14, 19, 52] and we infer that, if exploitative competition between corals and sponge occurs in nature, it will be during periods of low food concentration. Incubations with sponges and corals under natural concentrations of organic matter may confirm whether niche overlap will induce exploitative competition.

Species-specific differences in food incorporation

Our experiments did show a highly significant ‘species effect’, with incorporation and respiration rates of the isotopically-enriched food sources being more than an order of magnitude higher for the sponge as compared to the coral. We here discuss two, non-mutually exclusive, explanations for this striking difference.

The first explanation is that the higher uptake and processing of the isotopically-enriched food by the sponge is due to a higher natural metabolic rate as compared to the coral. We cannot verify this from the experiment as we unfortunately have no metabolic rate data such as oxygen consumption. We are unaware of metabolic rate data for *H. coriacea* from the literature. However, a compilation of respiration rate data from organisms from a cold-water coral reef at Rockall Bank and literature data cold-water sponges, suggests that the metabolic rate of cold-water sponges are slightly higher (range of 0.001 – 0.006 d^{-1}) than that of the cold-water coral *L. pertusa* (i.e. 0.0015 d^{-1}) [53]. Additional metabolic rate data are obviously needed to verify this, but if true, sponges would have a disproportionately higher role in the metabolism of a cold-water coral ecosystem. Accurate biomass estimations of corals and sponges on natural cold-water coral reefs are needed to assess their role in the cycling of organic carbon within these ecosystems.

The second explanation for the different processing rates of the isotopically-enriched food sources may be related to the feeding mode of these sessile species. The coral *L. pertusa* is a passive suspension feeder that captures particles from the water column following physical contact with nematocyst-laden tentacles [29, 54, 55], while the demosponge *H. coriacea* is an active suspension feeder that uses flagellated cells to pump ambient water through its filtration system to capture suspended (and dissolved) resources [56, 57]. Active suspension feeders are hypothesized to feed more effectively on smaller particles, while passive feeders are better at retaining larger, more energy-rich particles [58]. In our experiment, however, roughly equally-sized food sources were used and no differences in incorporation rates for algae and bacteria were observed for both species. Another key difference between these two feeding modes is that the particulate food flux is an important factor affecting the food uptake of passive

suspension feeders as it increases encounter rates between particles and feeding appendages, whereas particulate food *concentration* is more relevant for active suspension feeders as this increases uptake rate at a given pumping activity [59]. As an example, Sebens [60] found that the colony size of the passive suspension feeder *Alcyonium siderium* (Linnaeus, 1758) was higher at more exposed sites as compared to a site with hydrodynamic calmer conditions, despite having similar seston concentration. Sponges may take advantage of current-induced flow to enhance feeding [61], a mechanism that would blur this generalization. Recent evidence for glass sponges however indicates that passive flow is energetically favorable only for porous, thin-walled sponges living in high flow environments [56]. We therefore argue that the difference in feeding mode may partly explain the observed difference in uptake of labelled resources, as the experimental setup (i.e. a circular incubation chamber) apparently favored the active suspension feeding modus of the sponge. Experiments in which flux and concentration are varied in hydrodynamic controlled conditions, e.g., flumes, are needed to verify this, but if found valid, this may set feeding constraints on suitable habitats for cold-water corals and sponges in nature.

The energetic costs involved in pumping activity have been considered to be negligible [$<4\%$ of the daily energy expenditure, 62], but recent estimates of pumping activity by glass sponges suggest that the head loss over the filter system was previously underestimated with a factor 10, warranting an upward revision of the pumping costs in the energy budget to almost 30% [56]. Costs for active sponge pumping did however not translate into lower net growth efficiency for the sponge as compared to the coral.

In conclusion, the cold-water coral and its associated sponge both utilized the suspended bacteria and algae as food source indicating niche overlap between these taxa. A clearly higher food assimilation and respiration was found for the sponge as compared to the cold-water coral. We hypothesize that the active suspension feeding mode of the sponge explains the observed differences in resource uptake as opposed to the passive suspension feeding mode of the cold-water coral. These feeding mode differences may set constraints on suitable habitats for cold-water corals and sponges in nature.

Acknowledgments

Lisbeth Jonsson and Ann Larsson are thanked for their assistance with sampling and coral maintenance. Pieter van Rijswijk and the analytical lab of NIOZ-Yerseke is thanked for their skilled assistance with sample processing. This output reflects only the author's view and the European Union cannot be held responsible for any use that may be made of the information contained therein.

Author Contributions

Conceptualization: Dick van Oevelen, Christina E. Mueller, Tomas Lundälv, Fleur C. van Duyl, Jack J. Middelburg.

Data curation: Christina E. Mueller, Fleur C. van Duyl.

Formal analysis: Dick van Oevelen, Jasper M. de Goeij.

Funding acquisition: Jack J. Middelburg.

Investigation: Christina E. Mueller, Tomas Lundälv, Fleur C. van Duyl, Jack J. Middelburg.

Methodology: Tomas Lundälv, Fleur C. van Duyl, Jasper M. de Goeij, Jack J. Middelburg.

Project administration: Jack J. Middelburg.

Supervision: Dick van Oevelen, Jack J. Middelburg.

Validation: Jasper M. de Goeij.

Visualization: Dick van Oevelen.

Writing – original draft: Dick van Oevelen, Christina E. Mueller.

Writing – review & editing: Dick van Oevelen, Christina E. Mueller, Tomas Lundälv, Fleur C. van Duyl, Jasper M. de Goeij, Jack J. Middelburg.

References

- Bell JJ (2008) The functional roles of marine sponges. *Estuarine Coastal and Shelf Science* 79: 341–53
- Cathalot C, van Oevelen D, Cox T, Kutti T, Lavaleye M, duineveld G, et al. (2015) Cold-water coral reefs and adjacent sponge grounds: Hotspots of benthic respiration and organic carbon cycling in the deep sea. *Frontiers in Marine Science* 2: 37
- Long MH, Berg P, de Beer D, Ziemann JC (2013) In situ coral reef oxygen metabolism: an Eddy Correlation study. *PLOS ONE* 8: e58581 <https://doi.org/10.1371/journal.pone.0058581> PMID: 23536798
- Dame R, Dankers N, Prins T, Jongsma H, Smaal A (1991) The influence of mussel beds on nutrients in the Western Wadden Sea and Eastern Scheldt Estuaries. *Estuaries* 14: 130–8
- Dayton PK (1971) Competition, disturbance, and community organization: the provision and subsequent utilization of space in a rocky intertidal community. *Ecological Monographs* 41: 351–89
- Jones CG, Lawton JH, Shachak M (1994) Organisms as ecosystem engineers. *Oikos* 69: 373–86
- Suess E (1980) Particulate organic-carbon flux in the oceans—surface productivity and oxygen utilization. *Nature* 288: 260–3
- Roberts JM, Wheeler AJ, Freiwald A (2006) Reefs of the deep: The biology and geology of cold-water coral ecosystems. *Science* 312: 543–7 <https://doi.org/10.1126/science.1119861> PMID: 16645087
- van Weering TCE, de Haas H, de Stigter HC, Lykke-Andersen H, Kouvaev I (2003) Structure and development of giant carbonate mounds at the SW and SE Rockall Trough margins, NE Atlantic Ocean. *Marine Geology* 198: 67–81
- Lavaleye M, Duineveld G, Lundälv T, White M, Guihen D, Kiriakoulakis K, et al. (2009) Cold-water corals on the Tisler reef: Preliminary observations on the dynamic reef environment. *Oceanography* 22: 76–84
- Fosså JH, Mortensen PB, Furevik DM (2002) The deep-water coral *Lophelia pertusa* in Norwegian waters: distribution and fishery impacts. *Hydrobiologia* 471: 1–12
- Mohn C, Rengstorf A, White M, Duineveld G, Mienis F, Soetaert K, et al. (2014) Linking benthic hydrodynamics and cold-water coral occurrences: A high-resolution model study at three cold-water coral provinces in the NE Atlantic. *Progress in Oceanography* 122: 92–104
- Cyr F, van Haren H, Mienis F, Duineveld G, Bourgault D (2016) On the influence of cold-water coral mound size on flow hydrodynamics, and vice versa. *Geophysical Research Letters* 43: 775–83
- Duineveld GCA, Jeffreys RM, Lavaleye MSS, Davies AJ, Bergman MJN, Watmough T, et al. (2012) Spatial and tidal variation in food supply to shallow cold-water coral reefs of the Mingulay Reef complex (Outer Hebrides, Scotland). *Marine Ecology Progress Series* 444: 97–115
- Findlay HS, Artioli Y, Navas JM, Hennige SJ, Wicks LC, Huvenne VAI, et al. (2013) Tidal downwelling and implications for the carbon biogeochemistry of cold-water corals in relation to future ocean acidification and warming. *Global Change Biology* 19: 2708–19 <https://doi.org/10.1111/gcb.12256> PMID: 23666812
- Soetaert K, Mohn C, Rengstorf A, Grehan A, van Oevelen D (2016) Ecosystem engineering creates a direct nutritional link between 600-m deep cold-water coral mounds and surface productivity. *Scientific Reports* 6: 35057 <https://doi.org/10.1038/srep35057> PMID: 27725742
- Mueller CE, Larsson AI, Veuger B, Middelburg JJ, van Oevelen D (2014) Opportunistic feeding on various organic food sources by the cold-water coral *Lophelia pertusa*. *Biogeosciences* 11: 123–33
- Gori A, Grover R, Orejas C, Sikorski S, Ferrier-Pages C (2014) Uptake of dissolved free amino acids by four cold-water coral species from the Mediterranean Sea. *Deep-Sea Research Part II-Topical Studies in Oceanography* 99: 42–50
- Duineveld GCA, Lavaleye MSS, Bergman MIN, De Stigter H, Mienis F (2007) Trophic structure of a cold-water coral mound community (Rockall Bank, NE Atlantic) in relation to the near-bottom particle supply and current regime. *Bulletin of Marine Science* 81: 449–67

20. Kiriakoulakis K, Fisher E, Wolff GA, Freiwald A, Grehan A, Roberts JM. Lipids and nitrogen isotopes of two deep-water corals from the North-East Atlantic: initial results and implication for their nutrition. In: Freiwald A, Roberts JM, editors. *Cold-Water Corals and Ecosystems*. Berlin Heidelberg: Springer-Verlag; 2005. p. 715–29.
21. Roberts JM, Wheeler A, Freiwald A, Cairns S. *Cold-water corals. The biology and geology of deep-sea coral habitats*. Cambridge: Cambridge University Press; 2009.
22. Wulff JL (2006) Ecological interactions of marine sponges. *Canadian Journal of Zoology* 84: 146–66
23. Van Soest RWM, Lavaleye MSS (2005) Diversity and abundance of sponges in bathyal coral reefs of Rockall Bank, NE Atlantic, from boxcore samples. *Marine Biology Research* 1: 338–49
24. Witte U, Brattegard T, Graf G, Springer B (1997) Particle capture and deposition by deep-sea sponges from the Norwegian-Greenland Sea. *Marine Ecology Progress Series* 154: 241–52
25. Yahel G, Whitney F, Reiswig HM, Eerkes-Medrano DI, Leys SP (2007) In situ feeding and metabolism of glass sponges (Hexactinellida, Porifera) studied in a deep temperate fjord with a remotely operated submersible. *Limnology and Oceanography* 52: 428–40
26. Leys SP, Kahn AS, Fang JKH, Kutti T, Bannister RJ (In press) Phagocytosis of microbial symbionts balances the carbon and nitrogen budget for the deep-water boreal sponge *Geodia barretti*. *Limnology and Oceanography*????
27. Pile AJ, Patterson MR, Witman JD (1996) In situ grazing on plankton. *Marine Ecology Progress Series* 141: 95–102
28. van Soest RWM, de Voogd NJ (2013) Sponge species composition of North-East Atlantic cold-water coral reefs compared in a bathyal to inshore gradient. *Journal of the Marine Biological Association of the United Kingdom* 95: 1461–74
29. Buhl-Mortensen P (2001) Aquarium observations on the deep-water coral *Lophelia pertusa* (L., 1758) (Scleractinia) and selected associated invertebrates. *Ophelia* 54: 83–104
30. Mueller CE, Lundalv T, Middelburg JJ, van Oevelen D (2013) The symbiosis between *Lophelia pertusa* and *Eunice norvegica* stimulates coral calcification and worm assimilation. *PLOS ONE* 8: e58660 <https://doi.org/10.1371/journal.pone.0058660> PMID: 23536808
31. Middelburg JJ, Mueller CE, Veuger B, Larsson AI, Form A, van Oevelen D (2015) Discovery of symbiotic nitrogen fixation and chemoautotrophy in cold-water corals. *Scientific Reports* 5: 9, <https://doi.org/10.1038/srep17962> PMID: 26644069
32. van Oevelen D, Mueller CE, Lundalv T, Middelburg JJ (2016) Food selectivity and processing by the cold-water coral *Lophelia pertusa*. *Biogeosciences* 13: 5789–98
33. Wagner H, Purser A, Thomsen L, Jesus CC, Lundalv T (2011) Particulate organic matter fluxes and hydrodynamics at the Tisler cold-water coral reef. *Journal of Marine Systems* 85: 19–29
34. Guihen D, White M, Lundalv T (2012) Temperature shocks and ecological implications at a cold-water coral reef. *Marine Biodiversity Records* 5: 1–10
35. Miller JH. *Experiments in molecular genetics*: Cold Spring Harbor Laboratory; 1972.
36. Guillard RRL. Culture of phytoplankton for feeding marine invertebrates. In: Smith WL, Chanley MH, editors. *Culture of Marine Invertebrate Animals*: Springer US; 1975. p. 29–60.
37. Dodds LA, Roberts JM, Taylor AC, Marubini F (2007) Metabolic tolerance of the cold-water coral *Lophelia pertusa* (Scleractinia) to temperature and dissolved oxygen change. *Journal of Experimental Marine Biology and Ecology* 349: 205–14
38. Nieuwenhuize J, Maas YEM, Middelburg JJ (1994) Rapid analysis of organic carbon and nitrogen in particulate materials. *Marine Chemistry* 45: 217–24
39. Tanaka Y, Miyajima T, Koike I, Hayashibara T, Ogawa H (2007) Imbalanced coral growth between organic tissue and carbonate skeleton caused by nutrient enrichment. *Limnology and Oceanography* 52: 1139–46
40. Adkins JF, Boyle EA, Curry WB, Lutringer A (2003) Stable isotopes in deep-sea corals and a new mechanism for “vital effects”. *Geochimica et Cosmochimica Acta* 67: 1129–43
41. Orejas C, Gori A, Rad-Menendez C, Last KS, Davies AJ, Beveridge CM, et al. (2016) The effect of flow speed and food size on the capture efficiency and feeding behaviour of the cold-water coral *Lophelia pertusa*. *Journal of Experimental Marine Biology and Ecology* 481: 34–40
42. Tsounis G, Orejas C, Reynaud S, Gili JM, Allemand D, Ferrier-Pages C (2010) Prey-capture rates in four Mediterranean cold-water corals. *Marine Ecology Progress Series* 398: 149–55
43. Maier C, de Kluijver A, Agis M, Brussaard CPD, van Duyl FC, Weinbauer MG (2011) Dynamics of nutrients, total organic carbon, prokaryotes and viruses in onboard incubations of cold-water corals. *Biogeosciences* 8: 2609–20

44. De Goeij JM, Moodley L, Houtekamer M, Carballeira NM, Van Duyl FC (2008) Tracing ^{13}C -enriched dissolved and particulate organic carbon in the bacteria-containing coral reef sponge *Halisarca caerulea*: Evidence for DOM feeding. *Limnology and Oceanography* 53: 1376–86
45. Ribes M, Coma R, Gili JM (1998) Seasonal variation of in situ feeding rates by the temperate ascidian *Halocynthia papillosa*. *Marine Ecology Progress Series* 175: 201–13
46. van Soest RWM, Cleary DFR, de Kluijver MJ, Lavaleye MSS, Maier C, van Duyl FC (2007) Sponge diversity and community composition in Irish bathyal coral reefs. *Contributions to Zoology* 76: 121–42
47. Turon X, Galera J, Uriz MJ (1997) Clearance rates and aquiferous systems in two sponges with contrasting life-history strategies. *Journal of Experimental Zoology* 278: 22–36
48. van Duyl FC, Hegeman J, Hoogstraten A, Maier C (2008) Dissolved carbon fixation by sponge-microbe consortia of deep water coral mounds in the northeastern Atlantic Ocean. *Marine Ecology Progress Series* 358: 137–50
49. Rix L, de Goeij JM, Mueller CE, Struck U, Middelburg JJ, van Duyl FC, et al. (2016) Coral mucus fuels the sponge loop in warm- and cold-water coral reef ecosystems. *Scientific Reports* 6: 18715 <https://doi.org/10.1038/srep18715> PMID: 26740019
50. Wild C, Mayr C, Wehrmann L, Schöttner S, Naumann M, Hoffmann F, et al. (2008) Organic matter release by cold-water corals and its implication for fauna-microbe interaction. *Marine Ecology Progress Series* 372: 67–75
51. Kiriakoulakis K, Freiwald A, Fisher E, Wolff GA (2007) Organic matter quality and supply to deep-water coral/mound systems of the NW European Continental Margin. *International Journal of Earth Sciences* 96: 159–70
52. Van Engeland T, Godø OR, Johnsen E, Duineveld GCA, van Oevelen D (Submitted) Food supply mechanisms to a cold-water coral reef on the Norwegian continental shelf revealed by continuous measurements from a cabled ocean observatory. PLEASE REMOVE
53. Van Oevelen D, Duineveld GCA, Lavaleye MSS, Mienis F, Soetaert K, Heip CHR (2009) The cold-water coral community as hotspot of carbon cycling on continental margins: a food web analysis from Rockall Bank (northeast Atlantic). *Limnology and Oceanography* 54: 1829–44
54. Frederiksen R, Jensen A, Westerberg H (1992) The distribution of the scleractinian coral *Lophelia pertusa* around the Faroe islands and the relation to internal tidal mixing. *Sarsia* 77: 157–71
55. Zetsche E-M, Baussant T, Meysman FJR, van Oevelen D (2016) Direct visualization of mucus production by the cold-water coral *Lophelia pertusa* with digital holographic microscopy. *PLOS ONE* 11: e0146766 <https://doi.org/10.1371/journal.pone.0146766> PMID: 26840074
56. Leys SP, Yahel G, Reidenbach MA, Tunnicliffe V, Shavit U, Reiswig HM (2011) The sponge pump: the role of current induced flow in the design of the sponge body plan. *PLOS ONE* 6:
57. Larsen PS, Riisgard HU (1994) The sponge pump. *Journal of Theoretical Biology* 168: 53–63
58. Gili J-M, Coma R (1998) Benthic suspension feeders: their paramount role in littoral marine food webs. *Trends in Ecology & Evolution* 13: 316–21
59. Lesser MP, Witman JD, Sebens KP (1994) Effects of flow and seston availability on scope for growth of benthic suspension-feeding invertebrates from the Gulf of Maine. *Biological Bulletin* 187: 319–35 <https://doi.org/10.2307/1542289> PMID: 29281393
60. Sebens KP (1984) Water flow and colony size—interhabitat comparisons of the octocoral *Alcyonium siderium*. *Proceedings of the National Academy of Sciences of the United States of America* 81: 5473–7 PMID: 16593509
61. Vogel S (1977) Current-induced flow through living sponges in nature. *Proceedings of the National Academy of Sciences of the United States of America* 74: 2069–71 PMID: 266728
62. Riisgård HU, Larsen PS (1995) Filter-feeding in marine macroinvertebrates—pump characteristics, modeling and energy-cost. *Biological Reviews of the Cambridge Philosophical Society* 70: 67–106 <https://doi.org/10.1111/j.1469-185X.1995.tb01440.x> PMID: 21545387