Supporting Information

Single-cell visualization indicates direct role of sponge host in uptake of dissolved organic matter

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Figure S1

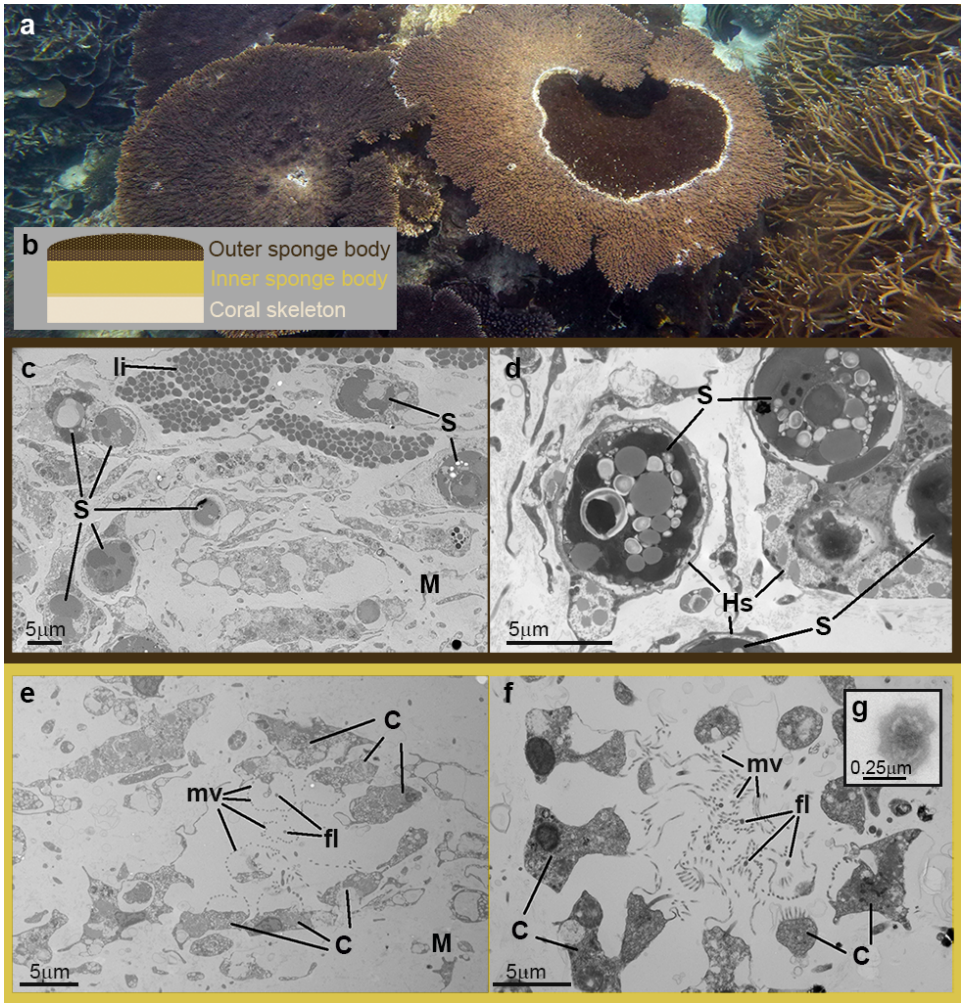

Figure S1: Macro and microscopic observations of the bioeroding sponge *Cliona orientalis* from the Great Barrier Reef, Australia. **a** *C. orientalis* (brown patch) in competition with a plating Acroporid coral. **b** Vertical cross section of the experimental sponge cylinders (35 mm diameter, colour scheme applies to figures of the main article). The outer sponge body hosts most of the endosymbiotic dinoflagellates of the genus *Gerakladium*, but the inner sponge body where light does not penetrate is relatively void of *Gerakladium*. The underlying coral skeleton has not yet been eroded by the sponge. **c** Transmission Electron Micrograph (TEM) of the outer sponge body, showing the dinoflagellate symbiont cells (S) and dense lipid accumulations (li) in the sponge mesohyl (M). **d** Further magnification shows the intracellular positioning of dinoflagellate symbionts (S) inside archaeocyte-like host cells (Hs). **e** TEM image of the inner sponge body showing the heterotrophic feeding cells or choanocytes (C) that line the aquiferous canals. Choanocytes bear a flagellum (fl, seen in cross section), which is surrounded by a collar of cellular extensions called microvilli (mv). Beating of the flagellae creates a water flow across the microvilli enabling trapping of food particles which are later phagocytosed by the cell, or passed on to other cells in the surrounding mesohyl (M). **f** Further magnification showing a cross section of choanocytes forming a so-called choanocyte chamber. **g** TEM image of an unidentified bacterium (potentially *Rhodothalassium* sp., an Alphaproteobacterium; Ramsby et al, 2018, *Mol Ecol* 27:2124–2137) found occasionally in the inner but also the outer sponge body.

Figure S2

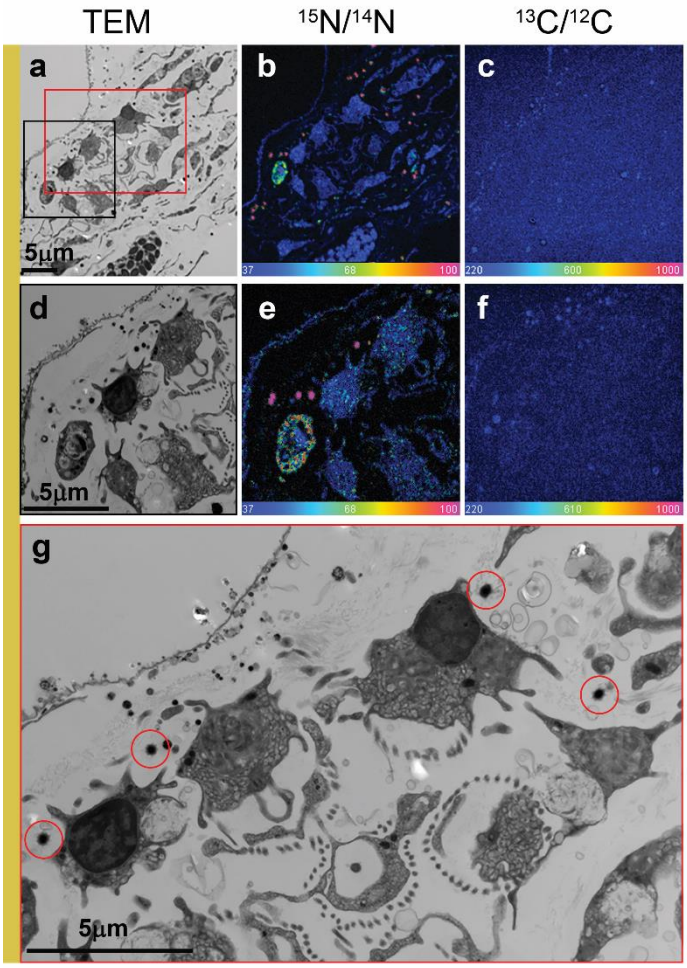

Figure S2: Visualization of ^{15}N and ^{13}C isotopic distribution in the inner body of the *Cliona orientalis* holobiont at the end of the chase period (t=24h). **a, d, g** TEM displaying sponge choanocytes and other host material, as well as unidentified prokaryotic microorganisms (circled in red in **g,** also shown in higher resolution in Fig. S1g). Selected areas displayed in **a** are enlarged in **d** (black rectangle) and in **g** (red rectangle). **b, e** Corresponding ^{15}N-distribution showing enriched prokaryotic microorganisms ($\Delta\delta^{15}N$ = 1684 ± 107‰, mean ± SEM of the 27 microorganisms detected in **b**), and other sponge material. **c,f** Corresponding ^{13}C-distribution of the same microorganisms ($\Delta\delta^{13}C$ = 120 ± 22‰). Note that in this case the carbon enrichment is substantially less profound than the nitrogen enrichment. Images **b**, **c**, **e**, and **f** show the deviation of the ratios from the natural abundance ratio, using a rainbow scale in hue saturation intensity. The rainbow scale ranges from natural abundance in blue (0.0037 for $^{15}N/^{14}N$ and 2x 0.0110 for $^{13}C/^{12}C$) to several-fold enrichment above natural abundance in red (approx. 3-fold for $^{15}N/^{14}N$ and 5-fold for $^{13}C/^{12}C$). The ochre yellow bar on the far left of the panel refers to the colour scheme in Fig. S1b.

Table S1

Results of two-factorial permutational analysis of variance testing the Euclidean dissimilarities between the $^{15}N/^{14}N$ and $^{13}C/^{12}C$ ratios of each area of interest [AOI, 1st factor with 5 levels: (I) Dinoflagellate symbionts=S, (II) Cytoplasm of dinoflagellate-hosting sponge cells=Hs, (III) nearby outer-body mesohyl cells or intercellular areas=Mo, (IV) Choanocytes=C and (V) nearby inner-body mesohyl cells and intercellular space=Mi] between the three time-points (Time, 2nd factor with 3 levels: 0h, 3h and 24h). Bold face indicates statistical significance ($P < 0.05$). Dispersions differed amongst the compared groups, with the greatest dispersion found in the largest group, rendering the permutational tests relatively conservative (Anderson and Walsh, 2013, *Ecol Monograph* 83: 557-574). df=degrees of freedom; SS=sum of squares; MS=mean sum of squares; Pseudo-F=F value by permutation, $P_{(perm)}$=P-value by permutation.

		Main Permanova Test						Pairwise Comparisons		
Parameter	Source	df	SS	MS	Pseudo-F	$P_{(perm)}$	Level	Groups	Pseudo-T	$P_{(perm)}$
$^{15}N/^{14}N$	Time	2	**3.14E-03**	**1.57E-03**	**3.4404**	**0.0380**	S	**0h versus 3h**	**4.6753**	**0.0002**
	AOI	4	**7.85E-03**	**1.96E-03**	**4.3003**	**0.0108**		**0h versus 24h**	**4.1256**	**0.0006**
	Time x AOI	8	**1.54E-02**	**1.92E-03**	**4.2036**	**0.0036**		**3h versus 24h**	**2.4791**	**0.0146**
	Res	722	0.32969	4.57E-04			Hs	**0h versus 3h**	**2.2993**	**0.0004**
								0h versus 24h	**3.9715**	**0.0010**
								3h versus 24h	**2.8916**	**0.0042**
							Mo	**0h versus 3h**	**9.1998**	**0.0002**
								0h versus 24h	**3.6583**	**0.0002**
								3h versus 24h	**2.6775**	**0.0054**
							C	**0h versus 3h**	**4.0446**	**0.0002**
								0h versus 24h	**8.0880**	**0.0002**
								3h versus 24h	**2.8360**	**0.0042**
							Mi	**0h versus 3h**	**8.2188**	**0.0002**
								0h versus 24h	**5.1020**	**0.0002**
								3h versus 24h	0.5716	0.5686
$^{13}C/^{12}C$	Time	2	1.11E-04	5.55E-05	2.6311	0.0738	S	**0h versus 3h**	**2.4770**	**0.0154**
	AOI	4	**5.13E-04**	**1.28E-04**	**6.0761**	**0.0154**		**0h versus 24h**	**2.4800**	**0.0176**
	Time x AOI	8	**6.20E-04**	**7.75E-05**	**3.6694**	**0.0246**		3h versus 24h	0.8709	0.3790
	Res	722	1.52E-02	2.11E-05			Hs	**0h versus 3h**	**2.0907**	**0.0420**
								0h versus 24h	**2.7301**	**0.0096**
								3h versus 24h	0.6242	0.5370

Mo	**0h versus 3h**	**3.4711**	**0.0012**
	0h versus 24h	**2.0989**	**0.0384**
	3h versus 24h	**2.0808**	**0.0396**
C	**0h versus 3h**	**4.4125**	**0.0002**
	0h versus 24h	**8.6999**	**0.0002**
	3h versus 24h	**2.7601**	**0.0044**
Mi	**0h versus 3h**	**2.2346**	**0.0276**
	0h versus 24h	**4.3404**	**0.0002**
	3h versus 24h	1.6421	0.1088

Table S2

Results of permutational analysis of variance testing the Euclidean dissimilarities between the levels of ^{15}N- and ^{13}C-enrichment (Δδ values above background) of each area of interest (AOI abbreviations as explained in the legend of Table S1) between treatment time-points (3h and 24h). Bold face indicates statistical significance ($P < 0.05$). Dispersions differed amongst the compared groups, with the greatest dispersion found in the largest group, rendering the permutational tests relatively conservative (Anderson and Walsh, 2013, *Ecol Monograph* 83: 557-574). df=degrees of freedom; SS=sum of squares; MS=mean sum of squares; Pseudo-F=F value by permutation, $P_{(perm)}$=P-value by permutation.

		Main Permanova Test					Pairwise Comparisons (3h versus 24h)		
Parameter	Source	df	SS	MS	Pseudo-F	$P_{(perm)}$	AOI Level	Pseudo-T	$P_{(perm)}$
^{15}N-enrichment	Time	1	9.97E+06	9.97E+06	0.2169	0.6412	S	**2.4791**	**0.0122**
	AOI	4	**1.07E+09**	**2.68E+08**	**5.8416**	**0.0006**	Hs	**2.8916**	**0.0042**
	Time x AOI	4	**7.74E+08**	**1.93E+08**	**4.2106**	**0.0048**	Mo	**2.6775**	**0.0044**
	Res	534	2.45E+10	4.60E+07			C	**2.8360**	**0.0056**
							Mi	0.57164	0.5738
^{13}C-enrichment	Time	1	1.23E+05	1.23E+05	1.9340	0.1630	S	0.8709	0.3842
	AOI	4	**1.63E+06**	**4.07E+05**	**6.3833**	**0.0004**	Hs	0.6242	0.5298
	Time x AOI	4	**7.25E+05**	**1.81E+05**	**2.8463**	**0.0322**	Mo	**2.0808**	**0.0380**
	Res	534	3.40E+07	63698			C	**2.7601**	**0.0026**
							Mi	1.6421	0.0980