

# **Overcalcified forms of the coccolithophore *Emiliana huxleyi* in high CO<sub>2</sub> waters are not pre-adapted to ocean acidification.**

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## **SUPPLEMENTARY MATERIALS**

## **Supplementary Section S1**

### *Variation in relative abundance of E. huxleyi morphotypes with depth.*

We note that the dominant morphotype of *Emiliana huxleyi* was usually the same at the surface and deeper in the water column (Fig. S1-S2). One exception was a station near Punta Lengua de Vaca (Tongoy Station 18) where lightly calcified morphotypes dominated below the thermocline and R/overcalcified morphotypes dominated above (Fig. S1f). Another exception was the station 2 in the JF survey, where the lightly calcified morphotypes were dominant within and below the picnocline but the A morphotype was dominant, although at the lower total abundance (Fig. S1h). Table S3 (Supplementary section S3) gives abundances with depth at the stations shown in Fig. S1-S2.

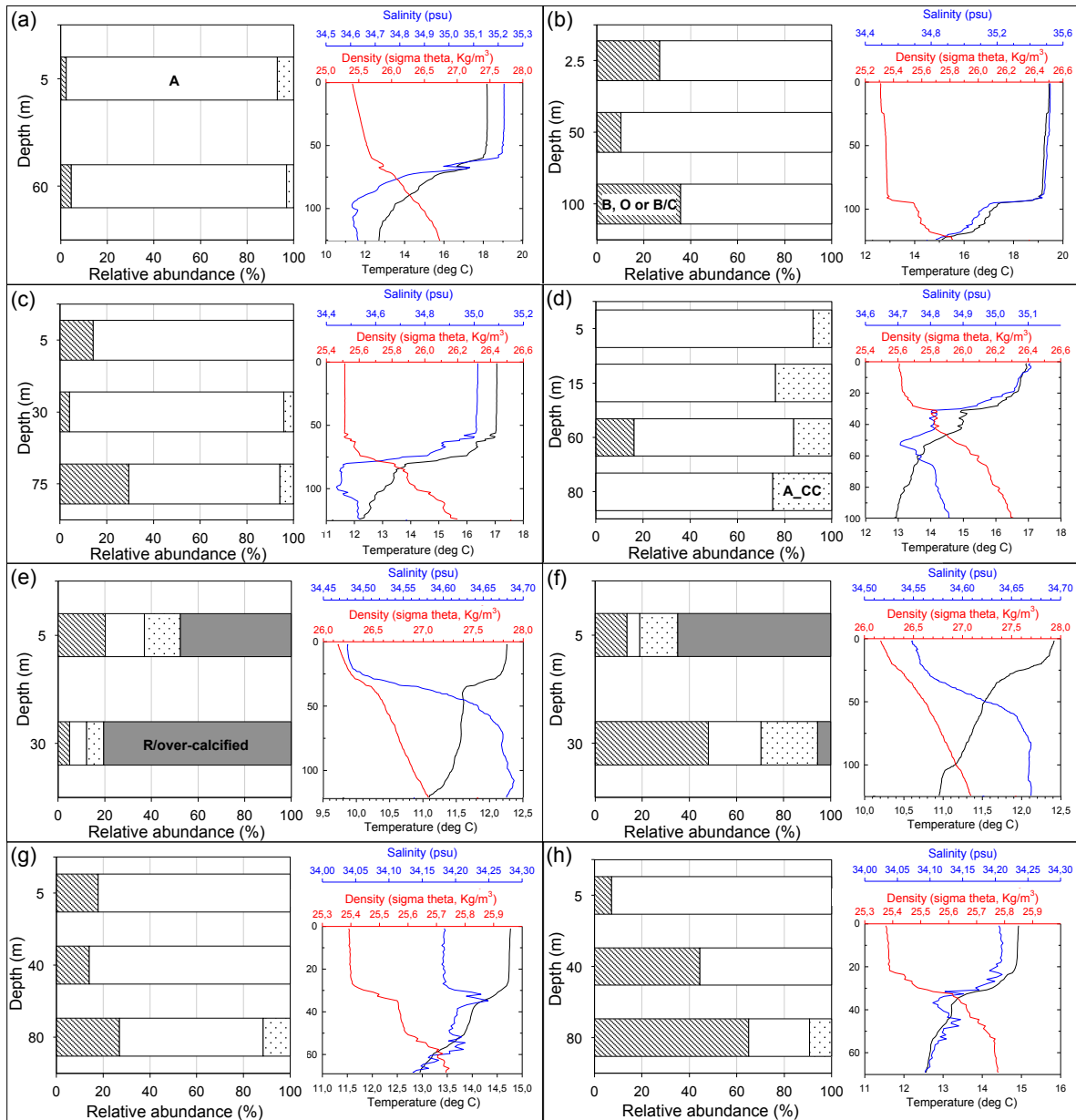


Figure S1. Relative abundances of *Emiliana huxleyi* morphotypes in the upper water column by study site. In a-d), e-f) and g-h) the relative abundances yielded by *E. huxleyi* morphotypes in NBP cruise (st. H04, H13, H19, BB2f), Tongoy Bay (st. 01 and 18) and Juan Fernandez surveys (st. 01, 02) are shown, respectively. Temperature (black), salinity (blue) and density (red) profiles for each station are shown at the right. Morphotypes are indicated on the bars.

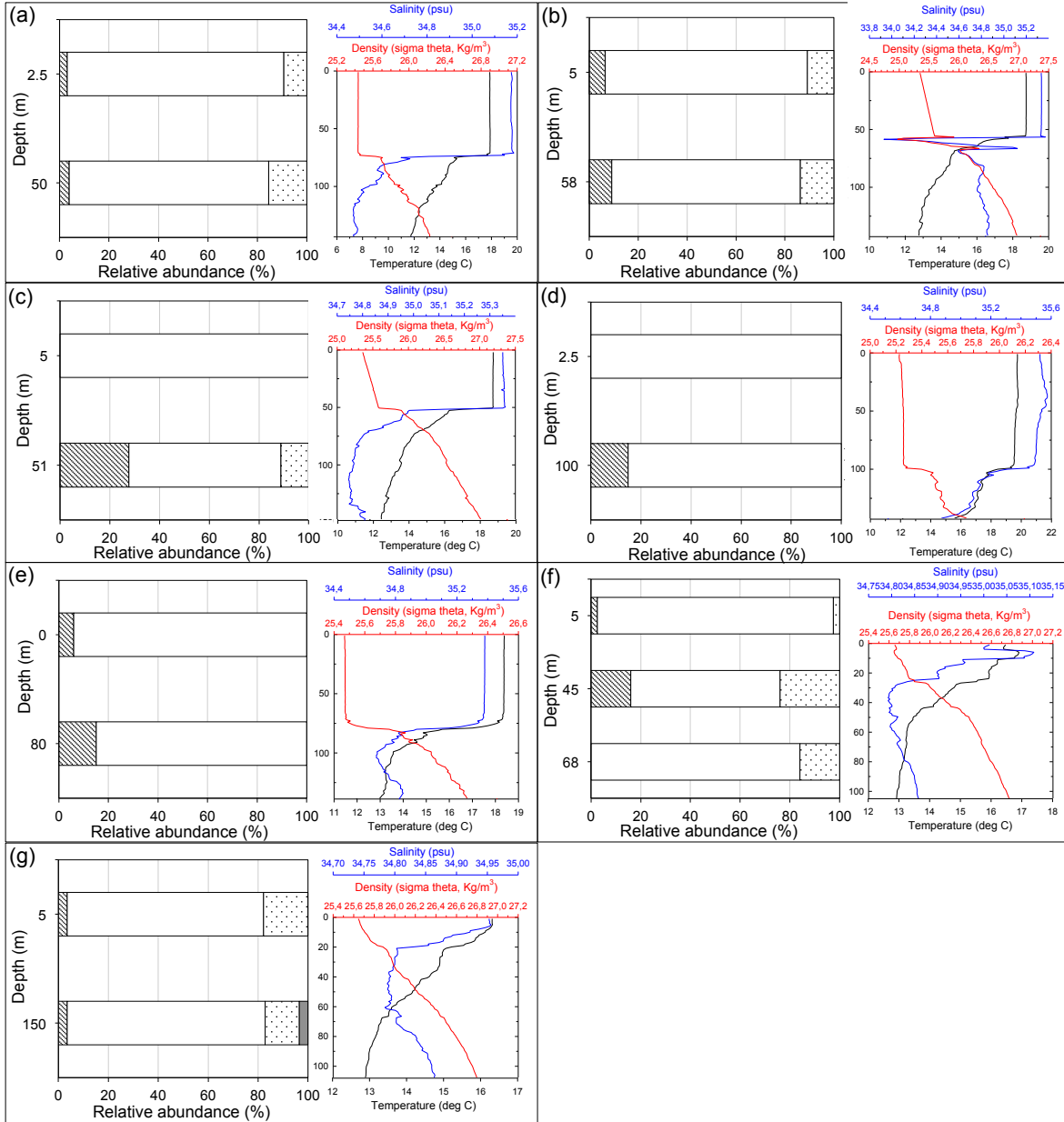


Figure S2. Relative abundances of *Emiliana huxleyi* morphotypes in the upper water column. In a-g) the relative abundances yielded by *E. huxleyi* morphotypes in NBP cruise (st. H01, BB1a, BB1b, H10, H17, BB2b, BB2c) are shown in panels a-g. Temperature (black), salinity (blue) and density (red) profiles for each station are shown at the right. Morphotypes are as in Fig. S1. A conductivity sensor error in BB1a caused a spike that was not filtered out successfully.

## Supplementary Section S2

### *Redundancy analysis (RDA) methodology used and RDA results for *Emiliana huxleyi* morphotype distributions constrained by environmental variables.*

To determine the abiotic variables driving the *Emiliana huxleyi* populations a redundancy analysis (RDA) was performed (rda function in vegan package Oksanen et al., 2007, performed in RStudio version 1.0.143 for mac OS). RDA is a direct constrained method that combine multivariate multiple linear regression with principal component analysis (Borcard et al., 2011). To RDA analyses we followed the methodology provide by Borcard et al. (2011). The variation in *E. huxleyi* morphotypes (matrix composed by relative abundances) were regressed on environmental conditions (temperature, salinity and  $p\text{CO}_2$ ), while controlling for sampling location (vector of offshore distances in km). To test for significance of RDA model and axis the pseudo-F statistic was calculated by set a minimal number of 1,000 sample permutations (Borcard et al., 2011). As linear dependencies between environmental variables can inflated the regression coefficient (Borcard et al., 2011), variance inflation factors were checked after each RDA analysis (vif.cca function in vegan package). RDA results are plotted in Fig. S3.

#### References.

Borcard, D., Gillet, F. and Legendre, P. 2011. Numerical ecology with R. Springer Science+Business Media. pp. 306.

Oksanen, J., Blanchet, F., Kindt, R., Legendre, P., Minchin, P., O'Hara, R., Simpson, G., Solymos, P., Stevens, M. and Wagner, H. 2007. Vegan: Community Ecology Package. R package version 2.3-1. Available at: <https://cran.r-project.org/web/packages/vegan/index.html> (last accessed 16 July 2017).

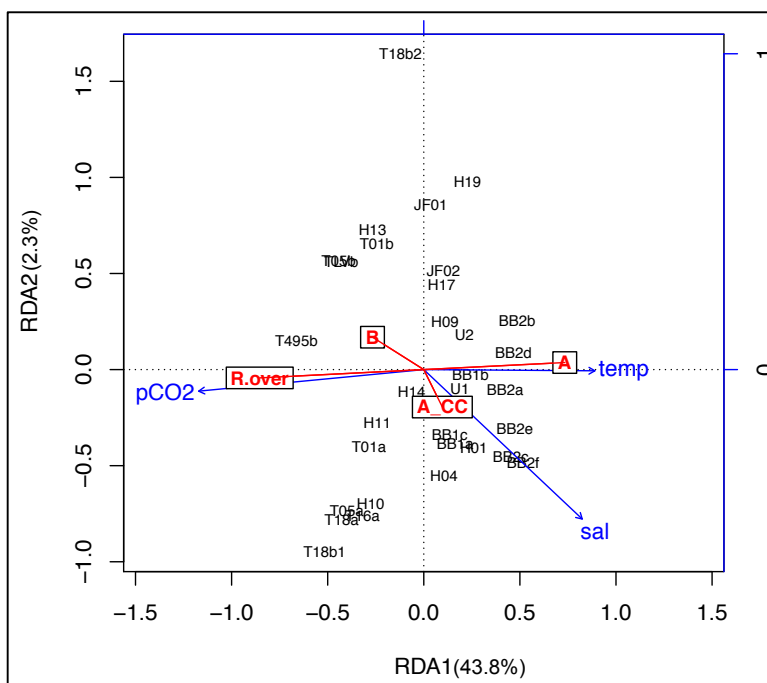


Figure S3. Redundancy analysis results for *Emiliana huxleyi* morphotype distributions constrained by environmental variables. The relative abundances of *E. huxleyi* morphotypes (red labels) from surface stations (black labels) were constrained by three environmental variables (blue arrows). Percentage of variance explained by each RDA axis are displayed. Only the first RDA axis appeared to be significant ( $p < 0.05$ ). RDA triplot was performed with site scores and scaling 2.

## Supplementary Section S3

### *Tables of environmental parameters and coccolithophore species.*

Table S1. Environmental data for the Eastern South Pacific corresponding to spring 2011 (JF and TONa surveys), 2012 (TONb,c and QUI surveys) and winter 2013 (NBP cruise) periods. na – no available data. No obs – Not observed.

Station ID	Latitude (South)	Longitude (West)	Z (m)	Offshore distance (km)	Temp. (°C)	Sal. (PSU)	MLD (m)	O <sub>2</sub> (μmol Kg <sup>-1</sup> )	Fluoro. (mg m <sup>-3</sup> )	pCO <sub>2</sub> (uatm)	pH	HCO <sub>3</sub> <sup>-</sup>	CO <sub>3</sub> <sup>2-</sup> (μmol kg SW <sup>-1</sup> )	Revelle factor	Ωcal	SiO <sub>4</sub> (μM)	PO <sub>4</sub> <sup>3-</sup> (μM)	NO <sub>3</sub> <sup>-</sup> (μM)
JF01	-33.596	-78.630	5	650	14.76	34.18	34	201.0	na	353.9	8.09	1847.8	164.1	11.124	3.93	na	na	na
JF02	-33.660	-78.602	5	645	14.91	34.21	31	195.1	na	333.5	8.11	1840.0	173.2	10.808	4.15	na	na	na
TON01a	-30.117	-71.619	5	23	12.25	34.48	no obs.	185.6	na	718.0	7.81	2035.6	87.9	15.681	2.10	12.70	na	17.12
TON05a	-30.181	-71.573	5	11	12.72	34.49	no obs.	174.0	na	658.9	7.85	2032.0	97.2	15.032	2.32	na	na	na
TON16a	-30.248	-71.650	5	1	12.35	34.54	no obs.	164.9	na	614.0	7.88	2031.5	103.3	14.665	2.47	18.24	na	17.06
TON18a	-30.247	-71.694	5	6	12.40	34.55	no obs.	161.2	na	620.8	7.87	2033.4	101.9	14.742	2.43	17.90	na	16.32
TON01b	-30.117	-71.619	5	23	14.10	33.83	na	na	na	635.2	7.86	1990.1	101.7	14.604	2.44	1.55	1.63	8.83
TON05b	-30.181	-71.573	5	11	13.90	34.18	na	na	na	637.7	7.86	2009.6	103.4	14.569	2.47	2.11	1.51	9.06
TON18b	-30.247	-71.694	5	6	14.10	34.44	na	na	na	559.7	7.92	1991.8	117.6	13.663	2.81	2.02	1.38	5.70
TON495c	-30.495	-71.774	5	8	13.54	34.50	na	na	na	809.4	7.77	2073.9	85.6	15.995	2.05	na	na	na
TONLVc	-30.238	-71.652	5	2	14.40	34.00	na	na	na	723.6	7.82	2037.8	95.2	15.231	2.28	na	na	na
TON18c	-30.247	-71.694	5	6	14.46	34.60	na	na	na	618.7	7.88	2010.7	110.1	14.138	2.63	na	na	na
QUI01	-33.379	-71.734	15	4	14.12	34.41	na	na	na	934.0	7.71	2089.2	77.2	16.601	1.84	na	2.2	11.03
HYDRO01	-22.216	-74.227	2.5	414	17.88	35.18	73	221.0	0.895	396.0	8.06	1896.3	180.7	10.776	4.31	0.98	0.64	0.71
HYDRO04	-17.092	-78.635	5	701	18.19	35.22	66	184.1	0.928	412.2	8.04	1905.7	177.8	10.886	4.24	1.56	0.77	1.45
BB1a	-13.999	-81.199	5	532	18.75	35.34	57	177.9	0.867	413.5	8.04	1902.7	181.8	10.759	4.33	na	na	na
BB1b	-13.921	-81.277	5	527	18.76	35.35	51	177.3	1.075	416.6	8.04	1905.6	180.9	10.793	4.31	1.70	0.78	2.20
BB1c	-13.710	-81.389	5	560	18.82	35.39	66	181.4	1.564	418.1	8.04	1906.8	180.9	10.798	4.31	na	na	na
HYDRO09	-12.992	-82.198	5	622	19.24	35.42	na	183.4	0.916	405.2	8.05	1892.0	187.4	10.572	4.46	na	na	na
HYDRO10	-16.749	-85.998	2.5	1424	19.77	35.53	99	218.0	0.459	398.6	8.05	1882.7	193.8	10.375	4.62	0.96	0.59	0.45
HYDRO11	-16.749	-84.998	0	1307	19.77	35.50	79	215.1	0.103	409.7	8.04	1890.2	190.0	10.494	4.53	1.16	0.64	0.74
HYDRO13	-16.750	-83.000	2.5	1094	19.43	35.52	94	217.8	0.530	403.9	8.05	1891.4	189.7	10.507	4.52	1.17	0.66	1.14
HYDRO14	-16.749	-82.000	0	1010	19.24	35.51	99	229.4	0.434	405.9	8.05	1895.3	188.0	10.563	4.48	1.27	0.70	1.82
HYDRO17	-16.749	-79.000	0	674	18.38	35.38	79	222.6	0.335	402.5	8.05	1900.0	183.0	10.720	4.36	1.23	0.66	1.38
U1	-19.467	-76.150	5	623	17.53	35.14	na	na	na	444.8	8.01	1932.8	164.9	11.396	3.93	na	na	na
HYDRO19	-21.499	-73.499	5	355	17.06	35.02	74	226.9	0.705	389.7	8.06	1897.1	175.9	10.923	4.20	1.01	0.61	1.48
U2	-21.502	-73.246	5	329	16.68	34.89	na	na	na	396.0	8.05	1903.4	171.4	11.087	4.09	na	na	na
BB2a	-20.419	-70.675	5	53	17.20	35.11	na	na	na	448.0	8.01	1939.2	161.6	11.514	3.85	na	na	na
BB2b	-20.769	-70.659	5	48	16.81	35.09	26	179.3	1.296	464.3	8.00	1951.2	155.3	11.784	3.70	1.98	0.99	3.23
BB2c	-20.755	-70.650	5	47	16.30	34.96	19	174.4	0.996	490.7	7.97	1968.6	145.8	12.217	3.48	2.08	1.15	4.24
BB2d	-20.742	-70.644	5	47	16.69	34.98	na	na	na	467.9	7.99	1953.5	153.1	11.876	3.65	na	na	na
BB2e	-20.748	-70.657	5	47	17.14	35.10	na	na	na	457.2	8.00	1945.4	158.9	11.630	3.79	na	na	na
BB2f	-20.732	-70.682	5	52	16.92	35.10	30	217.6	1.527	453.0	8.01	1942.0	160.3	11.568	3.82	na	na	na









<i>Acanthoica quattrosolina</i>	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Algirosphaera robusta</i>	0.00	0.00	1.30	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

<i>Syracosphaera ossa</i>	Syracosphaeraceae	1.41	5.33	1.30	2.86	2.63	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Syracosphaera prolongata</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Syracosphaera squamigera</i>		1.41	1.33	0.00	0.00	1.32	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Syracosphaera molischii</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.02	0.00	0.00	0.00	0.00	0.98	0.00	0.00	0.00	0.00	0.00	0.00
<i>Syracosphaera pulchra</i>		0.00	0.00	1.30	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Syracosphaera cf. bannockii</i>		0.00	0.00	1.30	0.00	0.00	0.00	0.00	1.02	0.00	0.00	0.00	1.87	0.00	0.00	1.49	0.00	0.00	0.00	0.00
<i>Syracosphaera histrica</i>		0.00	0.00	0.00	1.43	1.32	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Syracosphaera anthos</i>		0.00	0.00	1.30	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Syracosphaera lamina</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.67	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Michaelsarsia elegans</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Ophiaster formosus</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Calciosolenia brasiliensis</i>	Calciosoleniaceae	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Calciosolenia murrayi</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Oolithotus antillarum</i>	Calcidiscaceae	1.41	0.00	1.30	1.43	0.00	0.00	1.61	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Calcidiscus leptoporus</i>		0.00	2.67	2.60	2.86	3.95	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Hayaster perplexus</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Umbilicosphaera sibogae</i>		0.00	0.00	0.00	4.29	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Alisphaera unicornis</i>	Alisphaeraceae	0.00	1.33	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Alisphaera pinnigera</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Umbellosphaera irregularis</i>	Umbellosphaeraceae	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Umbellosphaera tenuis</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Pontosphaera syracusana</i>	Pontosphaeraceae	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Scyphosphaera apsteinii</i>		0.00	1.33	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Helicosphaera carteri</i>	Helicosphaeraceae	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Helicosphaera hyalina</i>		0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Pappomonas</i> sp. type 2	Pappomonasphaeraceae	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
<i>Florisphaera profunda</i>		a	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Gladiolithus flabellatus</i>			0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Coronosphaera mediterranea</i>		b	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
<i>Tetralithoides quadrilaminata</i>		c	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

a – nannoliths *incertae sedis*, b – genus *incertae sedis*, c – narrow-rimmed placoliths.

Table S3. Absolute abundances of coccolithophores recorded in stations that appear in Fig. S1-S2.

Station ID	Depth (m)	Total coccolithophore abundances ( $10^3$ cells $L^{-1}$ )
H04	5	20.1
	60	17.3
H13	2.5	33.3
	50	33.8
	100	5.0
H19	5	19.8
	30	44.5
	75	17.7
BB2f	5	38.9
	15	26.1
	60	na
	80	na
TON01	5	25.7
	30	7.6
TON18	5	5.8
	30	30.4
JF01	5	29.7
	40	14.7
	80	3.1
JF02	5	3.3
	40	24.5
	80	3.4
H01	2.5	44.5
	50	51.6
BB1a	5	22.6
	58	14.4
BB1b	5	10.3
	51	2.7
H10	2.5	43.3
	100	14.0
H17	0	42.7
	80	13.6
BB2b	5	48.5
	45	2.0
	68	3.1
BB2c	5	39.3
	150	na

na – no available data

