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Ocean acidification does not affect magnesium composition or dolomite formation in living crustose coralline algae, *Porolithon onkodes* in an experimental system

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Supplementary information for Nash et al.

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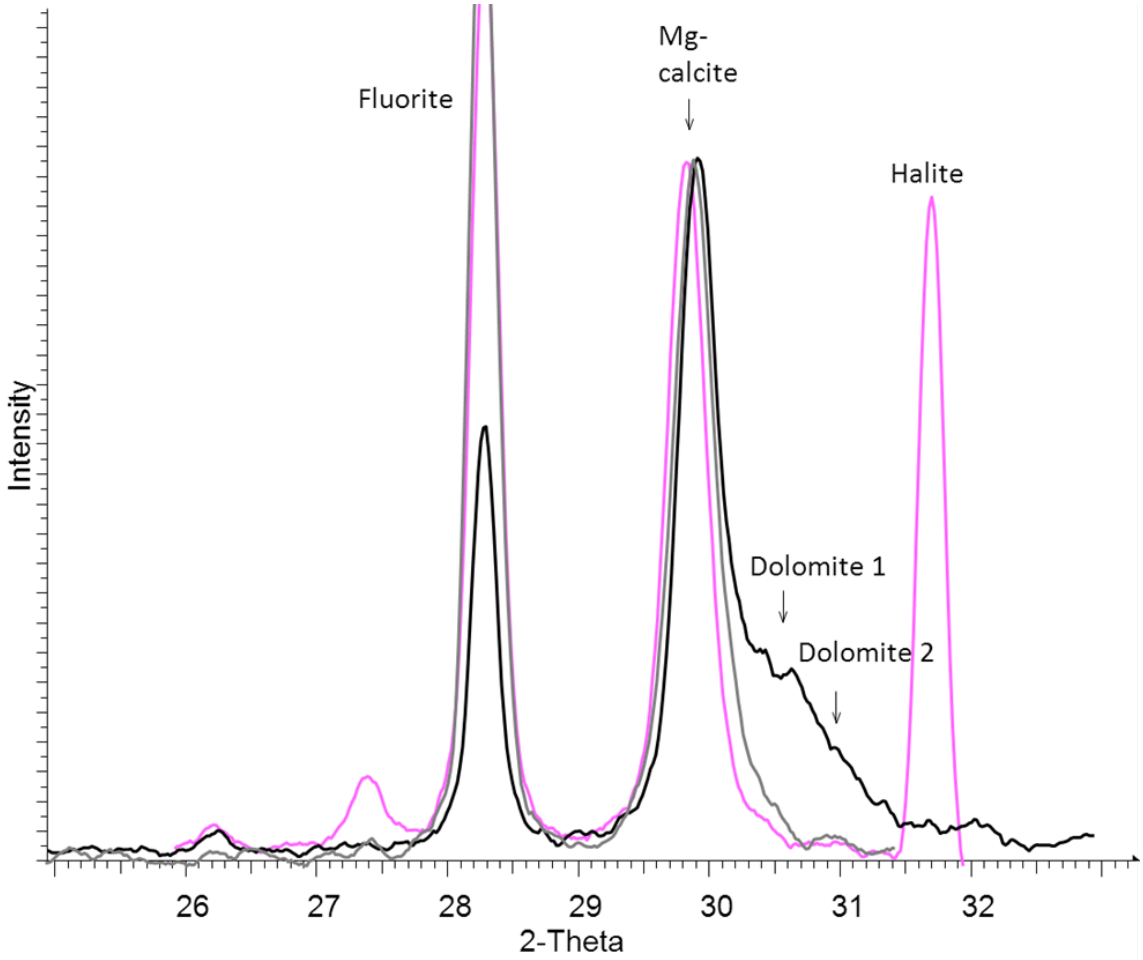

Figure S1: Example XRD scans. New settlement (pink line)(control tank 24, sample1, mol% 14.4, Asymm mol% 14.6, SI Table 3) has no peak asymmetry, the 3 month growth crust (grey line) (control 2, mol% 16.5, Asymm mol% 17.3, SI Table 1) has slight asymmetry and the bulk crust (black line) (control 2, mol% 17.6, Asymm mol% 26.7, SI Table 4) incorporating pre-experimental crust has asymmetry with an overlapping but clear peak for dolomite. Dolomite 1 is the peak position for ~50 mol% $MgCO_3$ for disordered dolomite (as identified by Zhang et al. 2010, discussed in Nash et al. 2011 and expanded in Diaz-Pulido et al. 2014). Dolomite 2 is the peak position for stoichiometric fully ordered dolomite (ICSD file card PDF 00-036-0426).

Full data tables for XRD results

	Mol % $MgCO_3$	**Asymm mol%**
Pre-I 1	16.2%	17.1%
Pre-I 2	16.6%	17.0%
Pre-I 3	17.1%	17.9%
Pre-I 5	16.3%	17.5%
Pre-I 6	16.8%	17.9%
Average	**16.6%**	**17.5%**
St. Dev	**0.39%**	**0.42%**
Control 1	16.0%	17.3%
Control 2	16.5%	17.3%
Control 3	16.4%	17.5%
Control 4	16.8%	18.6%
Control 6	16.8%	*23.0%
Average	**16.5%**	**18.8%**
St. Dev	**0.31%**	**2.45%**
Med 1	17.3%	18.0%
Med 2	16.5%	17.3%
Med 3	16.2%	16.8%
Med 4	16.2%	17.3%
Med 5	15.9%	16.7%
Average	**16.4%**	**17.2%**
St. Dev	**0.53%**	**0.51%**
High 1	16.5%	18.1%
High 2	17.5%	18.9%
High 4	16.9%	17.5%
High 5	16.2%	17.1%
High 6	16.4%	18.2%
Average	**16.7%**	**17.9%**
St. Dev	**0.45%**	**0.68%**

Table S1: Magnesium composition for new *P. onkodes* crust grown during first 3 months in pre-industrial (Pre-I), control and high CO_2. Asymm mol% incorporates the influence of any peak asymmetry toward dolomite. Net asymmetry is the difference between the

asymmetry mol% and the standard mol% $MgCO_3$. Net asymmetry > ~ 1 suggests dolomite is present. There was no statistically significant difference between any of the treatments for either mol%.

*This sample had halite and the XRD peak overlapped a low magnesite shoulder skewing the calculated mol% to higher values- sample excluded from average calculation for statistical analysis.

Treatment	**Sample**	**Mol% $MgCO_3$**	**Asymm mol%**
Pre-I	Tank 19	17.1%	18.8%
Pre-I	Tank 22	16.8%	20.6%
Pre-I	Tank 17	17.0%	19.1%
Pre-I	Tank 19, S2	16.1%	19.9%
Pre-I	Tank 19, S3	16.3%	17.6%
Average		**16.7%**	**19.2%**
St. Dev.		**0.42%**	**1.12%**
Med	Tank 18	16.3%	17.8%
Med	Tank 23	16.2%	17.5%
Med	Tank 15	16.5%	19.1%
Average		**16.4%**	**18.1%**
St. Dev.		**0.15%**	**0.84%**
High	Tank 21, S1	16.4%	18.9%
High	Tank 21, S2	15.7%	18.5%
High	Tank 21, S3	16.7%	18.6%
High	Tank 16, S1	16.2%	17.7%
High	Tank 16, S2	16.8%	18.8%
High	Tank 16, S3	17.0%	18.3%
Average		**16.5%**	**18.5%**
St. Dev.		**0.46%**	**0.44%**

Table S2: Mol% $MgCO_3$ for new *P. onkodes* crust after 6 months. There is no difference between treatments.

Treatment	Sample no.	mol% $MgCO_3$	Asymm mol%
Pre-I	Tank 19, S1	15.8%	16.0%
Pre-I	Tank 19, S2	14.3%	14.9%
Pre-I	Tank 17, S1	14.4%	14.5%
Average		**14.8%**	**15.1%**
St. Dev.		**0.83%**	**0.77%**
Control	Tank 20, S1	15.3%	15.2%
Control	Tank 20, S2	14.7%	14.8%
Control	Tank 24, S1	14.4%	14.6%
Control	Tank 24, S2	14.5%	14.5%
Average		**14.7%**	**14.8%**
St. Dev.		**0.4%**	**0.3%**
Medium	Tank 18, S1	14.2%	14.2%
Medium	Tank 18, S2	14.7%	14.7%
Medium	Tank 18, S3	15.2%	15.5%
Medium	Tank 23, S1	15.0%	15.0%
Medium	Tank 15, S1	14.4%	14.6%
Average		**14.7%**	**14.8%**
St. Dev.		**0.41%**	**0.48%**
High	Tank 16, S1	15.1%	*14.7%
High	Tank 16, S2	15.0%	16.0%
High	Tank 16, S3	14.9%	14.8%
High	Tank 13, S1	15.6%	15.8%
High	Tank 13, S2	13.8%	13.8%
Average		**14.9%**	**15.0%**
St. Dev.		**0.7%**	**0.9%**

Table S3: Mol% $MgCO_3$ for new settlement -unidentified CCA- after 6 months. CCA had not reached reproductive stage and were not able to be identified. In order to have enough material for XRD, each XRD sample comprised multiple (2-4) small growth patches. The range of 0.2 mol% $MgCO_3$ for the averages, is within measurement precision. There is no significant difference between treatments for mol% $MgCO_3$. There is little to no asymmetry influence on the mol% $MgCO_3$, this in agreement with the observation that thick crusts had not formed.

*The XRD pattern for this sample had a low calcite peak overlapping the Mg-calcite peak and accounts for the shift to lower mol% in the asymmetry measurement.

3 Month	Sample	mol% $MgCO_3$	Asymm mol%	Aragonite
Pre-I	Tank 2	17.4%	20.6%	0
Pre-I	Tank 5	17.8%	19.9%	1.3%
Pre-I	Tank 6	17.9%	23.9%	1.3%
Control	Tank 3	17.9%	20.4%	0
Control	Tank 4	17.2%	23.1%	11.9%
Control	Tank 2	17.6%	26.7%	0.8%
Control	Tank 1	17.5%	19.3%	0.6%
Control	Tank 6	17.9%	24.4%	3.0%
Medium	Tank 1	17.6%	21.4%	10.2%
Medium	Tank 2	16.9%	18.7%	1.3%
Medium	Tank 3	17.8%	22.9%	0
Medium	Tank 4	16.9%	20.2%	0
Medium	Tank 5	17.5%	18.7%	1.5%
High	Tank 2	17.3%	21.7%	0
High	Tank 5	17.3%	20.7%	0
High	Tank 4	17.1%	18.5%	1.7%
Average		**17.5%**	**21.3%**	
St. Dev		**0.33%**	**2.34%**	
6 Month	**Sample**	**Mol% $MgCO_3$**	**Asymm mol%**	**Aragonite**
Pre-I	Tank 19	18.2%	25.1%	0
Pre-I	Tank 22	17.6%	23.2%	0
Pre-I	Tank 17	18.1%	18.9%	4.10%
Pre-I	Tank 19, S2	17.9%	23.7%	0
Pre-I	Tank 19, S3	16.4%	18.9%	9.50%
Med	Tank 18	17.4%	20.6%	0.90%
Med	Tank 23	16.9%	19.1%	9.40%
Med	Tank 15	17.6%	23.7%	0
High	Tank 16, S1	17.9%	20.1%	0
High	Tank 16, S2	17.6%	25.7%	0
High	Tank 16, S3	17.6%	22.8%	0
High	Tank 21, S1	17.1%	21.5%	1.80%
High	Tank 21, S2	17.9%	24.1%	0
High	Tank 21, S3	18.2%	20.5%	1.20%
Average		**17.6%**	**22.0%**	
St. Dev		**0.51%**	**2.34%**	

Table S4: Mineralogy for pre-experimental *P. onkodes* crust after 3 and 6 months experimental duration. There is no difference in mol% $MgCO_3$ or Asymmetry mol% for 3 and 6 months. The range of mol% $MgCO_3$ is from 16.4 to 18.2 mol% $MgCO_3$, whereas the Asymm. mol% which incorporates the dolomite influence, has a greater range, from 18.5 to 26.7 mol% $MgCO_3$. This indicates that samples range from having no or minimal dolomite up to being considered as dolomite -rich (as per Nash et al. 2013). Aragonite ranges from zero up to nearly 12%.

Treatment	Spectrum	mol% $MgCO_3$	Treatment	Spectrum	mol% $MgCO_3$	Treatment	Spectrum	mol% $MgCO_3$
Control	Spectrum 14	59.8%	Control	Spectrum 23	28.3%	High	Spectrum 93	17.9%
High	Spectrum 39	58.6%	Control	Spectrum 56	28.3%	Pre	Spectrum 42	17.9%
High	Spectrum 19	56.5%	Pre	Spectrum 57	28.1%	High	Spectrum 4	17.5%
High	Spectrum 16	55.9%	Pre	Spectrum 51	28.0%	Control	Spectrum 32	17.1%
Control	Spectrum 66	53.1%	Pre	Spectrum 60	27.6%	High	Spectrum 5	16.5%
High	Spectrum 37	51.4%	High	Spectrum 104	27.5%	Control	Spectrum 13	16.4%
Control	Spectrum 18	46.0%	High	Spectrum 18	27.0%	High	Spectrum 8	16.0%
Pre	Spectrum 72	45.7%	Pre	Spectrum 53	26.5%	High	Spectrum 34	15.6%
Pre	Spectrum 71	45.4%	Pre	Spectrum 55	26.0%	Control	Spectrum 41	15.4%
Pre	Spectrum 70	44.8%	Pre	Spectrum 61	25.9%	High	Spectrum 12	15.0%
Control	Spectrum 50	44.7%	Pre	Spectrum 45	24.9%	Pre	Spectrum 67	14.8%
Control	Spectrum 68	44.7%	Pre	Spectrum 54	24.6%	High	Spectrum 22	14.4%
Control	Spectrum 11	44.4%	Control	Spectrum 9	24.5%	High	Spectrum 89	14.3%
Control	Spectrum 30	43.7%	Control	Spectrum 33	24.5%	High	Spectrum 99	14.3%
Control	Spectrum 37	43.1%	Control	Spectrum 45	24.4%	Pre	Spectrum 81	14.1%
Control	Spectrum 47	41.9%	Pre	Spectrum 73	23.2%	Control	Spectrum 53	14.0%
High	Spectrum 38	41.5%	High	Spectrum 84	23.1%	High	Spectrum 3	13.6%
Control	Spectrum 29	41.2%	Pre	Spectrum 66	23.1%	Control	Spectrum 61	13.5%
Pre	Spectrum 59	40.8%	High	Spectrum 90	23.0%	High	Spectrum 7	13.5%
High	Spectrum 103	40.6%	High	Spectrum 91	22.5%	High	Spectrum 32	13.3%
Control	Spectrum 46	40.6%	High	Spectrum 98	22.3%	Pre	Spectrum 69	13.2%
Pre	Spectrum 41	40.0%	High	Spectrum 30	22.2%	High	Spectrum 25	12.5%
Control	Spectrum 21	39.2%	Pre	Spectrum 47	22.2%	High	Spectrum 31	12.0%
Control	Spectrum 22	37.6%	High	Spectrum 92	22.1%	Pre	Spectrum 76	11.9%
High	Spectrum 13	37.6%	Pre	Spectrum 68	22.0%	High	Spectrum 9	11.8%
Control	Spectrum 55	37.4%	Control	Spectrum 12	21.7%	High	Spectrum 28	11.6%
High	Spectrum 108	37.3%	Pre	Spectrum 65	21.5%	Control	Spectrum 39	11.0%
Pre	Spectrum 43	37.2%	Pre	Spectrum 64	21.3%	High	Spectrum 35	11.0%

Treatment	Spectrum	mol% $MgCO_3$	Treatment	Spectrum	mol% $MgCO_3$	Treatment	Spectrum	mol% $MgCO_3$
Pre	Spectrum 58	36.3%	High	Spectrum 106	21.2%	Pre	Spectrum 74	10.5%
Pre	Spectrum 44	36.2%	Control	Spectrum 48	21.1%	Control	Spectrum 49	10.5%
Control	Spectrum 31	35.9%	High	Spectrum 23	21.1%	High	Spectrum 105	10.2%
Pre	Spectrum 46	35.8%	High	Spectrum 87	20.9%	Control	Spectrum 35	9.8%
High	Spectrum 15	34.7%	Pre	Spectrum 78	20.4%	Control	Spectrum 63	9.8%
High	Spectrum 107	34.6%	High	Spectrum 88	20.4%	Control	Spectrum 38	9.4%
High	Spectrum 17	34.1%	High	Spectrum 96	20.3%	Control	Spectrum 44	9.3%
High	Spectrum 109	34.0%	Pre	Spectrum 62	20.3%	Control	Spectrum 60	8.1%
Pre	Spectrum 52	33.3%	High	Spectrum 36	20.2%	Pre	Spectrum 79	7.9%
Pre	Spectrum 48	32.0%	High	Spectrum 27	19.8%	Control	Spectrum 27	7.8%
Pre	Spectrum 50	31.8%	High	Spectrum 97	19.8%	High	Spectrum 33	7.5%
Control	Spectrum 15	31.5%	High	Spectrum 85	19.3%	High	Spectrum 10	7.2%
Control	Spectrum 25	31.1%	High	Spectrum 26	19.2%	Pre	Spectrum 63	7.0%
Control	Spectrum 20	30.9%	High	Spectrum 102	19.2%	Control	Spectrum 34	6.9%
Control	Spectrum 19	30.2%	Control	Spectrum 40	19.0%	Control	Spectrum 52	6.9%
Control	Spectrum 43	30.2%	Pre	Spectrum 56	18.9%	Control	Spectrum 58	6.9%
Control	Spectrum 16	29.5%	High	Spectrum 29	18.7%	Control	Spectrum 59	6.9%
Control	Spectrum 10	29.3%	High	Spectrum 6	18.6%	Control	Spectrum 57	6.7%
Control	Spectrum 42	29.2%	High	Spectrum 94	18.5%	Control	Spectrum 67	6.3%
Control	Spectrum 17	29.1%	Pre	Spectrum 77	18.4%	Control	Spectrum 26	6.2%
High	Spectrum 14	29.0%	Pre	Spectrum 75	18.2%	Control	Spectrum 36	6.1%
High	Spectrum 20	28.8%	High	Spectrum 86	18.0%			

Table S5: SEM-EDS spectrum results for mol% $MgCO_3$. Dolomite range 36.3 – 59.8 mol% $MgCO_3$. Mg-calcite range 10.5 – 28.3 mol% $MgCO_3$.

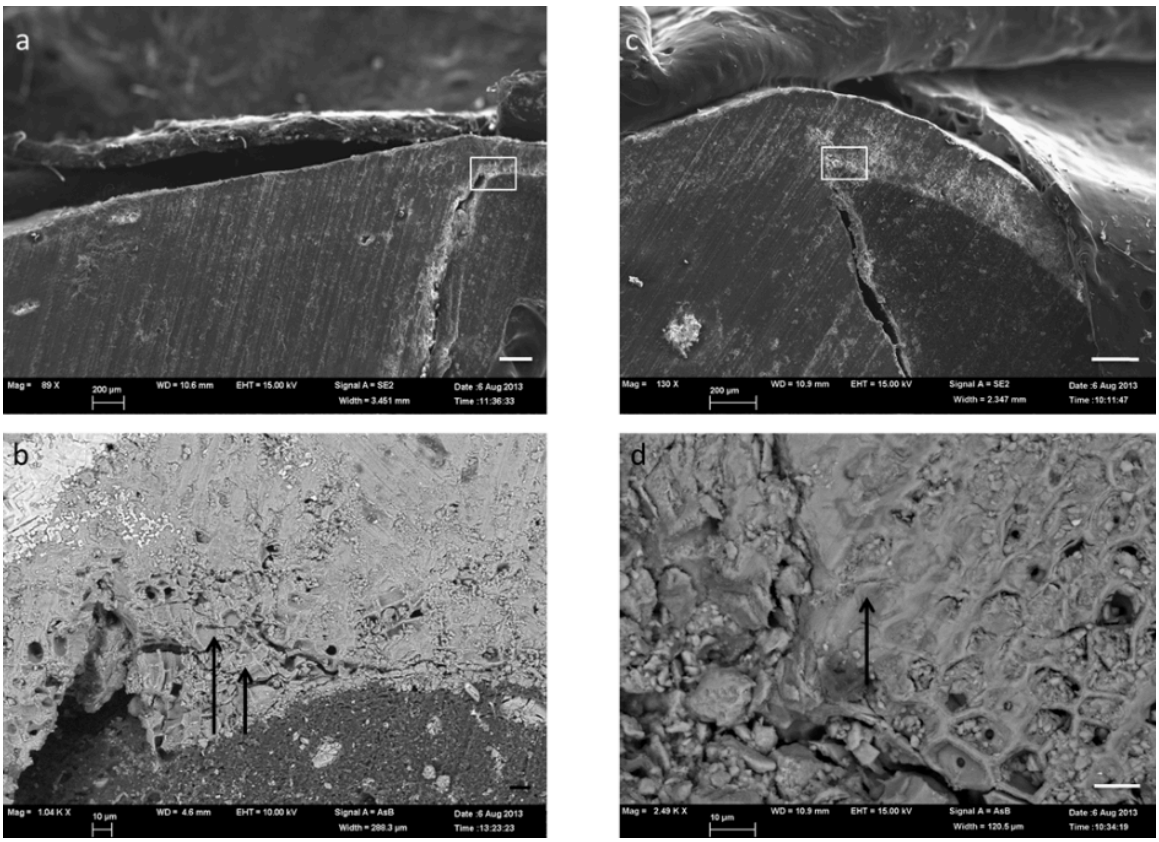

Figure S2. SEM of Pre-industrial **(a, b)** and High CO_2 **(c, d)** new *P. onkodes* crust with dolomite. **(a)** Overview of experimental growth on resin (right hand side) Pre-industrial CCA. Box is enlarged in B. **(b)** Experimental growth with dolomite (measured by EDS as 37.6 and 55.9 mol% $MgCO_3$) (arrows). Similarly to the control samples, experimental cells are not generally in-filled with dolomite to the extent observed in the old growth. **(c)** Overview of experimental growth on resin for High CO_2 CCA. Box enlarged in D. **(d)** Experimental growth with dolomite (EDS 40.0 mol% $MgCO_3$) (arrows). Similarly to control and Pre-industrial, the new cells are generally not in-filled with dolomite to the extent observed in the old growth. Scale bars: A and C = 200 microns, B and D = 10 microns.

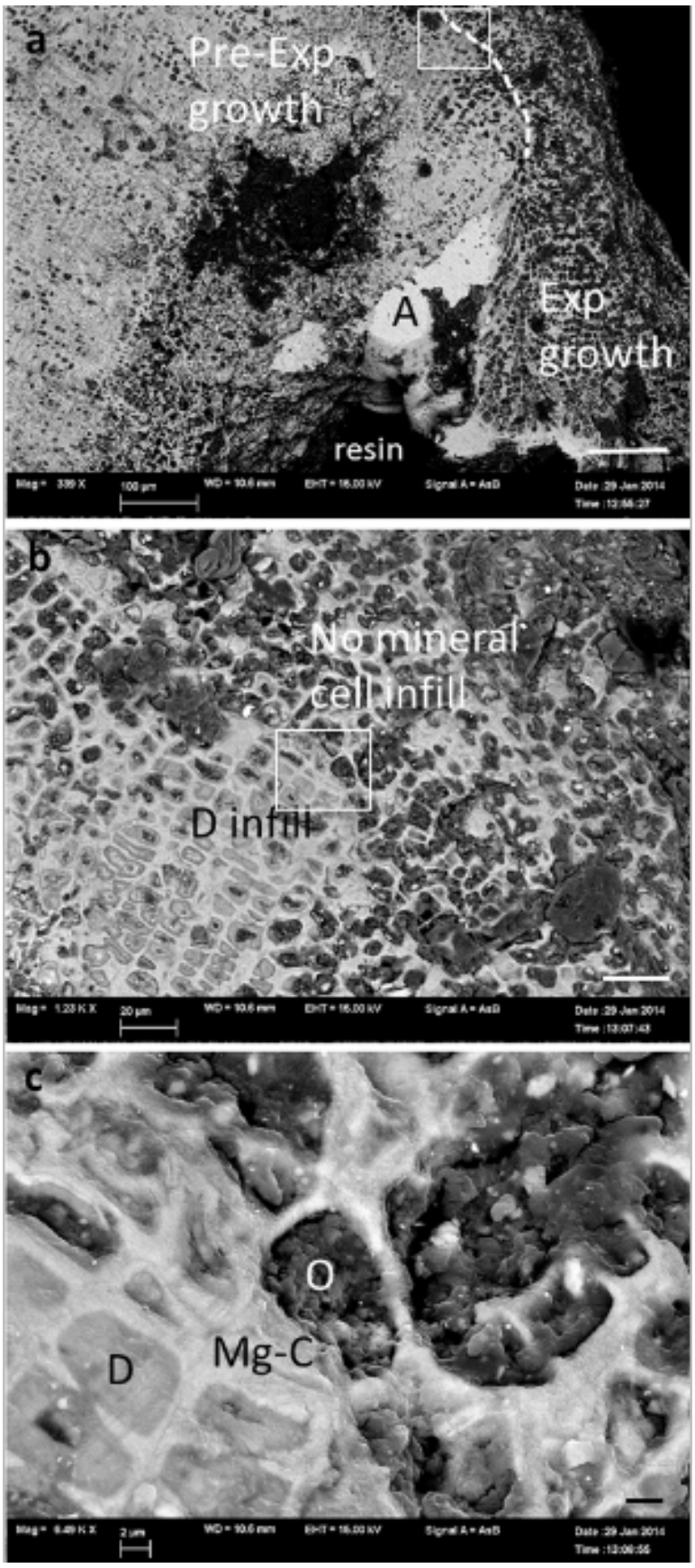
a
Pre-Exp growth
A
Exp growth
resin
b
No mineral cell infill
D infill
c
O
Mg-C
D

Figure S3: Control *P. onkodes* experimental growth over resin. **(a)** The experimental growth over the resin can be traced back to determine the boundary for the pre-experimental growth and the experimental growth. Beneath the experimental growth on resin, part of the crust has altered to aragonite (A). White box enlarged in B. **(b)** Transition from pre-experimental growth with dolomite infill (D infill) to experimental growth with organic infill but no mineral infill. **(c)** close up of transition showing dolomite in-fill cells **(d)** and cells in experimental growth with organic material (O), Mg-C – Mg-calcite, D - dolomite. Scale bars A = 100 microns, B = 20 microns, C = 2 microns.

Supplementary reference

Zhang, F., Xu, H., Hironi, K., and Roden E, E.: A relationship between d104 value and composition in the calcite-disordered dolomite solid-solution series, Am. Mineral, 95, 1650, 2010.