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Determining how biotic and abiotic variables affect the shell condition and parameters of *Heliconoides inflatus* pteropods from a sediment trap in the Cariaco Basin

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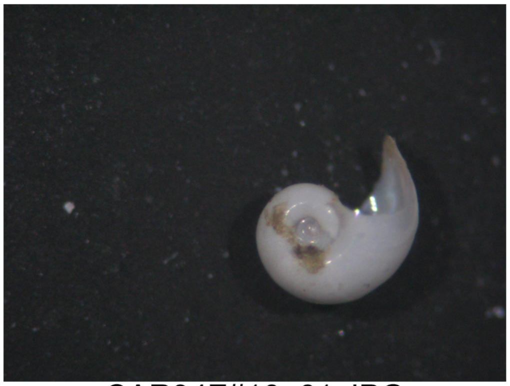
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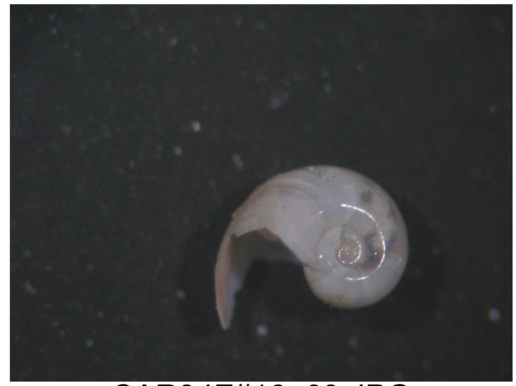
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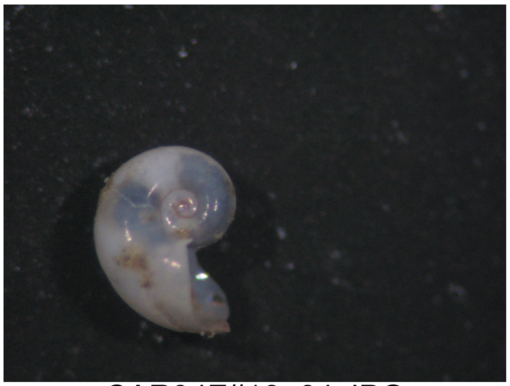
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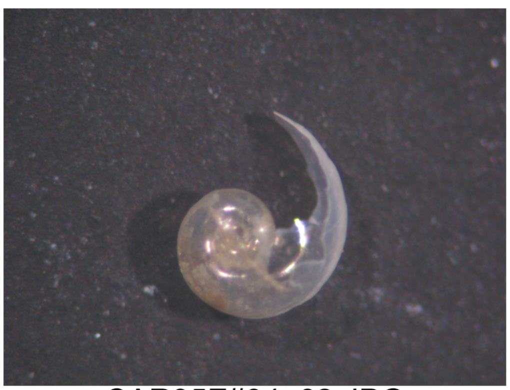
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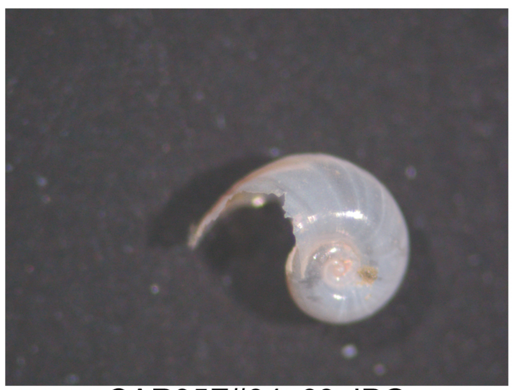
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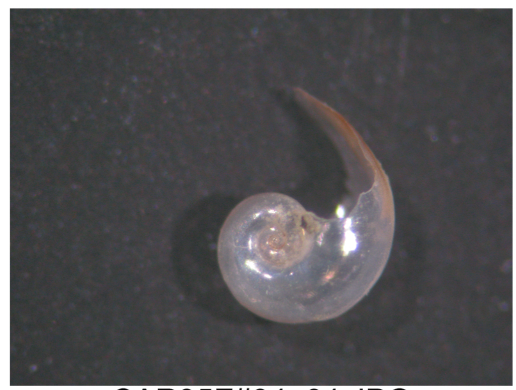
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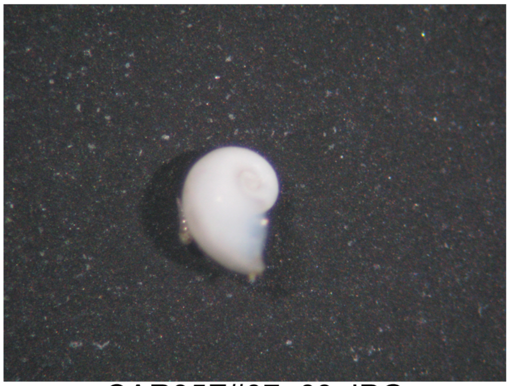
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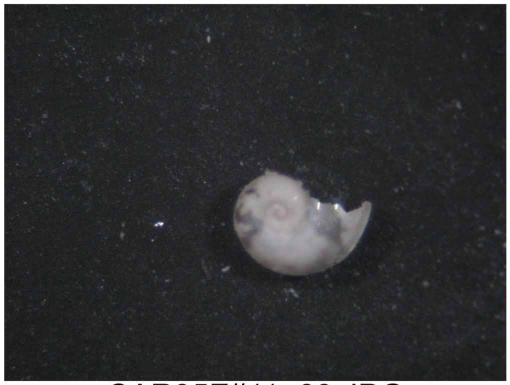
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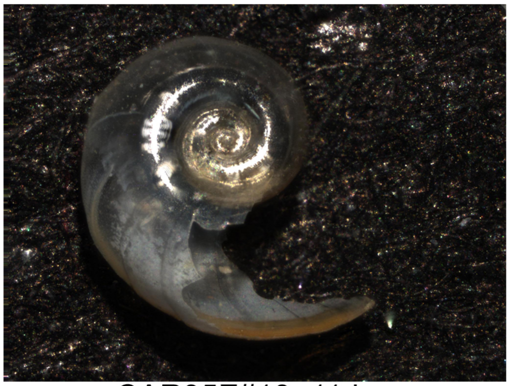
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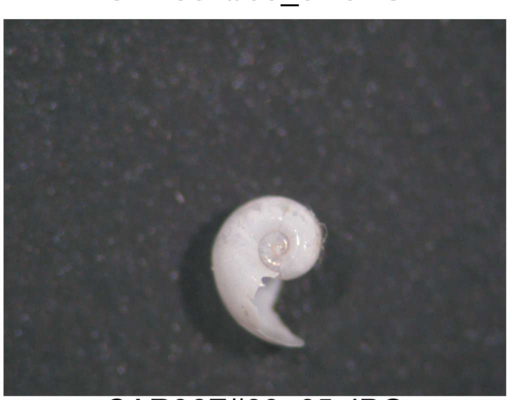
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CAR36Z#08_03.JPG


CAR36Z#08_04.JPG

Figure S1: Light microscope images

Methods

Table S1: Shell whorl, length, thickness, and volume measurements, and a record of which machine the specimen was scanned on. GE = v tome|x m, General Electric, Lewistown, PA. AMNH = v tome|x s, American Museum of Natural History, New York, NY.

Sample name	date (DD/MM/YYYY)	Number of whorls	Length (mm)	Volume (mm3)	thickness (mm)	Scanned at	ANSP Catalog No.
CAR34Z#10_01	21/03/2013	2 5/8	1.549	0.109	0.018	GE	477912
CAR34Z#10_02	21/03/2013	2 1/2	1.709	0.099	0.016	GE	477912
CAR34Z#10_03	21/03/2013	2 5/8	1.876	0.112	0.018	GE	477912
CAR34Z#10_04	21/03/2013	2 1/2	1.829	0.142	0.022	GE	477912
CAR34Z#10_05	21/03/2013	2 3/8	1.534	0.060	0.014	GE	477912
CAR35Z#04_01	20/06/2013	2 1/2	0.832	0.017	0.008	GE	477913
CAR35Z#04_02	20/06/2013	2 3/4	1.048	0.024	0.012	GE	477913
CAR35Z#04_03	20/06/2013	2 3/4	0.805	0.030	0.012	GE	477913
CAR35Z#04_04	20/06/2013	2 3/4	1.256	0.030	0.013	GE	477913
CAR35Z#04_05	20/06/2013	2 3/4	1.018	0.032	0.011	GE	477913
CAR35Z#04_06	20/06/2013	2 7/8	1.135	0.037	0.012	GE	477913
CAR35Z#04_08	20/06/2013	2 3/4	1.092	0.029	0.011	GE	477913
CAR35Z#07_01	01/08/2013	2 3/4	1.145	0.028	0.012	GE	477914
CAR35Z#07_02	01/08/2013	2 3/4	0.923	0.015	0.009	GE	477914
CAR35Z#07_04	01/08/2013	2 1/2	0.843	0.013	0.010	GE	477914
CAR35Z#07_05	01/08/2013	2 3/8	0.788	0.010	0.011	AMNH	477914
CAR35Z#07_08	01/08/2013	2 3/8	0.821	0.007	0.007	AMNH	477914
CAR35Z#07_09	01/08/2013	2 3/8	0.775	0.011	0.010	AMNH	477914
CAR35Z#07_10	01/08/2013	2 1/4	0.694	0.006	0.006	AMNH	477914
CAR35Z#07_11	01/08/2013	2 1/4	0.741	0.008	0.005	AMNH	477914
CAR35Z#07_14	01/08/2013	2 5/8	1.062	0.019	0.007	AMNH	477914
CAR35Z#11_02	26/09/2013	2 1/4	0.735	0.007	0.008	GE	477915
CAR35Z#11_03	26/09/2013	2 1/4	0.681	0.005	0.007	GE	477915
CAR35Z#11_04	26/09/2013	2 7/8	1.187	0.026	0.016	GE	477915
CAR35Z#13_01	24/10/2013	2 7/8	1.164	0.028	0.009	GE	477916
CAR35Z#13_02	24/10/2013	2 5/8	1.042	0.020	0.009	GE	477916
CAR35Z#13_05	24/10/2013	2 1/2	0.886	0.017	0.011	GE	477916
CAR35Z#13_06	24/10/2013	2 5/8	0.902	0.012	0.011	AMNH	477916
CAR35Z#13_07	24/10/2013	2 7/8	1.270	0.030	0.011	AMNH	477916
CAR35Z#13_10	24/10/2013	2 3/4	1.070	0.022	0.010	AMNH	477916
CAR35Z#13_11	24/10/2013	2 7/8	1.400	0.038	0.010	AMNH	477916
CAR35Z#13_13	24/10/2013	2 1/2	0.974	0.018	0.009	AMNH	477916
CAR35Z#13_14	24/10/2013	2 1/2	0.729	0.010	0.009	AMNH	477916
CAR36Z#03_01	08/12/2013	2 3/4	0.988	0.025	0.013	GE	477917
CAR36Z#03_02	08/12/2013	2 3/4	0.985	0.023	0.009	GE	477917

CAR36Z#03_03	**08/12/2013**	**2 5/8**	**0.985**	**0.023**	**0.011**	**GE**	**477917**
CAR36Z#03_04	**08/12/2013**	**2 5/8**	**1.063**	**0.029**	**0.014**	**GE**	**477917**
CAR36Z#06_01	**19/01/2014**	**2 5/8**	**0.931**	**0.018**	**0.011**	**GE**	**477918**
CAR36Z#06_02	**19/01/2014**	**2 7/8**	**0.979**	**0.029**	**0.011**	**GE**	**477918**
CAR36Z#08_01	**16/02/2014**	**2 7/8**	**1.117**	**0.036**	**0.012**	**GE**	**477919**
CAR36Z#08_02	**16/02/2014**	**2 3/4**	**1.016**	**0.026**	**0.015**	**GE**	**477919**
CAR36Z#08_03	**16/02/2014**	**2 5/8**	**0.938**	**0.023**	**0.010**	**GE**	**477919**
CAR36Z#08_04	**16/02/2014**	**2 3/4**	**0.972**	**0.027**	**0.015**	**GE**	**477919**
CAR36Z#08_05	**16/02/2014**	**2 3/4**	**1.150**	**0.022**	**0.009**	**AMNH**	**477919**

Not all 44 specimens could be scanned using the same equipment due to the closure of the GE facility in Lewistown, and limited availability of the scanner at AMNH. To test the impact of these changes on the analyses conducted in this paper, one specimen, CAR35Z#04_01, was scanned 4 times. The details of each scan are highlighted in Table S2.

Table S2: Scan parameters for the repeat scans run on specimen CAR35Z#04_01 to test the impact of different scanners and scan settings on the measurements of the *Heliconoides inflatus* shells in this study.

	Location	Scanner (GE Phoenix)	Target	Power (kV)	Current (μA)	Total # images	Exposure time (ms)	# images per view
Original scan	GE	v|tome|x m	diamond	65	230	1000	500	Average 5 Skip 1
AMNH scan	AMNH	v|tome|x s	diamond	65	230	1500	400	Average 3 Skip 1
AMNH rescan	AMNH	v|tome|x s	diamond	65	230	1500	400	Average 3 Skip 1
AMNH scan with GE setup	AMNH	v|tome|x s	diamond	65	230	1000	500	Average 5 Skip 1

The four repeat scans did show some variability (Fig. S2). However, despite this, the modal shell thickness of the shell analysed in all four scans is 0.008 mm. This demonstrates that modal shell thickness is a robust measurement and was not impacted by the different scanners and scan parameters used in this study.

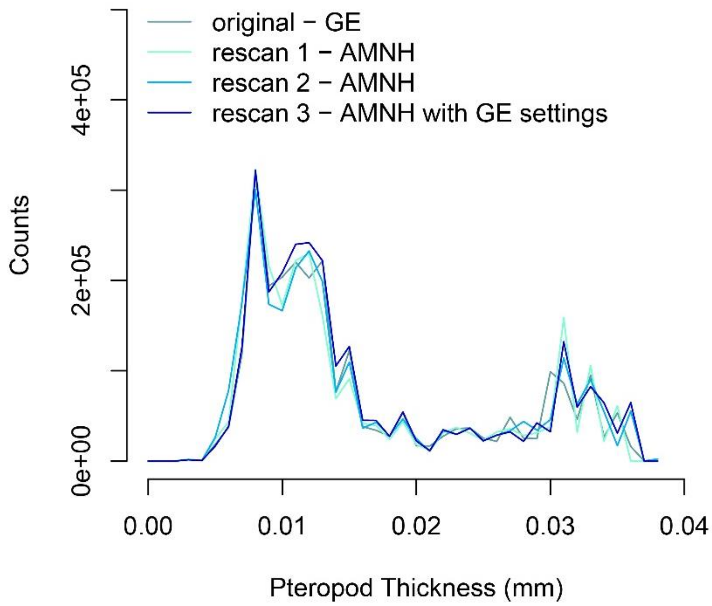

Figure S2: Shell thickness histograms from the four repeat scans of specimens CAR35Z#04_01. Although there is some variability between the four records, the modal shell thickness is recorded as 0.008 mm in all four scans. This suggests that modal shell thickness is a robust measurement and was not affected by the different scanners and scan parameters used in this study.

Results

Table S3: Pteropod shell condition scores assessed using the LDX and opacity methods

			Shell condition	
Sample	trap #	day	LDX	Opacity
CAR34Z#10_01	10	79	2.0	0.39
CAR34Z#10_02	10	79	2.0	0.40
CAR34Z#10_03	10	79	1.5	0.31
CAR34Z#10_04	10	79	2.0	0.33
CAR34Z#10_05	10	79	2.5	0.43
CAR35Z#04_01	4	170	2.5	0.40
CAR35Z#04_02	4	170	0.0	0.17
CAR35Z#04_03	4	170	1.0	0.36
CAR35Z#04_04	4	170	0.0	0.23
CAR35Z#04_05	4	170	1.0	0.39
CAR35Z#04_06	4	170	1.5	0.40
CAR35Z#04_07	4	170	0.5	0.31
CAR35Z#04_08	4	170	1.0	0.38
CAR35Z#07_01	7	212	2.0	0.43
CAR35Z#07_02	7	212	2.0	0.59
CAR35Z#07_03	7	212	2.0	0.59
CAR35Z#07_04	7	212	2.0	0.74

CAR35Z#07_05	7	212	2.0	0.36
CAR35Z#07_09	7	212	2.0	0.35
CAR35Z#07_10	7	212	2.0	0.33
CAR35Z#07_11	7	212	2.0	0.57
CAR35Z#07_14	7	212	2.5	0.45
CAR35Z#11_01	11	268	4.0	0.43
CAR35Z#11_02	11	268	2.0	0.46
CAR35Z#11_03	11	268	3.0	0.40
CAR35Z#11_04	11	268	2.5	0.46
CAR35Z#11_05	11	268	4.0	0.48
CAR35Z#13_01	13	296	4.0	0.44
CAR35Z#13_02	13	296	3.0	0.60
CAR35Z#13_03	13	296	2.0	0.51
CAR35Z#13_04	13	296	2.0	0.58
CAR35Z#13_05	13	296	3.0	0.64
CAR35Z#13_06	13	296	2.5	0.33
CAR35Z#13_07	13	296	2.0	0.53
CAR35Z#13_10	13	296	3.0	0.58
CAR35Z#13_11	13	296	0.5	0.19
CAR35Z#13_13	13	296	2.0	0.51
CAR35Z#13_14	13	296	3.0	0.27
CAR36#03_01	3	341	1.0	0.49
CAR36#03_02	3	341	0.5	0.35
CAR36#03_03	3	341	1.0	0.38
CAR36#03_04	3	341	1.5	0.43
CAR36#03_05	3	341	2.0	0.50
CAR36Z#06_01	6	383	3.0	0.50
CAR36Z#06_02	6	383	2.0	0.46
CAR36Z#08_01	8	411	1.5	0.45
CAR36Z#08_02	8	411	2.0	0.49
CAR36Z#08_03	8	411	2.0	0.44
CAR36Z#08_04	8	411	2.0	0.57

Table S4: Water chemistry and hydrographic measurements at 55 m depth from 12 time points through the year. The measurements were made on monthly cruises that did not directly correspond to the closing times of the sediment trap cups. Water chemistry parameters from 55 m were selected as this is the depth closest to the calcification depth of this species (75 m; Keul et al., 2017).

Month	Year	Temp. at 55m (°C)	Salinity at 55m (PSU)	Alkalinity at 55m (µmol/kg)	pH at 55m	Ωarag (calculated from TA and pH)	PO4 at 55 m (µM)	NO3 and NO2 at 55 m (µM)
3	2013	20.741	36.726	2390	7.893	2.39	0.48	8.12
4	2013	21.816	36.903	2418	7.949	2.80	0.27	4.94
5	2013	21.278	36.864	2419	7.928	2.64	0.32	6.16
6	2013	22.574	36.873	2422	7.959	2.93	0.25	4.43
7	2013	22.798	36.932	2436	7.959	2.97	0.17	3.83
8	2013		36.907	2425	7.957		0.19	3.52
9	2013	24.844	36.66	2416	8.033	3.59	0.06	0.00
10	2013	23.125	36.813	2425	7.947	2.92	0.35	5.37
11	2013	24.152	36.738	2420	7.989	3.25	0.18	2.92
12	2013	23.092	36.787	2420	7.952	2.93	0.32	4.90
1	2014	20.643	36.734	2390	7.884	2.34		8.84
2	2014	20.64	36.723	2420	7.865	2.28	0.52	8.81

Table S5: Relationships among different shell parameters (whorls, diameter, thickness, and shell condition (LDX)), were examined relative to each other and to the sample collection date using a simple linear model. To account running multiple comparisons, *p* values were corrected using both the more conservative Bonferroni correction and the less conservative false discovery rate FDR)

test	R_2	*p*	Bonferonni	FDR
amount~diameter	0.819	0.000	0.000	0.000
amount~whorls	0.012	0.047	0.425	0.055
thickness~diameter	0.582	0.000	0.000	0.000
thickness~amount	0.680	0.000	0.000	0.000
thickness~whorls	0.089	0.049	0.444	0.055
whorls~diameter	0.095	0.042	0.374	0.055
LDX~day	0.028	0.251	1.000	0.251
LDX~trap	0.357	0.000	0.000	0.000
LDX~opacity	0.208	0.010	0.090	0.018

Table S6: Results of a Welch's t-test run to assess whether there is a significant difference between specimens sampled during times of upwelling (December - April, and times when there is no upwelling (i.e., during the rainy season (August - November)).

Category	Mean rainy season (no-upwelling)	Mean upwelling	*t*	*p*	
# whorls	2.6	2.7	-1.752	0.0891	
shell diameter	0.94	1.23	-2.906	0.0080	*
amount of shell material	0.0166	0.0502	-3.23	0.0052	*
modal shell thickness	0.009	0.014	-4.0597	0.0004	*
residual thickness	-0.236	0.266	-2.337	0.0260	*

* = significant

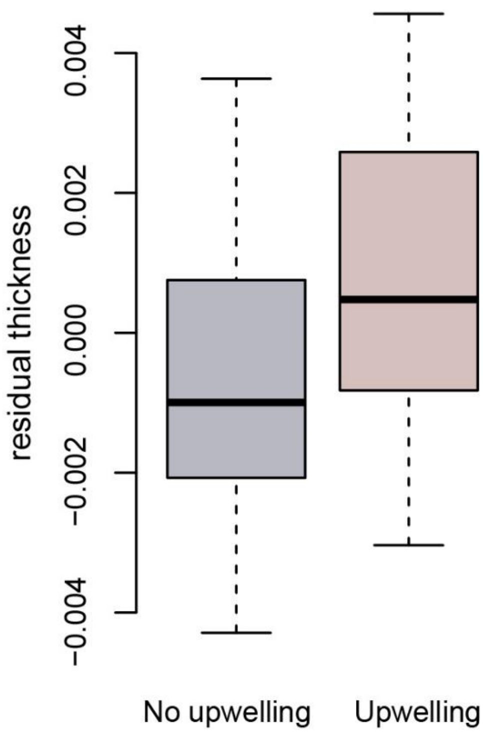

Figure S3: The calculated residual shell thickness (i.e., the thickness once the influence of length on thickness has been removed) of specimens from times of upwelling (red) and times of no upwelling (grey) in the Cariaco Basin. Specimens collected during times of upwelling are significantly thicker than those which formed at times with no upwelling (Welch's t-test: *p* = 0.0260)