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1 **Rapid multi-generational acclimation of coralline algal reproductive structures to ocean**
2 **acidification**

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28 Keywords: acclimation, conceptacles, coralline algae, multi-generational, ocean acidification, pH
29 variability

30

31 **Abstract**

32

33 The future of coral reef ecosystems is under threat because vital reef-accreting species such as
34 coralline algae are highly susceptible to ocean acidification. Although ocean acidification is known to
35 reduce coralline algal growth rates, its direct effects on the development of coralline algal
36 reproductive structures (conceptacles) is largely unknown. Furthermore, the long-term, multi-
37 generational response of coralline algae to ocean acidification is extremely understudied. Here, we
38 investigate how mean pH, pH variability and the pH regime experienced in their natural habitat affect
39 coralline algal conceptacle abundance and size across six generations of exposure. We show that
40 second generation coralline algae exposed to ocean acidification treatments had conceptacle
41 abundances 60% lower than those kept in present day conditions, suggesting that conceptacle
42 development is initially highly sensitive to ocean acidification. However, this negative effect of ocean
43 acidification on conceptacle abundance disappears after three generations of exposure. Moreover, we
44 show that this transgenerational acclimation of conceptacle development is not facilitated by a trade-
45 off with reduced investment in growth, as higher conceptacle abundances are associated with crusts
46 with faster growth rates. These results indicate that the potential reproductive output of coralline algae
47 may be sustained under future ocean acidification.

48

49 **Introduction**

50 Ocean acidification is a threat to many marine organisms, with calcifying taxa and the ecosystems
51 they support particularly at risk [1,2]. As a hotspot for marine biodiversity [3,4], and an annual source
52 of billions of dollars to the global economy [5] coral reefs are one of the most valuable ecosystems
53 threatened by ocean acidification. These ecosystems are vulnerable because framework-building
54 corals and the coralline algae that cement this framework together may both be impaired by changes
55 in seawater carbonate chemistry [1,2]. Furthermore, coralline algae are predicted to be highly

56 susceptible to these altered conditions, as they precipitate one of the most soluble calcium carbonate
57 polymorphs, high magnesium calcite [6,7]. Coralline algae are important to the functioning of coral
58 reefs as they maintain the structural integrity of reefs by cementing them together, whilst also
59 providing a number of ecosystem services such as facilitating coral larval settlement [8,9] and
60 providing food for invertebrates [10]. Therefore, if coralline algal survival, growth or reproductive
61 output is reduced by future ocean acidification these services that are vital to the persistence of coral
62 reefs could be lost. Previous research has demonstrated reduced calcification and growth in mature
63 coralline algae under ocean acidification [11–13], as well as significant reductions in coralline algae at
64 natural analogues of future ocean acidification [14,15]. Moreover, coralline algal recruitment and
65 calcification during early life-stages appear to be highly susceptible to reduced seawater pH lower
66 than 7.91 as demonstrated through decreasing calcification rates and ~90% reduced recruitment rate
67 (or success) [16–18]. If such large reductions in recruitment are realized under ocean acidification,
68 coralline algal populations will inevitably experience significant declines, with severe negative
69 consequences for coral reefs [19].

70

71 When assessing the impacts of environmental change on marine organisms it is important to consider
72 time scales that match the timespan organisms will have to respond to external changes, as previous
73 studies have demonstrated differing responses under long and short term exposure [20,21]. Such
74 considerations are also required as a host of transgenerational processes can act over multiple
75 generations to improve fitness. For example, tropical reef fish are initially sensitive to ocean warming
76 but can rapidly acclimate over two generations [22], whilst Rock Oyster larvae exposed to elevated
77 pCO₂ grew larger and developed faster when their parents also experienced elevated pCO₂ [23]. When
78 discussing generational improvements in performance it is important to define the use of terms such
79 as acclimation or adaptation. Here, we use the term acclimation to refer to changes in performance
80 driven by non-genetic plasticity in responses [22,24,25], and the term adaptation to refer to changes in
81 performance driven by genetic change [26,27]. A recent study by Cornwall *et al.*, [18] was the first to
82 assess the response of a reef-accreting calcifier to ocean acidification across multiple (>two)
83 generations. They report that although coralline algae are initially highly sensitive to ocean

84 acidification, growth was no longer affected by ocean acidification after six generations of exposure.
85 Yet, this novel result should be interpreted with caution, as increases in growth after multiple
86 generations under ocean acidification may be at the expense of other processes such as reproduction.
87 Similar trade-offs have been observed in coralline algae, where maintaining calcification rates under
88 ocean acidification was coupled with harmful changes in skeletal ultrastructure [28,29], although such
89 trade-offs are not always observed [30]. Therefore, whilst measuring growth is valuable, a multi-
90 generational assessment of reproductive structures in response to ocean acidification will provide
91 further insights into how coralline algae respond to ocean acidification.

92

93 The pH variability within any one habitat can dictate the sensitivity of resident calcifying species to
94 ocean acidification. Exposure to variable pH regimes is common in shallow-water reef ecosystems
95 where local metabolic activity and long water residence times combine to cause large diurnal
96 oscillations in seawater chemistry [31,32]. Such variable pH regimes may mitigate the effects of
97 ocean acidification on coralline algae by facilitating enhanced calcification rates during the day
98 [13,33], by selecting for individuals with low night pH tolerance due to phenotypic plasticity [34,35]
99 or natural transgenerational acclimation [25,36]. However, such hypothesised benefits are not always
100 apparent, with roughly half of past studies indicating coralline algae respond negatively to greater pH
101 variability [37], and that pre-exposure to low pH due to pH variability does not facilitate acclimation
102 to ocean acidification [18,38–40]. Contrasting results such as these suggest the effect of pH variability
103 on coralline algal responses to ocean acidification may be species or population specific. To-date, the
104 impacts of pH variability on the conceptacles of coralline algae from sites with vastly different pH
105 regimes has not been assessed

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107 To understand the vulnerability of coralline algal recruitment and early life stages to ocean
108 acidification, it would be ideal to isolate and investigate the various stages of reproduction and early
109 life stage development. Coralline algae theoretically have a triphasic life cycle that allows
110 reproduction via both sexual and asexual pathways. In coralline algae, like in many rhodophytes, the

111 asexual phase may be the only one ever observed [41] and occurs through tetraspore-to-tetraspore-
112 cycling, in which tetrasporophytes produce self-perpetuating diploid spores [42]. Additionally,
113 tetrasporophytes have been observed to produce diploid bispores, which can also grow directly into
114 tetrasporophytes [42]. In coralline algae, many reproductive structures, including the spore producing
115 sporangia, are housed within reproductive conceptacles found on the surface of intergenicula or crusts
116 [42]. Thus, the development and abundance of these conceptacles (Figure 1) have a key role in
117 sustaining coralline algal populations and can be used as a proxy for reproductive output. However, it
118 is important to note that measurements of conceptacle abundance do not exactly equal fecundity.
119 Despite their importance, there has been limited research into the direct effects of ocean acidification
120 on coralline algal conceptacles, with the single study conducted finding no impact of reduced pH
121 (7.81, 7.60) on *Pneophyllum* sp. conceptacle numbers [43].

122

123 Here, we investigate how reduced mean pH, differing levels of pH variability and the pH regime
124 experienced by coralline algae in their natural habitat impact the abundance and size of conceptacles
125 of the reef building coralline algae *Hydrolithon reinboldii*, across six generations of exposure. We
126 hypothesize that, (1) coralline algal conceptacle abundance and size will decline under ocean
127 acidification, (2) prior exposure to low pH will not influence *H. reinboldii*'s tolerance to ocean
128 acidification, (3) diurnal pH variability will not reduce the effect of ocean acidification, and (4) any
129 impact of ocean acidification on *H. reinboldii* conceptacles will be reduced after multiple generations
130 of exposure.

131

132 **Materials and Methods**

133

134 **Treatment Design and Experimental Set-up**

135 Coralline algal rhodoliths of the species *Hydrolithon reinboldii* were collected from two sites with
136 contrasting pH regimes in the macrotidal Kimberley region of Australia in October 2016 (see figure
137 S1 for a map of these sites and figure S2 for information on the pH regimes). See supplementary

138 methods for site description and collection information. To investigate the effects of mean pH and
139 different levels of pH variability on *H. reinboldii* four pH treatments were employed within aquaria.
140 Two treatments had a mean pH_T of 8.00, selected to represent the present day mean surface seawater
141 pH_T detailed in ocean acidification studies best practices guide [44], and two treatments had a mean
142 pH_T of 7.70, designed to simulate potential future pH under ocean acidification (Representative
143 Concentration Pathway 8.5; 37). For the two treatments with the same mean pH, one was
144 characterised by high pH variability and the other low pH variability, designed to emulate the pH
145 regimes of Tallon and Shell Island respectively. Thus, the four experimental pH treatments were
146 denoted: (1) present-day low variability (8.00 mean pH_T, mean daily range of 0.14), (2) present-day
147 high variability (8.00 mean pH_T, mean daily range of 0.58), (3) ocean acidification low variability
148 (7.70 mean pH_T, mean daily range of 0.20) and (4) ocean acidification high variability (7.70 mean
149 pH_T, mean daily range of 0.89). Figure S3 highlights the pH regime of all four treatments and Table
150 S1 displays the achieved pH for each of the four pH treatments used. Coralline algae collected from
151 the two sites were divided amongst the four treatments in 48 experimental tanks, resulting in six
152 independent experimental tanks for each of the eight pH treatment × site of origin combinations. See
153 supplementary methods for full details of experimental set-up and design.

154

155 **Multi-generational Experiment**

156 In this study only the conceptacles of coralline algae that recruited during the experiment were
157 assessed, therefore the generation one mature rhodoliths collected from the wild (F0), were used
158 solely as source populations. The juvenile coralline algae assessed in this study are the same as those
159 studied in Cornwall *et al.*, [18]. Whilst in the experimental tanks the generation one wild coralline
160 algae released spores which went on to form recruits on the inside walls of the tanks. After 78 days
161 these generation two coralline algal recruits were visible on the walls of the experimental tank.
162 Mature wild individuals were removed from the tanks on day 128 and two 4.5 cm × 5.0 cm (length ×
163 width) sections were cut out of each tank. These two sections of each tank were primarily cut from the
164 area near the water outflow where possible, as this area contained the highest cover of coralline algal

165 recruits. These plates were then placed into completely new tanks (in the same treatment as the
166 previous generation), allowing for assessment of this second generation. Within these new
167 experimental tanks generation two algae formed conceptacles and released spores, which again settled
168 onto the walls of the tank. Two 4.5 cm × 5.0 cm sections containing generation 3 recruits were cut
169 from the experimental tank and placed into a new experimental tank (See figure S4 for schematic
170 representation of this experimental approach). This process was repeated every 41–51 days, allowing
171 five generations (generation numbers 2–6) of plate-based coralline algal conceptacles to be studied.
172 Coralline algal recruits across all generations possessed uniporate conceptacles, containing multiple
173 tetrasporangial spores which are characteristic of *H. reinboldii* [41], and suggests each new generation
174 was a result of asexual reproduction [46]. Contamination by foreign spores from local environments
175 was unlikely due to the consistent times to visible recruitment across generations, the absence of
176 coralline algae recruiting in the header tanks, and the observation that all recruits had similar
177 anatomical features [41]. To investigate whether any observed differences across generation were due
178 to an altered response to treatments or an interaction between altered response to treatments and
179 laboratory effects [47], a reciprocal transplant experiment was conducted. For full details of this
180 reciprocal transplant experiment see supplementary methods.

181

182 **Conceptacle Measurements**

183 At the end of each generation, the coralline algae plates were removed from the tanks and stored in
184 silica gel for up to three years, before three random locations on each plate were blindly selected and
185 photographed using a compound microscope for analysis. For each plate conceptacle abundance was
186 measured by counting all the visible conceptacles in the area of analysis. Individual conceptacle
187 diameters were measured as the distance between the two most distant points of each conceptacle. For
188 full details of microscopic conceptacle imaging and analysis see supplementary methods.

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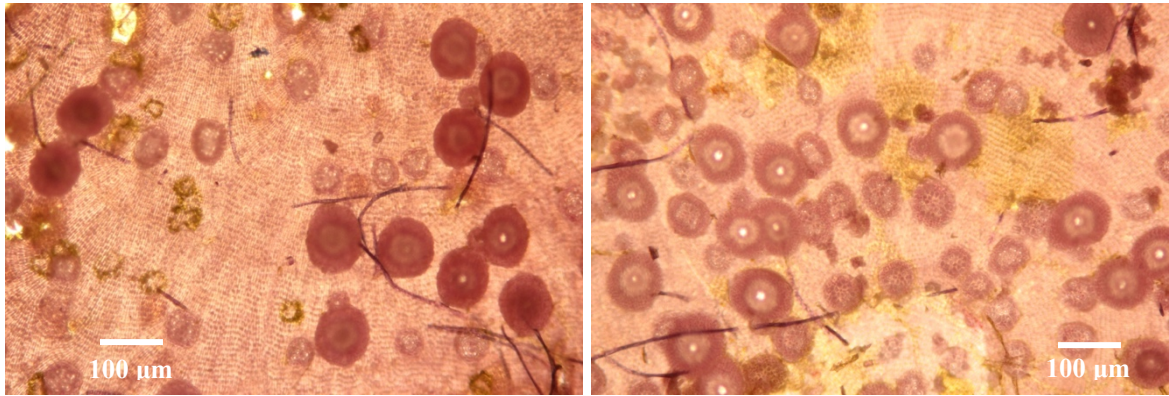


Figure 1. Images of the *Hydrolithon reinboldii* conceptacles quantified in this study. Shown are examples of the microscopic images used to quantify conceptacle abundance and diameter.

Growth and Recruitment Measurements

Multi-generational growth and total recruit area of the same coralline algae populations were investigated in relation to the conceptacle abundances measured in this study. Full methods and data are reported in Cornwall *et al.*, [18], but see supplementary methods for details of these measurements and their comparison with conceptacle abundances.

Statistical Analysis

Conceptacle abundance per plate and mean conceptacle diameter per plate was analysed using generalised linear mixed models with generation, mean treatment pH, pH variability and site of origin as fixed factors and header tank and water bath as random effects. As data was not normally distributed a Poisson distribution was specified for conceptacle abundances and a Gamma distribution was specified for conceptacle diameter measurements. For the reciprocal transplant experiment, both conceptacle abundance and diameter were assessed using the same model structure, except that generation and mean treatment pH factors were replaced with the factors; generation 2–6 mean treatment pH and transplant treatment mean pH. Following the reciprocal transplant experiment, Wilcoxon rank sum tests were used to test for pairwise differences in conceptacle abundances between the different generation 2–6 and transplant treatment combinations.

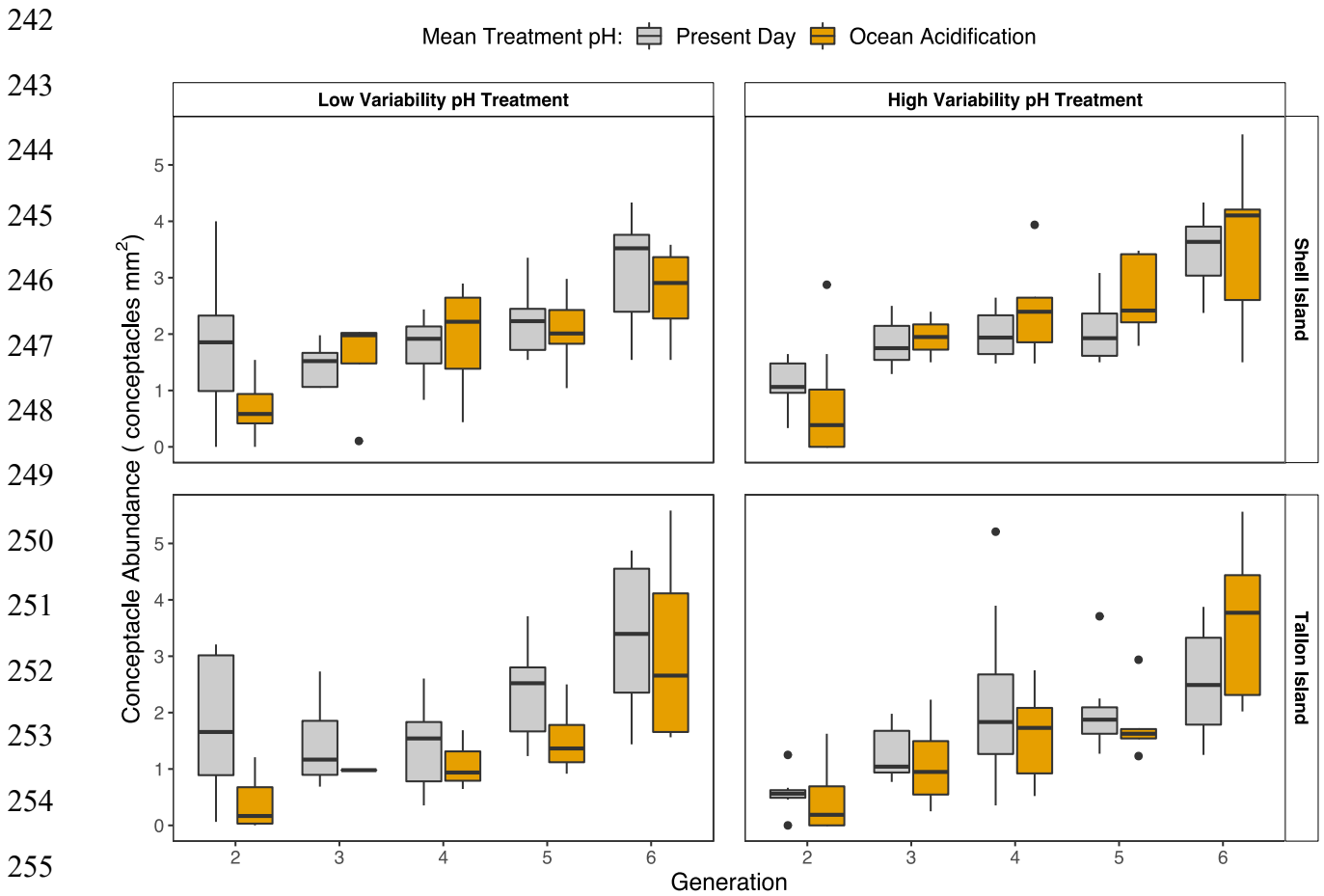
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To allow for comparison with growth and recruitment data, mean conceptacle abundances per plate were averaged for each experiment tank (total of 48 per generation). The effect of conceptacle abundance on the total recruit area of the following generation, as well as the influence of treatment mean pH was assessed using generalised linear mixed models with conceptacle abundance and mean treatment pH as fixed factors. A quasi-Poisson model was used to account for non-normality of data and overdispersion greater than 1. The same model was used to investigate whether changes in growth rates impacted the abundance of conceptacles, however in this model growth and mean pH were specified as fixed factors. Assumptions of all models were checked via visual inspection of standard model diagnostic plots. All analyses were performed in R version 3.6.1.

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240 **Results**

241 **(a) Conceptacle Abundance**



256 **Figure 2.** *H. reinboldii* conceptacle abundance after exposure to present-day (mean pH 8.00) and ocean
257 acidification (mean pH 7.70) treatments for between two and six generations. Figures are split by site of
258 origin (Shell or Tallon Island) and variability of pH treatment (low or high). Median, 25% and 75%
259 quartiles are presented.

260

261 The abundance of conceptacles varied with generation ($F = 206.882, p < 0.001$), treatment mean pH

262 ($F = 8.241, p = 0.004$) and coralline algae site of origin ($F = 5.354, p = 0.022$) (Figure 2). No

263 interactive effects between site of origin, mean treatment pH and level of pH variability were

264 observed, therefore abundance and size data from different populations and pH variability treatments

265 were grouped for subsequent analyses of mean treatment pH and generational effects (Figure S5).

266 Coralline algae exposed to ocean acidification treatments for two generations had a mean conceptacle

267 abundance that was 60% lower than coralline algae kept in present-day treatments (Figure S5) (Mean
 268 conceptacle abundances, standard error and n for each generation by treatment combination are
 269 detailed in Table S2). However, after three generations of exposure the reduction in mean conceptacle
 270 abundances under ocean acidification declined to 7% (Figure S5). This differing effect of treatment
 271 mean pH across generations is supported by a significant interaction between generation and mean pH
 272 ($F = 8.204, p = 0.005$).

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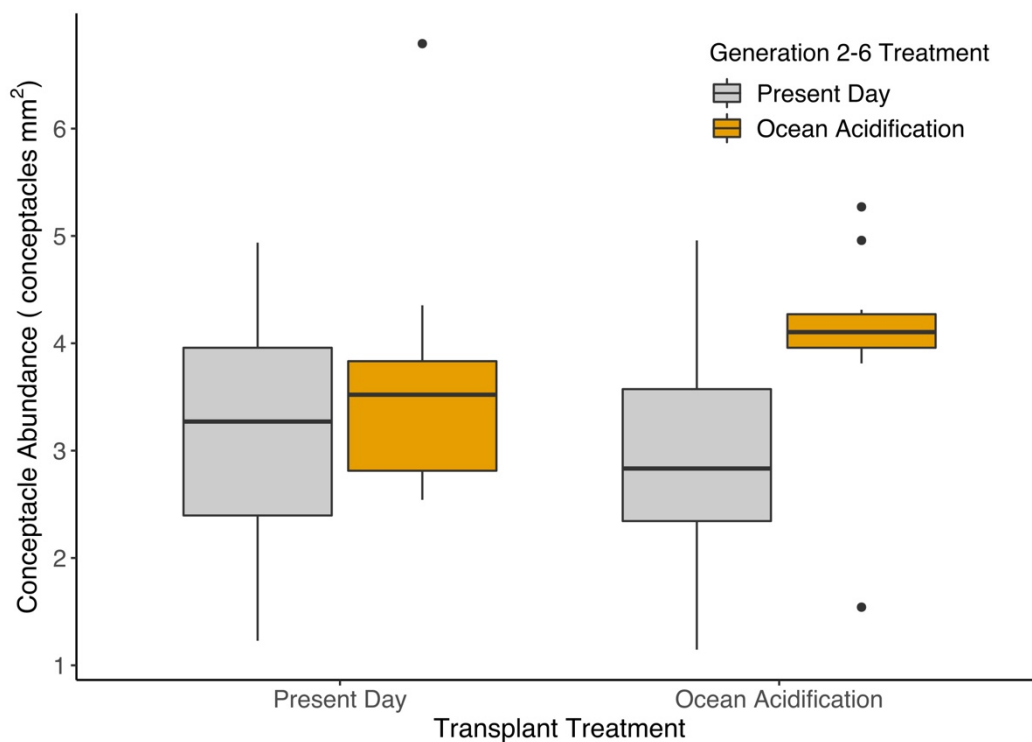
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286 **Figure 3.** Results from reciprocal transplant experiment. *H. reinboldii* conceptacle abundance
 287 after seven generations of exposure to present-day and ocean acidification treatments (same
 288 generation 2–6 treatment and transplant treatment), and those transplanted to novel treatments
 289 in generation seven (different generation 2–6 and transplant treatment). Responses to low and
 290 high variability pH treatments are combined. Median, 25% and 75% quartiles are displayed.

291

292 During the reciprocal transplant experiment, coralline algae exposed to ocean acidification treatments
 293 throughout generations 2–6 had higher conceptacle abundances than those in present-day treatments
 294 in generations 2–6 ($F = 7.797, p = 0.012$) (Figure 3). However, transplant treatment ($F = 0.059, p =$

295 0.810) and the interaction between generation 2-6 treatment and transplant treatment ($F = 0.303$, $p =$
296 0.589) did not affect conceptacle abundance. Populations exposed to ocean acidification treatments
297 for seven generations had a mean conceptacle abundance of $4.04 \text{ conceptacles mm}^2$, whereas those in
298 present day treatments throughout generations 2–6, had a mean conceptacle abundance of 3.06
299 conceptacles mm^2 after being transplanted to ocean acidification treatments in generation seven. This
300 represents a 25% decline in conceptacle abundance in populations transplanted to ocean acidification
301 treatments compared with those in ocean acidification treatments for seven generations. This
302 difference is demonstrated by a significant pairwise difference between conceptacle abundances in
303 these two reciprocal transplant groups ($W=38.5$, $p = 0.024$). Mean conceptacle abundance, standard
304 error and n for each generation 2–6 treatment by transplant treatment combination is detailed in Table
305 S3.

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323 **Conceptacle Abundance vs Growth and Recruitment**

324 In each generation an increase in mean coralline algal growth per tank has a positive effect on the
325 mean conceptacle abundance of the same coralline algae (Figure S6). Increasing conceptacle
326 abundance with increasing growth rates are evidence of this ($F = 110.937$ $p < 0.001$). The relationship
327 between growth and conceptacle abundance is not affected by mean treatment pH ($F = 0.006$, $p =$
328 0.937) or generation ($F = 1.958$, $p = 0.163$). As the conceptacle abundance of the plate-based coralline
329 algae increases, the total recruit area (of the next generation) derived from that population also
330 increases ($F = 39.671$, $p < 0.001$) (Figure S7). Mean treatment pH ($F = 18.848$, $p < 0.001$) and
331 generation ($F = 7.610$, $p = 0.006$) have a significant effect on total recruit area ($F = 18.848$, $p <$
332 0.001). The relationship between conceptacle abundance and next generation recruitment is not
333 affected by mean treatment pH ($F = 0.006$, $p = 0.937$) yet it is affected by generation ($F = 5.100$, $p =$
334 0.0250).

335

336 **(b) Conceptacle Size**

337 Conceptacle diameter increased with generation ($F = 14.099$, $p < 0.001$, $df = 1$) (Figure S8), however
338 diameter did not vary with mean treatment pH, site of origin or level of pH variability (Table S7,
339 Figure S9). The reciprocal transplant experiment showed no effect of generation 2–6 treatment,
340 transplant treatment or any interaction between the two on diameter (Figure S10).

341

342 **Discussion**

343 Here, we demonstrate that coralline algal conceptacle abundance is initially impacted by ocean
344 acidification, but conceptacle size is not. This result indicates that the initial formation of
345 conceptacles, rather than their later development is impaired by altered carbonate chemistry.
346 However, as the first study to assess the impact of ocean acidification on conceptacles across multiple
347 generations, this study also provides evidence that coralline algae may be more robust to the effects of
348 ocean acidification than widely predicted, as from the third generation onwards there is no impact of
349 ocean acidification on conceptacles (Figure 2). Such an observation indicates that coralline algal
350 conceptacles will continue to develop in a more acidic ocean. This is important as conceptacles are

351 vital reproductive structures, without which the production of spores and subsequent production of
352 next-generation recruits would not be possible. Thus, if conceptacles were impacted by future ocean
353 acidification it would have severe consequences for the persistence and proliferation of coralline algal
354 populations. This ability to maintain conceptacle abundances and size under ocean acidification will
355 also be beneficial to other coral reefs organisms that are threatened by environmental change [1,2].
356 For example, continuing spore production and the formation of new coralline algal crusts will provide
357 substrate for future coral larval settlement [8,9] and will also aid in binding calcium carbonate reef
358 structures together, as they are increasingly weakened by ocean acidification [7,28]. Therefore,
359 through such downstream effects the maintenance of coralline algal conceptacles under ocean
360 acidification could be vital to the future persistence of coral reefs.

361

362 We consider it likely that the reduced effect of ocean acidification across generations observed in this
363 study is due to non-genetic acclimation, as coralline algae across all generations possessed features
364 that are indicative of asexual reproduction [46]. However, we cannot definitively state that the
365 changes observed here were not driven by selective genetic adaptation across generations. The
366 transgenerational acclimation of coralline algal conceptacle abundances to ocean acidification
367 observed here follows the trend observed for growth rates [18]. Moreover, the positive relationship
368 between conceptacle abundances and growth rates (Figure S6) across all generations and treatments
369 demonstrates that trade-offs between these two processes are not underlying the multi-generational
370 acclimation of conceptacle abundance and growth to ocean acidification. For example, if such a trade-
371 off was driving this acclimation you would expect to see a negative relationship between growth and
372 conceptacle abundance within the later “acclimated” generations. This observation that multi-
373 generational acclimation is observed for conceptacle abundance and growth [18] further indicates that
374 coralline algae are capable of acclimating to ocean acidification. However, despite the overall
375 similarity in responses, acclimation of growth rates to ocean acidification were stepwise/linear in
376 nature (reported in Cornwall *et al.*, [18]), and therefore not as rapid as the acclimation of
377 conceptacles. Whilst the acclimation of conceptacle formation observed here is deemed rapid
378 compared with growth, similar rapid transgenerational acclimation has been observed in differing

379 marine taxa. For example, the reproductive output of marine invertebrates such as polychaetes [48]
380 and copepods [49] is initially impaired under ocean acidification, however this effect disappears by
381 the third generation, as the reproductive output of populations exposed to ocean acidification does not
382 differ from controls. Additionally, transgenerational acclimation of growth rates to ocean acidification
383 has been observed within three generations of exposure in calcifying organisms such as oysters [50]
384 and clams [24], suggesting various biological processes are capable of rapid acclimation to ocean
385 acidification. However, our results and those of Cornwall *et al.*, [18] suggest that different processes
386 may exhibit contrasting acclimation times.

387

388 The varying rates of acclimation of different biological processes in coralline algae are likely driven
389 by the physiological mechanisms underpinning these processes. Here, we propose two mechanisms
390 that may have driven the faster multi-generational acclimation of conceptacle development, compared
391 with growth [18]. Firstly, linear extension of coralline algae is dictated by both growth of organic
392 tissue and calcification of the cell walls. The latter is impacted by factors such as the control of
393 calcification site chemistry and the production of organic matrices, both of which may be disrupted by
394 ocean acidification [51,52]. Thus, the complexity of calcification and its dependence on specific
395 chemical conditions may represent a physiochemical bottleneck to the rate at which calcification, and
396 therefore growth can acclimatise to ocean acidification. In contrast, the primarily uncalcified [53,54],
397 conceptacles are not subject to the same chemical constraints, potentially facilitating faster
398 acclimation. An alternate and not mutually exclusive explanation is that coralline algae prioritise
399 energetic investment in reproduction over calcification. Energetic trade-offs between reproduction and
400 other processes such as growth commonly occur in nature [55,56], and are enhanced due to increased
401 maintenance costs under stressful conditions [57]. Therefore, rapid acclimation of reproductive
402 conceptacles may come from prioritizing energetic investment in reproduction, as ensuring the
403 production of offspring under stress is of the greatest benefit to Darwinian fitness. Similar multi-
404 generational trends in resource allocation towards reproduction under ocean acidification have also
405 been observed in copepods [58]. Here, our novel comparison of acclimatory responses (growth rates

406 vs conceptacle development), indicates that the persistence of populations under ocean acidification
407 will be constrained by the biological process that undergoes the slowest rate of acclimation.
408

409 The reduction in second generation conceptacle abundances under ocean acidification mirror the
410 widely reported negative impacts of ocean acidification on the calcification, growth and recruitment
411 of first and second generation coralline algae [11,16,59]. This initial reduction in conceptacle
412 abundance observed here, highlights reduced conceptacle formation as one potential mechanism
413 driving these previously reported declines in coralline algae recruitment under ocean acidification
414 [16,17]. However, in contrast to the initial negative effect observed here, Ordoñez *et al.*, [43] found
415 that conceptacle numbers in *Pneophyllum* sp. coralline algae were not reduced when exposed to
416 medium (pH 7.81) or low (pH 7.60) pH treatments. Such a result is unexpected, as these populations
417 were subject to neither transgenerational nor developmental exposure to low pH conditions, as they
418 recruited into experimental conditions from the wild. However, species-specific responses to ocean
419 acidification have been documented in coralline algae [37,60], therefore this opposing result may be
420 due to differences between the thin crusted *Pneophyllum* sp. studied by Ordoñez *et al.*, [43], and the
421 thick-crusted *H. reinboldii* assessed here. Evidence of such species effects are displayed in Ordoñez *et*
422 *al.*, [43] as they observed a significant decline in the relative density of thick crusted *Porolithon*
423 *onkodes* under high pCO₂, but not *Pneophyllum* sp., suggesting that morphology can influence
424 response to ocean acidification. Alternatively, these populations may have had natural resilience to
425 this magnitude of ocean acidification, and further research is required to elucidate which factors could
426 be driving such differences.

427

428 The rapid acclimation of reproductive conceptacles observed here is not translated to rapid increases
429 in final recruit area of the next generation, as recruit area acclimation was slower and stepwise [18].
430 Therefore, although the trend of increasing recruit area with increasing conceptacle abundance (Figure
431 S7) suggests that higher conceptacle abundances may aid in buffering the impacts of ocean
432 acidification on recruitment, it is likely that final recruitment will be determined by the cumulative
433 impact of ocean acidification on reproductive structure formation, spore settlement, and early

434 development processes. For example, coralline algae display delayed and weakened spore attachment
435 [61], as well as reduced spore germination [62] in response to ocean acidification, potentially
436 highlighting the later sporulate stage as a bottleneck in final recruitment.

437

438 Our results also demonstrate that pH variability does not modify the impact of ocean acidification on
439 coralline algal conceptacles. This is shown as coralline algae grown in both high variability and low
440 variability pH treatments were not differently affected by the mean pH corresponding to ocean
441 acidification. This lack of effect of pH variability is at odds with some past literature, where pH
442 variability has been shown to reduce the impact of ocean acidification [33,38], while it can also
443 enhance the negative impacts of ocean acidification [37]. These contrasting and inconsistent effects of
444 pH variability demonstrate that its interaction with future ocean acidification will be complex and can
445 potentially elicit a full range of effects from negative to positive, with the lack of effect observed here
446 in between the gamut of responses. As previous studies have highlighted that the influence of pH
447 variability varies amongst species [20,63], it appears that the effect of ocean acidification on
448 *H.reinboldii* is not dictated by pH variability as the calcification and photosynthesis of mature
449 rhodoliths [40], the growth rates and recruitment of juveniles [18] and the size and abundance of
450 juvenile conceptacles are all unaffected.

451

452 The effect of ocean acidification on coralline algal conceptacles is not influenced by the pH variability
453 experienced by the population in their natural environment here. This was hypothesized *a priori*, as
454 similar responses were observed with adult calcification rates [40] and juvenile growth rate [18].
455 Nevertheless, this result contrasts with the often-posed hypothesis in the literature that environments
456 with naturally high variations in an abiotic factor, will increase the resistance of resident organisms to
457 future mean changes in that abiotic factor [37]. Though this trend has recently been observed in corals
458 from habitats with greater thermal variability [64,65], mean conditions may be as important or more
459 important than variability [66]. For example, mean pH is higher at Tallon Island, while pH variability
460 is greater than Shell Island. Therefore, both could be acting against each other in conditioning
461 coralline algal responses to low pH. Our results suggest a similar priming effect of mean pH, as three

462 generations of exposure to reduced mean pH was enough to eliminate the effects of reduced mean pH
463 on the conceptacles of later generations.

464

465 Overall, this study demonstrates that the abundance of coralline algal conceptacles is greatly reduced
466 in second generation coralline algae reared under simulated ocean acidification conditions. However,
467 in just three generations of exposure to these same ocean acidification conditions the coralline algal
468 reproductive structures can rapidly acclimate, as we observe no effects of ocean acidification beyond
469 the second generation. This acclimation is much faster than that previously reported for growth rates,
470 as growth took six generations to acclimate. Despite differing rates of acclimation, the acclimation of
471 conceptacle development observed here is not facilitated by reduced growth, as both processes
472 acclimate together across all generations. Therefore, this study further supports the possibility that
473 coralline algae could acclimate to the effects of ocean acidification. As multiple studies have reported
474 the ability of marine organisms to acclimate to environmental change, our observation that
475 acclimation rates vary for different biological processes introduces a new consideration for studies
476 assessing how marine organisms will respond to future environmental change.

477

478 **Ethics**

479 All local regulations and permit requirements were followed during this study.

480

481 **Data accessibility**

482 Data is accessible in the Dryad Digital Repository:

483 https://datadryad.org/stash/share/BVDT9xgRvmfMvs-ruppJEYdEBSB81PoKG8c_1UmetTQ

484

485 **Authors Contributions**

486 B.M. wrote the paper. C.E.C and S.C. conceived the research and designed the experiment. C.E.C.,
487 S.C. and B.M. conducted fieldwork. B.M., C.E.C., S.C., F.P. and E.L. ran the experiment. B.M., A.C.,
488 M.B. and A.P. conducted photograph analysis. M.T.M. provided essential laboratory equipment,

489 facilities and analyses. B.M. conducted the statistical analyses. All authors edited the manuscript, or
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491

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498

499 **Conflict of Interest**

500 The authors declare no conflicts of interest.

501

502 **References**

- 503 1. Chan NCS, Connolly SR. 2013 Sensitivity of coral calcification to ocean acidification: A
504 meta-analysis. *Glob. Chang. Biol.* **19**, 282–290. (doi:10.1111/gcb.12011)
- 505 2. Kroeker KJ, Kordas RL, Crim R, Hendriks IE, Ramajo L, Singh GS, Duarte CM, Gattuso JP.
506 2013 Impacts of ocean acidification on marine organisms: Quantifying sensitivities and
507 interaction with warming. *Glob. Chang. Biol.* **19**, 1884–1896. (doi:10.1111/gcb.12179)
- 508 3. Mouillot D *et al.* 2014 Functional over-redundancy and high functional vulnerability in global
509 fish faunas on tropical reefs. *Proc. Natl. Acad. Sci. U.S.A.* **111**, 13757–13762.
510 (doi:10.1073/pnas.1317625111)
- 511 4. Roberts CM *et al.* 2002 Marine biodiversity hotspots and conservation priorities for tropical
512 reefs. *Science*. **295**, 1280–1284. (doi:10.1126/science.1067728)
- 513 5. O’Mahoney J, Simes R, Redhill D, Heaton K, Atkinson C, Hayward E, Nguyen M. 2017 At
514 what price? The economic, social and icon value of the Great Barrier Reef.
- 515 6. Busenberg E, Plummer LN. 1989 Thermodynamics of magnesian calcite solid-solutions at
516 25°C and 1 atm total pressure. *Geochim. Cosmochim. Acta* **53**, 1189–1208.

- 517 7. Ries JB. 2011 Skeletal mineralogy in a high-CO₂ world. *J. Exp. Mar. Bio. Ecol.* **403**, 54–64.
518 (doi:10.1016/j.jembe.2011.04.006)
- 519 8. Price N. 2010 Habitat selection, facilitation, and biotic settlement cues affect distribution and
520 performance of coral recruits in French Polynesia. *Oecologia* **163**, 747–758.
- 521 9. O’Leary JK, Potts DC, Braga JC, McClanahan TR. 2012 Indirect consequences of fishing:
522 reduction of coralline algae suppresses juvenile coral abundance. *Coral Reefs* **31**, 547–559.
- 523 10. Kawamura T. 1998 A review of feeding and growth of postlarval abalone. *J. Shellfish Res.* **17**,
524 615–625.
- 525 11. Anthony KRN, Kline DI, Diaz-Pulido G, Dove S, Hoegh-Guldberg O. 2008 Ocean
526 acidification causes bleaching and productivity loss in coral reef builders. *Proc. Natl. Acad.*
527 *Sci. U.S.A.* **105**, 17442–17446. (doi:10.1073/pnas.0804478105)
- 528 12. Jokiel PL, Rodgers KS, Kuffner IB, Andersson AJ, Cox EF, Mackenzie FT. 2008 Ocean
529 acidification and calcifying reef organisms: A mesocosm investigation. *Coral Reefs* **27**, 473–
530 483. (doi:10.1007/s00338-008-0380-9)
- 531 13. Semesi IS, Kangwe J, Björk M. 2009 Alterations in seawater pH and CO₂ affect calcification
532 and photosynthesis in the tropical coralline alga, *Hydrolithon sp.* (Rhodophyta). *Estuar. Coast.*
533 *Shelf Sci.* **84**, 337–341. (doi:10.1016/j.ecss.2009.03.038)
- 534 14. Fabricius KE, Klübenschedl A, Harrington L, Noonan S, De’Ath G. 2015 In situ changes of
535 tropical crustose coralline algae along carbon dioxide gradients. *Sci. Rep.* **5**, 1–7.
536 (doi:10.1038/srep09537)
- 537 15. Hall-Spencer JM, Rodolfo-Metalpa R, Martin S, Ransome E, Fine M, Turner SM, Rowley SJ,
538 Tedesco D, Buia M-C. 2008 Volcanic carbon dioxide vents show ecosystem effects of ocean
539 acidification. *Nature* **454**, 96–99.
- 540 16. Kuffner IB, Andersson AJ, Jokiel PL, Rodgers KS, MacKenzie FT. 2008 Decreased
541 abundance of crustose coralline algae due to ocean acidification. *Nat. Geosci.* **1**, 114–117.
542 (doi:10.1038/ngeo100)
- 543 17. Crook ED, Kroeker KJ, Potts DC, Rebolledo-Vieyra M, Hernandez-Terrones LM, Paytan A.
544 2016 Recruitment and succession in a tropical benthic community in response to in-situ ocean

- 545 acidification. *PLoS One* **11**, 1–16. (doi:10.1371/journal.pone.0146707)
- 546 18. Cornwall CE, Comeau S, DeCarlo TM, Larcombe E, Moore B, Giltrow K, Puerzer F,
547 D’Alexis Q, McCulloch MT. 2020 A coralline alga gains tolerance to ocean acidification over
548 multiple generations of exposure. *Nat. Clim. Chang.* **10**, 143–146. (doi:10.1038/s41558-019-
549 0681-8)
- 550 19. Fabricius KE, Noonan SHC, Abrego D, Harrington L, De’Ath G. 2017 Low recruitment due to
551 altered settlement substrata as primary constraint for coral communities under ocean
552 acidification. *Proc. R. Soc. B.* **284**. (doi:10.1098/rspb.2017.1536)
- 553 20. McCoy SJ, Kamenos NA. 2015 Coralline algae (Rhodophyta) in a changing world: integrating
554 ecological, physiological, and geochemical responses to global change. *J. Phycol.* **51**, 6–24.
- 555 21. Sordo L, Santos R, Barrote I, Silva J. 2018 High CO₂ decreases the long-term resilience of the
556 free-living coralline algae *Phymatolithon lusitanicum*. *Ecol. Evol.* **8**, 4781–4792.
557 (doi:10.1002/ece3.4020)
- 558 22. Donelson JM, Munday PL, McCormick MI, Pitcher CR. 2012 Rapid transgenerational
559 acclimation of a tropical reef fish to climate change. *Nat. Clim. Chang.* **2**, 30–32.
560 (doi:10.1038/nclimate1323)
- 561 23. Parker LM, Ross PM, O’connor WA, Borysko L, Raftos DA, Pörtner H. 2012 Adult exposure
562 influences offspring response to ocean acidification in oysters. *Glob. Chang. Biol.* **18**, 82–92.
- 563 24. Zhao L, Yang F, Milano S, Han T, Walliser EO, Schöne BR. 2018 Transgenerational
564 acclimation to seawater acidification in the Manila clam *Ruditapes philippinarum*: Preferential
565 uptake of metabolic carbon. *Sci. Total Environ.* **627**, 95–103.
566 (doi:10.1016/j.scitotenv.2018.01.225)
- 567 25. Goncalves P, Anderson K, Thompson EL, Melwani A, Parker LM, Ross PM, Raftos DA. 2016
568 Rapid transcriptional acclimation following transgenerational exposure of oysters to ocean
569 acidification. *Mol. Ecol.* **25**, 4836–4849. (doi:10.1111/mec.13808)
- 570 26. Sunday JM, Calosi P, Dupont S, Munday PL, Stillman JH, Reusch TBH. 2014 Evolution in an
571 acidifying ocean. *Trends Ecol. Evol.* **29**, 117–125.
- 572 27. Lohbeck KT, Riebesell U, Reusch TBH. 2012 Adaptive evolution of a key phytoplankton

- 573 species to ocean acidification. *Nat. Geosci.* **5**, 346–351.
- 574 28. Ragazzola F, Foster LC, Form A, Anderson PSL, Hansteen TH, Fietzke J. 2012 Ocean
575 acidification weakens the structural integrity of coralline algae. *Glob. Chang. Biol.* **18**, 2804–
576 2812. (doi:10.1111/j.1365-2486.2012.02756.x)
- 577 29. McCoy SJ, Ragazzola F. 2014 Skeletal trade-offs in coralline algae in response to ocean
578 acidification. *Nat. Clim. Chang.* **4**, 719–723. (doi:10.1038/nclimate2273)
- 579 30. Kamenos NA, Perna G, Gambi MC, Micheli F, Kroeker KJ. 2016 Coralline algae in a
580 naturally acidified ecosystem persist by maintaining control of skeletal mineralogy and size.
581 *Proc. R. Soc. B.* **283**, 20161159. (doi:10.1098/rspb.2016.1159)
- 582 31. Anthony KRN, A. Kleypas J, Gattuso JP. 2011 Coral reefs modify their seawater carbon
583 chemistry - implications for impacts of ocean acidification. *Glob. Chang. Biol.* **17**, 3655–3666.
584 (doi:10.1111/j.1365-2486.2011.02510.x)
- 585 32. Hofmann GE *et al.* 2011 High-frequency dynamics of ocean pH: A multi-ecosystem
586 comparison. *PLoS One* **6**. (doi:10.1371/journal.pone.0028983)
- 587 33. Wahl M, Schneider Covachã S, Saderne V, Hiebenthal C, Müller JD, Pansch C, Sawall Y.
588 2017 Macroalgae may mitigate ocean acidification effects on mussel calcification by
589 increasing pH and its fluctuations. *Limnol. Oceanogr.* (doi:10.1002/lno.10608)
- 590 34. Schaum CE, Collins S. 2014 Plasticity predicts evolution in a marine alga. *Proc. R. Soc. B*
591 *Biol. Sci.* **281**, 20141486.
- 592 35. Vargas CA, Lagos NA, Lardies MA, Duarte C, Manríquez PH, Aguilera VM, Broitman B,
593 Widdicombe S, Dupont S. 2017 Species-specific responses to ocean acidification should
594 account for local adaptation and adaptive plasticity. *Nat. Ecol. Evol.* **1**, 1–7.
595 (doi:10.1038/s41559-017-0084)
- 596 36. Schunter C, Welch MJ, Ryu T, Zhang H, Berumen ML, Nilsson GE, Munday PL, Ravasi T.
597 2016 Molecular signatures of transgenerational response to ocean acidification in a species of
598 reef fish. *Nat. Clim. Chang.* **6**, 1014–1018. (doi:10.1038/nclimate3087)
- 599 37. Rivest EB, Comeau S, Cornwall CE. 2017 The role of natural variability in shaping the
600 response of coral reef organisms to climate change. *Curr. Clim. Chang. Reports* **3**, 271–281.

- 601 38. Comeau S, Edmunds PJ, Spindel NB, Carpenter RC. 2014 Diel pCO₂ oscillations modulate the
602 response of the coral *Acropora hyacinthus* to ocean acidification. *Mar. Ecol. Prog. Ser.* **501**,
603 99–111. (doi:10.3354/meps10690)
- 604 39. Johnson MD, Moriarty VW, Carpenter RC. 2014 Acclimatization of the crustose coralline alga
605 Porolithon onkodes to variable pCO₂. *PLoS One* **9**. (doi:10.1371/journal.pone.0087678)
- 606 40. Cornwall CE, Comeau S, DeCarlo TM, Moore B, D’Alexis Q, McCulloch MT. 2018
607 Resistance of corals and coralline algae to ocean acidification: physiological control of
608 calcification under natural pH variability. *Proc. R. Soc. B.* **285**, 20181168.
609 (doi:10.1098/rspb.2018.1168)
- 610 41. Townsend RA, Huisman JM. 2018 *Algae of Australia: Marine Benthic Algae of North-*
611 *Western Australia.*
- 612 42. Johansen HW. 1981 *Coralline algae: a first synthesis.* CRC press.
- 613 43. Ordoñez A, Doropoulos C, Diaz-Pulido G. 2014 Effects of ocean acidification on population
614 dynamics and community structure of crustose coralline algae. *Biol. Bull.* **226**, 255–268.
615 (doi:10.1086/BBLv226n3p255)
- 616 44. Riebesell U, Fabry VJ, Hansson L, Gattuso J-P. 2011 *Guide to best practices for ocean*
617 *acidification research and data reporting.* Office for Official Publications of the European
618 Communities.
- 619 45. IPCC IPOCC. 2019 Special report on global warming of 1.5°C (SR15).
- 620 46. Harvey A. 2005 Coralline algae of central New Zealand: an identification guide to
621 common ‘crustose’ species. *NIWA Inf. Ser.* **57**, 1–145.
- 622 47. Torda G *et al.* 2017 Rapid adaptive responses to climate change in corals. *Nat. Clim. Chang.* **7**,
623 627–636.
- 624 48. Rodríguez-Romero A, Jarrold MD, Massamba-N’Siala G, Spicer JI, Calosi P. 2016 Multi-
625 generational responses of a marine polychaete to a rapid change in seawater pCO₂. *Evol. Appl.*
626 **9**, 1082–1095.
- 627 49. Thor P, Dupont S. 2015 Transgenerational effects alleviate severe fecundity loss during ocean
628 acidification in a ubiquitous planktonic copepod. *Glob. Chang. Biol.* **21**, 2261–2271.

- 629 50. Parker LM, O'Connor WA, Raftos DA, Pörtner HO, Ross PM. 2015 Persistence of positive
630 carryover effects in the oyster, *Saccostrea glomerata*, following transgenerational exposure to
631 ocean acidification. *PLoS One* **10**, 1–19. (doi:10.1371/journal.pone.0132276)
- 632 51. Cornwall CE, Comeau S, McCulloch MT. 2017 Coralline algae elevate pH at the site of
633 calcification under ocean acidification. *Glob. Chang. Biol.* **23**, 4245–4256.
634 (doi:10.1111/gcb.13673)
- 635 52. Allison N, Cole C, Hintz C, Hintz K, Rae J, Finch A. 2018 The effect of ocean acidification on
636 tropical coral calcification: Insights from calcification fluid DIC chemistry. *Chem. Geol.* **497**,
637 162–169. (doi:10.1016/j.chemgeo.2018.09.004)
- 638 53. Mu X, Riding R. 1999 Skeletal ultrastructure of the calcified red alga *Galaxaura oblongata*,
639 Hainan Island, China. *Rev. Palaeobot. Palynol.* **104**, 205–212.
- 640 54. Adey WH, Halfar J, Williams B. 2013 The coralline genus *Clathromorphum* foslie emend.
641 adey: biological, physiological, and ecological factors controlling carbonate production in an
642 arctic-subarctic climate archive.
- 643 55. Zera AJ, Harshman LG. 2001 The Physiology of Life History Trade-Offs in Animals. *Annu.*
644 *Rev. Ecol. Syst.* **32**, 95–126. (doi:10.1146/annurev.ecolsys.32.081501.114006)
- 645 56. Harshman LG, Zera AJ. 2007 The cost of reproduction: the devil in the details. *Trends Ecol.*
646 *Evol.* **22**, 80–86. (doi:10.1016/j.tree.2006.10.008)
- 647 57. Jager T, Ravagnan E, Dupont S. 2016 Near-future ocean acidification impacts maintenance
648 costs in sea-urchin larvae: identification of stress factors and tipping points using a DEB
649 modelling approach. *J. Exp. Mar. Bio. Ecol.* **474**, 11–17.
- 650 58. Fitzer SC, Caldwell GS, Close AJ, Clare AS, Upstill-Goddard RC, Bentley MG. 2012 Ocean
651 acidification induces multi-generational decline in copepod naupliar production with possible
652 conflict for reproductive resource allocation. *J. Exp. Mar. Bio. Ecol.* **418–419**, 30–36.
653 (doi:10.1016/j.jembe.2012.03.009)
- 654 59. Johnson MD, Carpenter RC. 2012 Ocean acidification and warming decrease calcification in
655 the crustose coralline alga *Hydrolithon onkodes* and increase susceptibility to grazing. *J. Exp.*
656 *Mar. Bio. Ecol.* **434–435**, 94–101. (doi:10.1016/j.jembe.2012.08.005)

- 657 60. Comeau S, Carpenter RC, Nojiri Y, Putnam HM, Sakai K, Edmunds PJ. 2014 Pacific-wide
658 contrast highlights resistance of reef calcifiers to ocean acidification. *Proc. R. Soc. B.* **281**,
659 20141339.
- 660 61. Guenther R, Miklasz K, Carrington E, Martone PT. 2017 Macroalgal spore dysfunction: ocean
661 acidification delays and weakens adhesion. *J. Phycol.* **54**, 153–158. (doi:10.1111/jpy.12614)
- 662 62. Ordoñez A, Kennedy E V., Diaz-Pulido G. 2017 Reduced spore germination explains
663 sensitivity of reef-building algae to climate change stressors. *PLoS One* **12**, 1–19.
664 (doi:10.1371/journal.pone.0189122)
- 665 63. Noisette F, Egilisdottir H, Davoult D, Martin S. 2013 Physiological responses of three
666 temperate coralline algae from contrasting habitats to near-future ocean acidification. *J. Exp.*
667 *Mar. Bio. Ecol.* **448**, 179–187.
- 668 64. Oliver TA, Palumbi SR. 2011 Do fluctuating temperature environments elevate coral thermal
669 tolerance? *Coral reefs* **30**, 429–440.
- 670 65. Schoepf V, Stat M, Falter JL, Mcculloch MT. 2015 Limits to the thermal tolerance of corals
671 adapted to a highly fluctuating , naturally extreme temperature environment. *Sci. Rep.* 1–14.
672 (doi:10.1038/srep17639)
- 673 66. Rivest EB, Chen C-S, Fan T-Y, Li H-H, Hofmann GE. 2017 Lipid consumption in coral larvae
674 differs among sites: a consideration of environmental history in a global ocean change
675 scenario. *Proc. R. Soc. B.* **284**, 20162825.
- 676