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Interactive effects of ocean acidification and rising sea temperatures alter predation rate and predator selectivity in reef fish communities.

Short title: ocean acidification interacts with global warming

Maud C.O. Ferrari¹, Philip L. Munday², Jodie L. Rummer², Mark I. McCormick², Katherine Corkill², Sue-Ann Watson², Bridie J.M. Allan², Mark G. Meekan³, Douglas P. Chivers⁴

¹Department of Biomedical Sciences, WCVN, University of Saskatchewan, Saskatoon, SK S7N 5B4 Canada (maud.ferrari@usask.ca)

²ARC Centre of Excellence for Coral Reef Studies, and School of Marine and Tropical Biology, James Cook University, Townsville, QLD 4811 Australia (philip.munday@jcu.edu.au, jodie.rummer@jcu.edu.au, mark.mccormick@jcu.edu.au, katherine.corkill@my.jcu.edu.au, sueann.watson@jcu.edu.au, bridie.allan@my.jcu.edu.au)

³Australian Institute of Marine Science, UWA Ocean Sciences Centre (MO96), Crawley, WA, 6009 Australia (m.meekan@aims.gov.au)

⁴Department of Biology, University of Saskatchewan, SK S7N 5E2 Canada (doug.chivers@usask.ca)

*Corresponding author: Maud C.O. Ferrari, Department of Biomedical Sciences, WCVN, 52 Campus Drive, Saskatoon, SK S7N 5B4 Canada. Phone: 306-966-4317, Fax: 306-966-7376 Email: maud.ferrari@usask.ca

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ABSTRACT

Ocean warming and acidification are serious threats to marine life. While each stressor alone has been studied in detail, their combined effects on the outcome of ecological interactions are poorly understood. We measured predation rates and predator selectivity of two closely-related species of damselfish exposed to a predatory dottyback. We found temperature and CO₂ interacted synergistically on overall predation rate, but antagonistically on predator selectivity. Notably, elevated CO₂ or temperature alone reversed predator selectivity, but the interaction between the two stressors cancelled selectivity. Routine metabolic rates of the two prey showed strong species differences in tolerance to CO₂ and not temperature, but these differences did not correlate with recorded mortality. This highlights the difficulty of linking species-level physiological tolerance to resulting ecological outcomes. This study is the first to document both synergistic and antagonistic effects of elevated CO₂ and temperature on a crucial ecological process like predator-prey dynamics.

INTRODUCTION

Ocean warming and acidification have the potential to dramatically change the structure and function of marine ecosystems (Doney *et al.*, 2012, Harley *et al.*, 2006, Hoegh-Guldberg & Bruno, 2010). Many studies have tested the effects of either elevated temperature or higher CO₂ concentrations on individual performance, measuring key physiological or life-history traits such as metabolism, development and growth, calcification, behaviour and reproduction (Briffa *et al.*, 2012, Doney *et al.*, 2009, Kroeker *et al.*, 2010). Some of these studies show a surprisingly large variation in the effects of those stressors among closely-related species (Ferrari *et al.*, 2011a, Johansen & Jones, 2011). Fewer studies, however, have examined the consequences of such environmental change on the resulting ecological interactions among species (Connell *et al.*, 2013, Diaz-Pulido *et al.*, 2011, Ferrari *et al.*, 2011b, McCormick *et al.*, 2013). While the effects of projected future temperatures and CO₂ concentrations have been studied individually for many years, both stressors will act simultaneously in the future. Consequently, it is necessary to consider how they interact to reliably predict their impacts on marine ecosystems (Kroeker *et al.*, 2013, Wernberg *et al.*, 2012). However, rare are the studies that focused on ecological endpoints. One study has looked at the combined effects of warming and acidification on the foraging efficacy of a predator: Landes and Zimmer (2012) investigated the combined effects of acidification and warming on the handling time of a green crab predator (*Carcinus maenas*) feeding on periwinkles (*Littorina littorea*) but failed to find any interactions between the two stressors. More studies on ecological processes, such as competition and predation, are needed to understand how species-level sensitivity to warming and

acidification will manifest themselves in the ecosystem. Not all sensitivities at the species-level may be ecologically meaningful, and inversely, small species-level alterations in some conditions may be exacerbated by the addition of other stressors. More integration between species-level and ecological-level effects are needed, and this integration needs to focus on multiple stressors (Russell *et al.*, 2012).

The first goal of the present study was to investigate the interactive effects of increased CO₂ and temperature on the outcome of predator-prey encounters, as predation is a key process shaping communities. We used a simplified community of two common damselfish species, *Pomacentrus amboinensis* and *P. nagasakiensis*, and one mesopredator, the dottedback *Pseudochromis fuscus*. Fishes were exposed to ambient (440 µatm) or elevated (995 µatm) CO₂ conditions crossed with ambient or elevated temperature (+3°C), in a full factorial design, to examine both the individual and combined effects of the stressors on trophic interactions. Elevated CO₂ and temperature were chosen to match the end-of-century projections for the tropical oceans based on the RCP8.5 emission scenario (Stocker *et al.*, 2013).

Ectotherms, whose performances are tightly linked to environmental temperature, will be particularly sensitive to ocean warming (Gilbert *et al.*, 2014). Tropical species are known to live near their thermal optimum, and as temperature rises, their capacity for oxygen transport or aerobic capacity is reduced, affecting behaviour, growth and swimming abilities (Johansen & Jones, 2011, Portner & Farrell, 2008). Increased CO₂ levels have also been shown to provide physiological constraints on these same traits (Briffa *et al.*, 2012, Kroeker *et al.*, 2010). These constraints at the species level are thought to be somewhat translatable into changes at the ecological level (Portner & Farrell, 2008). Hence, our second goal was to identify the possible prey-related physiological mechanisms responsible for the changes we observed. We did this by investigating any alterations in routine metabolic rates of prey under the four combinations of temperature and CO₂. Metabolic costs could either increase or decrease in the presence of the stressors, and the energy surplus or deficiency would then indicate the individual's capacity to exhibit costly but ecologically-relevant traits such as activity level, habitat choice, aggression, or predator avoidance (Portner & Farrell, 2008). Increased temperature should increase foraging demands as fish have to bridge the energetic gap caused by increased routine metabolic rate (Gilbert *et al.*, 2014). In turn, this may lead to increased risk-taking behaviour to have access to more or higher value food items. Recent studies

showed that elevated CO₂ conditions resulted in altered risk assessment and antipredator behaviour via neurotransmitter interference (Chivers *et al.*, 2014, Nilsson *et al.*, 2012) but also affect metabolic scopes, although the direction of this change is variable among species (Munday *et al.*, 2009, Rummer *et al.*, 2013). Hence, predictions on the interactive effects of the two stressors are difficult to make. In addition, the differential effects of the stressors on the two species may matter more than the absolute level of handicap they provide.

We focused on coral reef communities for three reasons. First, coral reef ecosystems may be among the most threatened by changes in temperature, via habitat loss (Hoegh-Guldberg & Bruno, 2010). Second, some reef fishes are living close to their thermal optimum and may be particularly vulnerable to relatively small increases in temperature (Nilsson *et al.*, 2009). Damselfish species have become a model organism for the study of temperature and CO₂ effects on fish and lots of information is already known on their response to both CO₂ and temperature for a number of endpoints, including physiology, growth, behaviour, cognitive function, and even *in situ* survival. Finally, our juvenile prey are tested just at their transition from the open ocean to coral reef habitat, a time when the population is subject to a severe predation-induced bottleneck; this transition is likely to be the period when most of the CO₂-tolerance phenotypic selection will occur (Munday *et al.*, 2010a). Our two prey species are known to exhibit different sensitivities to elevated CO₂ conditions (Ferrari *et al.*, 2011a), but share the same ecological niche, compete for the same food items, and share the same mesopredators, including *Pseudochromis fuscus*. The predator, *P. fuscus* has altered food preference (Cripps *et al.*, 2011) and decreased strike success (Allan *et al.*, 2013) in high CO₂ conditions. Elevated CO₂ levels can alter trophic interactions in a coral fish system (Ferrari *et al.*, 2011b), but it is unknown how the addition of elevated temperature might influence these dynamics.

MATERIALS AND METHODS

Fish maintenance and CO₂ and temperature treatments

Wild-caught pre-settlement juvenile *P. amboinensis* and *P. nagasakiensis* (16-21 days old) were maintained in the laboratory (<2 weeks) until needed, and were then transferred into 35-L aquaria and assigned to one treatment combination of the following 2x2 design: fish were exposed to control or elevated CO₂ (440 vs 995 µatm) crossed with control or elevated temperature (28 vs 31°C). Control CO₂ and temperature matched ambient summer conditions at Lizard Island, where the study was conducted (see online supplement for details on location and fish capture). The fish were

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fed freshly-hatched *Artemia* nauplii three times a day. Wild-caught adult *P. fuscus* were kept individually in mesh baskets placed in flow-through tanks and fed daily with squid pieces and fish pellets. They underwent the same CO₂ and temperature treatment protocols as the damselfishes, but were kept separated from the prey.

The treatment consisted of 2 phases: a 6-day temperature acclimation period and a 4-day CO₂ treatment period. The first phase consisted of slowly acclimating the fish to elevated temperature, by increasing the temperature at a maximum rate of 0.5°C per day, while the controls were maintained in identical conditions at ambient temperature. The fish were then transferred into their respective treatment containers, which were either maintained at ambient or elevated temperature and ambient or elevated CO₂ levels for 4 days. Previous experiments demonstrated that the behavioural effects of elevated CO₂ are manifest within 4 days of exposure to relevant CO₂ treatments, but longer durations of exposure do not further alter behavioural responses (Munday *et al.*, 2014). Descriptions of the CO₂ treatments can be found in the online supplement. Water chemistry for different treatments is summarized in Table 1.

As in previous CO₂ studies limited to short-term exposures, our exposure to CO₂ was rapid. However, damselfish larvae exposed to elevated CO₂ over a few days showed identical behavioural impairment as larvae raised under the same CO₂ levels from hatching (Munday *et al.*, 2010b) or those occurring at natural CO₂ seeps (Munday *et al.*, 2014), indicating that alterations in behaviour were not due to a sudden CO₂ exposure. In addition, our larval fish naturally experience a change in CO₂ environments as they transition from the open ocean to coral reef habitats and experience natural fluctuating CO₂ concentrations on a daily basis (Shaw *et al.*, 2013).

Mesocosm experiment

Mesocosm setup: All mesocosms were placed outdoors under a shade cloth, so the fish would experience natural light cycles. Each mesocosm consisted of an insulated 368-L circular food-grade pool (111 cm diameter, 45 cm high) containing a 1-cm deep sand substrate, air-stone, and a high-precision water heater. The heater could potentially provide shelter from predator attacks so every pool was equipped with one, although only the heaters in pools at elevated temperature were functioning. Average treatment temperatures were: ambient temperature: 28.3°C (range: 25.7-30.1

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$\pm 1.0^{\circ}\text{C}$ – SD); elevated temperature: 31.1°C (range: $29.4\text{--}32.3 \pm 1.0^{\circ}\text{C}$), based on 4 daily measurements of each replicate mesocosm (see Supp Figure). Two pieces of clean dead bushy hard coral (*Pocillopora damicornis*) were placed beside each other in the middle of each pool, forming a coral patch of ~ 90 cm in circumference and ~ 20 cm in height. Live corals provide a better quality habitat, but dead coral were used to avoid coral stress and bleaching in high temperature pools. To allow a better control of temperature, there was no inflow of water in the pools during the trials.

Experimental setup: Six prey (3 per species) of matching treatment were randomly chosen and placed in a mesocosm of matching temperature and left to acclimate to their new environment. One hour later, a predator of matching CO_2 and temperature treatment was introduced, which marked the start of the trial. After 22 h, all the fish were removed from the pool and we recorded the number and species of damselfishes that survived the predation trial. Each day, two replicates from each of the 4 treatments were performed and we switched the treatment associated with each pool the next day to avoid position bias. Seawater was maintained at control CO_2 levels due to the difficulty of dosing mesocosms with CO_2 and because previous experiments have shown the effects of CO_2 levels similar to those used here last for at least 3 days and that fish exposed to high CO_2 exhibit the same behavioural responses when tested in either control or elevated CO_2 conditions (Munday *et al.*, 2010b). Each day, the coral patches from all the pools were mixed and rebuilt randomly to avoid patch structure biases in predation success/failure. The fish were fed twice daily (1100 and 1700 h) with 60 mL of a solution of freshly hatched *Artemia sp* (~ 250 per mL). We performed 17-18 replicates in each of the 4 treatment groups. Each animal was only used once in the experiment. The mean (\pm SD) standard length of *P. amboinensis*, *P. nagasakiensis* and *P. fuscus* were $1.26 (\pm 0.08)$, $1.40 (\pm 0.07)$ and $6.75 (\pm 0.73)$ cm respectively.

Respirometry

Intermittent-flow respirometry was used to determine routine O_2 consumption rates ($\dot{M}\text{O}_{2\text{Routine}}$), as it has been found a reliable estimate of resting and routine metabolic rates in fish (Roche *et al.*, 2013). The O_2 consumption rates were measured for a total of 32 individual fish for each of the two damselfish species, which included 8 replicates per species in each of the 4 treatments. Prior to the start of each trial, all fish had been fasted for 24 hours (McLeod *et al.*, 2013, Rummer *et al.*, 2013). The methodology for the respirometry technique can be found in the online supplement. Unfortunately, our system was not adequate for larger species like our predators.

Statistical analyses

Fish size: Our two prey species differ naturally in their size. However, differences in size variation within species or variation in size between species among treatment groups could influence the results. Thus, we used 2-way ANOVAs to test if the size of each species, or their difference, differed among CO₂ and temperature groups. We also tested to ensure the size of the predators did not differ among our treatment groups.

Predation rate: For each trial, we computed a predation rate (#fish eaten/6). These were used as raw data in a 2-way ANOVA, testing the effect of CO₂ (control vs. elevated) and temperature (control vs. elevated) on predation rate.

Prey selectivity: We computed a prey selectivity index for *P. fuscus* following Chesson (1983):

$$s_i = \frac{r_i/n_i}{\sum_{j=1}^m (r_j/n_j)}, i = 1, \dots, m$$

Where n_i represents the number of prey type i at the beginning of the experiment, r_i the number of prey type i consumed by the predator, and j the number of different prey types. This selectivity can be interpreted as the preference of the predator for a prey type relative to the average preference for alternative prey types. The selectivity value ranges from 0 (total avoidance of prey type) to 1 (only prey type selected). If both prey species are selected equally by the predator, the selectivity for each prey species is 0.5. Trials where predators ate none of the prey ($N = 7$ across all treatments without treatment bias) were removed, given that no selectivity could be computed, leaving us with $n=15-16$ /treatment for this response variable. Using the two indexes as response variables, we performed a 3-way RM MANOVA to test the effect of CO₂, temperature and species on the selectivity value for each trial. The repeated-measures approach accounted for the dependency of the selectivity among species (the score for the two species are related), while still allowing us to compare selectivity among species (Ferrari *et al.* 2011b).

Respirometry: We used a 3-way ANOVA, followed by subsequent 2-way ANOVAs to determine the effect of CO₂, temperature, and species on the metabolic rate of the fish. The data for all tests described above met parametric assumptions.

RESULTS

Fish size: The size of the fish did not differ among treatment groups, for either *P. nagasakiensis*: (CO₂: F_{1,114}=0.6, P=0.4, temperature: F_{1,114}=0.1, P=0.9, CO₂*temp: F_{1,114}=0.2, P=0.6) or *P. amboinensis* (CO₂: F_{1,141}=0.1, P=0.9, temperature: F_{1,141}=0.6, P=0.4, CO₂*temp: F_{1,114}=0.1, P=0.9). Further, the size difference between the two species did not vary among treatment groups (CO₂: F_{1,102}=2.2, P=0.12, temperature: F_{1,102}=0.2, P=0.6, CO₂*temp: F_{1,114}=2.6, P=0.11). In addition, the size of the predator did not vary among treatment groups (CO₂: F_{1,65}=2.7, P=0.1, temperature: F_{1,65}=0.1, P=0.8, CO₂*temp: F_{1,65}=1.0, P=0.3).

Predation rate: The 2-way ANOVA revealed a significant synergistic interaction between CO₂ and temperature on overall predation rate (F_{1,65}=4.9, P=0.03, figure 1). Elevated temperature alone (F_{1,33}= 0.1, P=0.69) or CO₂ alone (F_{1,33}= 0.1, P=0.99) did not affect the overall predation rate compared to controls, but the rate increased from 30% to 70% when both stressors were present (F_{1,33}= 10.5, P=0.003)

Selectivity: The 3-way RM ANOVA revealed a significant antagonistic interaction of CO₂ and temperature on predator selectivity (species*CO₂*temperature: Pillai's trace: F_{1,58} = 12.9, P=0.001, figure 2). An increase in temperature alone reversed the selectivity patterns of the two species (Pillai's trace: species*temp: F_{1,28}=8.3, P=0.007), and so did an increase in CO₂ alone (Pillai's trace: species*temp: F_{1,29}=11.5, P=0.002). However, when both stressors were present, the selectivity pattern disappeared and both species were selected equally (F_{1,15}=0.1, P=0.99).

Respirometry: The 3-way ANOVA revealed a significant interaction among CO₂, temperature and species (F_{1,56}=5.2, P=0.026). We performed 2-way ANOVA on each species, and found that while the metabolic rate of *P. amboinensis* increased as a response to increased temperature only (temp: F_{1,28}=4.7, P=0.04; CO₂: F_{1,28}=0.001, P=0.99, temp*CO₂: F_{1,28}=0.27, P=0.61), the metabolic rate of *P. nagasakiensis* was affected by an interaction between temperature and CO₂ (F_{1,28}=9.0, P=0.006). The

metabolic rate did not increase with increased CO₂ or temperature alone (P=0.5 and 0.7 respectively), but increased when both CO₂ and temperature were elevated (P=0.003).

DISCUSSION

For the first time, we have been able to show a clear interactive effect of two climate change-related stressors on predator-prey interactions in a coral reef ecosystem, with a synergistic interaction on predation rate and an antagonistic interaction on selectivity. As an individual stressor, we found that elevated CO₂ concentrations completely reversed predator selectivity for *P. amboinensis* and *P. nagasakiensis* compared to ambient conditions, but did not cause a change in overall predation rate, results that are consistent with our previous work examining the effects of elevated CO₂ on trophic dynamics (Ferrari *et al.*, 2011b). Interestingly, an increase in water temperature had similar effects. However, when both stressors were present, a new pattern emerged. Together, these stressors caused total predation rate to increase dramatically from 30 to 70% and for any selectivity to disappear, with roughly equal numbers of each prey consumed by the predator. Our results also demonstrate that conclusions reached about the potential impacts of ocean acidification or warming may be inaccurate if these co-occurring stressors are not investigated together. Our results differ from those of Landes and Zimmer (2012) who failed to find an interaction between CO₂ and temperature on the foraging behaviour of crabs on periwinkles. However, their experiment did not allow to investigate the dynamics between predators and prey, given that the predator was given only one semi-sessile prey from which to choose. While the success of a predators on a prey may not be impacted, community-level effects may still appear in such systems if predators switch to easier alternate prey that are more affected by each or both of the stressors. Increasing the realism of such experiments will uncover patterns, such as switch in predator selectivity, that cannot be picked up by simple predator-prey systems.

In order to understand the ecological impact of a changing climate, studies designed to address the combined effects of increasing CO₂ and temperature are particularly valuable. So far, studies with reef fishes that have tested both stressors together have been limited to individual-level outcomes. Some demonstrated additive effects of the two stressors on some endpoints like metabolic rate (Munday *et al.*, 2009), while others showed interactive effects of the two stressors on behavioural lateralization (Domenici *et al.*, 2014) and foraging (Nowicki *et al.*, 2012). Our results provide the first ecological-level insights into the complex interactive effects of increased temperature and CO₂ on

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predator-prey dynamics in fishes. When both stressors were present, we saw that the overall rate of predation increased dramatically. We have considerable evidence from field observations that fish exposed to elevated CO₂ maintain a greater distance from shelter and are bolder (Ferrari *et al.*, 2011a, Munday *et al.*, 2010b), taking greater risks to acquire food. If an increase in temperature were to shift the foraging/risk trade-off even further towards feeding, then we should expect much higher mortality with the combined stressors, consistent with our results. However, we cannot partition the role played by predator from that of prey. Ferrari *et al.* (2011a) documented that there is considerable variation in the tolerances of fish within the genus *Pomacentrus* to elevated CO₂, with *P. amboinensis* being more affected than *P. nagasakiensis*. However, our results suggest that such ranked sensitivities might not hold when additional stressors are added, and interspecific variation need to be re-evaluated in conditions of elevated temperature.

Our most striking finding is the effects on predator selectivity. Elevated CO₂ alone completely reversed predator selectivity, as did temperature alone. However, when elevated CO₂ and temperature were combined, the pattern of selectivity totally disappeared. This result highlights that predicting the outcome of predator-prey encounters under a changing climate will be challenging. In order to gain a full appreciation of how dynamics may change, it will be imperative that we consider both the relevant interacting species and the relevant co-occurring environmental stressors. There were clear effects of elevated CO₂ and temperature on resting metabolic rates of the two species; however the species-level responses did not match the change in mortality of prey or the prey selectivity of the predator. The condition under which the greatest discrepancy in basal metabolic rate was found was for elevated temperature and ambient CO₂ levels, with *P. amboinensis* displaying a 15-20% increase in O₂ consumption compared to ambient conditions, while *P. nagasakiensis* still maintained the same metabolic rate as ambient conditions. Under these conditions, one could predict *P. amboinensis* to suffer higher predation rate, but this pattern did not emerge, indicating that other unmeasured factors are likely playing a more important role in dictating trophic outcomes. This demonstrates that it will be difficult to predict the outcome of ecological interactions, and thus the impacts of acidification and warming at the community and ecosystem level, simply based on species-level differences in physiological tolerances to these stressors.

A greater proportion of the predation was directed towards *P. nagasakiensis* when under high CO₂ than under ambient CO₂. The reason for this is unclear, however, a recent study by McCormick et al. (2013) revealed that elevated CO₂ reverses the outcome of competition for space in juvenile damselfish, *P. amboinensis* and *P. moluccensis*. This outcome was mediated by changes in aggression levels, affecting the distribution and vulnerability of each species to predation risk. Our results could fit this hypothesis, given that our species also compete for habitat. An alternative hypothesis is that difference of these stressors on energy balance between the two species could alter their susceptibility to predation. Our results indicate that increased CO₂ conditions did not lead to a significant difference in routine metabolic rate, but we did not measure maximum metabolic rate. Two pomacentrids have been found to exhibit a higher maximum metabolic rate under elevated CO₂ (Couturier et al., 2013, Rummer et al., 2013) while others exhibited a lower one under similar situations (Couturier et al., 2013, Munday et al., 2009). Fish that are more "active" or bold under elevated CO₂ could be easier targets for predators (Hamilton et al., 2014). In contrast, fish exhibiting an enhanced maximum metabolic rate could escape a predator more efficiently, but data are not yet available to support this hypothesis.

Somewhat surprisingly, we did not find that an increase in temperature alone led to an increase in predation rate. Higher temperatures, up to a point, should be associated with higher metabolic rates – as found here for *P. amboinensis* – and hunger level for both predators and prey. From the prey's perspective, an increase in hunger level should promote greater risk taking, meaning that the fish should maintain a greater distance from the coral where food is more abundant and this should lead to higher predation rate (McCormick & Weaver, 2012). It is possible that this mortality effect was not manifested in our study because of the relatively short duration of the test (22 hours). An increase in temperature for a greater duration of time may be required to change the foraging/risk trade-off such that the fish move away from the coral and increase feeding (Lima & Bednekoff, 1999), and it may be that the prey's relatively high body condition may have buffered this trade-off in the short term. Perhaps most interesting was that an increase in temperature alone changed prey selectivity. Again *P. nagasakiensis* was more susceptible to predation under high temperature than under low temperature. This difference could be the result of differential foraging/risk trade-offs between the two species, changing competition for space or a difference in aerobic capacity.

The occurrence of multiple stressors may not simply intensify the patterns emerging from each stressor alone, but may, in some cases, change the patterns observed altogether. Carefully controlled studies integrating physiology, behaviour and trophic ecology hold promise to understand how a changing climate will influence trophic relationships. Moreover, these endpoints need to be studied and contrasted within the same studies, if we are to understand the potential integration between species-level sensitivities and the resulting ecological alterations.

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AUTHORS CONTRIBUTIONS: MF, DC, MIM, PM conceived the project; MF, DC, JLR, and KC collected the data, MF and JLR analysed the data; PM and SAW provided water chemistry parameters and technical assistance with the CO₂ system, MGM and BA provided assistance with the implementation of the experiment. MF and DC wrote the initial draft of the manuscript; all authors contributed to the final version.

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Table 1: Mean (\pm SD) seawater parameters in the experimental system. Temperature, salinity pH, and total alkalinity were measured directly. pCO₂ was estimated from these parameters using CO2SYS

| Treatment | Temperature (°C) | Salinity (ppt) | pH _{NBS} | Total alkalinity ($\mu\text{mol.kg}^{-1}$ SW) | pCO ₂ (μatm) |
|---|-------------------|----------------|---------------------|--|--------------------------------------|
| Control CO ₂ Control temp | 27.8 (\pm 0.1) | 35.2 | 8.15 (\pm 0.01) | 2272.9 (\pm 13.8) | 436.2 (\pm 13.6) |
| Control CO ₂ Elevated temp | 31.1 (\pm 0.1) | 35.2 | 8.15 (\pm 0.01) | 2272.9 (\pm 13.8) | 441.9 (\pm 13.9) |
| Elevated CO ₂ Control temp | 27.8 (\pm 0.1) | 35.2 | 7.85 (\pm 0.004) | 2265.2 (\pm 5.2) | 984.7 (\pm 10.6) |
| Elevated CO ₂ Elevated temp | 31.0 (\pm 0.1) | 35.2 | 7.85 (\pm 0.004) | 2265.2 (\pm 5.2) | 1006.7 (\pm 10.6) |

Figure 1: Mean (\pm SE) predation rate (proportion of prey consumed in 22h) when prey and predator were maintained under ambient (440 μ atm) or elevated (995 μ atm) CO₂, crossed with either ambient temperature (28°C light bars) or elevated temperature (31°C, solid bars).

Figure 2: Mean (\pm SE) predator selectivity for *P. amboinensis* (light bars) and *P. nagasakiensis* (dark bars) when prey and predator were maintained under ambient or elevated CO₂ (440 vs 995 μ atm), and under ambient temperature (28°C - top panel) or elevated temperature (31°C - bottom panel). A selectivity of 0.5 indicates no preference for a particular prey species by the predator.

Figure 3: Mean (\pm SE) routine metabolic rates (estimated from oxygen consumption rates) for *P. amboinensis* (light bar) and *P. nagasakiensis* (dark bars) maintained under ambient or elevated temperature (28 vs 31°C) and ambient and/or elevated CO₂ (440 vs 995 μ atm).





