Effects of high temperature and CO_2 on intracellular DMSP in the cold-water coral *Lophelia pertusa*

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Running title: Effect of OA on DMSP in *Lophelia*

Abstract

Significant warming and acidification of the oceans is projected to occur by the end of the century. CO_2 vents, areas of upwelling and downwelling, and potential leaks from carbon capture and storage facilities may also cause localised environmental changes, enhancing or depressing the effect of global climate change. Cold-water coral ecosystems are threatened by future changes in carbonate chemistry, yet our knowledge of the response of these corals to high temperature and high CO_2 conditions is limited. Dimethylsulphoniopropionate (DMSP), and its breakdown product dimethylsulphide (DMS), are putative antioxidants that may be accumulated by invertebrates via their food or symbionts, although recent research suggests that some invertebrates may also be able to synthesise DMSP. This study provides the first information on the impact of high temperature (12°C) and high CO_2 (817 ppm) on intracellular DMSP in the cold water coral *Lophelia pertusa* from the Mingulay Reef Complex, Scotland (56°49'N, 07°23'W), where *in situ* environmental conditions are meditated by tidally-induced downwellings. An increase in intracellular DMSP under high CO_2 conditions was observed, whilst water column particulate DMS+DMSP was reduced. In both high temperature treatments, intracellular DMSP was similar to the control treatment, whilst dissolved DMSP+DMS was not significantly different between any of the treatments. These results suggest that *L. pertusa* accumulates DMSP from the surrounding water column; uptake may be up-regulated under high CO_2 conditions, but mediated by high temperature. These results provide new insight into the biotic control of deep-sea biogeochemistry and may impact our understanding of the global sulphur cycle, and the survival of cold water corals under projected global change.

Keywords: climate change; cold-water coral; dimethylsulphide (DMS); dimethylsulphoniopropionate (DMSP); global warming; ocean acidification

Introduction

Since the Industrial Revolution, atmospheric CO_2 emissions have continued to rise as a direct result of industrial and agricultural growth, from pre-industrial levels of 280 parts per million (Intergovernmental Panel for Climate Change, IPCC 2007), to current levels of ~390 ppm (NOAA 2012). Increased levels of anthropogenic CO_2 are projected to cause two major climatic changes that may threaten marine ecosystems: global warming and ocean acidification (OA).

Over the last century, the average temperature of the surface ocean has risen by 0.74°C (Hoegh-Guldberg et al. 2007; Solomon et al. 2007). Future IPCC projections suggest further temperature increases by as much as 3.6°C (IPCC 2007). The world's oceans act as a sink for atmospheric CO_2, with surface waters absorbing around 30% of anthropogenic CO_2 emissions (Form and Riebesell 2012). Whilst this has played an important role in helping to mitigate the atmospheric effects of climate change, there have been significant effects on seawater carbonate chemistry. During the last 200 years, oceanic pH has dropped by 0.1 units and is projected to continue to decrease by a further 0.3-0.5 units by 2100 (Caldeira and Wickett 2005) – a process known as OA. Although the rate at which global surface warming and OA will occur can be modelled with a degree of accuracy in the open ocean, the naturally high environmental variability of coastal regions makes projections difficult and the biological impacts unclear (Duarte et al. 2013). Natural environmental variability may be driven by downwelling of surface water (Findlay et al. 2013), upwelling of acidified deep water (e.g. the California Current system; Feely et al. 2008; Gruber et al. 2012), the presence of CO_2 vents (e.g. Hall-Spencer et al. 2008) and biotic composition (e.g. Anthony et al. 2011). The development of under-sea carbon capture and storage (CCS) facilities may also represent a new, anthropogenic, source of environmental variability (Blackford et al. 2009). There is growing concern that leaks from CCS infrastructure, thereby acting as a CO_2 point-source, may significantly affect oceanic carbonate chemistry at a local to regional scale (Blackford et al. 2009). CO_2 vents, biotic control, upwelling and CCS leaks may enhance or depress OA effects, necessitating a requirement to understand the effects of acute and chronic changes in carbonate chemistry on marine biota.

Increases in oceanic CO_2 cause the carbonate saturation of the water to decrease. The aragonite saturation horizon (ASH) is a naturally occurring boundary, below which the dissolution of aragonite, a calcium carbonate polymorph, is favoured. Future OA projections suggest that the ASH will shoal (i.e. become shallower) throughout the 21st century (Caldeira and Wickett 2005). Scleractinian cold-water corals (CWCs) are aragonitic organisms that may be found in water depths of 1000s metres, and thus may be highly susceptible to OA. CWCs can form extensive reef frameworks in fjord, continental shelf, slope and seamount settings (Roberts et al. 2009); the habitats they provide are classified as Ecologically and Biologically Significant Areas by the Convention on Biological Diversity (C.B.D. 2008). Today, only ~5% of CWC reefs are below the ASH (Form and Riebesell

2012). It is estimated that as much as 70% of the reefs could be below the ASH by 2100 (Form and Riebesell 2012), threatening the survival of CWCs and the biodiversity they support (Roberts et al. 2009). Hence, there is a pressing need for further research on the impacts of climate change on cold-water corals.

The impact of OA on cold-water corals is poorly understood, although it has been suggested that calcification rates of cold-water corals have already declined in response to elevations in CO_2 since the Industrial Revolution (Maier et al. 2012). Under short-term (1-week) exposure to high-CO_2 (up to 981 µatm), calcification rates in the cold-water coral *Lophelia pertusa* decreased by up to 30% (Form and Riebesell 2012). However, under longer (6 months) exposure to high-CO_2 (up to 984 µatm), the corals were able to continue to calcify beneath their tissue (Form and Riebesell 2012). Thus, there is some evidence for resilience in cold-water corals against OA (McCulloch et al. 2012), but studies have not yet taken into account the potentially synergistic effects of acidification and increased temperature, despite recorded diel environmental variability in carbonate chemistry at the Mingulay Reef CWC Complex, western Scotland (Findlay et al. 2013).

Research suggests that invertebrates may use algal-derived metabolites, such as dimethylsulphoniopropionate (DMSP) in a similar way to algae, for example in response to environmental challenges such as oxidative stress (Van Alstyne et al. 2006). A number of functions have been described for DMSP, an algal secondary metabolite (Stefels 2000). DMSP is also the major precursor to the climatically active gas dimethylsulphide (DMS), which has been linked to local climate regulation through the production of aerosol particles and cloud formation (Charlson et al. 1987; Ayers and Cainey 2007). Azooxanthellate invertebrates have been observed to accumulate DMSP from their diets, whilst zooxanthellate organisms acquire DMSP primarily from their symbionts (Van Alstyne and Puglisi 2007), but may also be capable of synthesising DMSP internally (Raina et al. 2013). It has been proposed that DMSP and its breakdown products may form an 'antioxidant cascade' (Sunda et al. 2002), able to scavenge a number of reactive oxygen species (ROS), thus minimising oxidative damage. Recent research has shown that intracellular DMSP concentrations in temperate (Burdett et al. 2012) and tropical (Burdett et al. 2013) calcifying macroalgae may be elevated in response to high CO_2 / low carbonate saturation conditions.

The aim of this study was to assess the short-term effect of high temperature and high CO_2 on the intracellular DMSP concentration of the cold-water coral *Lophelia pertusa* at the Mingulay Reef Complex, western Scotland, where short-term variability in carbonate chemistry has recently been recorded, and where significant changes in the carbonate chemistry are expected in the future (Findlay et al. 2013). In this study, it was hypothesised that intracellular DMSP would increase in corals exposed to high temperature and high CO_2, due to an increase in ROS production and a requirement for antioxidant mechanisms.

Material and methods

Study organism and sample collection location

Lophelia pertusa is the predominant species of CWC in the north-east Atlantic (Cairns 1994; Fosså et al. 2002), forming biodiversity hotspots in deep-ocean ecosystems worldwide. Coral colonies were collected from the Mingulay Reef Complex in the Sea of the Hebrides, west of Scotland (56°49'N, 07°23'W) during the 'Changing Oceans Expedition' in June 2012 on the Royal Research Ship *James Cook* (cruise JC073) (Roberts and participants 2013). The Mingulay Reef Complex is characterised by diel variability in carbonate chemistry, driven by tidally-induced downwellings of surface water (Findlay et al. 2013). At the time of the experiment, the Mingulay Reef was exposed to in situ diel pCO_2 changes of >60 µatm, equivalent to ~0.1 pH units (Findlay et al. 2013). Coral samples were obtained using the *Holland-1* Remotely Operated Vehicle (ROV) 180 m below chart datum. Coral colonies were acclimated to laboratory conditions for 2 days prior to starting the experiment in a holding tank maintained at ambient seabed temperature, which was sufficient for physiological stability in the control treatment, as demonstrated by Hennige et al. (2014).

Experimental setup

Coral colonies were fragmented and distributed between four abiotic treatments for 10 days, following Hennige et al. (2014) (Table 1): control (9°C / 396 ± 2 ppm CO_2), high temperature (12°C / 396 ± 2 ppm CO_2), high CO_2 (9°C / 817 ± 3 ppm, ~pH 7.8) and high temperature+CO_2 (12°C, 817 ± 3 ppm CO_2). Fragments from individual colonies were split between treatment tanks to prevent colony pseudo-replication within treatments. Once time zero (T0) measurements for DMSP were taken (see below), air enriched with CO_2 was initiated in the high CO_2 treatment tanks, controlled by a Wusthoff Digimix system and monitored by a LiCor 820 gas analyser. Ambient salinity (~35.3) was maintained throughout the experiment. Mesocosm water was partially (25-30%) changed every 2 days with freshly collected, unfiltered, seawater, pumped from 70 m water depth, providing a food source to the corals.

Carbonate chemistry determination

Over the course of the experiment, target pH was monitored daily using a Mettler Toledo SevenGo pH meter (NBS). This data was checked independently by calculated carbonate chemistry data from total alkalinity at set timepoints, and from measured CO_2 gas inputs (Licor-820) (these results are presented in Table 1). Samples for total alkalinity (A_T) were collected in 40 ml EPA vials and immediately poisoned with a saturated solution of mercuric chloride (8 µl). A_T was measured on a Metrohm 702 SM Titrino using the open-cell potentiometric titration method on 20 ml sample volumes with 0.01M HCl (repeatability: max. ± 0.1% at Alkalinity ~2200 µmol kg^{-1}). All A_T samples

were analysed at 25 ± 0.1ºC with temperature regulation using a water-bath (Grant OLS 200). Certified Reference Materials (batch 109) from A.G. Dickson (Scripps Institution of Oceanography) were used to standardize the acid at the beginning and end of each day of analysis. Carbonate parameters were calculated from A_T and pCO_2 using CO2SYS (Pierrot et al. 2006) with dissociation constants from Mehrbach et al. (1973), refit by Dickson and Millero (1987) and KSO_4 using Dickson (1990).

Coral intracellular DMSP

Coral fragments (n = 4-5 per treatment from different colonies, 5-7 polyps per fragment) were sampled at T0 and T10 days. Polyps (tissue and skeleton) were sealed in gas-tight borosilicate glass vials (Wheaton) using Pharma-fix septa (Grace Alltech) in 0.33M sodium hydroxide (NaOH) solution, to hydrolyse intracellular DMSP into DMS. Samples were stored in the dark at room temperature until DMS analysis (within 2 months). Intracellular DMSP concentrations were normalised to percent biomass (see below) following DMS quantification.

Mesocosm water DMS/P

Mesocosm water samples collected on T0 and T10 days were fixed for total dissolved DMSP+DMS (DMS/P) and total particulate DMS/P. Water samples were filtered using GF/F 0.7-µm depth filters (Millepore) according to Kiene and Slezak (2006). The filtrate and filter were sealed in separate borosilicate glass vials (Wheaton) and crimped shut with Pharma-Fix septa (Grace Alltech) at a final NaOH concentration of 0.33 M. Samples were stored in the dark at room temperature until DMS analysis (within 2 months).

DMS Quantification

All samples were analysed using the purge-cryotrap-gas chromatography (GC) technique (Turner et al. 1990) using a Shimadzu 2014 gas chromatograph at the University of Glasgow. The GC was equipped with a 25 m capillary column (Restek RTx-5MS, injector and column temperature: 45°C, nitrogen carrier gas) and a sulphur-specific flame photometric detector (200°C). Sample concentrations were calculated from DMSP standard calibration curves (DMSP standard from Research Plus Inc). DMS detection limit for all samples was 0.64 ng; analytical precision was within 3%. Intracellular DMSP is presented as nmol g^{-1} biomass; water samples are presented as nmol L^{-1}.

Percent biomass

The mass of each *L. pertusa* polyp was recorded before (tissue and skeleton) and after (skeleton only) storage in 0.33 M NaOH. The difference between the initial and final mass was considered biomass and used to express the intracellular concentration of DMSP.

180 **Statistical Analyses**

181 To test for differences between T0 and T10 control treatments, a 2-sample t-test was used for
182 intracellular DMSP (no data transformation was required to meet test assumptions of normality and
183 homogeneity of variance) and dissolved DMS/P (x^2 data transformation). Particulate DMS/P data
184 could not be transformed to meet parametric test assumptions, so a non-parametric Mann-Whitney
185 test was used. To test for differences between treatments at T10, an ANOVA General Linear Model
186 was used for intracellular DMSP (no data transformation required) and dissolved DMS/P (x^2 data
187 transformation). Particulate DMS/P data could not be transformed to meet parametric test
188 assumptions, so a multiple-comparison Kruskall Wallis test was used. Minitab V14 was used for all
189 statistical tests.

190

191 **Results**

192 **Carbonate chemistry**

193 Table 1 presents the average (X ± SE) carbonate chemistry parameters associated with each
194 mesocosm treatment over the course of the 10-day experiment. Control and high CO_2 treatments were
195 maintained at 9.06 ± 0.02°C and 8.90 ± 0.11°C respectively, whilst the high temperature and high
196 temperature+CO_2 treatments were maintained at 12.3 ± 0.10°C and 12.3 ± 0.02°C respectively (X
197 ±SE, Table 1). Aragonite saturation state (ΩArg) in the control and high temperature treatments was
198 1.82 ± 0.05 and 2.01 ± 0.10 respectively (X ±SE, Table 1). ΩArg in the high CO_2 treatment was 1.09
199 ± 0.01 and 1.30 ± 0.04 in the high temperature+CO_2 treatment (X ±SE, Table 1). Measurements of pH
200 using the handheld meter (NBS scale) were 8.00 ± 0.02, 7.99 ± 0.02, 7.94 ± 0.03 and 7.90 ± 0.02 for
201 control, high temperature, high CO_2, and high temperature+CO_2 treatments respectively. The
202 relatively high pH means of the high CO_2 and high temperature+CO_2 treatments include
203 measurements following water changes with unfiltered deep seawater which was not pre-bubbled with
204 CO_2. The pH ranges for the high CO_2 and the high-temperature+CO_2 treatments were 7.76-8.07 and
205 7.75-8.03 respectively, driven by the frequent water changes (Hennige, pers. obs.). Whilst this did not
206 exactly mimic the observed diurnal variability at Mingulay (Findlay et al. 2013), the range in pH is
207 similar to that expected under projected future conditions.

208 **Intracellular DMSP in *Lophelia pertusa***

209 No significant difference in intracellular DMSP concentration was observed between T0 and
210 T10 control measurements ($t_6 = 0.12$, $P = 0.908$, Figure 1). Intracellular DMSP concentrations in the
211 T10 high CO_2 treatment (X ± SE = 35.36 ± 6.96 nmol g^{-1} biomass) were higher than the T10 control
212 treatment (16.03 ± 3.25 nmol g^{-1} biomass), although this was marginally insignificant ($F_{(3,18)} = 2.56$, P

= 0.094, Tukey's pairwise comparison: $P = 0.077$, Figure 2). The T10 high temperature (22.52 ± 3.05 nmol g^{-1} biomass) and high temperature+CO_2 treatments (20.43 ± 7.25 nmol g^{-1} biomass) were not significantly different to the T10 control (Tukey's pairwise comparison: $P = 0.81$ and $P = 0.94$ respectively; Figure 2).

Mesocosm dissolved DMS/P

A significant decline in dissolved DMS/P concentrations was observed between T0 and T10 control measurements ($t_4 = 6.89$, $P = 0.002$, Figure 1). In contrast, no significant difference between T10 treatments was observed ($F_{(3,15)} = 1.21$, $P = 0.349$, Figure 2).

Mesocosm particulate DMS/P

A significant decrease in particulate DMS/P concentrations was observed in the control treatment from T0 to T10 ($W = 26.0$, $P = 0.030$, Figure 1). A significant difference in particulate DMS/P concentrations was also observed between T10 treatments ($H_3 = 12.79$, $P = 0.005$, Figure 2): particulate DMS/P in the high temperature treatment (X ± SE = 15.0 ± 0.88 nmol L^{-1}) was significantly higher than the high CO_2 (4.68 ± 0.17 nmol L^{-1}; pairwise comparison: $P = 0.002$) and high temperature+CO_2 treatments (5.19 ± 0.54 nmol L^{-1}; pairwise comparison: $P = 0.005$; Figure 2). Particulate DMS/P in the T10 control (10.80 ± 0.18 nmol L^{-1}) was not significantly different to any of the T10 experimental treatments (Figure 2).

Discussion

Intracellular DMSP

This is the first study to assess the intracellular DMSP concentrations of *Lophelia pertusa*. Samples were taken from different colonies from within the Mingulay Reef Complex, accounting for, at least in part, the observed variations in DMSP concentrations. Similarly large variations in intracellular DMSP concentrations between coral colonies have also been reported for tropical, zooxanthellate species (Broadbent et al. 2002). Intracellular DMSP concentrations from this study were similar to other invertebrate Phyla such as Porifera and Mollusca (Van Alstyne and Puglisi 2007), but were, surprisingly, higher than other azooxanthellate Cnidaria, which are often below detection limits (Van Alstyne and Puglisi 2007; Van Alstyne et al. 2009). This highlights the potential importance of cold-water corals as a store for DMSP. Given previous studies of DMSP in invertebrates (e.g. Van Alstyne et al. 2006), intracellular DMSP in *L. pertusa* is likely to be nutritionally derived because of *L. pertusa's* azooxanthellate life mode. However, recent research suggests that tropical corals may be able to internally synthesise DMSP, independent of zooxanthallae

(Raina et al. 2013); the existence of this mechanism in *L. pertusa* cannot be ruled out. Here, a modest increase in intracellular DMSP in the high CO_2 treatment was observed, supporting previous research of CO_2-driven up-regulation of intracellular DMSP in some phytoplankton (Arnold et al. 2012) and macroalgae (Burdett et al. 2012; Burdett et al. 2013), although this has not been uniformly observed and may be species-specific (Hopkins et al. 2010; Kerrison et al. 2012; Spielmeyer and Pohnert 2012). High CO_2 may lead to an energetic imbalance in *L. pertusa* (Hennige et al. 2014), perhaps enhancing the requirement for secondary metabolites such as DMSP. However, because of the short experimental period of this study, it is still difficult to predict the longer-term acclamatory responses that will occur under an ocean acidification scenario.

High temperature had an apparently antagonistic effect on high CO_2 impacts, perhaps because of an increase in feeding rate under higher temperatures (Dodds et al. 2007). However, the high temperature used in this study, although above the coral's preferred temperature range in the north east Atlantic (6-9 °C; Cairns 1994), has previously been observed within the Mingulay Reef Complex (Davies et al. 2009). Additionally, the Mingulay Reef is known to be exposed to large seasonal and diel variations in temperature (Findlay et al. 2013), so a degree of thermal tolerance was perhaps to be expected.

Mesocosm DMS/P

DMS/P in the mesocosm water column may have been derived from algae, detritus and/or the corals, but does more accurately represent in situ conditions compared to the provision of an artificial food source for the corals. The method adopted here provides a total DMSP+DMS concentration; different ratios in source matter and / or variations in DMSP / DMS production will have contributed to the observed variations in water column DMS/P concentrations. Dissolved DMS/P did not differ significantly between treatments, although a decline in dissolved DMS/P and particulate DMS/P was observed from T0 to T10 in under control conditions, perhaps reflecting a change in the natural planktonic composition of the water pumped from 70 m depth. Nevertheless, comparisons between the control and treatment conditions may still be made at T10. In the high CO_2 and high temperature+CO_2 treatments, particulate DMS/P was depleted, suggesting that accumulation by the corals may have reduced mesocosm DMS/P concentrations, as has been observed in azooxanthellate shallow-water cnidarians (e.g. Van Alstyne et al. 2006). The extent to which DMSP accumulation may be regulated by invertebrates in response to physiological requirements (e.g. as an antioxidant) is only speculative at present (Van Alstyne and Puglisi 2007), but this study suggests that *L. pertusa* may up-regulate DMSP uptake under high CO_2 conditions. Further, rate-determinant studies should seek to quantify DMSP uptake by *L. pertusa.*

The impact of high CO_2 and high temperature on the planktonic food supply for *L. pertusa* is important to consider. The effect of projected environmental change on phytoplankton DMS/P

dynamics is likely to be highly species-specific (Wingenter et al. 2007; Vogt et al. 2008; Hopkins et al. 2010; Avgoustidi et al. 2012), affecting phytoplankton bloom community composition, and impacting the nutritional quality and DMSP content of the *L. pertusa* diet. This is particularly important in the Mingulay Reef complex given the daily downwellings of surface water (Findlay et al. 2013). This, combined with an impact on coral metabolism and feeding rate, may significantly affect rates of intracellular DMSP accumulation by *L. pertusa*, impacting the use of DMSP as an antioxidant by the coral.

Environmental implications and conclusions

This study provides new information on the potential for biotic control on deep sea biogeochemistry that may inform future biogeochemical studies on CWC reefs. A combination of short-term (e.g. vents, up/downwelling, CCS leaks) and long-term (ocean acidification) sources of high CO_2, combined with projected increases in water temperature may expose CWCs to high CO_2 and high temperature conditions for varying time periods. Despite the experimental limitations, this study suggests that the CWC *L. pertusa* may accumulate DMSP in its tissues in response to short-term, consistent exposure to high CO_2, perhaps for use as an antioxidant; further studies are, however, needed to confirm this. Consequently, in areas of high coral density, DMSP accumulation may remove particulate DMS/P from the surrounding water column, reducing the availability of DMS/P to other organisms such as microbes, which are known to utilise DMSP and DMS as a carbon and sulphur source (Green et al. 2011; Hatton et al. 2012), potentially affecting the ecosystem function of local habitats. Additionally, in shallow marine environments, intracellular DMSP acts as a settlement cue for a number of invertebrate larvae (Huggett et al. 2006; Steller and Cáceres-Martinez 2009). If this is also the case in CWC habitats, a high CO_2-driven up-regulation of DMSP by *L. pertusa* may alter the dynamics of the CWC ecosystem. Tidal downwellings connect the Mingulay Reef Complex to the surface ocean. Uptake of DMSP and DMS by the corals may therefore be a, previously overlooked, factor in the magnitude of surface ocean DMSP production vs sea-air DMS flux and the subsequent potential for climate regulation through cloud formation.

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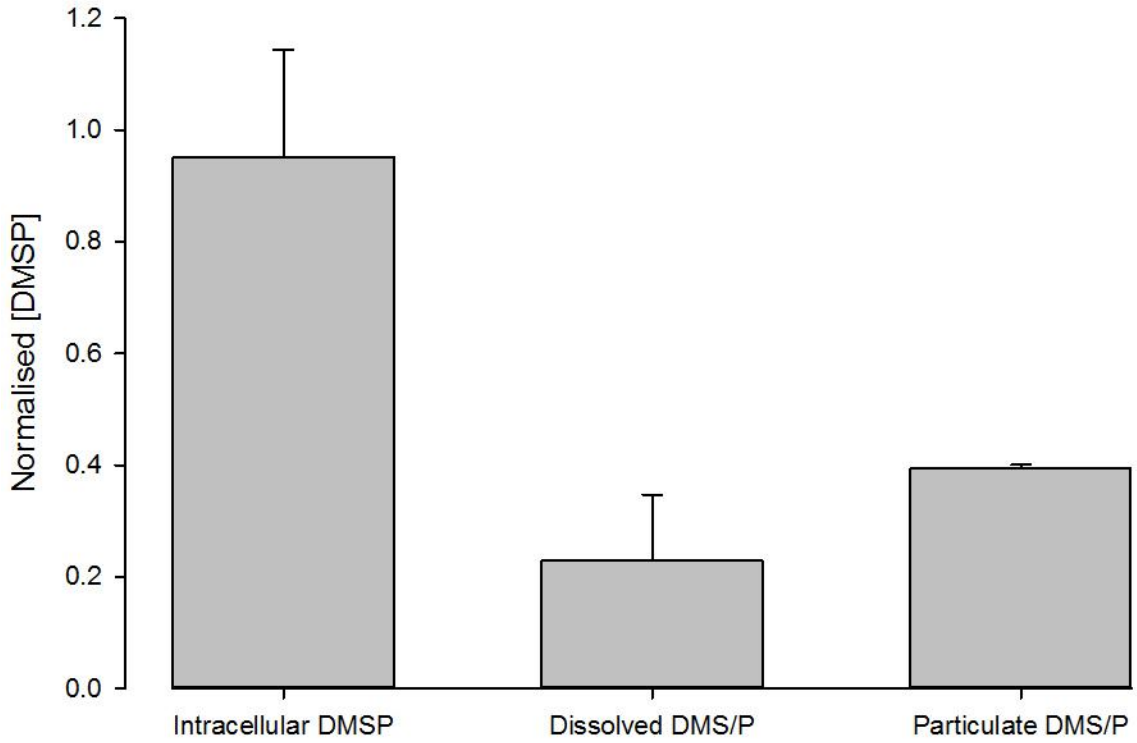
Table 1. Mean (±SE) values of measured temperature, salinity, injected CO_2 and total alkalinity, and derived dissolved inorganic carbon species (TCO_2), pH (Total scale) and aragonite saturation state (ΩArg) for control, high temperature, high CO_2 and high temperature+CO_2 treatments.

Parameter	**Treatment**			
	Control	**High temperature**	**High CO_2**	**High temperature+ CO_2**
Temperature (°C)	9.06 ± 0.02	12.30 ± 0.10	8.90 ± 0.11	12.30 ± 0.02
Salinity	35.3 ± 0.02	35.4 ± 0.03	35.2 ± 0.08	35.4 ± 0.05
Alkalinity (µmol kg^{-1})	2197 ± 34.9	2170 ± 57.3	2299 ± 8.52	2277 ± 27.0
TCO_2 (µmol kg^{-1})	2031 ± 30.5	1986 ± 49.5	2227 ± 7.86	2189 ± 24.8
pH_T	8.03 ± 0.01	8.02 ± 0.01	7.76 ± 0.00	7.76 ± 0.00
pCO_2 (ppm)	396 ± 2.08	396 ± 2.08	817 ± 2.49	817 ± 2.49
ΩArg	1.82 ± 0.05	2.01 ± 0.10	1.09 ± 0.01	1.30 ± 0.04



476

477 **Fig.1** *Lophelia pertusa* intracellular DMSP (n = 5) and dissolved and particulate DMS/P
478 concentrations (n = 4) in the control treatment at the end of the 10-day experiment, normalised against
479 control measurements at the beginning of the experiment. See Table 1 for a summary of the
480 experimental conditions. Data presented as mean ± SE

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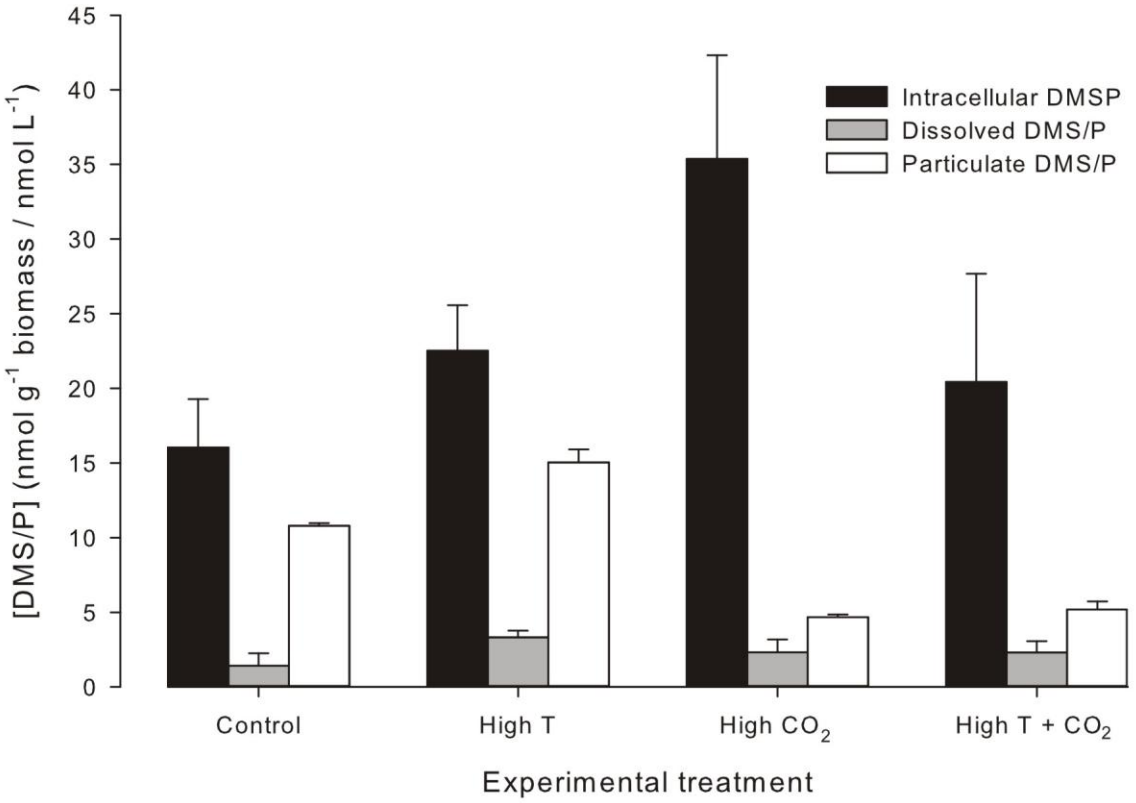

Fig.2 DMSP concentrations following 10-days incubation at control, high temperature, high CO_2 and high temperature+CO_2 conditions. *Lophelia pertusa* intracellular DMSP (normalised to g of coral tissue biomass; n = 5 except in high temperature+CO_2 treatment where n = 4) and dissolved (n = 4) and particulate (n = 4) DMS/P concentrations are presented. See Table 1 for a summary of the experimental conditions. Data presented as mean ± SE