Revealing the mechanistic basis of reef-building coral tolerance versus susceptibility to low O_2 stress

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Certificate of original authorship

I, Rachel Clare Alderdice declare that this thesis, is submitted in fulfilment of the requirements for the award of Doctor of Philosophy, in the Faculty of Science at the University of Technology Sydney.

This thesis is wholly my own work unless otherwise reference or acknowledged.

In addition, I certify that all information sources and literature used are indicated in the thesis.

This document has not been submitted for qualifications at any other academic institution.

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Thesis abstract

Exposure of marine life to low oxygen is accelerating worldwide as a consequence of climate change and localized pollution. Global coral populations have been in significant decline now since the 1950s, experiencing widespread fatal bleaching episodes (i.e., loss of their symbiotic algae). However, only recently have such events been proposed to be directly associated with an inadequate oxygen supply (hypoxia).

I first examined the mechanistic basis for coral hypoxia stress response systems between two common reef-building *Acropora* species reported to have differential bleaching thresholds to heat stress in the field. As expected, only the less stress-tolerant species bleached under night-time deoxygenated conditions, corresponding to contrasting gene expression profiles indicative of varied effectiveness of their hypoxia-inducible factor (HIF) hypoxia response system (HRS).

I next considered how the responses observed for adult corals applied to coral larvae, where the latter exhibit very different physiologies related to their predominant free-living planktonic (without photosynthetic algae symbionts) versus benthic stages. Despite exhibiting a consistent swimming phenotype compared to control samples, coral planulae demonstrated similar HIF-HRS expression to the adult and differential gene expression that reflected a disruption of pathways involved in developmental regulation, mitochondrial activity, lipid metabolism, and O_2-sensitive epigenetic regulators.

I then incorporated deoxygenated seawater into short-term heat assays and demonstrated that deoxygenation can lower the thermal limit of an *Acropora* coral species by as much as 0.4 ºC and 1ºC based on the maximum photosystem II (PSII) photosynthetic efficiency and bleaching index score, respectively. I showed that even heating alone activates putative genes key to the HIF-HRS, which may suggest that a hypoxic state is reached in the coral tissue under heat stress, possibly as a result of an O_2-intensive stress response.

Hypoxia stress associated genes I identified from model *Acropora* corals were then considered against genomic gene sets of a wider range of coral species based on the notion that a variation in gene copy number can result in differential gene expression with subsequent differences in the effectiveness of any given stress response. Therefore, I used an ortholog-based meta-analysis to investigate how the hypoxia gene set inventory differed amongst 24 coral species. Interestingly, the highest gene copy number was consistently presented by *Porites lutea*, which is considered to exhibit inherently greater stress tolerance to bleaching. As such, the unevenly expanded (or reduced) hypoxia genes presented here provide key genes of interest to target in examining (or diagnosing) coral stress thresholds.

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Thesis structure

This thesis is comprised of a series of four data chapters (Chapters 2 to 5) with their own full and chapter-focussed introduction sections and hence my general introduction (background literature review, Chapter 1) has been kept brief to avoid unnecessary repetition. At the time of thesis submission, all data chapters were published or accepted for publication.

Chapter 1: Contributed to the following published perspective article.

Hughes, D.J., **Alderdice, R.**, Cooney, C., Kühl, M., Pernice, M., Voolstra, C.R., Suggett, D.J. (2020) Coral reef survival under accelerating ocean deoxygenation. *Nature Climate Change* 10: 296–307. doi:10.1038/s41558-020-0737-9.

Chapter 2: Presented as the following published research article.

Alderdice, R., Suggett, D.J., Cárdenas, A., Hughes, D.J., Kühl, M., Pernice, M., Voolstra, C.R. (2021) Divergence of hypoxia response systems under deoxygenation in reef-forming corals aligns with bleaching susceptibility. *Global Change Biology* 27: 312-326. gcb.15436. doi:10.1111/gcb.15436.

Chapter 3: Presented as the following published research article.

Alderdice, R., Pernice, M., Cárdenas, A., Hughes, D. J., Harrison, P. L., Boulotte, N., Chartrand, K., Kühl, M., Suggett, D.J., Voolstra, C.R. (2022) Hypoxia as a physiological cue and pathological stress for coral larvae. *Molecular Ecology* 00, 1–17. doi:10.1111/mec.16259.

Chapter 4: Presented as the following research article accepted for publication.

Alderdice, R., Perna, G., Cárdenas, A., Hume, B., Wolf, M., Pernice, M., Kühl, M., Suggett, D. J., Voolstra, C. R. (2022) Deoxygenation lowers the coral thermal threshold. *Scientific Reports* (*Accepted*).

Chapter 5: Presented as the following published research article.

Alderdice, R., Hume, B.C., Kühl, M., Pernice, M., Suggett, D. J., Voolstra, C. R. (2022) Disparate inventories of hypoxia gene sets across corals. *Frontiers in Marine Science* 9, 834332. doi:10.3389/fmars.2022.834332.

Chapter 6: Discussion of results and insights gained from all data chapters.

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