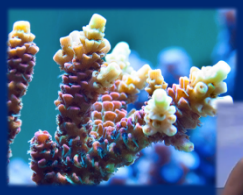


Oxidative stress-related gene expression in juveniles of the coral *Acropora millepora*

Melissa M Rocker[1,3], Bette L Willis[1,2], Line K Bay[4]

[1]School of Marine and Tropical Biology & [2]ARC Centre of Excellence for Coral Reef Studies, JCU, Townsville, QLD,
[3]AIMS@JCU, Townsville, QLD, [4]Australian Institute of Marine Science, PMB 3, Townsville MC, QLD

Objective:

The objective of this study was to determine if *Symbiodinium* type affects the gene expression response of *Acropora millepora* juveniles experimentally inoculated with C or D *Symbiodinium* types and exposed to thermal stress.

Approach:

- *Acropora millepora* juveniles were experimentally inoculated with *Symbiodinium* types C1 or D and grown in the field for one year.
- Juveniles were randomly placed in six aquaria (three ambient control and three experimental heat stress) and acclimated to 28°C .
- Temperatures in the heat treatment were raised 0.5°C every six hours until water temperature reached 32°C .
- Juveniles with C, D, and mixed symbiont associations were sampled every second day during the 14-day experiment.
- Post experimental *Symbiodinium* types were determined with PCR and SSCP of the 18S nuclear ribosomal DNA internal transcribed spacer 1 region.
- Reverse Transcription qPCR (RT-qPCR) and the GeXP system were used to quantify relative gene expression of 50 target genes in the coral juveniles.

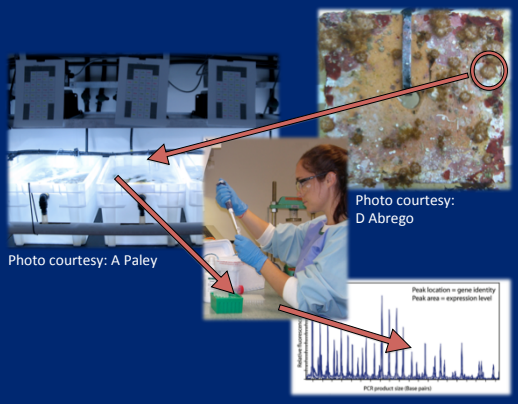


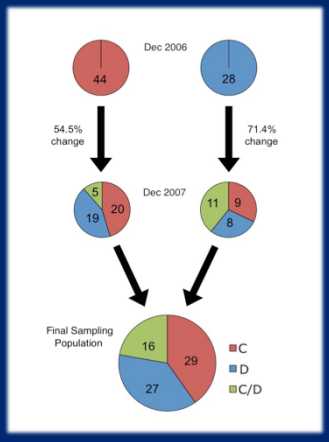
Photo courtesy: A Paley

Photo courtesy: D Abrego

Results:

- 59.7% of the 72 juveniles did not retain the originally inoculated *Symbiodinium* type, resulting in 40.3% ITS1 type C, 37.5% ITS1 type D, and 22.2% mixed ITS1 type C and D (Fig. 1).
- A NOS-interacting gene, regulating nitric oxide production, was up-regulated in the middle time period of the stress treatment (32°C; Fig. 2), but remained lower in the control treatment (28°C) until the last sampling time, when expression was up-regulated.
- Two heat shock proteins, HSP70 and HSP90, displayed significant differences between corals with single versus multiple symbiont types across the first two time periods of the experiment (Fig. 3), but were not differentially expressed at the end of the experiment.

Figure 1 (below): Successional changes of symbiont complements in juvenile *A. millepora* after a one-year grow-out period at Magnetic Island. Pie charts depict proportion and numbers of juveniles hosting type C (red), D (blue), and C/D mixed (green).



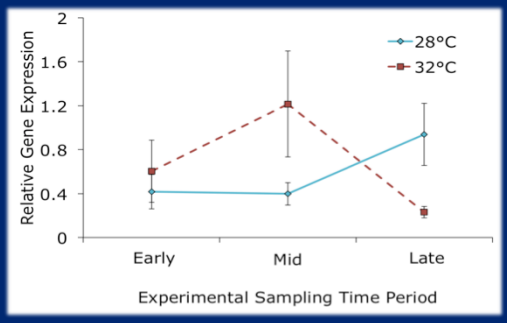


Figure 2 (above): Changes in juvenile *A. millepora* gene expression of a NOS-interacting gene during the heat stress experiment. Error bars represent +/- SE, n = 3, 7 and 8 for each treatment during early, mid and late times.

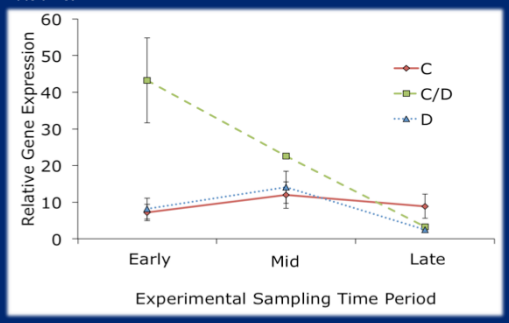


Figure 3 (right): Expression of HSP90 in juvenile *A. millepora* hosting different symbiont complements exposed to laboratory heat stress. Error bars represent +/- SE, n ranges between 1 and 8 per treatment. Expression patterns of HSP70 and HSP90 were almost identical and therefore only HSP90 is shown.

Conclusions:

- Coral-*Symbiodinium* associations change through early ontogeny, with coral juveniles possibly acquiring new *Symbiodinium* types from the environment. Results support an important role for symbiont complements in the transcriptomes of corals and highlight high variability among individuals.
- Two HSPs were differentially expressed between corals with single versus mixed symbiont communities, suggesting differences in the need for HSPs under homeostasis or early stress in corals with "pure" versus mixed symbiont complements.
- Only 3 out of 50 heat and oxidative stress genes were differentially expressed, raising the possibility that the 32°C (and 360 µM photon / m^2) treatment was not stressful for the coral juveniles, potentially because they were grown in a relatively warm location and inoculated with locally acclimatised/adapted *Symbiodinium* types.

JAMES COOK UNIVERSITY AUSTRALIA · AIMS@JCU · ARC Centre of Excellence Coral Reef Studies · Australian Institute of Marine Science