Biochemistry of the Coral Symbiosis by Vibrational Spectroscopy

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Introduction

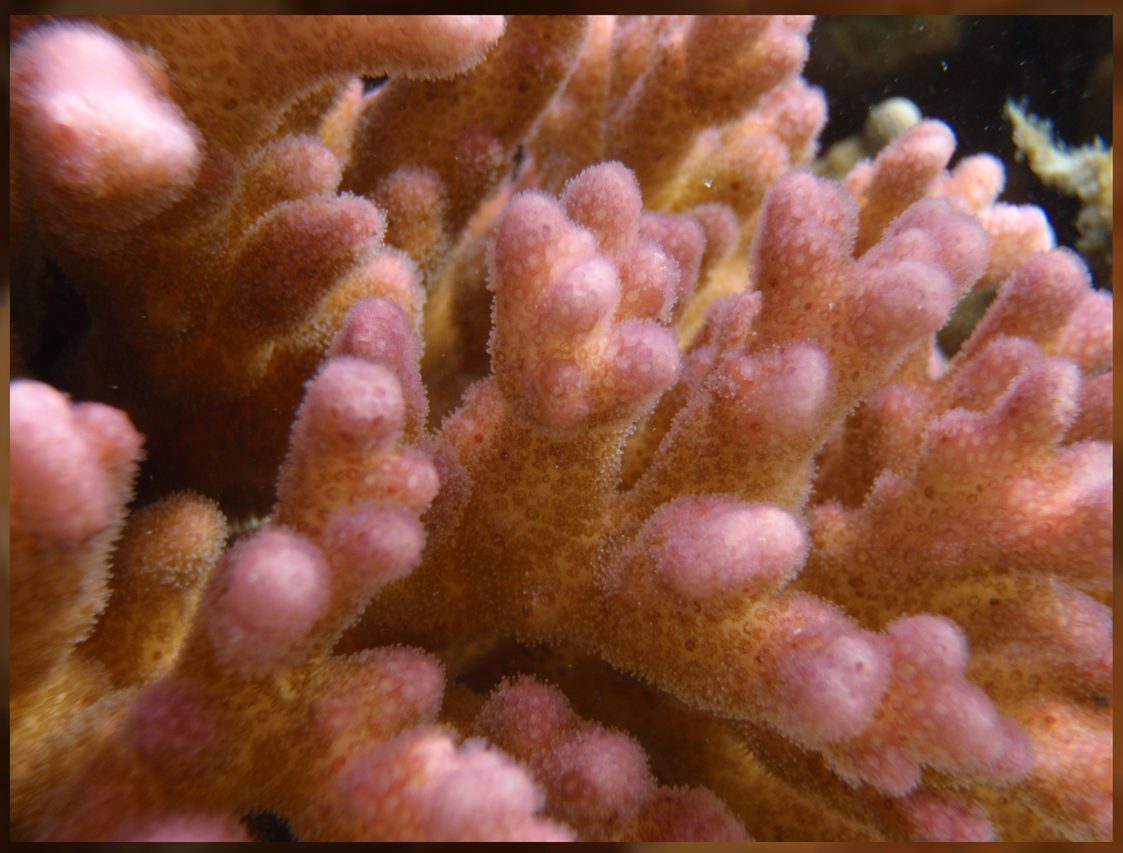
Research into the mechanism of bleaching in scleractinian corals, has suggested a breakdown in the symbiotic relationship between the cnidarian host and its symbiotic dinoflagellate "zooxanthellae" could be the cause[1,2]. Under stress, macromolecular pools shift in order to compensate for a lack of a particular nutrient[3]. Changes in elemental stoichiometry result in changes in macromolecular distribution. Understanding how this process occurs is an important part of the overall picture of coral bleaching.

Fourier Transform Infrared (FTIR) Spectroscopy is a form of vibrational spectroscopy that can quantify biochemical systems in the form of characteristic peaks in a spectrum based on the vibrations of the macromolecular components[3,4]

Method

For the purposes of this study, colonies of *Pocillopora damicornis* were subjected to extremely low flow conditions in order to cause severe tissue degradation. Samples were placed in a separate tank with no circulation and were left for 2 days under constant light at 250 μmol photons.$m^{-1}.s^{-1}$. After two days, the tissue showed signs of severe lysis; the tissue on the branches was partially separated from the skeleton. These corals were said to be moribund.

The tissue was removed from the skeleton with compressed air and the resulting slurry was then freeze-dried to remove water. The powder that remained was ground together with solid potassium bromide in an approximately 1:100 ratio (sample to KBr). The powder mixture was then pressed under extremely high pressure to obtain a transparent disc. Standard powder samples of bovine serum albumin (BSA), palmitic acid and potato starch were also prepared in the same manner for spectral comparison.



***Pocillopora damicornis* colony (Photo by Ross Hill)**

Table 1: Main Infrared band assignments for healthy coral tissue

Wavenumber (cm^{-1})	Nutrient Pool	Assignment
3295	**Protein**	N-H stretching (Amide A)
2960-2860	**Lipid**	CH_2 and CH_3 symmetric and antisymmetric stretching
1730	**Lipid**	Carbonyl stretching
1660	**Protein**	Carbonyl stretching (Amide I)
1550	**Protein**	N-H bending and C-N stretching (Amide II)
1410	**Protein**	CH_2 and CH_3 methylene chain stretching
1340	**Collagen**	CH_3 bending (mesoglea)
1235	**Carbohydrate**	C-O stretching, Phospholipid P-O-C stretching
1080	**Protein**	PO_2 stretching
935-900	**Phospholipid**	PO_2 bending

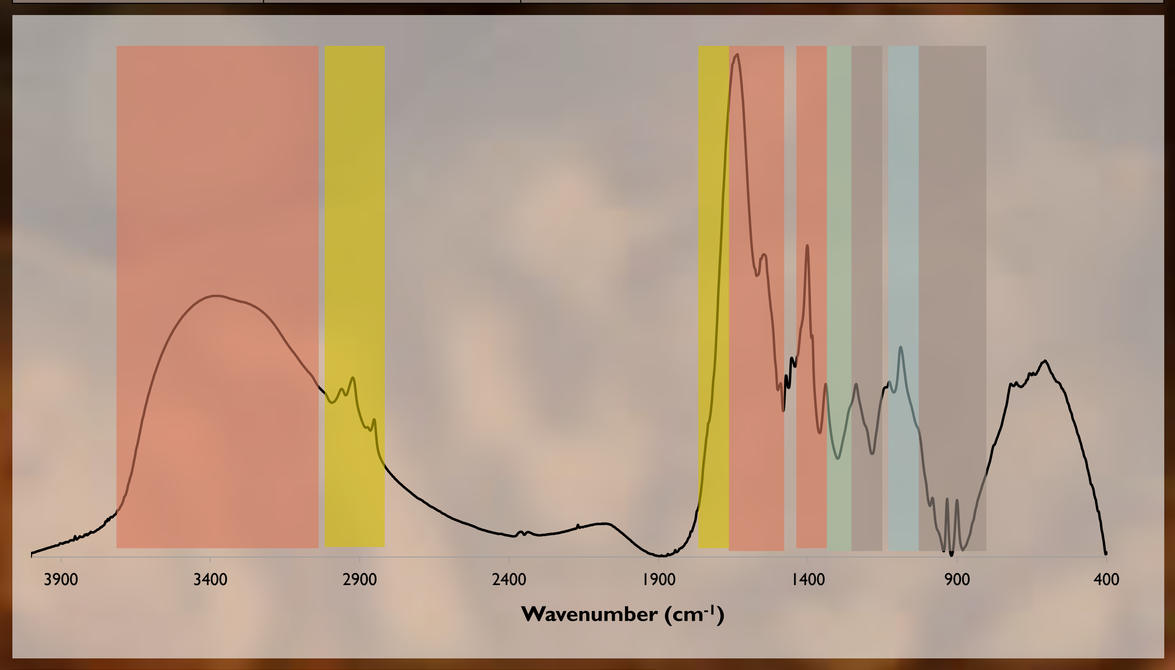


Figure 1: Infrared spectrum of healthy coral tissue

Figure 2: Comparison of 1800-400cm^{-1} region of infrared spectra of healthy and moribund *P. damicornis* tissue

Discussion

The spectra show a number of important changes in macromolecular pools. Storage lipids show a relative decrease in content which could indicate a corresponding decrease in organic carbon translocation from the symbiont. Phospholipid content also changes; the characteristic doublet at 900-930cm^{-1} is not present in the moribund tissue spectrum which suggests a decrease in cell function due to the breakdown of the cell membranes.

Protein structure shows signs of degradation; changes in shape and shifts in maximum wavenumber of the amide I band indicate protein secondary structure changes towards more disordered conformations. This could again point towards a breakdown in nucleic acid translocation from the symbiont to replace the degraded proteins needed for normal function.

Conclusions

FTIR spectroscopy has great potential as a tool for the analysis of stress in the coral holobiont. It is particularly suited to the characterisation of the macromolecular structure as it can analyse the biochemistry of the entire system simultaneously with very little sample preparation. Further development is needed in order to quantify and assess the relative changes in protein, carbohydrate and lipid contents, individually.

Great Barrier Reef Marine Park Authority collection permit number G09/31733.1

References

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Background image courtesy of Dr Ross Hill

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