Vibrio species induce pathogenic effects on tropical reef corals and their *Symbiodinium* clade phylo-types *in situ* and *in vitro*

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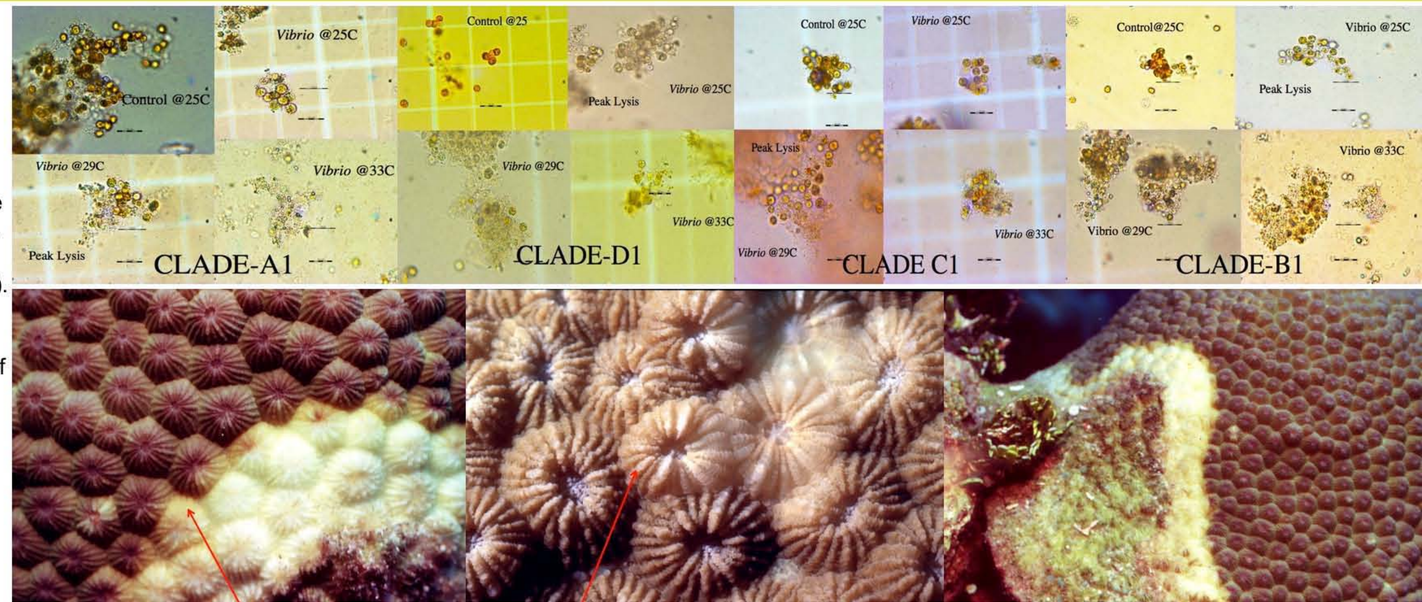
ABSTRACT

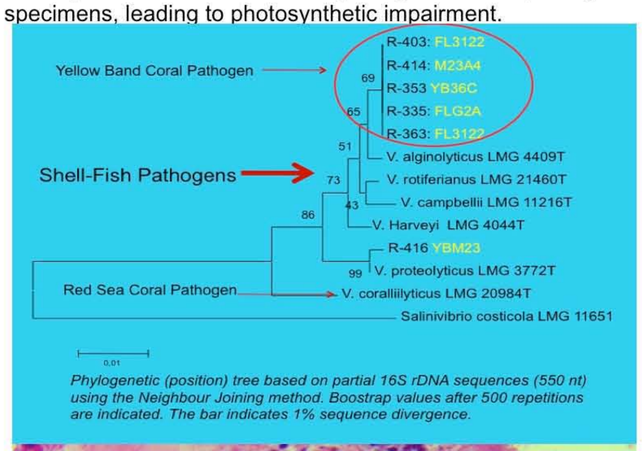
Vibrio strains coupled with thermal stress have been linked to Yellow Band Coral Disease (YBD) in Caribbean, Pacific and Red Sea reef building species, causing a breakdown between coral host and its endosymbiotic zooxanthellae (Cervino et al., 2004-2008). *Vibrio* strains isolated from YBD lesions specifically target symbiotic zooxanthellae found in hermatypic coral host tissues. Symbiotic zooxanthellae ie. *"Symbiodinium* clade phylotypes" A1, B2, C1, and D1, were inoculated with *Vibrio alginolyticus* and four novel yellow-band causing *Vibrio spp* isolated from the Indo-Pacific, Florida and the Caribbean. Heat treated phylotypes inoculated with the YB-consortium showed a significant decrease in cell density, as well as a sharp decrease in cell division from 24 to 48h, compared to non heat stressed inoculated zooxanthellae. The exposure of *Symbiodinium* to *Vibrio* is accompanied by a marked reduction in the rates of photosynthetic electron transport down Photosystem II (PSII). However, the efficiency of photochemical conversion in PSII (Fv/Fm) is affected only at later stages of stress development. These results suggest that *Vibrio* attack starts by impacting the function and integrity of the thylakoid membranes, followed by impairment of the PSII protein complexes. This pattern is in contrast to that of thermal stress that starts with impairment of PSII and a decrease in F_v/F_m (Warner et. al.,1999; Gorbunov et al., 2001; Tchernov et al., 2004). Non-heat treated controls indicate a decrease in density and a decrease in cell cycle mitotic index (MI%) over 48 hr. However, this is not as significant as when higher temperature is applied. These data are consistent with results from corals showing signs of YBD in the field. Thermal stress, when coupled with these *Vibrio* strains, inhibits energy production and severely impairs the cell-cycle of *Symbiodinium* clade phylotypes *in-vivo* and *in situ* (Cervino et al., 2004). Cytopathological examinations show a decrease in chlorophyll, cell size, increased cell lysis, and cell degeneration in all *Vibrio* inoculated heat treated zooxanthellae, compared to non-heat treated inoculated zooxanthellae. Variable fluorescence measurements provide further evidence that *Vibrio* toxin production and virulence is enhanced during infection under heat stress. Transmission electron microscope analysis also indicates that the photosynthetic apparatus within the chloroplasts are displaced to the outer margin of the zooxanthellae cell. "Plastoglobules", containing lipids and polysaccharide products inside chloroplasts, are seen to form. Their numbers increase during the upregulation of plastid lipid metabolism, in response to thermal oxidative stress and during senescence. Severe decompartmentalization of chloroplast grana occurs, as thylakoid membranes disappear in *Vibrio*-heat treated specimens, leading to photosynthetic impairment.



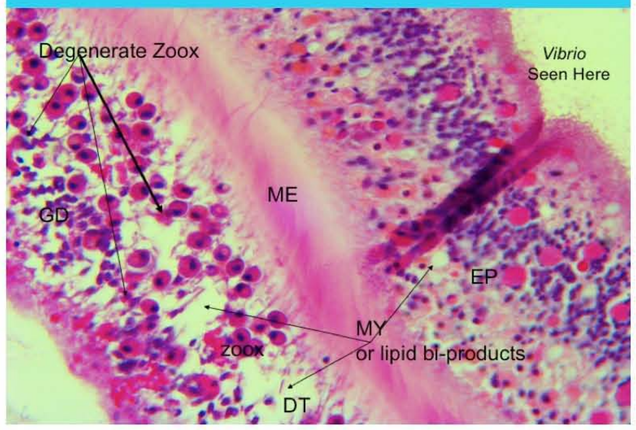
Photosynthetic symbioants are attacked inside the gastrodermal animal tisssue

"Translocation of energy from the symbioant to the coral animal disrupted"

Vibrio Induced Yellow Band Disease is Found in Almost Every Tropical Ocean



Phylogenetic (position) tree based on partial 16S rDNA sequences (550 nt) using the Neighbour Joining method. Boostrap values after 500 repetitions are indicated. The bar indicates 1% sequence divergence.



DT, tissue detachment; GD, gastrodermis; ME= mesoglea; MY=mucocytes; ZOOX, zooxanthellae, EP Epidermis.