

Ecotoxicological Investigation of Coral Planula Toxicity



ABOVE: FIG.2 - A SCUBA diver setting “planula traps” over a single coral colony. Traps are set on a coral colony at dusk during the spawning season, then collected at dawn. The trap does no damage to the coral colony, and allows the colony to remain fast to its reef.



LEFT: FIG.3 - An air bubble in the planula trap allows the trap housing to float above the coral colony. Planulae are positively buoyant, so they will float and collect into the trap housing.

BELOW: FIG.10 - Planulae of *Stylophora pistillata* exposed to 500 nanomolar of the fungicide, chlorothalonil for 5 hours. The planulae are dead and grossly deformed.



CORAL PLANULA TOXICITY ASSAY

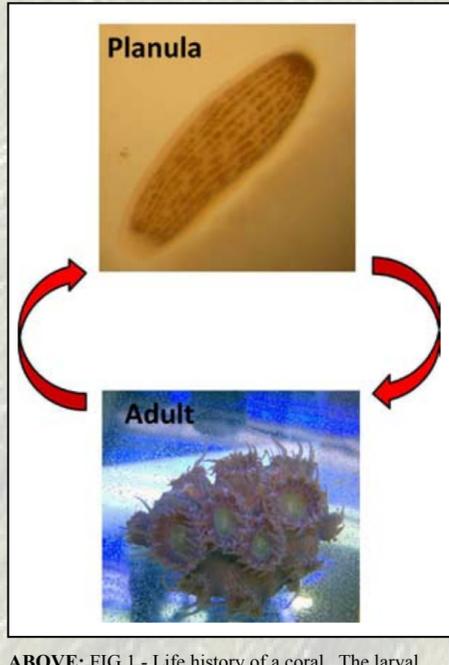
Coral planulae are the larval stage of a coral after it has undergone sexual reproduction – they are coral babies (Fig. 1). The general rule is that the juvenile stages of an organism are much more sensitive to the toxic effect of pollutants (as much as a 1,000 fold). Without coral recruitment to juvenile a reef, that reef is a coral zombie – it is only a matter of time before the reef system collapses. Hence, conducting ecotoxicological studies using coral planula is paramount for us to understand the magnitude of harm human activities can have on a coral reef.

One of the best ways of collecting coral planula without disturbing a natural reef is to collect them using planula traps (Figs. 2 & 3). Planula are brought into the lab, and then separated and counted (Fig. 4). Planula are then placed into wells of a Teflon microplate so that they can be used in a sound and rigorous toxicological experimental design (Fig. 5).



ABOVE: FIG.4 - Separation, counting, and inspection of coral planulae captured that morning.

The easiest and least expensive measures for toxicity are planula deformity and mortality. Healthy planulae are easy to score (Fig. 6). Planula exposed to various concentrations of different pollutants can manifest a number of deformities (Figs. 7-11), while death of a planula is obvious (Fig. 12). Unfortunately, deformity and death are the crudest measurement forms and the result of ACUTE and morbid exposure concentrations of a pollutant. One of the goals of ecotoxicology is to determine subtle changes of coral condition before they manifest themselves as gross morbidity or mortality. Coral planula can also be analyzed for a number of cellular biomarkers that indicate the condition of cellular health and homeostasis, thereby allowing us to determine and model the effects of coral being exposed to much lower concentrations of a pollutant, as well as helping us to understand the pathology of the coral associated with the pollutant (Fig. 13).



ABOVE: FIG.1 - Life history of a coral. The larval stage of a coral, the planula, will float through the ocean until it finds an acceptable place where it can settle and be recruited onto a coral reef. Once settled, the coral planula will metamorph into a coral polyp, and then replicate itself asexually into a coral colony. Coral polyps give rise to coral planula through sexual reproduction.



ABOVE: FIG.11 - Planulae of *Stylophora pistillata* exposed to 5 millimolar sodium cyanide for 24 hours in the dark. In the wild, corals are exposed to cyanide as a result of the illegal cyanide fishing practices. The planulae are dead and grossly deformed.

BELOW LEFT: FIG.8 - Planula of *Stylophora pistillata* exposed to 0.0125 micrograms of hydrogen sulfide per milliliter. The planula is grossly deformed, but not yet dead. Exposure was for four hours.

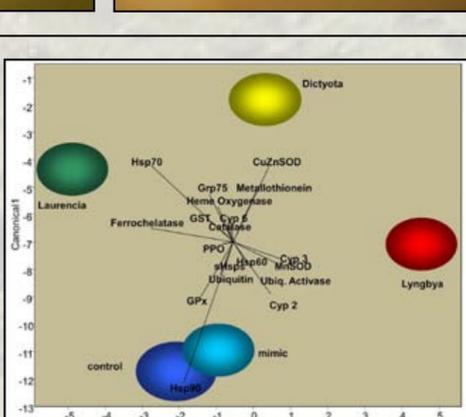
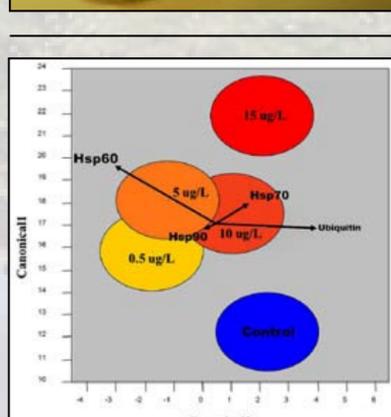
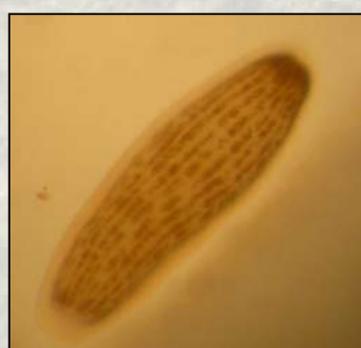
BELOW RIGHT: FIG.7 - Planula of *Stylophora pistillata* exposed to 0.05 micrograms of hydrogen sulfide per milliliter. The planula is dead and grossly deformed. Exposure was for four hours.



ABOVE: FIG.5 - Coral planula can be exposed to a pollutant using a Teflon 24-well microplate. Ten planula per well, and 4 wells per treatment. A control and five concentrations of a pollutant can be tested using a single Teflon microplate.

BELOW LEFT: FIG.9 - Planulae of *Stylophora pistillata* exposed to 25 micromolar of the anti-foulant herbicide, Irgarol 1051 for 24 hours. The planulae are dead and grossly deformed.

BELOW RIGHT: FIG.6 - A healthy planula from the coral *Stylophora pistillata*.



ABOVE LEFT: FIG.12 - Planulae of *Porites astreoides* were exposed to four different concentrations of the mosquito-control pesticide, Dibrom. Four different cellular biomarkers were measured in the planulae; Heat-shock protein 60, Heat-shock protein 70, Heat-shock protein 90, and ubiquitin. Data were analyzed using canonical correlation analysis. Centroids that do NOT overlap indicate significant differences in biomarker patterns, while centroids that do overlap indicate no significant difference in biomarker patterns. What this data tells us is that exposure to Dibrom induces a significant change in the protein metabolic condition to coral planula.

ABOVE RIGHT: FIG.13 - Planulae of *Porites astreoides* were exposed to three different types of algae that are found on coral reefs in the Caribbean: Lyngbya, Dictyota, and Laurencia. The demise of coral reefs in the Caribbean are often observed as a Community Phase-Shift: the ecological community goes from being a coral-dominated community to an algal-dominated community. When and as this shift occurs, the corals on the reef die. The big question is do the algae leach chemicals into the water to kill or repel corals, thereby allowing the algae to obtain more substrate surface area for them to live on. Coral planulae exposed to the three different algal species were assayed for 18 different cellular biomarkers. Data were analyzed using canonical correlation analysis. Centroids that overlap indicate no significant difference in biomarker patterns, while centroids that do NOT overlap indicate significant differences in biomarker patterns. What this data tells us is that exposure to the three different algal species causes a significant change in the cellular condition of the planula, and that these physiological conditions are pathological – the algae are releasing a chemical that is toxic to the coral. What does this mean ecologically? It means that algae could prohibit coral planula from recruiting onto a reef, potentially explaining why we see the drastic drop in coral recruitment in many areas of the Caribbean.