## Sensory abilities of coral reef fish larvae in the detection of their settlement habitat

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#### Abstract:

One of the great mysteries of fish ecology is how larvae locate the relatively rare patches of coral reef habitat on which they settle. The answer must lie partly in the sensory abilities of fish because it seems unlikely that successful settlement is solely a matter of chance. The objective of my post-doctorate attempts to estimate the sensory abilities of coral reef fish larvae when they search for a settlement habitat. Larval recognition of settlement habitat may involve the detection of conspecifics or of shelter characteristics determined by emissions of visual, chemical and mechanical cues. I conducted a series of laboratory experiments using larvae of *Chromis viridis* (damselfish). *C. viridis* larvae responded positively to visual, mechanical, and olfactory cues expressed by conspecifics. Larvae choose compartments of experimental arenas containing conspecifics in 75% of trials, and failed to show any significant directional responses to heterospecifics or coral substrates. Then, I conducted HPLC analyses of seawater containing *C. viridis* juveniles and isolated high concentrations of several organic compounds. Subsequent laboratory trials demonstrated that *C. viridis* larvae responded positively to only one of these organic compounds. This compound was characterized by a weak polarity and was detected at 230 nm with a 31-minute retention time in HPLC.

#### Résumé:

Un des grands mystères de l'écologie des poissons est de savoir comment les larves détectent les rares agrégats d'habitats sur lesquelles elles peuvent s'installer. La réponse doit se trouver dans le monde sensoriel des poissons car il est difficile d'imaginer que le succès de l'installation (trouver un habitat) soit seulement due à la chance. L'objectif du post-doctorat est d'estimer les capacités sensorielles des larves de poissons coralliens lorsqu'elles cherchent leur habitat d'installation. La reconnaissance de cet habitat implique la détection des conspécifiques ou de l'habitat grâce à l'émission de signaux visuels, chimiques et mécaniques. J'ai réalisé des expériences en laboratoire utilisant les larves de *Chromis viridis* (poisson demoiselle). Les larves de *C. viridis* sont attirées par les signaux visuels, chimiques et mécaniques émis par leurs conspécifiques. Les larves choisissent les compartiments des aquariums contenant les conspécifiques dans 75% des cas, et ne montrent aucune attraction vers les compartiments contenant des congénères ou l'habitat corallien. J'ai ensuite réalisé des analyses en HPLC sur de l'eau contenant des juvéniles de *C. viridis* et ainsi isolé des composés chimiques. Des expériences en aquarium ont ensuite démontrés que les larves de *C. viridis* étaient attirées par l'un de ces composés. Ce composé est caractérisé par une faible polarité et est détectable à 230 nm avec un temps de rétention de 31 minutes.

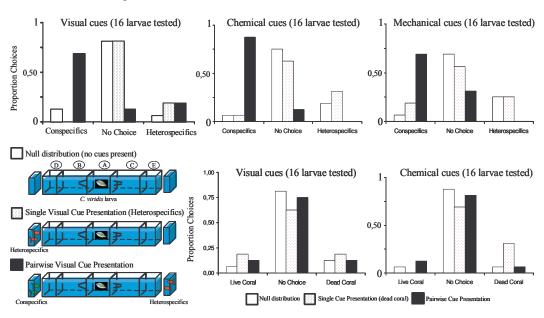
## 1) Introduction:

Perhaps one of the greatest challenges facing the majority of marine reef organisms with larval stages that potentially disperse and develop in offshore waters is how to relocate the relatively rare patches of coral reef habitat on which they settle and ultimately reside as adults. Once a larva of any taxon arrives upon a reef, it must locate a suitable micro-habitat in which to settle. Larval recognition of settlement habitat can be based on the detection of conspecifics already settled and/or of characteristics of coral habitat using visual (e.g. shape of a coral colony), chemical (e.g. an odour of anemone) and mechanical (e.g. vibratory or sound waves of fish) cues. The present study aimed to estimate the sensory modalities of coral reef fish larvae (example of *Chromis viridis*) for senses used in searching for their species' settlement habitat.

# 2) Laboratory experiments to determine ecological factors and sensory mechanisms underlying settlement choice

I used a series of laboratory experiments conducted on Kudaka Island (Ryukyu Archipelagos) to explore sensory abilities (visual, acoustic/vibratory, olfactory), and ecological determinants of settlement choices (conspecifics vs. heterospecific fishes vs. coral substrates) of *C. viridis* larvae. The experimental arena consists of an aquarium with five compartments (A–E), with A, B and C interconnected via funnels and D and E isolated from central compartments via plastic panels affixed with removable opaque barriers. Additional tanks on either side of arena are isolated from experimental arenas and mounted upon separate platforms to prevent transfer of vibratory signals. Experimental test subjects (fish larvae) are introduced into compartment A (one larva per arena), cues are presented in compartments B, C, D, E or tanks 1 or 2 (to test sensory mechanisms separately), and settlement choice is determined as the pattern of movement of test subjects to compartments B or C over 2-minute observation periods.

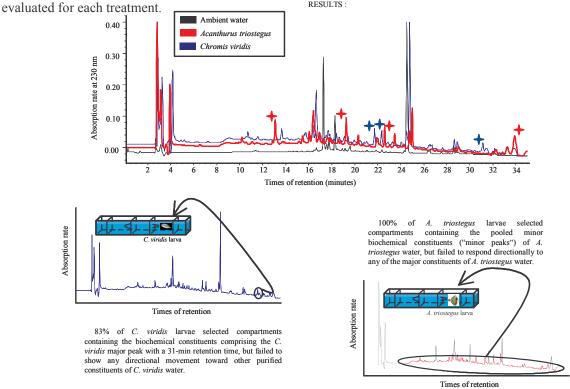
The main results acquired were illustrated below:



# 3) Biochemical analyses to detect, identify, isolate, and test specific chemical cues

I used high performance liquid chromatography (HPLC) to detect and isolate chemical compounds emitted by conspecifics and heterospecific fishes, and subsequently tested the attractive properties of these compounds on settling *C. viridis* in experimental arenas. I collected 2l of water from tanks containing (1) 10 juvenile *C. viridis* immersed for 6h, (2) 10 juvenile *Acanthurus triostegus* immersed for 6h, or (3) unoccupied ("ambient") filtered seawater. The three different seawater collections (*C. viridis* seawater, *A. triostegus* seawater and ambient seawater) were analyzed and compared using standard HPLC procedures. I collected dominant "peaks" of chromatograms (discrete biochemical constituents of water) in separate eppendorf tubes. These compounds (or sets of compounds of similar chromatographic properties) were then lyophilized and dissolved.

I identified three major peaks indicating compounds uniquely associated with *C. viridis* (asterisk symbol) four major peaks uniquely associated with *A. triostegus*, and I constructed three additional treatments comprised of (i) pooled minor peaks of *C. viridis*, (ii) pooled minor peaks of *A. triostegus*, and (iii) pooled major and minor peaks of ambient seawater. The five treatments of *C. viridis* (three major peaks and pooled minor peaks of *C. viridis* + pooled peaks of ambient seawater) were introduced singly into a compartment (either B or C) of the experimental arenas (filled with pure artificial water), and the responses of six individual test subjects (*C. viridis* larvae) were evaluated for each treatment (as described above). The same experiment was conducted with six treatments of *A. triostegus* (four major peaks and pooled minor peaks of *A. triostegus* + pooled peaks of ambient seawater) and the responses of six individual test subjects (*A. triostegus* larvae) were



# 4) Perpectives

There exists little doubt that the Earth's biodiversity is declining, and habitat destruction and degradation are commonplace. Areas experiencing perturbance exhibit declines in adult populations leading to extirpation at a higher rate than in pristine habitat, and the persistence of species in the area becomes reliant on the "rescue" effect of settlement. However, we do not know if the decline of marine organisms in degraded island is solely due to increased mortality of juveniles and adults of reef organisms, or to a decrease in the settlement potential of the reef. Many habitat cues are present on all reef states, but depending on the state of the reef, the cues present may vary and lead to vastly different outcomes in terms of the habitat choice of marine organism larvae. How larvae balance the use of available cues to choose a habitat, particularly in the absence of a key settlement inducer or preferred habitat (assumed to be present on pristine reefs), is not well described. This is a fruitful avenue for future research.