Ontogenic and ecological control of metamorphosis onset in a carapid fish, *Carapus homei*: experimental evidence from vertebra and otolith comparisons

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Introduction

In *Carapus homei*, reef colonisation is associated with a penetration inside a sea cucumber followed by heavy transformations during which the length of the fish is reduced by 60%. By comparing vertebral axis to otolith ontogenetic changes, **this study aimed (i)** to specify the events linked to metamorphosis and **(ii)** to establish to what extent these fish have the ability to delay it.

Fig.1

Fig.2

21 days 21 days 21 days

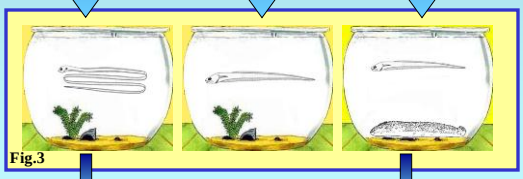

Fig.3

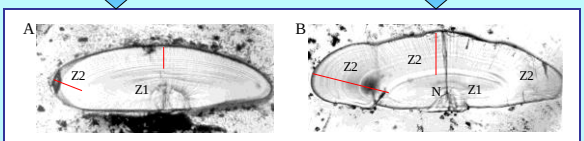

Fig. 4 Frontal sections of sagittae (A : 0,71mm – B : 2,38mm). Red line corresponds to the otolith growth since the day capture (= zone 2), N = nucleus, Z1 = zone 1.

Materials and Methods

27 Larvae of *Carapus homei* were collected when they arrived on the reef crest in front of the Opunohu bay (Moorea, French Polynesia) and on the Rangiroa atoll (French Polynesia).

Experimental conditions - duration : 7 – 14 - 21 days – **Fig.2**

1) Daylight: tank under natural lighting,

2) Dark: tank, shaded from natural daylight,

7) Holothurid: tank with presence of holothurian hosts (*Bohadschia argus*), which allowed fish to penetrate into them.

All the specimens were stained with alizarin Red S according to Taylor & Van Dyke's method (1985). The following data were collected: body and head lengths, sagitta length, numbers of vertebrae and of neural arches. In addition, more detailed observations were collected : the vertebra from which were noticed the early beginnings of the decalcification process and the number of non-calcified vertebrae.

Optical frontal sections of sagittae were obtained by grinding each slide and polishing them (Parmentier et al. 2003). All increments developed after capture were counted, irrespective of the numbers of days elapsed since day capture.

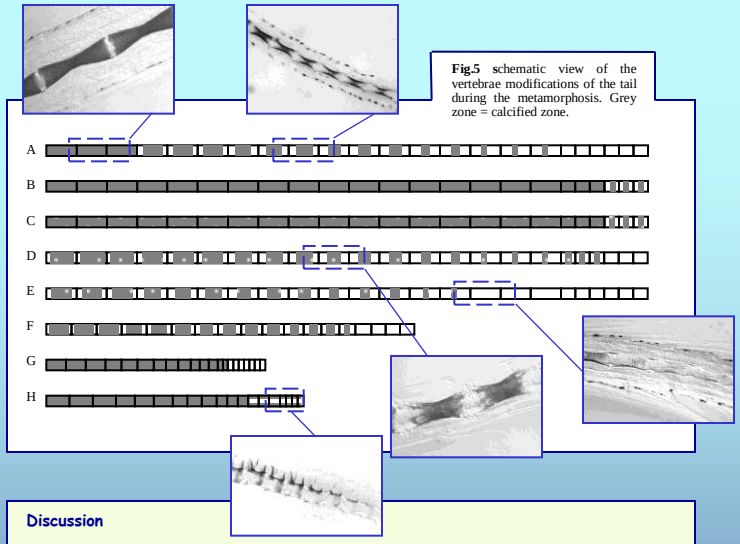

Fig.5 schematic view of the vertebrae modifications of the tail during the metamorphosis. Grey zone = calcified zone.

Results

Environnemental influence – Fig.3

After 21 days, the metamorphosis is complete for the larvae reared inside the sea cucumber whereas the fish reared in daylight are not metamorphosed. In dark condition, the metamorphosis is started but not yet completed.

Otolith Development – Fig.4 –

All the larvae used in the different experimental conditions have otoliths that show zone 1 (Z1) and a second zone (Z2) which corresponds to the metamorphosis stage. However, the number of increments seems dependent of the environmental rearing conditions : >80 increments in metamorphosed larvae and 21 increments in non- metamorphosed larvae.

Column development – Fig.5 –

For fish reared in the sea cucumber or in dark, it is possible to follow the metamorphosis of the column (stage A to H). Fish reared in daylight show only the stage B of development (= deeper calcification of the vertebral bodies).

1. The first vertebrae have the shape of a completely alizarin-stained diabolo. The alizarin staining is restricted to a single central band on the remaining vertebrae. Towards the caudal tip, this central band becomes progressively narrower until its complete absence on the last vertebrae.

2. The larvae of this group show vertebrae pricked with whitish spots , which corresponds to a decalcification process. (C)

3. The decalcification begins simultaneously at each tip of the vertebral body and seems to converge toward the centre. Depending on the specimen, the number of vertebrae showing only a thin alizarin-stained central band. Vertebrae of the caudal tip seem completely uncalcified (D et E).

4. It all seems as though the vertebra had undergone a process, analogous to the one observed in genetic, that could be called an ''excision-splicing'': the nonstained vertebral body parts are eliminated and the stained parts are brought closer together. (F and G)

5. New growth of the vertebrae where the calcification of the neural arches appears before that of the vertebral bodies. The latter show either an osseous matrix, or a start of calcification (H).

Discussion

1. Fish reared inside the sea cucumber and in darkness were the most developed. The sea cucumber could principally be considered as acting as a black box, and period of darkness seems sufficient to initiate metamorphosis whereas the natural light conditions delayed metamorphosis.

2. Comparaison between experiments 1 and 2 shows that the fish does not eat the sea cucumber during the metamorphosis. This suggests that fish could benefit from the organic material released by body reduction in order to use it in the development of the bones and the otoliths.

3. Whether the metamorphosis was initiated or not, otoliths were modified with the formation of a transition zone (Z2), whose structure varied depending on the experimental conditions. This study experimentally confirms that the formation of the transition zone is not induced by environmental variations linked to the settlement but depends on an internal signal since the zone 2 is present in tenuis reared in natural lighting and which showed a delayed metamorphosis.

➔ **First time** that a direct link between the **somatic development** and the **otolith development** is highlighted

➔ Sagittal development depends on an **internal signal** linked to the metamorphosis, somatic development depends also on **environmental conditions**

➔Onset in a Carapid Fish, Carapus homei: Experimental Evidence From Vertebra and Otolith **Comparisons. *JOURNAL OF EXPERIMENTAL ZOOLOGY* 301A:617–628 (2004)**

[1]Taylor WR, Van Dyke GC (1985). Cybium, 2 : 107-119.

[2]Parmentier E, Lagardère F & Vandewalle P (2002). Marine Biology, 141 : 491-501.