# THE EFFECT OF NUTRITIONAL CONDITION ON THE GROWTH TO POST-FLEXION OF BLUEFIN TUNA LARVAE UNDER CULTURED CONDITION

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## Introduction

The flexion of the notochord, as in many fish larvae, is considered a crucial point in the growth and mortality of scombrid larvae (Kaji 2003). The flexion, which it is associated with the caudal fin development, increases the swimming capacity of the larvae that benefits the capture of mobile preys (Margulies, 1993). Also, it coincides with the timing when the physiological capacities of larvae begin to develop significantly which allows an early shift to piscivory (Kaji 2003). Therefore, reaching the flexion as soon as possible can be considered beneficial for the growth and survival of the larvae and would probably determine which larvae reach the next crucial point, piscivory. However, scarce knowledge exists on which other characteristics could be associated with the different developmental rates under given culture condition.

In this study, we examined if the nutritional status of the larvae based on the RNA/DNA index could explain the differences in the development of the notochord flexion in laboratory reared Atlantic bluefin tuna (ABT) larvae. We also determined the daily ontogenetic developmental changes based on the timing of development of the notochord flexion at 28°C.

### **Materials and Methods**

ABT larvae were reared in four cylindrical tanks of 1.5m<sup>3</sup> volume, 1.5m diameter and 0.85m high. Water temperature was maintained constant at 28°C during the duration of the experiment. The larvae were only feed by enriched rotifers Origreen® (Skretting) from 2 dph to 17 dph and sea bream yolk-sac larvae from 14 dph to 20 dph when the experiment ended.

On days 8,10,11,12 and 13 dph 40 larvae were randomly sampled in all the tanks in order to follow at which age the different flexion phases appeared and to estimate the larval nutritional conditions. When >50 % of the sampled larvae were in the post-flexion developmental stage, the sub-sample was stopped and the diet was changed to larval fish to avoid cannibalism. To study the effect piscivory had on the nutritional condition of the larvae, another 40 larvae were sampled on 20 dph. Flexion phases were classified in four groups; F0: Pre-flexion, F1: development of the first caudal fin rays, F2: flexion and F3: post-flexion (Fig.1). Individually, sampled larvae were photographed and frozen at -80°C for further dry weight and RNA/DNA estimation. The quantity of RNA and DNA was determined of the whole body of the larvae following the method described by Caldarone et al., 2001, with slightly modifications. All statistical analyses were fitted using R statistical software.

## Results

At 28°C, caudal fins are first seen at 8 dph and from 10 dph, when the larvae start flexion of the notochord, three to four different developmental phases concur in the same tank on a given day. At 13 dph, more than the 80% of the larvae were already in the post-flexion phase. The largest larvae were typically those most developed (F3>F2>F1>F0). Mean length differences among the four developmental phases were evident.

A gradual increase of RNA and DNA contents per larva ( $\mu g$ /larva) was obtained until the end of the experiment. DNA content per larval dry weight ( $\mu g$ /mg) decreased with increasing development, unlike RNA content ( $\mu g$ /mg) where it is relatively constant (Fig.2a,b). RNA/DNA ratio increases with the development of the flexion phases (Fig.2c). The larvae that completed the flexion first show a higher RNA/DNA ratio than the others (Fig. 2c).

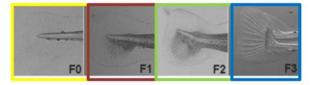


Figure 1. Different classified flexion phases. F0: pre-flexion, F1: first caudal fin rays, F2: flexion and F3: post-flexion.

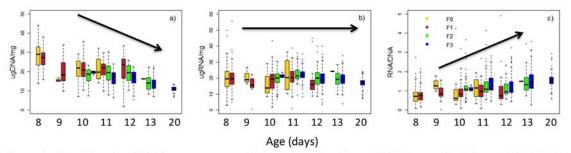


Figure 2. Boxplots for fish larvae content of a) µgDNA/mg b) µgRNA/mg and of the c) RNA/DNA index, sampled from 8 dph to 20 dph and at different developmental phases: F0, F1, F2 and F3.

#### Discussion

In our study, ABT larvae start the flexion (F2) on 10 dph at 28°C, which coincides with those seen in Pacific buefin tuna at 27°C (Fujimoto et al., 2008).

The decrease of the DNA content ( $\mu$ gDNA/mg) with the size of the larvae could be explained as a decrease in cell differentiation (hyperplasia) with the ontogeny of the larva. During the first days of life of bluefin tuna, the digestive system rapidly develops in order to be ready to start with the piscivory (Kaji et al., 2002). These rapid ontogenetic changes might mainly be caused by hyperplasia, while after, growth by cell enlargement (hypertrophy) might be predominant. The stability of the amount of RNA regardless the flexion developmental phase, indicates that all larval cell had similar protein synthesis capacity. The RNA/DNA index show differences of the nutritional condition depending on the developmental phases.

These findings indicate that protein synthesis increase in proportion to the flexion stage which is strongly associated with the development of the digestive system and therefore the increase in the nutritional condition. Ontogenetic differences in the RNA and DNA content will be discussed in more detail. Also, the pattern obtained for Atlantic bluefin tuna larva will be compared with those of other species.

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