

# RELATIONSHIP BETWEEN RNA, DNA AND PROTEIN CONTENT AND LARVAL DAILY GROWTH IN ALBORAN SEA SARDINE (*SARDINA PILCHARDUS*)

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## Abstract

A population of 124 sardine (*Sardina pilchardus*) larvae from the Bay of Málaga (Alborán Sea) were sampled during the peak spawning period (January, 1995) and analyzed individually for daily growth, nucleic acid and protein content. The daily growth pattern fits well a potential relationship, as well as, the allometric relationship between SL and otolith radius. The estimated daily growth rate was 0.71 mm/day for a 10 mm larvae and 0.42 mm/day for a 17 mm larvae. Larval size expressed in terms of SL, DNA and protein content follow likewise good potential relationships with otolith radius, wet weight and age. RNA/DNA and protein/DNA ratios show an increasing trend with size/age of the larvae.

**Key words:** larvae, growth, analytical methods, fishes, Alboran Sea

## Introduction

The Alborán Sea sardine (*Sardina pilchardus*) starts to spawn during early autumn (October), attains its peak spawning season in winter (December-January) and steadily declines its spawning activity till late spring (May). The spawning grounds are located off the coasts of Málaga, from the littoral waters attaining maximum densities at 100 m depth. The inshore waters of the Bay of Málaga (Fig. 1) is considered as their nursery grounds (1), and its coastal waters had been the most heavily exploited by an artisanal fleet targeting on the late larval stages of sardine (2).

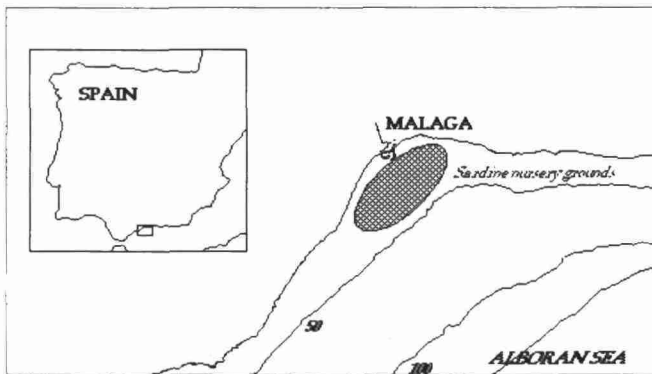


Fig. 1. Sardine larvae sampling area.

The fishing activity targeting on this resource normally works during nocturnal periods, particularly during the early morning hours before sunrise. Thus, most of the sampling was carried out at these times. Nevertheless, it was observed that sardine larvae abundance decreased as daylight hours progressed.

Sardine larvae were analyzed individually for daily growth analysis, RNA, DNA and protein content estimates. A total of 124 larvae were analyzed ranging from 8.4 to 28.3 mm (mean size at 17 mm).

Otolith microstructure analysis has proven in the past years a useful tool for recruitment studies. On the other hand, the analysis of larval condition through the estimation of nucleic acid content as a measurement of the nutritional status can aid in the assessment of the environmental impact on the biological variables of individuals. Well fed larvae and fast growing larvae show higher RNA/DNA ratios and wider daily increment deposition than starving larvae.

## Material and methods

The plankton sampling was carried out by means of short duration superficial tows ( $\approx 5$  min) with a WP-2 plankton net equipped with 1mm mesh. The depth range of the area of sampling varied from 10 m to 30 m off the littoral waters of the Bay of Málaga. During January 1995, sardine larvae were by means of a series of repetitive tows with a WP-2 plankton net (1 mm mesh) towed superficially. The depths covered ranged from 10 to 30 m depths, although generally most tows were in 15 m depth. The sampling was carried out during the early morning hours (sunrise) and late evening hours (dusk). The tows were in general around 5 minutes long. On board, the plankton sample was sorted for sardine larvae and conserved in liquid nitrogen.

The method followed for the joint analysis of otolith microstructure and nucleic acid and protein quantification was as follows. Larvae

were defrosted, measured and ulteriorly, otoliths extracted and mounted with common nail lacquer on glass slides. The same larvae were used for determining the nucleic acid content and protein content. The analytical methods followed are described in (3) and (4).

Sagittal otoliths were used for daily increment counts. Otoliths were projected to a video monitor and the increments were counted in the screen by means of a specifically designed software for increment counts (5). Increments were counted after the visualization of the check ring which defines the first feeding stages of the larvae. The software package gives an output of the resulting increment counts and the increment widths.

The statistical analysis carried out for the different regressions were done with the statistical package KaleidaGraph Version 2.1.0 running on a MacIntosh computer.

## Results and discussion

Daily growth fits a potential relationship ( $R=0.96$ ) (Fig. 2). The instantaneous growth rate calculated for larvae of 10 mm was 0.71 mm/day, while for larvae of 17 mm (mean size of analyzed larvae) was 0.42 mm/day. These values are higher than those observed for the Portuguese sardine (6). Nevertheless, the daily growth pattern of the Portuguese sardine was fit to a linear relationship because the size ranges sampled were limited to the early larval stages. However, this difference can also be due to the characteristics of the sampled area. The area has been traditionally considered as ideal for sardine and anchovy nursery grounds due to its particular hydrological conditions. It is characterized by well mixed waters. The wind regime enhances enrichment processes which eventually favours feeding availability.

The allometric relationship between size and otolith radius also followed a potential relationship ( $R=0.97$ ).

DNA and total protein content are intimately associated with larval cellular mass, and thus, their quantity is related to their relative growth status. Consequently, these show high correlations with larval size, otolith radius, wet weight and age (Table 1). On the other hand, RNA, as an index of protein production, shows a higher variability since it is mainly dependent on the success of feeding during the early life stages (Table 1).

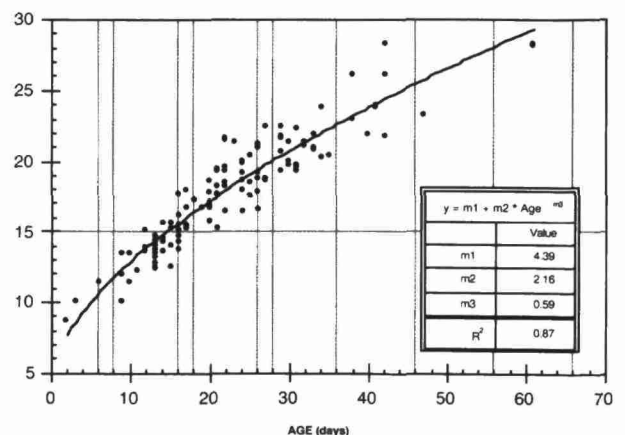


Fig. 2. Size (SL) of sardine larvae vs age.