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## Primary Growth Rings in Otoliths of Some Clupeids from Sri Lanka

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#### Abstract

This paper describes the otolith structure and the pattern of ring deposition of four species belonging to the family Clupeidae from the coastal waters of Sri Lanka.

The otoliths of all these species were found to contain primary growth rings which could be used in age determination. The structure of the otolith and the ring deposition pattern of the clupeids studied were similar to each other and to Sardinella spp.

Age estimates were made by assuming that these rings are formed daily. Dussumieria acuta and Ilisha melanostoma seem to grow similar to Sardinella, attaining a length of about 14 cm and 11 cm, respectively, in one year, while Escualosa thoracata reach a size of about 9 cm in one year. Opisthopterus tardoore grow relatively fast to a size of about 8 cm in 4 months. Calculated birthdates were in agreement with known spawning seasons except for O. tardoore. This could be the result of bias due to poor ring visibility.

## Introduction

The use of primary growth rings in otoliths for age determination is a recent development. Pannella (1974) recognized fine, closely arranged growth markings on the otoliths of tropical fishes and suggested that they were laid down on a daily basis. Since then, the number of tropical species found to have primary growth rings has steadily increased.

In a recent review Gjøsaeter et al. (1984) showed that all the tropical fish species studied contained primary growth rings. The daily periodicity of these rings has been firmly verified for 11 species. Another five species, although not directly verified, also appear to show daily rings.

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This paper includes observations on otoliths of four species of the family Clupeidae from the coastal waters of Sri Lanka. The main objective was to ascertain whether the primary growth rings can be counted and whether they can be used in age determination. Direct verification studies were not made. However, the results favor the hypothesis that the rings are formed daily.

## **Materials and Methods**

A total of 177 fish otoliths from four clupeid species collected from coastal fisheries were studied. Details are given in Table 1.

|                           | 10 11 10 10 10 10 10 10 10 10 10 10 10 1 |                    |                      |        |  |
|---------------------------|--|--------------------|----------------------|--------|--|
| Species                   | Sample<br>Place                          | collection<br>Date | Length range<br>(cm) | Number |  |
| Dussumieria<br>acuta      | Kalkudah                                 | 12.1.81            | 13.2-14.5            | 50     |  |
| Ilisha mela-<br>nostoma   | Negombo                                  | 16.6.83            | 10.2-12.3            | 41     |  |
| Escualosa<br>thoracata    | Negombo                                  | 16.6.83            | 7.6-9.1              | 45     |  |
| Opisthopterus<br>tardoore | Chilaw                                   | 16.6.83            | 5.9-9.8              | 41     |  |
|                           |  |                    |                      |        |  |

Table 1. Details of the material studied.

Length measurements and otolith extraction of all the samples were carried out while they were in fresh condition. The otoliths were stored dry in paper envelopes. The methods used for otolith extraction and preparation were as described for *Sardinella* spp. (Dayaratne and Gjøsaeter 1985).

Otoliths were viewed through a light microscope at  $250 \times and 400 \times magnification$ . Rings were counted from the nucleus towards the outer edge along the longest radius (Fig. 1). The radii of the otoliths were measured along the same axis. The number of rings in the nuclear region were noted for each species. The width of the rings in different regions of the otoliths was also measured in most species.





## **Results and Discussion**

## Fish Length/Otolith Radius Relation

The fish length/otolith radius regressions of the 4 clupeids studied are shown in Table 2. As the length ranges of the samples are very narrow, the regressions on fish length/otolith radius are not very satisfactory, as indicated by the low correlation coefficients and the high values of elevations (Table 2). The fish length otolith radius relation for Amblygaster sirm and Sardinella spp. with wider length ranges have shown linear relations passing through the origin with high correlation coefficients (Dayaratne and Gjøsaeter 1985). Forcing these regressions through the origin gave values of the resulting slopes shown in Table 2.

|                | Regression<br>analysis |     | Coefficient<br>of<br>determination |    | Value of b<br>obtained for<br>regression |  |
|----------------|------------------------|-----|------------------------------------|----|--|--|
| Species        | b                      | a   | r2                                 | N  | through origin                           |  |
| D. acuta       | 3.4                    | 8.4 | 0.22                               | 50 | 8.6                                      |  |
| I. melanostoma | 4.2                    | 5.1 | 0.74                               | 34 | 7.8                                      |  |
| E. thoracata   | 2.8                    | 5.6 | 0.24                               | 40 | 8.4                                      |  |
| 0. tardoore    | 5.4                    | 1.4 | 0.60                               | 41 | 6.6                                      |  |

Table 2. The regression analysis of the fish-length otolith-radius relation. Equation used was: L = bR + a, where L = fish length in cm and R = otolith radius in mm.

# Structure of Otoliths and Pattern of Ring Deposition

The otoliths examined were more or less similar in shape and structure to those of Sardinella. Dussumieri acuta otoliths were easy to read and the rings were clear from the central nucleus to the outer edge. The otoliths of Escualosa thoracata were transparent enough but the rings were not clear throughout. The otoliths of Ilisha melanostoma and Opisthopterus tardoore were less transparent and even after grinding it was difficult to see the rings in the central area.

The three regions of the otoliths (A, B and C in Fig. 1) were distinct in *E. thoracata*, *D. acuta* and *I. melanostoma*, while the demarcation of the nuclear region from the central region was difficult in *O. tardoore*. The number and the thickness of the rings in these regions differ considerably between the species (Table 3). The nuclear region (A) in the otolith is assumed to represent the period of larval growth. Accordingly, these fish take 14-22 days to complete metamorphosis. This is true only if rings are deposited daily and if ring deposition begins just after hatching.

The first assumption is reasonable as the daily nature of these rings has been verified for a similar species (Amblygaster sirm) from the same waters (Dayaratne and Gjøsaeter 1985). The exact time of the first ring formation is not known for these species. It has been shown that for *Clupea harengus* the otolith ring deposition is initiated about 5 days after hatching at the end of yolksac resorption (Rosenberg and Lough 1977; Gjøsaeter 1981). Brothers et al. (1976) found that the first daily growth ring in *Engraulis mordax* was formed only after yolksac absorption.

| Species        | - 1708-11              |                         |    | No. of rine               | 76   |     | Average ring             |        |  |  |
|----------------|------------------------|-------------------------|----|---------------------------|------|-----|--------------------------|--------|--|--|
|                | Fish<br>length<br>(cm) | Otolith<br>size<br>(mm) | N  | in nuclear<br>region<br>X | s.D. | A   | width (µm<br>region<br>B | )<br>C |  |  |
| D. acuta       | 14                     | 3.3                     | 10 | 21.2                      | 1.1  | 91  | 16.8                     | 2.7    |  |  |
| I. melanostoma | 11.5                   | 3.1                     | 10 | 17.9                      | 1.9  | 7.4 | 8.0                      | 2.7    |  |  |
| E. traracata   | 8.0                    | 1.9                     | 10 | 14.9                      | 1.2  | 6.6 | 8.8                      | 2.2    |  |  |
| 0. tardoore    | 7.5                    | 2.2                     | 10 | 21.0                      | 2.3  | 7.5 | 12.0                     | 5.7    |  |  |

Table 3. Otolith size and ring width in different regions of the otolith.

The present results are therefore only a tentative measure of the length of the larval period for each species. The variance observed within the species (Table 3) could be an indication that the length of this period is species specific characteristic.

If these rings are formed daily, their thickness could be used as an indication of the growth rate of the otolith and consequently that of the fish. Accordingly, *D. acuta* larvae (with an average ring width of 9.1  $\mu$ m) may grow faster than the other species (Table 3).

The central region (B) of the otoliths of all the species consisted of dark broad rings. This region represents a period of fast growth which lasts about 1-2 months. As observed from the average thickness of the rings in this region, *D. acuta* and *O. tardoore* with average ring width of 16.8  $\mu$ m and 12.0  $\mu$ m, respectively, seem to grow faster than the others. In these two species, 3-4 narrow rings were seen within the thick broad rings. This feature is common in many tropical fishes which grow faster than those from cold temperate waters (Pannella 1974). These narrow rings are thought to be subdaily increments but their environmental or physiological significance is unknown.

Subdaily rings have been observed in the early growth records of *Lepomis gibbosus* (Taubert and Coble 1977) and of *Oncorhyncus nerka* (Marshall and Parker 1982). According to Brothers et al. (1976), these subdaily patterns are present in sagittae of larval fishes and often are not distinguished from daily increments. In clupeids, it is easy to distinguish the subdaily increments from the daily increments. The former are found only within the thicker rings which correspond to the fast growing periods.

A gradual reduction in the thickness of the rings was observed from region B towards the region C. This was the case in all the species, indicating a gradual decrease in growth rate.

According to the present age estimates made by counting these rings, O. tardoore were about four months old. The other species had estimated ages of around 10-12 months (Table 4). Their very narrow rings (2.2-2.7  $\mu$ m) (Table 3) may be an indication of a reduced growth rate at this age.

Ring interruption was not very common among the clupeids studied, although few interruptions were occasionally seen in *D. acuta* and *E. thoracata*. An interesting feature observed in region C of some species is the occurrence of periodical markings. Groupings of about seven rings ( $\overline{X} = 7.2$ , S.D. = 0.83) and at times fourteen rings ( $\overline{X}$ = 13.2, S.D. = 2.2) were frequently seen in *O. tardoore*. Because of their periodicity the factors producing these patterns have been suggested to be related to or controlled by tidal fluctuations (Pannella 1980).

|                | Length<br>(cm) |      |      | Estimated age |       |      |  |
|----------------|----------------|------|------|---------------|-------|------|--|
| Species        | N              | X    | S.D. | N             | Ť     | S.D. |  |
| D. acuta       | 50             | 13.8 | 0.36 | 50            | 339.2 | 30.5 |  |
| I. melanostoma | 40             | 11.2 | 0.64 | 35            | 357.0 | 23.4 |  |
| E. thoracata   | 40             | 8.3  | 0.32 | 40            | 293.3 | 29.8 |  |
| 0. tardoore    | , 41           | 7.6  | 0.96 | 37            | 124.7 | 21.0 |  |

Table 4. Estimated age and mean length of the fish studied.

## Length-at-age Data

The available material is not sufficient to give direct growth estimates. Ages were estimated by assuming that the primary rings found in the otoliths are formed daily. The mean age estimates are given in Table 4.

According to the present growth estimates, *D. acuta* seems to grow as fast as the *Sardinella* spp. (Dayaratne and Gjøsaeter 1985), attaining a length of about 14 cm in one year. The fact that the otolith structure and pattern of ring deposition is very similar to *Sardinella* spp. and that they are from the same environment bring some confidence on the age estimates.

The age of I. melanostoma studied (mean length 11.2 cm) is about one year (Table 4). No comparative information is available. However, the rings towards the outer edge (region C) of the otolith were very narrow and at times difficult to see. It may be that these narrow rings are under the detection power of the light microscope as discussed by Morales-Nin (1988). Therefore, there is a possibility for the present values to be an underestimate. Nevertheless, the presence of clear rings in the otolith from the nuclear region at least up to the end of the region B, suggests that they could be used for age determination at least to about 8-9 months.

According to the present growth estimates, *E. thoracata* with a mean length of 8.3 cm are around 10 months old (Table 4). This is in broad agreement with age estimates and growth studies using monthly length-frequency distributions (Nair 1952) which reveal that these fish are about 10-11 cm in length when they are around one year old.

The length range of O. tardoore used was 5.9-9.8 cm and the estimated ages showed that even within this range there was a clear increase in number of rings with length (Fig. 2). A direct estimation of growth was not possible with the material available. However, the present age estimates indicate that this species is relatively fast growing, attaining a length of around 8 cm in about 4 months. This is supported by the observations of fairly wide rings in region B of the otolith.

The minimum size at maturity of this species is given as 14 cm (Radhakrishnan 1963). It is probable that the fish used in the present study were immature and a faster growth could be expected in the juveniles.



Fig. 2. Opisthopterus tardoore. Plot of estimated age versus fish length.

## **Estimated Spawning Seasons**

The ages of the fish estimated by counting in primary growth rings were used to calculate their spawning season (birthdays). The results are shown in Fig. 3 compared with known spawning periods.

The samples of *D. acuta* were caught in January and were fully mature. This indicates that spawning takes place around this time of the year. The calculated birthdates also suggest a spawning period from December to March. This fits well with the known spawning for this species from the northern Australian waters (Okera 1982).

The estimated spawning season for *I. melanostoma* is from April to June. No comparative information is available on the spawning of this species.

E. thoracata has an estimated spawning from September to February which coincides with the known spawning from the Indian waters (Nair 1952).

The calculated spawning of *O. tardoore*, however, precedes the known spawning season by about two months. The estimated spawning is from December to March while the available information from the Indian waters shows that it is from February to July (Radhakrishnan 1963).

The age estimates of some of these fishes, especially of the older ones, could probably have been biased due to the poor ring visibility, counting errors, etc. This would have in turn biased the estimates of the spawning season.



Fig. 3. A comparison of the estimated spawning seasons of four Sri Lankan clupeids with known spawning seasons.

The present study reveals that juvenile fish (6 months to 1 year old depending on the species) can be reliably aged by using the light microscope. According to Campana and Neilson (1985) the identification and interpretation of the microstructural features is more important than the use of one observation technique or another.

The structures that can be confused with daily growth increments under a light microscope, such as subdaily increments and some checks, can also be confused when viewed by scanning electron microscopy. The validation of daily increment formation is, therefore, a prerequisite to any otolith microstructural study.

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