# DAILY LINES OF GROWTH AS COMPONENTS OF THE SPAWNING BANDS IN THE OTOLITHS IN A TROPICAL FISH (*SPARISOMA AUROFRENATUM*) (CUVIER AND VALENCIENNES, 1839)

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ABSTRACT. The chances of characterizing spawning bands, as formed by thinner and more refringent daily lines of growth are commented. The possibilities of establishing a reproduction calendar, and sex patterns are suggested.

## INTRODUCTION

Pannella (1974) indicated the confusion existing between the terms "hyaline", and "transparent" applied to bands or rings of growth. Since the difference between them is mainly derived from observations on otoliths using either transmitted or reflected light, Pannella (*op.cit.*) prefers to speak about zones of fast and slow growth.

The recent interest on determination of age through the use of daily lines of growth (Pannella 1971, 1974, 1977; Miranda 1977) opens a new and wide field on the analysis of growth checks. Struhsaker and Uchiyama (1976) gave direct evidence that these daily lines of growth are measures of daily increments in fishes from Hawaii. They actually substantiated a net correlation between age, and size of the fish.

Brother *et al.* (1976) reported that daily increments in *Engraulis mordax* appear after the reabsortion of the viteline portion in the larvae. Besides this, the number of the daily lines ($= 1_d$), and the quantity of checks coincide with the time (in days) controlled in a life span. The same was observed with *Leuresthes tenuis*, but the $1_d$ line appeared at the moment of ecclosion. A recent variable, added to the determination of age,

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using daily lines, is the subdaily increments commented by Tauber and Cobble (1977).

Heald and Griffiths (1967) claimed the possibility of detecting the age of tropical species using the "winter rings", in a similar manner as it has been done with species of cold and temperate waters. They referred to Menon (1953), Klima (1959) and Volpe (1959) utilizing the same concept for tropical, and subtropical species. Pannella (1974) doubted the validity of such conclusions suggesting that those marks were probably spawning instead of seasonal checks.

This last hypothesis is tested by controlling, and establishing the relationship between the hyaline mark of the edge of the otoliths of ripe or just spent females, with the counts of daily lines.

New approaches for the physiology and spawning pattern, of the ovary maturation for the anchovy (*Engraulis mordax*), are given by Hunter and Goldberg (1980), they establish a nigthly and discontinuous process. Similar observations are lacking for anchovy males.

The intention of this report is to show further research possibilities, derived from the application of the recognition of spawning lines and bands, in connection with the physiology of maturation, spawning, ejeculation and sexual cycles for females and males. The chances of establishing a sexual reproduction pattern, and sexual reversal in fish are also considered.

## MATERIAL AND METHODS

It is difficult to get samples of mature, or spent males and females from fishes of the tropical reef. The material used came from a collection of fishes sampled in front of La Parguera, Puerto Rico, from September 20 until December 10, 1977. Otolith from mature females were prepared using dorso-ventral, and sagittal sections. From these only 8 (Table 1) are clear enough to be considered as readable microscopic preparations. The method used in the replication of the otoliths sections using acetate peels are those described elsewhere by Pannella (*op.cit.*). The best, and clearest preparations of the samples were used for reading the $l_d$ (daily lines). The regression between standard length of the fish and age in days was fitted using a Texas SR-52 calculator. The time series analysis of the daily increments, was run at the Computation Center of the University of Puerto Rico, using programmes of Davis and Sampson (1973), including auto-correlation

and smoothing, in FORTRAN.

## Otolith description

The otolith is compact. Hyaline and opaque bands are evident in reflected light. After polishing, daily lines are visible.

The ventral edge of the sagitta is sharp and the dorsal one is waved. The acoustic channel is open at both ends. The angular ostium separates the rostrum and antirostrum. The dorsal area is smaller than the ventral one (Fig. 1).

## Reproduction checks

Standard $l_d$ lines are observed on figure 2. Daily lines are similar, and no refringence is noted. A sagittal preparation of a specimen from a distinct spawned female (Sa-32, table 1) showed a hyaline ring or band, at the margin. Besides the standard lines of growth (daily lines) of similar aspect,other daily lines of growth formed a cluster of bright refringence. These appeared conspicuous, clear and transparent in the replica. This pattern suggests the characteristic feature of a spawning band ($= s_b$), formed by reproduction lines (fig. 3). The reproduction lines ($= l_R$) were brighter and thinner than the $l_d$ lines. These results complement those given by Rollefsen (1934) with regard to the spawning bands. A disrupted disposition of the concentric rings, at the side of the spawning checks (fig. 3), suggests that reabsortion has occurred.

The existence of $l_R$ in otolith replicas of males was also observed. The reproduction lines appeared forming a translucent band. The specimen Sa-85 (table 1) showed very fine daily lines in clusters of 10 to 15 at the edge of the sagitta. In the rest of the surface of the otolith, neither bands nor reproduction lines were observed.

Regarding the otoliths of mature females, no reproduction checks (bands or $l_R$ lines) were observed in the specimen (Sa-100); (see table 1). The estimated age of this specimen was $\sim$ 240 days. The specimen Sa-90; (Table 1) had a hyaline edge of 15 $l_R$, with an estimated age of 200 days, this was obtained by counting the $l_d$ lines of growth. The specimen Sa-43 was estimated to be of an age of 356 days; there were no reproduction checks in any part of either surface or edge, but, another female of SL = 140 mm of an

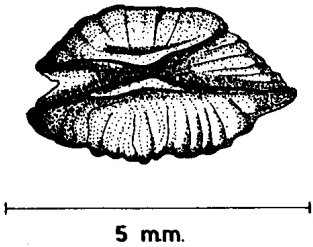


Fig. 1. Sagitta of *Sparisoma aurofrenatum*.

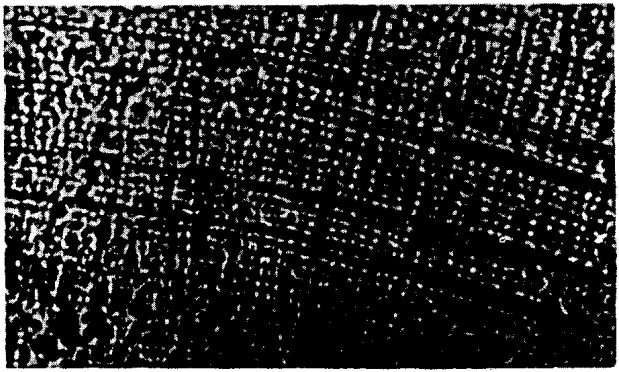


Fig. 2. Standard daily lines of growth.

age of 350-380 days had a reproduction band of 10-15 $1_R$; some other $1_R$ lines were observed in the antirostrum. The female Sa-31; of an estimated age of 170-180 days had a reproduction band close to the edge of the otolith with the following calendar of resting lines, $1_d$ lines, and reproduction $1_R$ lines:

| Margin | Oct. 12th 1977 | (Date of collection) |
|---|---|---|
| 2 $1_d$ | Oct. 10-11 | Period of resting |
| 1 $1_R$ | Oct. 9 | Period of reproduction |
| 6 $1_d$ | Oct. 3-8 | resting |
| 1 $1_R$ | Oct. 2 | reproduction |
| 2 $1_d$ | Sept. 30-Oct. 1 | resting |
| 1 $1_R$ | Sept. 29 | reproduction |
| 2 $1_d$ | Sept. 27-28 | resting |
| 1 $1_R$ | Sept. 26 | reproduction |
| 4 $1_d$ | Sept. 22-25 | resting |
| 12 $1_R$ | Sept. 10-21 | reproduction |
| 4 $1_d$ | Sept. 6-9 | resting |
| 1 $1_R$ | Sept. 5 | reproduction |
| 4 $1_d$ | Sept. 1-4 | resting |
| 2 $1_R$ | August 30-31 | reproduction |
| 6 $1_d$ | August 16-21 | resting |
| 1 $1_R$ | August 15 | reproduction |
| 1 $1_d$ | August 14 | reproduction |
| 13 $1_R$ | August 1°-13 | reproduction |
| 14 $1_d$ | July 28-31 | resting |
| 10 $1_R$ | July 18-27 | reproduction |
| 4 $1_d$ | July 14-27 | resting |
| 5 $1_R$ | July 9-13 | resting |

Two models of reproduction checks could be derived from this spawning calendar. One would have a daily pattern with resting periods from 2 to 6 days, the other one would be characterized by a continuous spawning of periods of 10 days or longer.

The series of daily line increments (Fig. 4) of a ripe male (Sa-38); age ∿247 $1_d$, showed hyaline margins (reproduction checks) wich formed the following calendar:

| Margin | Oct. 14 th. 1977 | (Date of collection) |
|---|---|---|
| 12 $1_R$ | Oct. 2-13 | reproduction |
| 25 $1_d$ | Sept. 7 - Oct. 1 | resting |
| 6 $1_R$ | Sept. 1-6 | reproduction |
| 38 $1_d$ | July 25 - August 31 | resting |
| 6 $1_R$ | July 19-24 | reproduction |

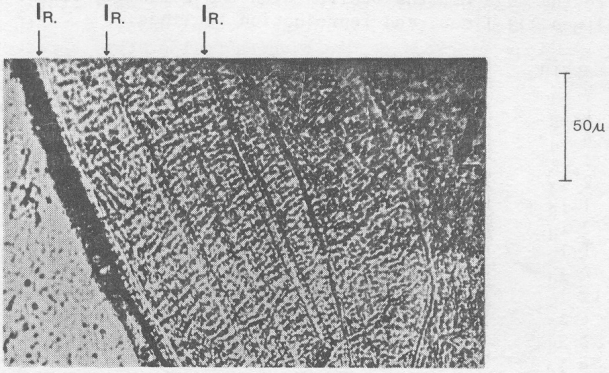


Fig. 3. Spawning bands ($S_b$) with reproduction lines ($1_R$) (female).

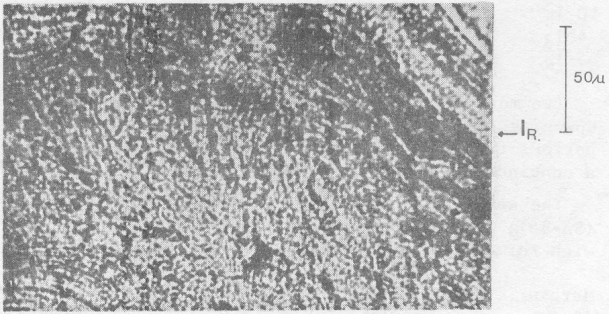


Fig. 4. Daily lines and reproduction lines in a male.

| | | |
|---|---|---|
| 18 $l_d$ | July 1-18 | resting |
| ±10 $l_R$ | ±June 21-30 | reproduction |

The reproduction of females could be characterized as a continuous process ocurring during several days. In both sexes there are long periods of rest which follow the reproduction events. Male and female patterns are different. In accordance with the continuity or discontinuity of the $l_R$ lines and the sequence of the $l_d$ lines it is possible to diferentiate sexes. As this species has a sex reversal, more experimental evidence is necessary to evaluate the chances of using the otoliths as a calendar of sex reversal.

A lag-correlation analysis of a periodogram of daily increments (Fig. 5) of these samples, and the spectrum of the smoothed power values (Fig. 6) showed harmonic numbers of multiples and submultiples of tidal or synodic periods, suggesting the existence of a daily growth rythm. In ripe specimens Sa-96, and Sa-79, $l_R$ this was not detected.

## Periods of reproduction

According to the suggested calendar of sexual activity, lengths of reproduction and resting periods were compared for both sexes. This was based on otoliths collected at similar dates. The sectors of otoliths extending from the edge (date of collection) to the last reproduction line were compared. For the female, the counts were made from July 27 to October 9, both days included, while for the male the count comprised July 1 to October 13; both days included.

For a female ∿240 days old a reproduction "season" of 74 days was found, while for a male 280-310 days old the reproduction "season" was 105 days. During this season the female had 35 days of active reproduction (35 $l_R$); the male had only 24 [(days of active reproduction)] (24 $l_R$). The female used only 33% of her "season" in actual spawning while the male used only 22%.

The mean number of reproduction lines per band (amount of reproduction lines) for the female was 45/11 = 4.09 lines; $s^2$ = 22.08; s = 4.69. The male had a mean of 34/4 = 8.50; $s^2$ = 6.75; s = 2.53. The variance shows a contagious distribution.

The mean number of resting lines for the female was 39 that is 52.7% of the reproduction season. For the male it was 81 that is 77.1% of the reproduction season. The mean number of $l_d$ per band

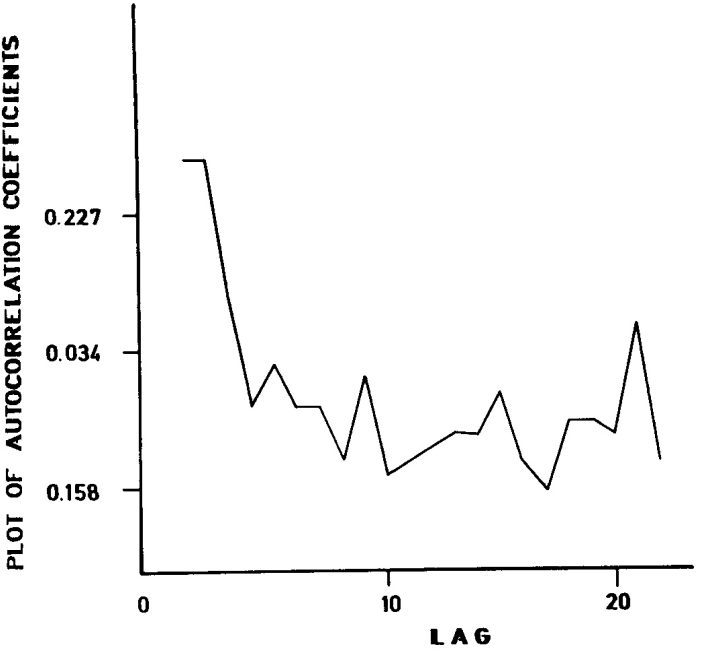


Fig. 5. Correlogram of daily increments.

was 39/10 = 3.9 ± 1.7 for the females, and the mean number of $1_d$ per band was 81/3 = 27.0; ± s = 8.28 for the males.

## Growth

An exponential growth curve (Fig. 7) was fitted to the 15 determinations of age in days. An r = 0.419 was found, which is not acceptable at p = 0.05. In any event the equation fitted for the tendency is: Y = 52.28 e o.011 Y (Y age in days; Y - SL in mm).

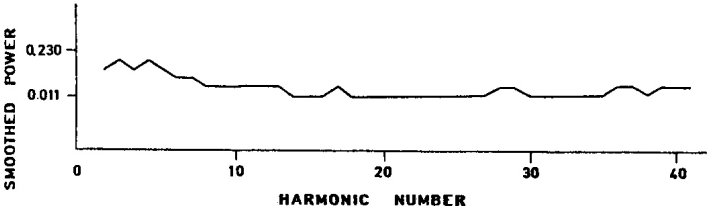


Fig. 6. Smoothed power of a series of daily increments.

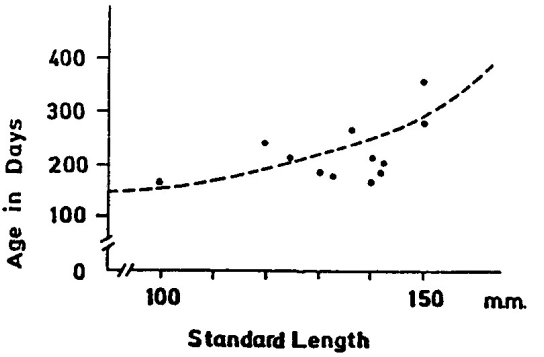


Fig. 7. Relationship between standard length and age in days.

## DISCUSSION

Several authors have demostrated the existence, and validity of daily lines as growth increments in otoliths. Since some authors have considered erroneously the existence of hyaline rings as annual marks, and due to the fact that these are similar to those found in temperate species, daily lines are essential in age determination of tropical fish. Through the description of these checks correlated with spawning activity, Pannella's warning (1974) not to confuse reproduction checks with annual rings becomes more evident.

Spawning lines or bands were not found in every fish considered mature. Perhaps the observation of the maturity phenomena was not precise enough to detect the coincidence of spawning or ejaculation with the registration of the mark on the otolith.

The existence of a spawning band in the tropical fish *Sparisoma aurofrenatum* (Cuvier and Valenciennes, 1839), which correlates with the hyaline margin of otoliths of just spawned or spent female and male fishes, is important. This spawning band, or reproduction check ($1_R$) has a variable quantity of normal daily lines ($1_d$) and/or reproduction lines ($1_R$). The reproduction lines, forming the spawning band are more refringent and thinner than the normal lines of growth, and through their counting and interpretation the possibility of establishing a reproduction calendar is suggested. In a calendar of this type the days of reproduction, spawning, and resting can be interpreted. In accordance with the "readable" checks in females and males the following observations can be made:

Females of age 240 days and SL= 118 mm had shorter reproduction "seasons" (74 days) than males, of age of 280-310 days and SL = 150 mm ($1_R$ = 105). The female used 33% of the reproduction season in 35 effective days of spawning, the male used 22.8% with a total of 24 effective days of spawning. The female had an average of four days of continued fertility and the male twice as much. The high variances observed indicated a contagious distribution of the reproduction. In other words, the female had a shorter period of continued resting, than the male.

The reproduction of females is concentrated in short periods having less fertile days. These alternate with resting days per period. Males would have a longer reproductive season and considerably more fertile days (per period) than females. Resting intervals of males are longer than those of females. Assuming that

there are different patterns for males and females, sex reversal could be detected.

A lag correlation, and power spectrum analysis of measurements of daily increments, suggests harmonic values as multiples and/or submultiples of tidal or synodic periods.

An exponential line of growth was fitted to the relationship between age in days (x) to standard length (Y in mm), but r was not significant at p = 0.05.

The statistical treatment and analysis of the observations, show great possibilities for further experimental design, for applications of the results, and for interpretations of reproductive patterns.

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* Not consultated by the author (mentioned by Heald and Griffiths, 1967).

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Table 1. Details of the samples of *Sparisoma aurofrenatum* (Cuvier and Valenciennes, 1839) ripe specimens; S.L. = standard length; $l_d$ = daily lines; $l_R$ = reproduction lines; G.W. = gonad weigth; B.W. Body weigth.

| N° | Sample # | Sex | S.L. mm | # $l_d$ (age) | (edge) $l_R$ # | G.W. g | B.W. g | Date of collection |
|---|---|---|---|---|---|---|---|---|
| 1 | Sa-100 | | 118 | ∿ 240 | none | 0.68 | .56 | Dec. 10, 1977 |
| 2 | Sa- 96 | | 158 | - | none | 2.66 | 1.32 | Dec. 8, 1977 |
| 3 | Sa- 90 | | 142 | 200 | 15 | 0.52 | 92.0 | Nov. 28, 1977 |
| 4 | Sa- 85 | | 148 | 225-285 | 10-15 | 0.07 | 101.0 | Nov. 21, 1977 |
| 5 | Sa- 79 | | 133 | 175 | none | 0.55 | 83.0 | Nov. 19, 1977 |
| 6 | Sa- 43 | | 150 | 356 | none | 1.00 | 116.0 | Oct. 14, 1977 |
| 7 | Sa- 38 | | 150 | 247 | 12 | 0.12 | 116.0 | Oct. 14, 1977 |
| 8 | Sa- 31 | | 142 | 170-180 | 7 | 0.42 | 100.0 | Oct. 12, 1977 |
| 9 | Sa- 32 | | 141 | 141-150 | 10-15 | 0.58 | 86.0 | Oct. 12, 1977 |
| 10 | Sa- 2 | | 140 | 350-380 | 10-15 | 1.3 | 90.0 | Sept.20, 1977 |