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# Note on Observations of Daily Rings on Otoliths of Deepwater Snappers



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# Introduction

To encourage the development of deep-sea fishing in Vanuatu, the Office de la Recherche Scientifique et Technique d'Outre Mer (ORSTOM) is studying growth in some of the major species: *Etelis carbunculus, E. coruscans, Pristipomoides multidens* and *P. flavipinnis.* Traditional techniques for estimating age from rings on hard structures (otoliths, scales and dorsal spines) cannot be used, because seasonal rings, if they exist, are not easily discernible and because the environment (temperature, salinity, oxygen level and concentration of other salts) varies little at depths between 200 and 300 m in the tropics.

We were unable to differentiate age groups of these fish by modal progressions in size distribution. Therefore, we decided to study rings on thin sections of otoliths. This technique, first suggested by Pannella (1971), is being used successfully in the tropics, notably in Hawaii (Uchida et al. 1982; Uchiyama, pers. comm.; Ralston 1976). The major difficulty lies in establishing the frequency at which the rings are laid. Results shown here depend on the validity of the hypothesis that the rings observed are produced daily.

## Methods and Equipment

Otoliths observed were from fish caught by the Fisheries Department in 1982 off Port-Vila. Fish were measured, weighed and sexed; the growth stage was identified and gonads were weighed. Otolith samples were taken only from the sagittas, not from the other two pairs of otoliths. As the catches were to be sold, otoliths were taken from the interior of the skull, after gills had been removed, so that the fish were not damaged. Otoliths were cleaned and kept dry in small paper bags without any other special care.

There were three stages in preparing otoliths: embedding, sectioning and acidification. A polyester resin (100 ml of Polylite 61203 resin and 4 ml methylethylketone catalyst), which hardens sufficiently in 24 hours, was used for embedding. Each embedded otolith was stuck to a glass slide (Plate 1).

A diamond saw (Plate 2) was used to make  $100-\mu$ m sections. Hydrochloric acid in 10% solution was then added for some thirty seconds to increase contrast between light and dark lines and so make counting easier.

The strongest magnification of the compound microscope (lens x 100) was used; a camera connected to a television screen facilitated observation of rings when they were wider than the definition of the televised picture; if not the observation was made directly through the microscope.

Rings were counted from the nucleus to the otolith's edge. They generally could not be counted in a straight line because all rings on the surface of the slide were not visible. If while counting, an opaque area appeared, there were two possibilities:

a) If an area of visibility was found following the last ring, counting could continue

b) If there was none, the unmarked area was measured and an estimate of rings made using the number of rings per unit length at the edge of the opaque area.



Plate 1. Otoliths prepared for sectioning. Various stages in the preparation of thin sections of otoliths after embedding in resin,



Plate 2. Circular saw with diamond blade.



Plate 3. Thin cross section of otolith of *Etelis carbunculus* observed in central zone. The nucleus (N) and, to the left, the beginning of the sulcus can be seen. Daily growth rings (J) are wide and clear. Scale:  $25 \text{ mm} = 20 \ \mu\text{m}$ .

# Observations

Two zones were distinguished on otoliths in these observations:

a) Zones exhibiting regular rings (Plates 3-6). These rings are thought to be laid down daily; their thickness decreases progressively from the nucleus to the edge of the otolith. However, superimposed on this general tendency are alternate increases and decreases of ring thickness (Plate 6). This phenomenon has not yet been analyzed, but might relate to lunar cycles, as described by Pannella (1980).



Plate 4. Thin cross section of otolith of *Etelis carbunculus* (same section as in Plate 3 and same scale) as seen at the thousandth ring. Rings are much wider near the nucleus. Scale: 25 mm = 20  $\mu$ m.



Plate 5. Thin cross section of otolith of *Etelis coruscans*. Areas of discontinuity (D) separate the series of daily growth rings. These marks may relate to spawning periods or other forms of stress. Scale: 25 mm = 20  $\mu$ m.

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b) Zones of discontinuity (Plates 5, 6). It was observed that there were occasional dark marks separating zones of daily rings which no longer appeared to be strictly parallel. The alignment of the daily rings was different in the areas of discontinuity. In these cases, it became difficult to count daily rings since the usual regularity was masked. When such areas were found, they were followed to where the succession of rings returned to its former direction.

The periodicity of discontinuous areas is being studied. It may be that they correspond to periods of spawning, but any major stress may be the cause. It is very difficult to decide how to distinguish various causes which may lead to the formation of such areas.



Plate 6. Thin cross section of otolith of *Pristipomoides flavipinnis*. Discontinuous areas (D) can be clearly seen as can an area (Z) within which the thickness of daily growth rings decreases and increases alternately. Scale: 25 mm = 20  $\mu$ m.

#### **Growth Parameters**

Growth curves for four deepwater snapper species have been obtained from observing daily rings, *Etelis carbunculus, E. coruscans, Pristipomoides multidens* and *P. flavipinnis.* Figs. 1 and 2 show these curves.



Fig. 1. Von Bertalanffy growth curves for *Etelis carbunculus (left)* and *E. coruscans (right)* with two forms of the von Bertalanffy growth equation, in which length (L) is in cm and time (t) is in days.



Fig. 2. Von Bertalanffy growth curves for *Pristipomoides flavipinnis* (*left*) and *P. multidens* (*right*) with two forms of the von Bertalanffy growth equation, in which length (L) is in cm and time (t) is in days.

#### Conclusion

Ageing fish by examination of hard parts, especially the study of otoliths, appears to be a particularly valuable source of information. The age of the fish may be established and information obtained which helps to provide parameters of its life history. This study is only beginning; it is continuing in a tripartite analysis: counting daily rings on larger numbers and more species; analyzing various categories of rings and their periodicity; and comparing the technique of using daily rings with other methods for estimating the age of fish. Growth parameters obtained are provisional, but furnish the first indispensable data for a study of the stocks.

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