International Council for the Exploration of the Sea.

CM 1968/Gen: 11

Tetracycline labelling as an aid to interpretation of otolith structures in age determination - A Progress Report

BY

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Introduction

Age determination of fish using otoliths, vertebrae (or other bones) or scales is now standard practice for a wide variety of species. The technique is dependent upon the correct interpretation of the structure of the material chosen. In the otolith, for example, this consists of a series of concentric shells of alternating hyaline (translucent) and opaque material. In many stocks of fish this material is laid down in a regular succession of zones, and it is usually assumed that the opaque material is deposited during the period of fast growth in the summer months and that the hyaline material is deposited during the period of slower growth in the winter. There is little doubt that this interpretation is usually correct, but some stocks of fish have structures in which the zones are irregular and confused and interpretation is difficult, e.g. the cod of the east Barents Sea, hake, and redfish.

Some confirmation of an interpretation may be obtained if the deposition of a particular zone in the structure can be definitely associated with a particular year or season. Experienced otolith readers studying regular and frequent samples from a particular stock of fish over a period of several years are able to identify zones in otoliths which are characteristic of a particular season or of a particular year-class of fish. Two examples of this are (i) the weak or faint 1946 opaque zone in many North Sea plaice of the 1944 year-class, which is still immediately recognizable in fish which are now 24 years old, and (ii) the two narrow opaque zones formed in 1955 and 1956 in many western Barents Sea cod of the 1954 year-class. It is our experience that if these atypical zones can be observed in formation and can be recognized in later years then they serve as a valuable reference point in time to which subsequent zones can be related, thereby aiding interpretation of the whole otolith structure.

It follows that a method which could label artificially the time of deposition of skeletal structures would be extremely valuable. This paper describes experiments in progress to determine the suitability of tetracycline for this purpose.

Tetracycline compounds are readily absorbed by vertebrate animals and are deposited in bony structures where calcification is taking place. Once deposited in skeletal tissue they can be detected by a yellow fluorescence in ultra-violet light. Kobayashi, Yuki, Furni, and Kosugiyama (1964) showed that tetracycline injected into the body cavity of small goldfish was deposited in skeletal structures and scales. More recently Weber and Ridgway (1967) have successfully used tetracycline for labelling fingerling trout and salmon by administering the antibiotic in food pellets. The results of Kobayashi et al. (1964) showed that goldfish given four injections, separated from each other by two-month intervals, had four clearly separated fluorescent layers in the bony structures, corresponding to each injection. This suggests that tetracycline deposition in the bone is completed within a short space of time after injection and that normal deposition is resumed subsequently.

Materials and Methods

The tetracycline compound used in these experiments was Achromycin (tetracycline hydrochloride). It was the intention that, after the administration of the tetracycline, the growth of the fish and the otolith should continue normally in the natural environment. Accordingly the main trials have been made in the field, with each fish being tagged and liberated after treatment. Up to the present time three field experiments have been made, using cod of the North Sea and the west of Scotland stocks. The cod were trawl-caught and were tagged in the normal way with the Lowestoft plastic flag tag (Williams 1963). The tetracycline was administered into the body cavity and the fish were returned immediately to the sea.

The three experiments were conducted using different dose rates of tetracycline:

	Experiment 1	Experiment 2	Experiment 3
	North Sea		West of Scotland
	November 1967	March 1968	June 1968
Achromycin solution	5% in distilled water	5% in Isotonic saline	2.5% in Isotonic saline
Dose rate (mg Achromycin per kg body weight)	100	50	25
Number of fish			
Tagged and treated	42	56	97
Tagged and not treated	83*	56	876

*includes 41 fish injected with distilled water only.

Tank experiments are also proceeding, designed to determine the minimum dose rate that will give a well defined mark in the otolith.

Results

From the results described by Kobayashi et al. (1964) we would expect that the examination of the otolith of fish that are eventually recaptured would reveal a thin fluorescent layer identifying that part of the otolith which was being deposited at the time of absorption of the tetracycline.

At the time of writing only a few of the treated fish have become available for examination; however, results from these have been encouraging. Tank experiments have shown that the deposition of the tetracycline in the bony tissues begins within two hours of injection. Confirmation that deposition begins soon after injection has been shown in the field trials by recaptures made a few days after liberation, the otoliths of these fish revealing a surface deposition of fluorescent material.

One fish treated and liberated in November 1967 and recaptured in the following July had grown normally. In the otolith there was a clear fluorescent band (as seen in transverse section) in the most recent hyaline zone, with a normal summer opaque zone deposited outside it. A clear band of fluorescence was also present in the centrum of the vertebrae and on the scales.

Two recoveries from the fish released in June 1968, recaptured after almost two months at liberty, showed a narrow band of fluorescence at the extreme edge of the otolith. The early deposition of the 1968 opaque zone was plainly visible in the area immediately preceding this fluorescent material. A band of fluorescence was also very clearly visible in the centrum of the vertebrae and in the scales.

Acknowledgement

The Achromycin used in these experiments was kindly supplied by Cyanamid of Gt. Britain Ltd. Our gratitude for this and for technical advice given is acknowledged here.

References

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