



Serial No. 898
(D. c. 2)

Document No. 37

ANNUAL MEETING - JUNE 1961

Problems in Developing a Standard Terminology for Ageing Techniques

by Albert C. Jensen

Fishery Research Biologist, U. S. Department of the Interior, Fish and Wildlife Service, Bureau of Commercial Fisheries, Biological Laboratory, Woods Hole, Massachusetts, U. S. A.

In recent years, the Standing Committee on Research and Statistics has been concerned with problems dealing with age assessment of the principal species of fish in the ICNAF Convention Area. An examination of one phase of the problem, through the exchange of cod otoliths, suggested that, while in the majority of the cases the age determinations were correct, important differences arose in the interpretation and reporting of the zone pattern in the otoliths. In an approach to the problem, the ad hoc Subcommittee on the ICNAF Cod Otolith Exchange Program recommended (1959):

That age readers should be given opportunity to meet when preparatory work has been done and be given sufficient time to study the age reading techniques used by different countries, to discuss the introduction of a standard terminology and symbol system. The possibility of using spawning zones as a means for stock assessment, and stock prediction, should be given attention.

That the Secretariat should approach the proper institutes in ICNAF member countries and request data on otolith terminology and symbols, and photographs of otoliths and scales with interpretations. This material should be distributed as soon as possible to age readers concerned, and treated at the proposed meeting of otolith specialists.

A guide for the Secretariat in the collection of data on age reading was included as an addendum to the report and subsequently circulated in the form of a questionnaire to 14 laboratory directors and biologists. Replies were received from 12 individuals in 4 different laboratories. The replies were collated and prepared as an ICNAF document with appendices (Keir, 1960). It clearly demonstrated that, while there was uniformity and agreement in most of the techniques and terms used in the various laboratories, there were still important differences in certain key terms and critical techniques. For example, the technique to prepare an otolith for reading ranged from simply breaking the otolith between the fingers with no subsequent treatment, to sawing the otolith with a circular saw dressed with diamond dust. Only three respondents offered a definition of spawning zone; several replied that they do not use the term or do not distinguish such zones from other otolith zones.

Keir's document was considered by the Subcommittee on Ageing Techniques at the 1960 Annual Meeting with a recommendation "... that a Working Party on Ageing Techniques should be established with a view to resolving these difficulties in the use of scales, otoliths and other bony structures in studies of age, growth and maturation, and stock separation and mixing." The subcommittee further outlined certain of the avenues of study that should be undertaken by the Working Party. Dir. Gunnar Rollefsen was appointed Convenor for the Working Party, with a meeting to be held in Bergen in the autumn of 1961, and the author was asked to prepare a set of terms, definitions, and symbols that he would take to Bergen for consideration by the Working Party (Anon., 1960).

A manuscript was prepared (Jensen, 1960) setting up a proposed standard terminology and notation for otolith readers. The manuscript synthesized the replies in Keir's (1960) document, setting down those terms in which there seemed to be general agreement among his respondents, and proposing terms that warranted consideration. The manuscript was then circulated among the author's colleagues at Woods Hole and also sent to 13 laboratory directors and biologists in ICNAF member nations. Ten replies were received, in addition to the comments offered by the Woods Hole staff, and again it was obvious that in general there was agreement among the respondents both in the use of the terms and the proposed definitions. There were important differences of opinion, however, in the definitions of, or the symbols for, the following terms:

- Type of edge growth (i. e. , narrow hyaline, wide opaque, etc.)
- Readability of otolith; clarity of zones
- Check marks
- Spawning zones
- Age assignation
- Nucleus

For the examples cited above, most respondents offered other terms to fit the definitions I had proposed, or suggested changes in the definitions, but in all cases the criticisms were well thought out and extremely constructive.

In addition to the terminology gleaned from Keir's questionnaire, and submitted in my proposal, there are the terms contained in the vast body of literature that has accumulated over the years. Many of the terms are obsolete and no longer in common usage among otolith readers, but a few of the terms may bear consideration by the Working Party and so have been included in the proposed terminology for otolith readers.

Where do we stand now in our efforts to establish standard terms and notation for use by those reporting age determination results? A glance at the literature, ICNAF reports, and the recent special correspondence, suggests that at least some problems have been resolved and agreement reached for many terms and techniques. It remains now to try to resolve the remaining problems. In some cases we may never be able to get complete agreement and in such cases perhaps the best we can hope for is to achieve mutual understanding of what is meant when one term or another is used in a report of otolith age determinations. Such mutual understanding may come about through face-to-face meetings of the biologists concerned. It may come about through actual demonstration of techniques or examples of special zones, for example, in certain otoliths. Certainly one way of achieving mutual understanding is through the use of photographs to illustrate instruments, or set ups, used to read otoliths, or to illustrate special otoliths through microphotographs.

It is my purpose here to outline some of the problems I encountered in preparing the proposed standard terminology for otolith readers. Perhaps a consideration of the problems by the Working Party will suggest possible solutions or at least provide a medium for the exchange of ideas on the general subject of age reading techniques.

Physiological Factors Causing Periodic Growth in Scales and Otoliths

Growth periodicity, causing the differential deposition of material in scales and otoliths, is the basis for age determination from these structures. In validation studies for many species it has been shown that growth zones in scales and otoliths are formed on a more or less regular annual basis, but what causes the growth periodicity is not so well understood. The cause may simply be a function of the animal, an external influence of the environment inducing physiological changes, or (as probably is the case) a combination of the two.

Basic Animal Physiology

1. What is the role of feeding cycles in the formation of zones in scales and otoliths? For many species, there is a diminution, if not cessation, of feeding at certain times of the year (particularly during the spawning period) followed by a recovery period of gluttony. In other species, however, the pattern of feeding does not change during the spawning period. Does the reduction of feeding cause a decline, or check, in the fishes' growth and a subsequent check in the growth of the hard parts?
2. What is the role of spawning in the formation of zones in scales and otoliths? Many workers recognize spawning zones in the otoliths of some species. Is the physical appearance of these zones caused by the maturation and discharge of sex products and the drain on physical reserves?

Environmental Considerations

1. What part does changing water temperature play in growth periodicity? It has been demonstrated, for haddock, at least, that so-called "winter growth" zones actually begin to form on the scales when the water temperatures are at the annual maximum and that "summer growth" zones begin to form following spawning but at the season when water temperatures are at the annual minimum.
2. What are some of the other environmental factors that may cause growth changes? Does disease cause a check in growth with a subsequent "check mark" on the hard parts? Are check marks formed (as is the case in certain bivalves) as the result of a shock? escape through the meshes of a trawl? or a close escape from a predator?

Techniques for Handling and Reading Scales and Otoliths

The manner in which scales and otoliths are stored may not have much influence on the resulting age determinations except in cases where, through improper storage, there is subsequent deterioration or actual destruction. Nevertheless, a discussion of the various methods used in different laboratories will serve as an exchange of information that could lead to more efficient handling methods.

Storage

It is desirable, of course, to read scales and otoliths soon after they are collected. This is not always possible, particularly when the collections are made during research cruises or aboard commercial vessels, or when an extensive back-log of materials is desired for a particular study. What then is the best method (or methods) for storing the materials?

1. Dry storage is probably best for scales, but in what manner? Many laboratories keep the scales from each individual fish in separate envelopes, with notations for sex, length, state of maturity, stomach contents, and other information on the face of the envelope. Frequently the envelopes are grouped in bundles and the bundles fastened with elastic fasteners, string, or paper-covered wire. Otoliths, too, are often stored dry in the same manner as scales. Are there better methods for such dry storage?
2. Wet storage, in an alcohol, alcohol-glycerine, or similar preservative, is necessary for otoliths from some species of fishes since the otoliths often become opaque in dry storage. This is particularly true for otoliths that are read whole under reflected light. How long can otoliths be kept in wet storage, and are there any undesirable changes in the physical or chemical makeup of the material under long-term wet storage?

Examination of materials for age determination

1. Scales are usually prepared for examination in one of three ways:
 - a. dry, untreated, under microscope: suitable for young fish with thin, easy to read scales; e. g. , scrod haddock;
 - b. impressed with heat and pressure in plastic, and impressions projected;
 - c. permanent, or semi-permanent mounts on slides, under microscope.

2. The scale or a projected image is examined by a trained reader who counts the year marks to determine the age. A recent electronic development, however, suggests that the time may not be far off when automation may help to materially reduce the amount of routine labor usually involved in complex age determination programs. The device, a semi-automatic electronic scale reading machine, was developed for use in a study of the biology of West Coast salmon (van Haagen and Dale, 1959). A pilot model of the device was built and is under test by biologists.

3. Otoliths are prepared and read in a variety of ways, some of which may be better than others, although each technique reported apparently satisfies the needs of the laboratory in which it is employed.

- a. Whole: The otolith is placed with the broad surface down in a watch glass (blackened or otherwise opaque) and moistened with a glycerine and water solution. The zones are read with the help of a magnifying glass or low-power microscope. This method is suitable for thin, less-dense, otoliths, such as those juvenile redbfish or silver hake.
- b. Cut otoliths are simply snapped in two with the fingers, although they may be hand "sawn" with a razor blade, scalpel or similar sharp instrument. A more elaborate method uses a powered, diamond-dust dressed saw.
- c. Low-power 'scopes (about 10X) are used to examine the cut surface of the otoliths. The otolith pieces may be held by partially embedding them in a cut cork holder, or in colored (usually medium to dark blue) plasteine modeling clay, and illuminated either with reflected or transmitted light. A glycerine-water solution is often used to moisten the cut surface of the otolith.

Photographing otoliths and scales

The value of good illustrations to explain particular or peculiar features in scales and otoliths was made evident in my recent correspondence for the standardized terminology. Several respondents included sketches, particularly in regard to the nucleus (nucleolus?) of the otolith, to demonstrate what they considered to be the nucleus under my definition. Of course, there is the obvious need for photographs to illustrate papers for publication. The study of otolith and scale photography could conceivably be a separate discussion session. Many laboratories have developed techniques for such photography, but I am most familiar with those used in Woods Hole. These are outlined below.

1. Microphotography
 - a. American Optical "Microstar," binocular microscope with built-in 35 mm. camera.
 - b. Other microscopes with camera attachments.
2. Extreme close-up photography
 - a. Leica 35 mm. camera with Focaslide attachment.
 - b. Direct photography using scale projector and Speed Graphic film holder.
 - c. Direct photography using scale projector and Land Polaroid camera.
3. Choice of films
 - a. Black and white film--several kinds allowing wide choice of film speed and contrast.
 - b. Color film--not as many kinds available as for black and white. Resulting photographs have more natural appearance.

Analysis of otolith or scale growth increments

1. Direct measurement from photographs.
2. Direct measurement on projected scale image.
3. Measurement under microscope.
 - a. Stage micrometer.
 - b. Eye-piece micrometer.

Interpretation and Reporting of Results

Age Determination

1. Assignment of ages--introduces problems of validation of method.
2. Other biological information--spawning, age at first maturity.

Stock definition

1. Variations in growth rate.
2. Differences in zonation within scales and otoliths.

Reporting results of scale and otolith readings

1. Standard terminology and notation.
2. Notation of reading method used.

Literature Cited

- Anon. 1959. Report of the ad hoc subcommittee on the ICNAF cod otolith exchange program. Report of the standing committee on research and statistics, ICNAF 1959 Annual Meeting. Appendix III, pp. 48-50.
- Anon. 1960. Report of the subcommittee on ageing techniques. Report of the standing committee on research and statistics, ICNAF 1960 Annual Meeting. Appendix X, p. 37
- Jensen, Albert C. 1960. A standard terminology and notation for otolith readers. U. S. Fish and Wildlife Service, Woods Hole (unpublished ms. report).
- Keir, Ronald S. 1960. Techniques for reading otoliths. ICNAF, 10th Annual Meeting, Ser. 714 (D. c. 2), Doc. 4, Appendices I-XII (mimeographed).
- van Haagen, Richard H. and Harry P. Dale. 1959. Fisheries instrumentation laboratory offers biologists new research tools. Commercial Fisheries Review 21(4): 27-31.

oo0oo