



Demersal Fish Committee

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The authors of the <u>"Report of the Workshop on Ageing of Redfish"</u> have asked us to attach to this report a new version of para. 5) on Radiology, as follows, which is to replace the one on page 4.

5) Radiology

Bennett et al. (1982) have used the decay rates of radionucleotides $(^{210} \text{ Pb}/^{226} \text{Ra})$ and claim to have confirmed estimates obtained by ageing sections of otoliths from the splitnose rockfish, S.diploproa. Otoliths were examined morphologically and were assayed for their natural radionucleotide concentrations. Four age groups of otoliths were identified based on growth-zone counting; in the first three, whole otolith and otolith section age estimates agreed while in the fourth, the otolith section age substantially exceeded the whole otolith age. Radiometric analysis demonstrated that all otoliths were deficient in 210Pb activity relative to 226 Ra activity with the deficiency decreasing with increasing number of growth zones. The magnitude of the $^{\rm 210Pb}/^{\rm 220Ra}$ radioactive disequilibrium in each otolith group, when compared to the number of growth zones and the otolith weight histories derived from the two techniques, identified the growth zones revealed by otolith sections as annual features. Thus when otolith section age exceeds whole otolith age (usually occurring after 20 to 25 years of age for this species), the otolith section technique was proposed to be the correct method. They concluded that estimates of longevity in the genus Sebastes near 80 yr. were therefore confirmed.

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International Council for the Exploration of the Sea

C.M.1984/G:2 Demersal Fish Committee

REPORT OF THE WORKSHOP ON AGEING OF REDFISH

Bremerhaven, 13-17 February 1984

This document is a report of a Workshop of the International Council for the Exploration of the Sea and does not necessarily represent the views of the Council. Therefore, it should not be quoted without consultation with the General Secretary.



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REPORT OF THE WORKSHOP ON AGEING OF REDFISH

As recommended by ICES (C.Res.1983/2:14), a Workshop on Ageing of Redfish was convened by Dr K Kosswig at the Institut für Seefischerei in Bremerhaven, Federal Republic of Germany, from 13-17 February 1984.

Participants

Dr K Kosswig Dr P Rubec Federal Republic of Germany Canada.

Introduction

A Workshop from 14-18 February 1983 recommended that further studies be conducted on ageing of redfish (ICES, C.M.1983/G:2). At the 71st Statutory Meeting of ICES it was recommended (C.Res.1983/2:14) that a Workshop on Ageing of Redfish be convened 13-17 February 1984. The terms of reference were to:

- compare results of age readings based on samples of scales and otoliths taken from the same fish. Samples from individual fish should be supplied by all participants;
- ii) investigate possible methods of age validation applicable for redfish.

Historical Perspective

A controversy concerning the best methods to age redfish has existed for over 25 years (Parrish, 1958). Kotthaus (1958) advocated rapid growth, while North American scientists advocated slow growth based on the interpretation of whole otoliths (Kelly and Wolf, 1959; Rasmussen, 1958; Sandeman, 1961; Parrish, 1958). Initially, most workers utilised otoliths for age determination. First the USSR and finally most European countries have adopted scale determination procedures due to difficulties in interpreting otoliths. North American scientists have continued to use otoliths. Uncertainty exists concerning the validity of age determinations for redfish species due to the various procedures being used and the different redfish stocks under consideration.

The age and growth of redfish was part of the ICES/ICNAF Redfish Symposium (Rollefsen, 1961). Most of the existing literature was summarized by the participants (Hansen, 1961; Sandeman, 1961; Surkova, 1961; Trout 1961). It was resolved that the conception of slow growth as advocated by experts at that time should be adopted (Rollefsen, 1961).

The approaches taken to age redfish and the age validation may be summarized as follows.

Ageing Procedures

a) Scales

USSR workers examine scales under polarized light (Surkova, 1961); Chekhova, 1971). Kosswig (1971, 1980) has perfected a technique of impregnating scales with silver nitrate and viewing them under polarized light. The procedure was first used by Savage (1919). Kosswig's procedure appears to give repeatable results.

b) Other Structures

Six and Horton (1977) examined 25 different structures from 3 species of <u>Sebastes</u> from the west coast of North America and found only the anal fin pterygiophore, opercle, scale, vertebra and otolith to provide consistency of results among multiple readers. Otoliths were of the preferred structure.

c) Otoliths

(i) Surfaces

Surface readings have been widely used by various workers (Kotthaus, 1958; Kelly and Wolf, 1959; Bratberg, 1956; Westrheim,1973; Kimura et al., 1979). This is generally acceptable for younger fish, but it has been shown (Irie, 1960) that growth of various surfaces of the otolith becomes allometric with age. When this is true, it cannot provide accurate age estimates regardless of the annular criteria used.

(ii) Break the otolith

By cracking the otolith or sawing it in half, various workers examine otoliths mounted in plasticene with reflected light. This is the procedure used for ageing redfish in St. John's, Newfoundland, generally with a 6 to JOX magnification (Sandeman, 1969; Clay and Clay). Various procedures such as wetting the surface with alcohol, toluene, clove oil or cooking oil can be used to enhance the visibility of the annuli. However, many checks and false annuli are visible. Hence much experience is needed to interpret these otoliths.

(iii) Break/burn

Methods have been developed by Chugunova (1959) and Christensen (1964) to account for the fact that most otoliths increase more in thickness than in length or breadth with age. The split otolith is heated in an alcohol flame (Chilton and Beamish, 1962) or a gas flame. Chilton (pers.comm.1983) believes this gives a better resolution of annuli than examining thin sections. Disadvantages are the uneven surface from cracked otoliths and possible changes of otolith surfaces if polished. High magnifications are used to examine burnt otoliths in Nanaimo, British Columbia (Chilton and Beamish, 1982). Many fine lines become visible using this procedure.

(iv) Cross sections

Thin cross sections of otoliths can be obtained through the use of either low speed (Buehler isomet) or high speed (Bronwill Sectioning Machine, TSM Model 77, Micromech Mfg.Corp. saws with diamond wafering blades)(Chilton and Beamish, 1982). A similar high speed saw has been developed by Federal Republic of Germany scientists in Hamburg. Personnel at Woods Hole, USA, obtain 0.178 mm sections using a microtome (Mayo <u>et al.</u>, 1981). The procedure involves mounting the otolith on cardboard tags with wax prior to sectioning (Nichy, 1977). This technique has been applied in St. Andrews and Moncton, N.B., Canada. A Buehler isomet is used to obtain 0.5 mm sections. Cross sections in Nanaimo are obtained by embedding the sections in epoxy, but this is more time-consuming (Chilton and Beamish, 1982). Consequently, most Ccean perch are aged using the break/burn procedure previously described (Chilton, pers.comm. 1984). Thin cross sections (0.4 - 0.5 mm) have been reported to provide better resolution of annular growth for older fish where blurring of annular zones occurs with burning (Leaman, pers. comm., 1984). Thin cross sections at high magnifications (80-100X) have been used to count annuli in various species of Pacific rockfish (<u>Sebastes</u> spp.). Using this technique, rockfish have been found to be much older than was previously accepted (Beamish, 1979; Shaw and Archibald, 1981; Archibald <u>et al.</u>, 1981; Chilton and Beamish, 1982). For example, Pacific Ocean Perch (<u>Sebastes alutus</u>) have been aged up to 90 years (Chilton and Beamish, 1982), when previously the maximum age determined from whole otoliths was 29 (Westrheim, 1973; Beamish, 1979).

Various stains have been tried unsuccessfully on otoliths (Six and Horten, 1977; Kosswig, pers.comm.,1984). Burning appears to enhance the visibility of annuli on sections. Various oils (vegetable, cedarwood, clove), alcohol, glycerine or toluene can be used to enhance the visibility of annuli. In Moncton, N.B., sections are embedded on black plastic trays using a liquid plastic (diatex) and toluene. The toluene appears to clear the opaque zones thus making the annuli more visible. This may be used on either unburnt or burnt sections. Good results have been obtained by burning 0.5 mm sections on a hot plate for 30 seconds and then mounting them in the 50:50 mixture of diatex and toluene.

Validation of Ageing Techniques

1) Following Known-Aged Fish

The only true method of validation is to follow known aged fish throughout their life. The 'validated' fish must have experienced the same biotic and abiotic environment as those for which the ageing method is to be used. Tank- or pond-held fish cannot be used to directly validate ages estimated from wild fish. It is important to validate the technique over the range of ages assigned to it. A common fault is to validate ages over the younger segment of the age range (Beamish and MoFarlane, 1983). One should differentiate between experiments designed to validate the ageing technique being used and those designed to actually validate (verify) the ages assigned.

Dominant year classes can be followed over a number of years. Westrheim (1973) and Gunderson (1977) provided good validation up to age 6 and partial validation to age 15 for otolith surface ageing for species of Pacific rockfish. Hansen (1959) reported a series of length frequency measurements of small redfish from Godthaab and West Greenland fjords, and Sandeman (1961) similarly for Hermitage Bay, Nfld. Both studies illustrate the progression of modes of different year classes during the early years of life of redfish. McKone and Legge (1979) used length frequency distributions to trace growth of the 1956 and 1958 year classes from the Gulf of St.Lawrence from 1959 to 1979. The data were used to confirm the reliability of ages of redfish (primarily <u>S. mentella</u>) determined from broken otoliths. By 1980, the Gulf of St. Lawrence redfish from the 1956 year class were 24 years old. Since the 1956 year class was very large, they were still detectable in large numbers.

2) Annular Zone Formation in Juveniles

Perlmutter and Clarke(1949) showed that redfish from the Gulf of Maine (S. fasciatus) have scales that display alternating zones of widely spaced and narrowly spaced circuli, the latter being formed between November and March. Kelly and Wolf (1959) reported similar results for <u>S. fasciatus</u> kellyi otoliths. Bratberg (1956) confirmed that the opaque zone of <u>S. marinus</u> from the Barents Sea was formed in May-September, while the hyaline zone of the otolith is formed in October-April. Similar observations were made on scales from the same fish. The data indicated only one annulus was laid down per year for fish up to age 4. The ages determined from otoliths were checked by scale-reading in 489 fish. Of these, 474 (96.9%) clearly showed the same number of annular rings as was seen on the corresponding otoliths. Similar results were found with annular ring formation in Pacific Ocean perch (<u>S. alutus</u>) studied by Westrheim (1973) and yellowtail rockfish (S. flavidus) by Kimura et al. (1979).

Mayo <u>et al.</u> (1981) confirmed age determinations up to age 7, using otolith sections for <u>Sebastes marinus</u> (this is probably <u>S. fasciatus</u>). Deposition of hyaline begins in August, and the formation of the opaque edge begins in August. A seasonal progression of modes of length frequencies for the 1971 year class was traced from 1971 to 1978.

3) Examination of Growth

Kimura <u>et al.</u> (1979) and Six and Horton (1977) used the criterion of monotonically increasing length at age as validation under the assumption that ageing errors would be detectable as anomalies in growth. They did not follow a single cohort to derive length at age. Comparison of their results with Archibald <u>et al.</u> (1981) and Beamish and Chilton (1982) demonstrates that monotonic growth is not a good criterion. Substantially higher age determinations for the same stocks also indicated monotonic growth.

4) Tagging of Known-Aged Fish

To our knowledge this has not been done for any species of Sebastes. It would involve very small fish because of the overlap in sizes. Alternatively, ages could be determined from scale samples or examination of other external structures such as fin spines or fin rays. Kelly and Barker (1961, 1963) tagged S. fasciatus kellyi at Eastport Maine. The fish were aged, by means of otoliths, after they had been recaptured. Comparisons were made with the otoliths of untagged fish of the same lengths taken at the time of tagging. There appeared to be a reduction in growth in the redfish due to the tags (Petersen disc tags). A total of 5.447 redfish were tagged from 1956 to 1958. A total of 1.876 (34.4%) were recaptured, measured and returned to the water. Some fish were recaptured as many as six times. This demonstrates the feasibility of such a study for this particular inshore population. Tagging redfish from deeper water is less feasible because the changes in pressure induce high mortalities when the fish are brought to the surface. While this study may not have replicated the observed growth of Gulf of Maine redfish, it is apparent from the number of annuli that only one annulus was deposited per year.

5) Radiology

Bennett <u>et</u> al. (1982) have used the decay rates of radionucleotides $({}^{210}\text{Pb}/{}^{226}\text{Ra})$ and confirmed estimates obtained by sections of <u>S. deploproa</u>. Leaman (pers.comm., 1984) notes that the fish aged were about 20 years old. Consequently, this does not confirm the high ages determined for other Pacific rockfish species (Shaw and Archibald, 1981; Chilton and Beamish, 1982; Leaman, in press).

6) Major Shifts in Mortality Rates

Pulsed changes in mortality rates, resulting from major shifts in fishing effort, that persist over time can be used in the same way as the progression of a dominant year class. This is difficult to find in <u>Sebastes</u> but an attempt is being made to generate such data for Vancouver Island Ocean perch (<u>S. alutus</u>) (Leaman, pers.comm., 1984).

7) Time Marks from Tagging Studies

Injection of time marks, such as oxytetracycline (OTC), allows the assessment on whether structures thought to be annuli are laid down on an annual basis. Only OTC injections have been tried with Pacific rockfish. Brian Culver of the Washington State Department of Fisheries has conducted a study on <u>S. melanops.</u> Five OTC-injected rockfish were recaptured, but their otoliths remain to be examined. The Department of Fisheries and Oceans at the Pacific Biological Station in Nanaimo, B.C. Canada, had tagged and OTC injected 13.984 yellowtail rockfish (<u>S. flavidus</u>) (Shaw <u>et al.</u>, 1981). (pers.comm., 1984) notes that 15 recoveries have been made. Five OTC-injected otolith pairs were recovered, 2 at 4-6 months, 2 at 11-13 months, and 1 at 29 months. The data are consistent with the idea that the marks on the fish were annual marks.The fish at liberty for 4 months was 104, the fish at liberty for 11 months was 21 and the fish released for 29 months was aged 13+. Several other species, <u>S. entomelas</u>, <u>S. pinniger</u> and <u>S. brevispinis</u>, were also tagged and OTC injected in lesser numbers (Shaw <u>et al.</u>, 1981). To date none of these fish have been recovered.

The methods described by Chilton and Beamish (1982) describe a fast growth phase in young fish and a slow growth phase in older fish. One can consider that the widely spaced marks in young fish represent true annuli from the above research. However, the data do not confirm slow growth represented by closely packed lines (Chilton and Beamish, 1982) visible on the otoliths of older <u>S. flavidus</u> (maximum age estimate 64).

8) Comparing Age Determinations from Various Structures on the Same Fish

Beamish (1979) has shown that surface age determinations were much lower than section age estimates (section/burn) for Pacific Ocean perch, <u>S. alutus.</u> He attributed this to the tendency of annuli to 'roll over' the edge of the otolith. Consequently, he believes that surface age estimates underestimate the true age. Sandeman (1969) compared the age and growth of <u>S. mentella</u> determined from cracked unburnt otoliths with scale-readings determined by Surkova (1962) from Hamilton Bank and Flemish Cap off Newfoundland. Deep-water redfish aged from otoliths had a slower growth and attained a greater age than those fish aged from scales.

These discrepancies may be due to several causes. Differences in the composition of the fish, from the same areas, taken at different times seem likely. Two species of redfish (<u>S. fasciatus</u> and <u>S. mentella</u>) occur in the area. It is possible that they have different growth rates. It is not known which are represented in the two studies. It is also possible that Sandeman overestimated or that Surkova underestimated their ages due to their ageing techniques. Surkova's data look inconsistent either in age determination or because two species were represented in the samples.

Unambiguous comparisons of age structures can only be obtained by comparing structures taken from the same individual fish. This has been done as previously described by Bratberg (1956) for young <u>S. marinus</u>. Chekhova (1971) conducted a comparison of scales and broken/burnt polished otoliths from a sample of 300 <u>S. mentella</u>. Otoliths of 274 redfish were suitable for the determination of age. Annual rings were counted along the longitudinal short axis of the otolith because the long axis was observed to have many extra rings. Chekhova concluded that no significant difference existed between the results obtained by the two methods. Back-calculations by the Lea and Lee (Frazer) formula gave similar results. Consequently, either method can be used and she preferred the use of scales.

METHODS

Scales and otoliths were supplied by the participants. Samples from Canada included 100 scales and otoliths collected in the Gulf of St. Lawrence during 1983. The otolith samples were sectioned, burned and mounted on plastic trays containing diatex and toluene (See review of ageing methods previously described). As controls, 25 unburned sections were also provided to match burned sections. These were taken from otolith pairs obtained from individual redfish. Scales from the 200 redfish were prepared by the procedures described by Kosswig (1980) with the modifications described in the 1983 report of the Workshop on Ageing of Redfish (Doc. C.M.1983/G:2).

The participants spent several days reviewing the material at various magnifications, low (6-30X) and high (100X). Ageing criteria for otoliths and scales were discussed in the light of previous research. Due to the limited time available only 25 otoliths and scales from the same individual fish were aged during the Workshop. The Canadian participant, P Rubec, aged most of the 25 burnt and 25 unburnt otoliths. K Kosswig aged the scales from the same redfish.

Additional material was exchanged. The Federal Republic of Germany participant agreed to age scales from Canadian redfish from the Gulf of St. Lawrence and material to be provided by Mr J Hunt obtained from the Scotian Shelf, off Nova Scotia. P Rubec obtained otoliths of <u>Sebastes</u> <u>marinus</u> off West Greenland for future comparison with scales. The redfish in the Gulf of St. Lawrence samples are <u>S. mentella</u> and <u>S. fasciatus</u>, S. fasciatus predominates on the Scotian Shelf.

RESULTS

Observations were made on burnt and unburnt otolith sections at low (6-30X) and high magnifications (100X). Many fine lines on burned otolith sections of older redfish (more than 15 years) were observed towards the outer edge of the otoliths examined at high magnification. These sections were very similar to photographs of Pacific rockfish species depicted by Chilton and Beamish (1982). The criteria for counting lines as annuli on otoliths were discussed. It was agreed by the participants that counting every line as done by Chilton and Beamish (1982) would give age estimates far in excess of ages determined from scales or otoliths from the same stock of redfish from the Gulf of St. Lawrence. It was observed that annuli could be interpreted on burned and unburned sections by the pattern of splitting. Burned sections had annuli which were more distinctive than other checks and false annuli. In older redfish, the many fine lines were interspersed on a regular basis (every 4-5 lines) by more well-defined lines which could be interpreted as annuli. The participants agreed that it was inconsistent of Chilton and Beamish (1982) to count only widely spaced annuli in young ages of otolith sections while ignoring intervening lines, but to insist on counting every line as an annulus for older ages on the same section.

Criteria were established by the participants for the ageing of otoliths. The criteria used for scales have been described by Kosswig (1980). It was decided to only examine the otoliths at low power magnifications of 20-30 times. It was also decided to interpret fainter lines, and lines which could not be traced along the edge of the otolith, as false annuli and splits in the annuli. In older redfish, of the many fine lines only the well-defined lines, which were regularly spaced, were counted as annuli. Using these criteria, redfish were aged which varied from 6-23 on otoliths and from 5-23 with scales from the same individual fish.

Out of the 25 otoliths examined, more than 2/3 were identical or in close agreement by one annulus with scale determinations. Initial samples of 4 unburned otoliths did not show close agreement. The inexperience of P Rubec as an age-reader may have contributed to this initial discrepancy. However, re-examination of these otoliths confirms that they were difficult to read in comparison to other sections. About 5 of the initial otoliths were embedded without the use of toluene. It is apparent that the toluene facilitated clearing of the otoliths, making toluene-cleared sections easier to age. It is believed that the use of toluene facilitated the interpretation of the remaining unburnt and burnt sections.

DISCUSSION

This initial comparison between redfish scales and otoliths found age determinations in close agreement. This is similar to the findings of Chekhova (1971). Further exchanges of material will be conducted to confirm whether this conclusion is valid. Both the scale and the otolith section/burn techniques were observed to yield clear observations of well-defined annuli.

Some previous studies may have yielded biassed data due to the techniques used for preparing samples. Faulty interpretation of criteria for annuli appears to account for excessively high ages in Pacific rockfish species (Chilton and Beamish, 1982).

Age validation procedures used in the studies of <u>Sebastes</u> have been critically reviewed. The technique of comparisons between different structures from the same fish is shown to yield valid comparisons. This technique is less expensive and less time-consuming than many of the other validation procedures described. It is up to West Coast researchers to validate their assertions of extreme longevity in Pacific species of <u>Sebastes</u>. The results of the present study indicate no revision of present techniques for ageing redfish is required. Some improvement in age determinations may be possible through burning otolith sections versus examining unburnt broken otoliths of redfish.

RECOMMENDATIONS

Exchanges of material between European and North American researchers should be continued and the results published. It would be worthwhile to have participants from the West Coast of North American in the USA and Canada. The ageing of redfish is seen as a common problem with ageing other species of <u>Sebastes</u> from the Pacific Ocean.

Future attempts at age validation should consider tagging studies of North American inshore populations or of pelagic stocks when or if they occur near the surface. Further studies with radionucleotides on a limited number of samples would be useful. Consideration should also be given to evaluations of whether age/length keys vary from year to year in a density-dependent manner. It may be possible to reduce ageing requirements by determining the adequacy of age/length keys used in cohort analyses. Computer modelling to analyse length-frequency data should be undertaken to validate ageing and possibly as a means to estimate ages from known length-frequency patterns.

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Literature referred to in this list are cited in the report of the Workshop. Not all of the documents discussed during the meeting necessarily appear here.

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