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Dear Age Reader:

The enclosed manual has been prepared to increase consistency of age readings among agencies using similar techniques. It has been published in loose-leaf form to facilitate revisions and additions which are likely to be made. This first edition principally addresses rockfish species, with an addendum on sablefish. As techniques improve or as other species are added, amendments will be published and distributed for insertion.

Pacific Marine Fisheries Commission
August 22, 1984

MANUAL

ON

GENERALIZED AGE DETERMINATION PROCEDURES

FOR

ROCKFISH

Prepared by:

Pacific Coast Groundfish Ageing Technicians

Under the Sponsorship of:

Pacific Marine Fisheries Commission

For:

The Canada-U.S. Groundfish Committee

August 1984

This manual was developed during two three-day workshops sponsored by PMFC at the NMFS Northwest Fisheries Center in Seattle, Washington on April 27, 28, & 29 and August 3, 4, & 5, 1983.

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GENERALIZED ROCKFISH AGEING PROCEDURES

The collection of age structures and their subsequent ageing by various facilities has typically been undertaken in a non-standardized fashion. Differences in method of collection, storage media (or lack of it), equipment used by different agencies, etc., have lead to possible discrepancies in age assigned to a given fish. Standardizing the ageing procedures used by all age determination facilities should reduce this variation.

The procedures outlined in this document may be applied to ageing all rockfish species, except where noted. The sagitta (subsequently termed 'otolith') is the preferred structure for ageing rockfish species. Two methods of ageing otoliths (surface and cross sectioning) are presented as well as two techniques for section ageing are described (burning and thin section). Recommendations are included for proper sample collection.

In order to ensure that the best and most representative ages are assigned to fishes, several important points should be borne in mind when ageing specimens from a new species, stock or locality. Specimens should be obtained, if possible, from at least a one-year period and over a broad spectrum of size classes so that the progression and pattern of annulus formation is apparent for the species in question. Difficulties with interpretation of the first annulus are common in many rockfishes, therefore, position of the first annulus should be validated by the examination of otoliths from juvenile specimens. In addition, novice ageing personnel should be cautioned that growth anomalies (and resulting interpretive difficulties) may arise in specimens from differing stocks and/or localities.

It has been demonstrated that examining the cross-section of the otolith either by burning or thin section is necessary to more accurately determine ages for those longer-lived and more difficult to read species of rockfish. It is therefore recommended that initial careful comparisons of ages be made using both surface and section methods to decide at what age, if any, it becomes necessary to section the otolith. In making this decision, consideration should be given to the early natural history of each species, particularly the age at reproductive maturity. For all species, the use of surface ageing as a supplement to section ages is beneficial in identifying the first several age zones in difficult specimens.

A basic approach to use when attempting to determine the age of a fish from otoliths, regardless of the specific method employed, requires the resolution of three potential problem areas; the interpretation of growth checks, determination of the first few annuli, and the determination of the type of growth at the otolith edge.

GROWTH CHECKS: During years of rapid growth expect growth checks to be apparent. It is useful to know the early life history of the species under study as growth checks may occur when there is a change in diet, depth or habitat. In young fish, before maturity, growth checks can easily be confused with annuli. To overcome this problem, either read along the ventral axis where you should be able to follow the annulus, or turn the otolith so the convex side faces upward and the growth checks will be less visible. Count the clear hyaline zones as annular rings. The width of white opaque zones formed during periods of rapid growth become narrower toward the outer edge. By recognizing the progression of opaque zone width, the reader can differentiate growth checks from annular rings.

FIRST ANNULUS: Once the growth checks have been resolved for the first year or two, micrometer measurements may be made on a sample of otoliths from young fish. The measurements are useful in judging the first and second year growth zones on otoliths from older fish when they are obscured.

EDGE TYPE: Interpretation of the growth zone at the edge of the otolith is critical and should be determined for each otolith. The month of collection for the otolith is necessary for interpreting the type of growth zone that is developing. Any opaque growth showing on the edge may be assigned to either the previous or current growth year, dependent upon the amount of growth seen. By convention, the birthdate of all fish is considered to be January 1.

SAMPLE COLLECTION

Samples may be collected and transported either with or without a liquid preservative medium. Collection procedures are outlined below. Wet Method:

- Collection of samples.
 - a. Samples must be properly identified and individually labeled.
 Required information includes species length, date and sex.
 Other collection information (i.e. area, cruise, etc.) can be specified by the individual study.
 - b. Extraction of both otoliths is recommended.
 - c. Cleanliness of otoliths is of critical importance to the ageing process. All external organic attachments must be removed; preferably by rubbing submerged otoliths in a container of water.
- Containers.
 - Leak-proof, break-proof containers (such as snap-top plastic vials) are recommended.

- b. Each pair of otoliths should be placed in individual containers.
 The discreteness of each fish sampled must be preserved.
- c. Individual specimen numbers should be assigned and placed inside containers. Specimen number labeling on the outside of the container is also useful. This will allow cross-referencing of the sample with appropriate length, sex, and weight data taken for each fish.

Preservatives.

- a. Ethanol/water (30-50% ethanol).
- b. Glycerine/water (50% glycerine). Thymol (approximately 1 tsp. per gallon) must be added to prevent fungal, bacterial, or algal growth. Complete dissolution of the thymol is important.

4. Storage.

- a. Otoliths stored prior to ageing must be clearly marked with appropriate information (i.e. species, cruise or sample identification).
- b. It is recommended that samples be aged within 6 months of collection. If a back-log of unaged otoliths accumulates, containers should periodically be checked for evaporation.

Dry Method:

- 1. Recommended collection methods are the same as described above.
- 2. Containers.
 - a. Coin envelopes may be used. Pertinent information must be written on the envelopes. Ungummed flaps may be desired; otherwise it is extremely important to dry otoliths thoroughly before insertion into the envelope.
 - b. Leak-proof, break-proof vials may also be considered.

- c. Individual specimens should be placed in separate containers, and specimen numbers should be assigned.
- d. If samples are mailed or otherwise transported to an ageing facility, care must be taken to prevent otolith breakage during shipment. Well padded, sturdy cardboard boxes may be required.

SURFACE AGEING

- 1. Equipment required and magnification.
 - a. Use high-quality optical equipment such as Wild binocular microscopes M3-M8 with 10-60x magnification.
 - b. Use high-intensity lighting, preferably fiber optic lamps.
 - An eyepiece micrometer may be useful.
- 2. Procedure for ageing.
 - a. For contrast between hyaline and opaque zones, submerge otolith in clear media (such as water or ethanol) in a petri dish with a black background makes hyaline zones appear darker. The purpose of liquid in the petri dish is to reduce glare.
- Axis of reading.
 - a. View the otolith surface with the concave surface facing upward.
 - b. Counts are most easily made from the center to the outer edge on young otoliths. On older otoliths, count hyaline zones from the edge toward the center. The areas of an otolith that are easiest to read depend on the species. In most cases, the posterio-dorsal, posterior and ventral axes are preferred (see Figure 1).

EXCEPTIONS: In chilipepper rockfish, the anterio- and posterioventral axes are preferred. In widow rockfish, the anterio-dorsal axis is preferred.

- 4. Criteria used in ageing.
 - a. To view older otoliths, it is necessary to tilt and roll them from side to side. Count the hyaline zones from the edge toward the center and obtain identical counts from two areas before assigning an age. When counts from two areas disagree, a third area is read to obtain an agreement.
 - b. Possible growth pattern irregularities:
 - Narrow opaque zones (a zone is narrower than the preceding and succeeding zones).
 - Doubling (two opaque zones appear close together with only a faint hyaline zone separating them).
 - 3. Strong hyaline zones in focus (apparent checks in the first year).
 - 4. Edge type notation in younger years (≤10); (0+, 0++, T-).
 0+ = relatively thin (narrow) opaque zone.
 0++ = relatively thick (wide) opaque zone.
 - T- = no complete translucent zone.

These problem areas are very subjective in nature and it is up to the discretion of the reader to judge the validity of such zones.

- c. By convention, the birthdate of all fish is considered to be January 1st.
- 5. Other procedures.
 - a. Dirty or chalky otoliths are dipped in 10% hydrochloric acid solution to remove film. Otoliths may be dipped for 2-3 seconds. Longer immersion may dissolve portions of the otolith and growth zones may thus be lost.

- 6. Efficiency.
 - a. An average of approximately 50-100 otoliths are assigned final ages daily.
 - b. Speed and accuracy depend on the species, clarity of the otoliths, and general age composition of the sample (old, young, or mixed group).
- 7. The rockfish species examined during the 27-29 April 1983 workshop have been rated according to their difficulty (surface reading). This rating appears to hold true for samples collected in California, Oregon, and Washington, but may not be indicative of the order of difficulty seen in other areas. Variations in environmental conditions seen in different collection areas result in a diversity of growth patterns.
 - a. Order of difficulty:
 - 1. Pacific Ocean Perch (\underline{S} . alutus) most difficult.
 - 2. Canary Rockfish (S. pinniger).
 - 3. Yellowtail Rockfish (\underline{S} . $\underline{flavidus}$).
 - 4. Chilipepper (S. goodei).
 - 5. Widow Rockfish (\underline{S} . entomelas).
 - 6. Black Rockfish (S. melanops).
 - 7. Bocaccio (S. paucispinis) least difficult.

BURNED SECTION AGEING

- Equipment required and magnification.
 - a. Use high-quality stereoscope (such as Wild microscope) with a range of 6-100x magnification.
 - 1. Cost approximately \$4000.
 - Good quality optical equipment results in a clearer picture and a reduction in eye strain; hence better production and precision.

- b. Good-quality microscope lights or double fiber optics are required (fiber optics are recommended).
 - Cost approximately \$250-500.
 - High-intensity light is necessary in order to clearly observe annuli at the outer edge of older, thicker otoliths.
 - Variable intensity lighting is necessary.
 - 4. Fiber optics are recommended due to the lack of heat generated during their operation and the high level of illumination possible.
 - c. Miscellaneous cost under \$30.
 - 1. Alcohol burner (\$4-10).
 - 2. Ethanol or denatured alcohol (non-toxic) (approximately \$10).
 - Forceps (to extract otoliths from vials and to hold during burning) (approximately \$10).
 - Lighter (approximately \$1).
 - 5. Small paint brush (50¢).
 - Modeling clay (\$2).
 - 7. Shallow watch glass (for holding clay) (\$2).
 - Corn or vegetable oil (\$1).
- Procedure for ageing.
 - a. Cleaning otolith prior to burning.
 - If stored in glycerine/water, the otolith may be washed in water and then dried thoroughly with a tissue.
 - If stored in ethanol/water, the otolith must be thoroughly dried with a tissue.
 - b. Breaking the otolith.
 - 1. The otolith must be broken dorsal-ventrally through the nucleus.

- a. Thinner otoliths may be broken between the reader's thumbs.
- Breaking of the otolith may also be accomplished using forceps.
- c. Thicker otoliths may require deep scoring prior to breaking. The use of a hand saw or an Isomet saw will accomplish this.
- Grinding of the broken surface may improve the surface if extremely uneven or irregular.
- 3. In order to more easily detect the first annulus on the burned surface, micrometer measurement of the width of the first year on the otolith surface after breaking is useful.
- c. Burning the otolith.
 - Burn the broken otolith (preferably the anterior half) over an ethanol burner (other alcohols may be toxic). Keep the flame low.
 - Pass the broken surface of the otolith back and forth through the yellow portion of the flame until a medium to dark brown color is attained. The annuli become darker than other checks when burned.
 - a. Avoid holding the broken otolith edge directly in the flame.
 - b. A lighter burn may be required for fish aged \leq 5 years.
 - c. Rockfish otoliths burn rapidly and overburned, charred surfaces may be removed by abrading the surface if needed, but an attempt to age the otolith should be made prior to grinding.
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- Cool the burned otolith approximately 15 seconds.
- 4. Place the burned otolith in modeling clay of a contrasting color (i.e. green, blue, or red and avoid yellow, white, and brown clays).
- 5. Paint the burned surface of the otolith with vegetable oil (non-toxic oil) with a small paintbrush. The reader may wish to use water on the burned surface rather than oil. This reduces glare on the reading surface, makes reburning easier if necessary, and allows the possible use of a sealant (e.g. nail mender) on the reading surface.
- Axis of reading.
 - a. The best areas for ageing are usually found on the dorsal (elongated) side.
 - b. No axis is equally legible on all otoliths.
- Criteria used in ageing.
 - a. The first annulus is the first distinct dark band from the nucleus (kernel). It could be a close grouping of lines usually merging on the sulcus edge.
 - b. Count every distinct dark band that can be followed to the sulcus (see Figure 3).
 - c. When viewing burned sections, it is helpful to initially scan the reading surface at a low power. In this way a reader may more easily perceive the growth pattern of the earlier years. A gradual increase of objective power enables the reader to derive a more accurate interpretation of the fine growth zones towards the edge.

5. Other procedures.

- a. A freshly broken and burned otolith is preferred for age determination.
- b. A previously burned and oil-painted otolith which has been stored reduces precision in age determination when a second attempt to age is made. Deterioration of a broken and burned otolith occurs through time.
- c. As an interesting experiment to produce a permanent record of a burned section it is suggested that nail mender be applied to the burned edge rather than oil. This will require good ventilation due to toxic fumes.

6. Efficiency.

- a. One experienced reader working 5-6 hours per day can optimally produce 50-100 burned section readings per day depending upon the difficulty of the sample. This does not include a second reading.
- b. The final age is assigned after the same age has been obtained for two readings.

THIN SECTION AGEING

- 1. Equipment required and magnification.
 - a. Either the Buehler Isomet Low-Speed Saw or Bronwill High-Speed Sectioning Machine may be used. With the resin embedding method, a jewelers saw may also be used.
 - b. Polishing sections, if desired, may be accomplished using a very fine grade of wet-dry sandpaper (400-600 grit carborundum) or jewelers rouge.

c. Either compound or dissecting microscopes may be used provided they have the necessary magnification. A range of 25-200x magnification is needed. High-quality optics are critical to achieve precision in otolith section reading.

Procedure for ageing.

- a. Preparation of the otolith prior to sectioning.
 - 1. The otolith must be thoroughly dry prior to sectioning:
 - a. If the otolith is stored in ethanol/water, towel and air dry.
 - b. If the otolith is stored in glycerine/water, towel dry, then dip in toluene to remove all traces of the storage medium. This will require good ventilation due to toxic fumes.

2. Mounting techniques:

- Mounting otolith on sectioning card method:
 - 1. Use Dennison double-thickness tags (No. 12-104-1).
 - 2. Draw intersecting lines on the tag (see Figure 2).
 - Label the tag with unique otolith identification data.
 - 4. Place a 0.5 inch piece of double-sided adhesive tape at the center of the tag.
 - 5. Place the otolith on the tape, centering the otolith's focus directly over the tag line intersection.
 - Otolith alignment on the tag is very important.
 Orient the otolith with its dorso-ventral axis
 on the long axis of the tag (see Figure 2).

- 7. Pour the mounting medium around and on top of the otolith. Any of the following mounting media are satisfactory:
 - a. Clear plastic casting resin.
 - b. Finger nail mender.
 - c. Clear, hard epoxy-based resins.
- 8. As the mounting media gels and hardens slightly, trim away excess media to the edges of the otolith with a razor blade.
- The mounting medium must be completely hardened and dry before sectioning (overnight or 24 hours).
- b. Embedding otolith in resin method:
 - Mark the otolith nucleus with a pencil to facilitate alignment on the sectioning saw.
 - Prepare a small specimen label (0.5 inch square)
 from paper and indelible ink, recording identification data.
 - 3. A semi-circular PVC trough-like mold (approximately l inch wide x 0.75 inch deep) is used in lengths of 25-30 cm (PVC mold accomodates at least 12 otoliths)
 - 4. The interior of the trough should be lightly waxed with a paste wax to facilitate removal of the cast resin, followed by the plugging of the trough ends with corks.
 - 5. A thin, 5-7 mm thick layer of clear casting resin is then poured into the mold and allowed to gel. This layer provides a base to support the otoliths.

- 6. Each otolith is then placed along its longitudinal axis on the resin base, along with its respective label. Care should be taken to align the otolith along this axis to ensure a perpendicular dorsal-ventral section.
- Casting resin should then be poured over the otolith, completely covering the structure. This resin should be allowed to harden overnight or for 24 hours.
- The resin bar can be readily removed by twisting the PVC mold slightly.
- The embedded otoliths in the resin bar may be sectioned, employing any of the thin-sectioning equipment described below.
- c. Epoxy-mounting method:
 - 1. Refer to Chilton and Beamish (1982) for a description of the epoxy-mounting method.
- b. Sectioning.
 - 1. Using the Buehler Isomet Low-Speed Saw to cut sections:
 - a. Place chuck in correct position (11-1184 single saddle chuck).
 - b. Place card holder in the chuck so that the saw blades will pass through the focus of the otolith on the card.
 - c. Place two blades (blue writing on the blades should face each other) on the saw with acetate or brass spacer between the blades. The separator should be about 1 1/2 cm smaller in diameter than the blades.

Use sufficient spacers to produce sections 0.5 mm thick. Thin sections may also be produced using a single blade. Following the first cut, position the otolith for the second cut by advancing the micrometer 20 microunits. This produces a section approximately 0.4 mm thick. A slow cutting speed of 3 is recommended.

- d. Adjust the stop on the card holder arm so that the machine stops just as it cuts into the Dennison marking tag (card).
- As you section, keep the cooling bath full of distilled water. The addition of a small amount of liquid soap aids in lubrication.
- f. Place a card with embedded otolith in the card holder.
- g. Adjust the holder so that the focus of the otolith will be centered on the cutting blades.
- h. Start the saw at a setting of about 7.
- i. Slowly lower the otolith onto the MOVING blades. A medium-sized weight should be on the chuck holding arm or the cutting will be too slow. Increase cutting speed to 10.
- j. The machine will stop automatically. When it stops, carefully lift up the arm and secure it with a rubber band.
- k. Remove the otolith card.
- Remove the section with foreceps. If a portion of the section is missing, it may be between the saw blades or in the coolant bath.

- m. Keep the blades sharp. The speed of sectioning will decrease if the blades are not sharp.
- 2. Using the Bronwill Sectioning Machine to cut sections:
 - a. Otoliths are encased in epoxy for support during the sectioning process. Otoliths may be sectioned 24 hours after embedding in epoxy.
 - b. Clamp the otoliths in the chuck of the Bronwill machine.
 - c. Several sections of about 0.5 mm are cut and placed on a slide.
 - d. For further detail, see Chilton and Beamish (1982).
- 3. Mounting otolith sections on glass slide.
 - a. Label glass slide with unique otolith identification data.
 - b. Place finger nail mender or histological mounting medium at the center of the slide, using about one drop.
 - c. Put otolith section on mounting medium. Allow to dry overnight.
 - d. Grind and polish if desired, prior to ageing.
 - e. Either put nail mender on top surface of the sectioned otolith OR brush with vegetable oil just prior to ageing.
- c. Ageing the otolith.
 - Age under a dissecting or compound microscope using transmitted light.
 - Increased magnification is often necessary to identify annuli deposited on older otoliths.
 - After ageing, slides should be cleaned and placed in slide boxes or trays for storage.

- Axis of reading.
 - a. The axis of reading may vary among rockfish species. The preferred counting area for most species is from the nucleus to the dorsal, internal (proximal) surface (area II in Figure 3).

 Age on each otolith section should be determined twice, preferably on different axes or areas.
- Criteria used in ageing.
 - a. A single year's growth in the section is interpreted as a pair of opaque-translucent zones; under transmitted light, the opaque zones deposited in later years appear as thin lines, the translucent zones are thicker.
- 5. Efficiency.
 - a. For all phases of the thin sectioning work (preparation, sectioning, ageing) it is estimated that 45-55 structures may be processed per person per day. Different methodologies may differ slightly in efficiency.

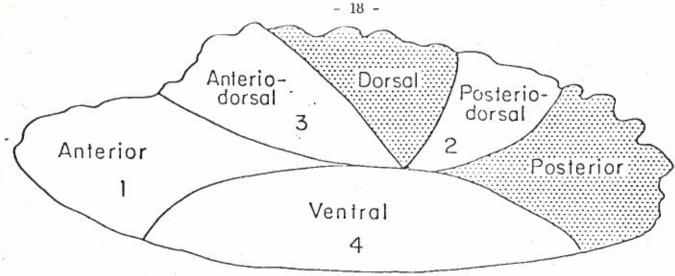
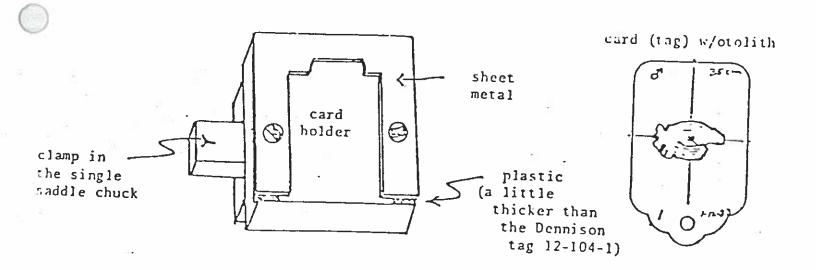


Figure 1. Drawing of a rockfish otolith showing various axes used in ageing.



Single saddle chuck and Dennison double-thickness tags used when mounting an otolith on a sectioning card. Note orientation of otolith on sectioning card.

DISTAL OR EXTERNAL EDGE

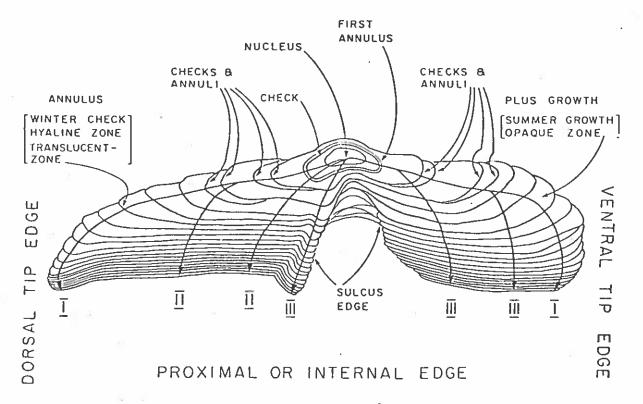


Figure 3. Drawing of an otolith cross section showing areas used for counting and pertinent ageing characteristics.

Taken from Chilton and Beamish (1982).

First Draft of Suggested Sablefish Addendum to the Proposed Rockfish Ageing Manual

General Comments on Growth/Ageing of Sablefish

It has been agreed that the preferred structure for ageing sablefish is the otolith and the preferred method for ageing is the burned section.

It appears that sablefish grow very rapidly in the early years of their life and rather dramatic slowing down can occur after four or five years. Whether the slowing down of otolith growth is related to environmental circumstances, sexual maturity or other factors has not been conclusively demonstrated. Agreement as to interpretation of checks laid down subsequent to the change in otolith growth has not been achieved; however, recognition of radical change in the pattern and size of rings after four to five years was agreed upon by all groups. It has also been suggested that these changes in growth pattern (rate) may vary between stocks of sablefish. It is hoped the work currently being conducted by several agencies will result in resolution of these questions in the future.

Recommended Procedures

It has been agreed that the general procedures developed for ageing rockfish otoliths can be applied to sablefish with the following exceptions:

- (1) Whereas rockfish may be aged using less than 100X magnification it is recommended that 100X or greater be available for use on sablefish. Fiber optics should be used for lighting.
- (2) Due to the very small size of a sablefish burned section it is possible to scan the entire surface. The ventral edge is usually the best to examine, however, any clear readable axis may be used. One should attempt to validate potential annuli by tracing to the sulcus edge.

(3) To define the first two (three) annuli it is recommended they be traced from the surface by tipping after b&b to trace them. It is suggested marking the first few years on the surface with a very sharp pencil will aid in locating these checks on the burned surface. Another suggestion is to embed the unburned half of the otolith section adjacent to the burned one in clay to compare the surface and b&b check marks.

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PROPOSED INTER-AGENCY AGE CALIBRATION MECHANISM

bу

Committee of Age Reading Experts (CARE)

Statement of Purpose: The purposes of this inter-agency groundfish age calibration mechanism are: 1) to determine the level of agreement (precision) of age determinations among the Pacific Coast fisheries management agencies for the principal rockfish (genus Sebastes) species of interest, 2) to ascertain the degree of ageing bias by a reader or agency and, 3) to work cooperatively to resolve differences of interpretation which lessen ageing agreement. It is not within the present scope of this mechanism to assess the accuracy of groundfish age assignments, for samples of known-age fishes are notably lacking.

Only those agencies actively ageing a particular species or who otherwise wish to be included in an exchange should participate in this cooperative exchange. There is little to be gained from the participation of a reader who is unfamiliar with the species in question.

These inter-agency age calibration procedures are presently restricted to the principal rockfish species that are aged on a production basis by Pacific Coast fisheries management agencies. If successful, the program's scope could be expanded to encompass other groundfish species.

Before implementing an inter-agency age calibration mechanism, we propose a pilot experiment to establish appropriate sample sizes for each species of concern and to resolve any potential logistical problems with an exchange of otoliths. This proposed mechanism will then be finalized and implemented by all participating fisheries agencies.

SAMPLE SIZE AND CHARACTERISTICS: Age determination agreement for many rockfish species is often negatively correlated with specimen age. To insure that candidates for all age groups are included in the pilot exchange, each species sample should include at least two specimens per each 2 cm size group commonly encountered to achieve a total sample size of 30 specimens. Within-reader and between-reader variance levels will be determined for the pilot sample, so that statistically meaningful sample sizes can be established.

Initially, one agency will be assigned the responsibility to prepare a species sample for distribution. This responsibility will then rotate among the agencies with an interest in that species. Each otolith pair should be closely scrutinized for crystallization or a poorly-cleaned surface and excluded if either condition will negatively affect the ageing comparison.

Date of capture and catch locality data should accompany the otoliths. Because otoliths will be aged with the burned-section technique, all samples should be sent dry.

AGEING TECHNIQUE: The burned-section technique has been recognized as the "state-of-the-art" technique and will be the principal technique employed in this otolith exchange. One intact whole otolith per specimen will be provided as an aid to the interpretation of the burned section or for the surface ageing of young specimens.

The surface ageing technique should be restricted to those individuals, primarily young fish, for which surface clarity is demonstrably superior to the burned section. Use of the surface technique to assign specimen age must be noted in the "Comments" section of the data sheet.

HANDLING OF OTOLITHS: The agency reader who initially provides a sample may burn and prepare both halves of an otolith from each pair and then may select the better section for inclusion in the exchange. The burned section should be coated with a thin layer of fingernail polish to preserve the burned surface, thus providing a relatively nonperishable structure. For each repeated reading, the preserved section may be coated with a non-toxic oil or immersed in a shallow dish of water to minimize reflection and glare.

In order to assess within-reader age assignment variance, replicate readings by the same reader are necessary. Therefore, each structure should be aged three times without access to prior age information.

Replicate readings should be recorded on separate data sheets. A minimum of one week should transpire between repeat readings. The independence of replicate readings is reliant upon the integrity of the reader. The sample should be retained by an agency for no more

than four weeks. The success of this age calibration mechanism relies to a significant degree on the timely circulation of ageing material among the participating agencies. Thus, upon completion of age assignments, each sample should be promptly forwarded to the next agency in the exchange.

TIME FRAME: Preparations for the pilot exchange should commence immediately following acceptance of this proposal by all participating agencies. Completion of the calibration mechanism protocol, based upon the pilot exchange, should be accomplished by mid-1984.

Implementation of this age calibration mechanism should commence at least six months before the Parent Committee meeting to allow for proper analysis and dissemination of results.

AGE DESIGNATIONS: Age determinations (number of annual rings) will be recorded as described in Chilton and Beamish(1982), using the internationally accepted birthdate of January 1. To aid in the resolution of ageing conflicts, the structure edge should be described with one of the following descriptions:

- (1) Translucent (hyaline) zone on edge
- (2) Opaque zone at edge just starting
- (3) Opaque zone at edge moderately to well developed The quality (ease of reading) of otoliths can vary greatly and may unduly influence age determinations. As such, a precision or "readability" index is valuable in the analysis of the comparative age data.

The following readability scale is adapted from that used by Doris Chilton for sablefish.

GOOD- same age for each repeated reading with no doubt FAIR- within two years for each reading

POOR- readings vary considerably or an educated guess As a reader ages a specimen, we recommend that the reader assess the quality of the otolith and its preparation using the readability scale.

DATA ANALYSIS: We recommend analysis of the resulting data employing three primary techniques. All are valuable and complementary diagnostic tools. The first is a standard analysis of variance, testing for within-reader and between-reader age determination differences. Each species sample will be stratified into three length groups of ten specimens each for analysis of age-dependent variation. The second is an index of "average percent error" as developed by Beamish and Fournier (Fisheries and Oceans-Canada). Staff from the Pacific Biological Station-Nanaimo have agreed to evaluate exchange results with this method. The third is the traditional percent agreement method for which George Hirschhorn (NWAFC-Seattle) has an excellent computer program designed to graphically present results of two-reader comparisons. The plots produced by this program are useful in identifying sources of ageing error. These plots, in concert with edge type and readability indices, will be valuable for post-testing training purposes.

One agency should act as a "clearing-house" for completed data sheets to expedite analysis. The Northwest and Alaska Fisheries Center is a logical choice for this role, for NWAFC personnel will conduct some of the analyses of the exchange results.

FUTURE WORK: An additional goal of CARE is to develop a set of nonperishable reference age structures for each rockfish species of interest. The feasibility of such a reference collection hinges upon
the development of a treatment which preserves the quality of the
burned otolith surface. Initial results using fingernal polish on
otoliths appear promising. Further evaluation of this treatment is
necessary before long-term preservation of burned sections can be assured.

SPECIES OF INTEREST BY AGENCY:

Sebastes alutus

Alaska Dept. of Fish and Game (ADFG)

NMFS-Northwest and Alaska Fish. Center (NWAFC)

Oregon Dept. of Fish and Wildlife (ODFW)

Pacific Biological Station (PBS)

Washington Dept. of Fisheries (WDF)

S. entomelas

NMFS-Tiburon

WDF

S. flavidus

NMFS-NWAFC

NMFS-Tiburon

PBS

WDF

S. goodei

California Dept. of Fish and Game (CDFG)

NMFS-Tiburon

S. melanops

ADFG

NMFS-Tiburon

WDF

S.pauciapinia

NMFS-Tiburon

PBS

S. pinniger

NMFS-NWAFC

NMFS-Tiburon

ODFW

PBS

WDF

Committee of Age Reading Experta

Age Calibration Mechanism Subcommittee

Francis D. Henry

Doris Chilton

Ruth Mandapat

Julie Lyons

REFERENCES CITED

Chilton, D.E., and R.J. Beamish, 1982. Age determination methods for fishes studied by the Groundfish Program at the Pacific Biological Station, California. Spec. Pub. Fish Aq. Sci. No. 60, 102 p.