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Age Determination Methods for Northwest Atlantic Species

Judy Penttila
Louise M. Dery (editors)

U.S. Department of Commerce

NOAA TECHNICAL REPORT NMFS

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U.S. DEPARTMENT OF COMMERCE
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National Marine Fisheries Service
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ABSTRACT

The successful application of techniques to enhance detection of age marks in biological specimens is of vital importance in fisheries research. This manual documents age determination techniques used by staff at the Woods Hole Laboratory, National Marine Fisheries Service. General information on procedures for preparing anatomical structures is described, together with criteria used to interpret growth patterns and assign ages. Annotated photographs of age structures are provided to illustrate criteria. Detailed procedures are given for the following species: Atlantic herring (*Clupea harengus*), haddock (*Melanogrammus aeglefinus*), Atlantic cod (*Gadus morhua*), pollock (*Pollachius virens*), silver hake (*Merluccius bilinearis*), red hake (*Urophycis chuss*), black sea bass (*Centropristis striata*), weakfish (*Cynoscion regalis*), Atlantic mackerel (*Scomber scombrus*), butterfish (*Peprilus triacanthus*), redfish (*Sebastes fasciatus*), summer flounder (*Paralichthys dentatus*), winter flounder (*Pseudopleuronectes americanus*), witch flounder (*Glyptocephalus cynoglossus*), American plaice (*Hippoglossoides platessoides*), yellowtail flounder (*Limanda ferruginea*), surf clam (*Spisula solidissima*), and ocean quahog (*Arctica islandica*).

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Introduction

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The ability to perform age determinations based on examinations of hard anatomical parts is of fundamental importance in fisheries research. As for trees, for which an age may be determined by counting annual rings in a cross section of the trunk, certain structures of finfish and bivalve molluscs taken from temperate waters also show alternating structural marks caused by changes in growth rates. Validation of a regular periodicity in these marks permits assigning a time scale and determination of age. The successful application of techniques to enhance detection of age marks in biological specimens is of vital importance in estimating growth and mortality rates, population age structure, and other parameters needed for understanding the dynamics of fishery resources and their response to natural phenomena and exploitation.

A wide variety of age-determination techniques have been developed for finfish and bivalve molluscs which depend on detection of contrasting bands in body parts such as scales, otoliths, fin rays, spines, and bones of fish, as well as external and internal structures of mollusc valves. At the Woods Hole Laboratory, such studies have been conducted for decades and a considerable body of information has been compiled for a variety of Northwest Atlantic species. In many cases, however, these methods have not been formally published (or were published in an incomplete form). The purpose of this manual is to document the techniques used by staff at Woods Hole for researchers dealing with similar species and problems in other regions.

A brief history of the various investigations and units responsible for age assessment at the Woods Hole Laboratory is given as background information. The Laboratory was first established in 1885, although studies of age and related research was fairly limited in the early years. The Laboratory was closed during World War II, and significant progress on age research did not resume until the Laboratory was reopened in 1947 and the North Atlantic Fishery Investigation of the Fishery Biology Branch, U.S. Fish and Wildlife Service, was established. Groundfish resource surveys were initiated to investigate the biology and resource potential of various fish stocks, with age reading conducted within "species" investigations by project leaders and their scientific aids and technicians. Age determinations for most species, however, were sporadic and were completed to answer specific research needs at the time, in contrast to a sustained production mode which has been characteristic of more recent years. Development and validation of techniques concurrently supported programs of the International Commission for the Northwest Atlantic Fisheries (ICNAF) which was organized in 1951 for the management of the groundfish fisheries of the Northwest Atlantic. Age determination studies conducted from 1951 through 1964 focused on haddock (*Melanogrammus aeglefinus*), redfish (*Sebastes fasciatus*), Atlantic herring (*Clupea harengus*), silver hake (*Merluccius bilinearis*), yellowtail flounder (*Limanda ferruginea*), Atlantic cod (*Gadus morhua*), scup (*Stenotomus chrysops*), summer flounder (*Paralichthys dentatus*), winter flounder (*Pseudopleuronectes americanus*), and Atlantic sea scallops (*Placopecten magellanicus*).

In 1965, species investigations at the Laboratory were aggregated into the Population Dynamics Program and a separate age determination unit was established. The work of the program involved collection of catch information, processing and determining the age of biological specimens, automatic data processing, and research on vital statistics, yield, and population processes. The new Age Reading Unit initiated routine ageing of haddock and yellowtail flounder and conducted preliminary studies from 1965 to 1970 to develop and validate ageing techniques for species such as fourspot flounder (*Paralichthys oblongus*), American plaice (*Hippoglossoides platessoides*), red hake (*Urophycis chuss*), and pollock (*Pollachius virens*). Experiments with staining otoliths were also conducted. Some species (e.g., redfish and Atlantic sea scallops), however, were still aged by individual investigators in other units.

During the early to mid-1970's many new techniques for preparing structures for age determination were developed, e.g., thin-sectioning and baking otoliths, and using laminated plastic for scale impressions. The number of species routinely examined for age (i.e., in a production type of mode) gradually increased through the 1970's to a current total of 18, and methods for an additional ten species have also been developed.

In 1978, the Fishery Biology Investigation was created. Currently, the Investigation is part of the Conservation and Utilization Division (CUD) of the Northeast Fisheries Center (NEFC). The primary function of the Investigation is to provide biological information required for assessing the status of selected fishery resources, including population age compositions, mortality and maturation rates, growth and fecundity parameters, and physiological and behavioral characteristics.

Major emphasis, however, is on routine assessment of age for 45,000 to 50,000 specimens, representative of commercially and recreationally important finfish and bivalve molluscs in the Northeast region. Current studies focus on growth analyses; age validations; development of new and more cost-efficient methods; studies of size and age at sexual maturity; automatic image analyses of age structures (Cambridge Instrument Company, Inc. 1980), including optical Fourier transform analyses for stock identification (Almeida et al. 1987); and examination of daily growth increments on larval otoliths (Jearld 1983, Campana and Neilson 1985).

This manual describes methods currently in use for biological sample preparation and age determination of most finfish and bivalve species for which the Investigation has responsibility. The various techniques used for preparing anatomical structures are described as well as criteria used to interpret growth patterns and to assign ages. Many of these methods and criteria have not been formally validated and must be considered "experimental." The age determination process consists of the following steps: collection and storage of age samples, preparation of structures for age determination, examination, interpretation, and assessment of the validity and reliability of the resulting data. Most specimens examined are from samples taken during routine NEFC bottomtrawl and shellfish resource surveys; specimens from commercial landings are, however, also collected at dockside by NEFC port samplers.

The first part of this manual contains general information on processing specimens for age determination and a glossary of terms. The remainder of the manual describes specific procedures developed for individual species. The species descriptions include information on biology and distribution; former studies of age and structures used for age determinations; sample storage, preparation, and methods of examination; and descriptions of growth patterns and problems related to age determination.

Table 1 lists the species considered in this manual, the age structure examined, specimen preparation method generally in use, average number of specimens aged each year (if the species is aged routinely), and the time series available in each case.

Note: In the figures for all sections which follow, black dots indicate annuli; black dashes indicate checks, splits, or false annuli.

Table 1

Age structures, preparation methods, average number of specimens examined each year, and time series of available data for species included in this manual.

Species	Age structure	Preparation method	Average number aged/year	Year samples first collected[1]
Atlantic herring	otoliths	embedded	4,800	1973
Haddock	scales	impressions	3,500	1931
Atlantic cod	otoliths	baked	4,450	1960
Pollock	otoliths	sectioned	2,550	1966
Silver hake	otoliths	sectioned	2,750	1955
Red hake	otoliths	sectioned	1,450	1964
Black sea bass	otoliths	sectioned	none	1980
Weakfish	scales	impressions	none	1978
Atlantic mackerel	otoliths	embedded	2,200	1973
Butterfish	otoliths	whole	2,550	1964
Redfish	otoliths	sectioned	2,500	1964
Summer flounder	scales	impressions	3,300	1974
Winter flounder	scales	impressions	3,250	1973
Witch flounder	otoliths	sectioned	1,450	1973
American plaice	otoliths	sectioned	3,200	1971
Yellowtail flounder	scales	impressions	5,500	1955
Surf clams	chondrophore	sectioned	2,950	1978
Ocean quahogs	valve	acetate peels	50	1978

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[1] In each case, sampling has been continued to the present.

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Thanks are also due to Ambrose Jearld, Jr., who originated the idea for the project.

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Glossary of Terms

Age determination notation

5	Five annuli counted (only one clear interpretation)
5(6)	Probably five, but possibly 6 annuli (moderately difficult to age, two interpretations possible)
5?	Five annuli is the best estimate (difficult to age, more than two interpretations possible)
5+	Five annuli counted with an additional seasonal growth increment

Annulus Any zone which forms once each year, usually the "winter" growth zone which marks the end of a year of growth.

Bivalve terminology for age determinations See figures 9 (p. 13) and 11 (p. 13).

Check Zone of slow "winter" type growth which is not a true annulus. Such rings are distinguished by the width of the zone relative to annuli, location relative to annuli, and incomplete formation or poor definition. Checks may also be differentiated from annuli on some scales by differences in platelet shape.

Chondrophore "Pit or large spoon-shaped form projecting from the hinge plate, usually supplemented by a prop extending to the surface of the valve"[1]

Circulus A concentric ridge formed on a scale by the periodic addition of material to the edge of the basal plate. The circuli on scales may be continuous or segmented by the scale radii, in which case the individual segments are termed platelets. Circuli are formed only on the outer surface of the scale; the inner surface is smooth. Circuli formed on bivalves are concentric, scalloped ridges which become crowded together at an annulus.

Collum Interruption in the sulcus acusticus which marks the location of the nucleus (Fig. 5, p. 12).

Crystallized otolith An otolith displaying inadequate calcification (Fig. 19, p. 15). An age determination is not possible because of missing annuli.

Ctenoid scale Type of scale having ctenii, or spine-like projections resembling the teeth of a comb, on its posterior edge.

"Cutting over" ("crossing over," erosion marks) Disruption of the circulus pattern on scales from erosion of the edge results in circuli formed after erosion that appear to intersect or "cross over" others that had been formed earlier. If scale edge erosion is an annual event, the "cutting-over" marks may be used to detect annuli.

Cycloid scales Scales that are oval or elliptical in shape.

Edge Outer periphery of the age structure.

Edge type Summer/winter or opaque/hyaline deposition occurring on the outer edge of the age structure representing the most recent growth.

End of annulus Outermost edge of a winter growth zone designated as an annulus.

False annulus Sometimes used synonymously with "check," refers to a zone of slow growth that is not counted as an annulus; also, a characteristic check ring on scales or otoliths which occurs before the first annulus and fairly close to the focus (scales) or nucleus (otoliths).

Focus Center or origin of a scale.

Hinge "Interlocking toothed devices in a bivalve; hinge plate is the dorsal margin carrying the hinge teeth; hinge teeth are interlocking teeth that unite the valves" (Arnold 1965). Annuli occurring in the hinge teeth may correspond in number and relative location to annular lines seen in the valves of molluscs.

Hyaline Zone that allows the passage of light (also referred to as translucent). On otoliths, the "hyaline" zone is composed primarily of organic material (otolin) with a reduced amount of inorganic material in the form of short, thin calcium aragonite needles. With transmitted light, hyaline zones appear bright; with reflected light, they appear dark. "Winter" zones are normally composed of hyaline material.

Lumen Central cavity of a spine.

Margin Edge of a valve. The ventral valve margin of a bivalve is often referred to, since it represents the most recent accretion of shell growth.

Nucleus Central portion of an otolith; sometimes used synonymously with the terms core, kernel, or primordium.

Opaque Zone that inhibits the passage of light. On otoliths, the "opaque" zone is composed primarily of inorganic calcium aragonite needles which are long and thick relative to those formed in hyaline zones. With transmitted light, opaque zones appear dark; with reflected light, they appear bright. "Summer" zones are normally composed of opaque material.

Otolith terminology for age determinations See figures 5 (p. 12) and 20 (p. 16).

Platelets Individual segments of a circulus on some types of scales which are separated by the scale radii.

Regenerated scale Scale which replaces one previously lost. These cannot be used for age determination because the central area has no circuli or annular growth features (Fig. 18, p. 15).

Sagittae Largest of three pairs of otoliths located in the sacculus of the inner ear of a fish; referred to simply as "otoliths" in the following sections.

Settling check Characteristic check ring on some marine groundfish otoliths. It occurs just outside the nucleus and is believed to form when the fish first become benthic in habit.

Shifted otolith Otolith which has moved in the sacculus; recognized by additional growth occurring along a different axis from previous growth. Annuli may thus be present only on certain parts of a shifted otolith, and absent on other parts. Shifting often occurs in conjunction with crystallization of an otolith.

Split Discontinuity in an annular zone, analogous to a "check." This causes the annulus to appear as two or more closely spaced "winter" zones.

Sulcus acusticus (referred to simply as "sulcus" in the following sections) Longitudinal groove extending down the convex surface of an otolith (Fig. 5, p. 12).

Umbo "That point of a bivalve situated immediately above the hinge, the beak, the first formed part of a bivalve" (Arnold 1965) [(See figures 9 (p. 13) and 11 (p. 13)].

Valve "One of the separable portions of a shell; bivalve, a shell in two sections" (Arnold 1965).

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[1] Arnold, W.H. 1965. A glossary of a thousand-and-one terms used in conchology. Veliger 7 (Suppl.), 50 p.

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Methods and Equipment

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Collection and storage

The primary sources of age samples processed by the Investigation are Northeast Fisheries Center (NEFC) bottom trawl and shellfish resource surveys and commercial landings. Additional samples are periodically collected during various state-conducted research surveys and by fisheries observers who serve on foreign fishing vessels.

Scales and otoliths are the anatomical structures most frequently collected from finfish. Scales are preferred because they are easier to collect and process, providing, of course, that clearly defined growth patterns are consistently formed. Young-of-year specimens and samples of certain species with fragile or difficult-to-remove age structures, e.g., Atlantic herring (*Clupea harengus*) and Atlantic mackerel (*Scomber scombrus*), are frozen whole for later dissection and processing at the laboratory. Other anatomical structures, such as fin rays or vertebrae, may be collected and used for special studies, such as age validation.

While scales are the easiest structure to collect, they must be taken from an area on the fish known to exhibit complete and clear growth patterns. For gadids and flounders, this area is on either side of the lateral line anterior to the caudal peduncle, the area where the first and largest scales develop. For other species, such as bluefish (*Pomatomus saltatrix*), black sea bass (*Centropristis striata*), striped bass (*Morone saxatilis*) and scup (*Stenotomus chrysops*), scales are removed from the area behind the pectoral fin where the largest scales are located. The area is first scraped with a blunt knife from the head towards the tail of the fish to remove adhering slime and dirt. The knife is then cleaned and used to remove a sample of scales by scraping firmly towards the head of the fish. The knife blade with adhering scales is then placed between the sheets of a folded absorbent paper liner in a coin envelope and wiped clean of the scales.

Otoliths are removed by dissection of the head of the fish with a sharp knife or a bone saw. Only the sagittal otoliths, the largest of the three pairs found in the sacculi of the inner ear located posterior to the brain, are removed for examination. Otoliths to be stored dry are removed from the sacculi (enveloping membranes) before being placed in an envelope. For some species (e.g., Atlantic mackerel, alewife (*Alosa pseudoharengus*) and Atlantic herring), otoliths are stored for one or two days in water-filled vials after dissection, since they require careful cleaning under a microscope at the laboratory.

The following information is recorded on the envelope for each specimen sampled on resource surveys: cruise, station, species, length, sex, and maturity. Corresponding information for specimens collected from commercial sources includes: vessel name, date, statistical area, latitude and longitude, port, depth, gear, species, market category, sampling method, length, and sex.

Surf clams (*Spisula solidissima*) and ocean quahogs (*Arctica islandica*) collected during NEFC surveys are shucked at sea. Whole paired valves are stored in cloth bags with labels referring to station location, date, and number of specimens. To minimize valve damage, small specimens are frozen whole for later processing at the laboratory.

Preparation of age structures

The following are general descriptions of methods for preparation of structures commonly used for age determination in the Fishery Biology Investigation. Other techniques, such as staining, may be used for special studies but are not presented in detail here. Modifications of certain techniques for particular species are described where appropriate. A complete list of equipment used, with specifications and possible commercial suppliers, is given in Appendix A.

Most of the procedures currently in use for finfish were developed in the early 1970's under the direction of Mr. Fred Nichy, then head of the Age and Growth Unit. From 1970 to 1975, he conducted numerous experiments to enhance otolith growth patterns by heating (e.g., burning, "deep frying" in hot oil) and baking in toaster and microwave ovens. Baking otoliths proved to be best for our purposes. His experiments with various plastics from 1972 to 1974 led to the use of laminated plastic for making scale impressions. Other experiments from 1972 to 1974 with low-speed saws and different types of blades led to development of procedures for thin-sectioning otoliths.

Scales

Age determinations using scales may be made from direct observations, scale impressions, or photographs. Actual fish scales are rarely used, however, because they are often covered with dirt or dried and pigmented residue. In addition, they are generally translucent because of their thickness, internal structure, and coloration, rendering growth zones difficult to interpret with transmitted light. Also, scales are not flat, resulting in uneven light diffraction and distortion of the image during microscopic examination.

Scale impressions in laminated plastic film usually avoid the above mentioned problems so that age marks are more detectable. Cellulose acetate plastics are avoided since they require heat or chemicals to soften the surface for adequate scale impressions. However, studies of fish species with thick scales (e.g., striped bass) may require the use of the heavier cellulose acetate plastic. Several types of laminated plastic film, consisting of a thin, soft polyethylene or surlyn layer over a thicker, harder vinyl or polyester substrate, are simpler to use and produce consistent results for most species. Representative compositions are as follows (Dery 1983):

Substrate	Middle Layer	Surface Layer
0.203 mm (0.0080 inch) semi-rigid polyester	0.002 mm (0.0001 inch) saran	0.032 mm (0.0013 inch) polyethylene
0.203 mm (0.0080 inch) semi-rigid polyester	0.002 mm (0.0001 inch) saran	0.019 mm (0.0008 inch) surlyn
0.190 mm (0.0075 inch) polyvinyl chloride	none	0.051 mm (0.0020 inch) surlyn
0.190 mm (0.0075 inch) polyvinyl chloride	none	0.051 mm (0.0020 inch) polyethylene
0.185 mm (0.0073 inch) vinyl chloride	0.018 mm (0.0007 inch) saran	0.051 mm (0.0020 inch) polyethylene

Any of the above laminated plastics produce impressions of scale surface features, but those having a middle layer of saran and a surface layer of polyethylene are generally superior.

Scale impressions have several advantages over the direct use of scales. They may be viewed by either transmitted or reflected light, and several scales may be impressed at the same time on one slide allowing for the selection of scales with the clearest features. The impressions are clean, even if the original scale used was not, and are easily stored and handled. The image of the scale is also flatter than the original scale and causes minimal depth-of-focus problems at high magnification. Impressions are fairly easy to prepare and simplify the handling of a large number of specimens.

Scale impressions are prepared by placing several scales, sculptured side up, on a heavy base slide of 1-mm thick (0.040 inch) cellulose acetate plastic. A laminated plastic slide, with the soft side down, is then placed over the scales (Fig. 1). Another heavy plastic slide (0.65-1 mm thick) is placed on top of the laminated plastic slide, and the whole "sandwich" of slides is rolled through a jeweler's press (Fig. 2). The two heavy acetate slides act as cushions to help equalize pressure over the thin and thick areas of the scales, resulting in a more uniform impression. The scales are then removed from the plastic slide and the resulting impression is stored in the original specimen envelope. The impressing procedure must be done in one smooth, continuous motion to avoid distortion of the finished impression. The two rollers of the press must be carefully adjusted to obtain a complete, clear impression. The upper roller of the press is usually canted slightly for impressing ctenoid scales. This applies slightly more pressure to the thin anterior edge of the scale. Also, two laminated slides may be used to "sandwich" very small scales, if it is difficult to distinguish between the sculptured and smooth sides.

Otoliths

Otolith preparation for microscopic examination includes whole, baked and broken, or thin cross-sectioned specimens. Species-specific methodology has been developed and is described more fully in the individual species sections of the manual.

Whole otoliths are microscopically viewed individually in ethyl alcohol or placed in depressions of black plastic trays. Embedding in resin improves contrast and enhances detection of growth zones under reflected light. Whole otoliths of some species, such as the short-lived butterfish (*Peprilus triacanthus*), are examined in ethyl alcohol in the unmounted condition, since they are thin enough for detection of early annuli and prominent, widely spaced later annuli. Pairs of otoliths from such species as Atlantic mackerel, Atlantic herring, and alewives are positioned in circular depressions in black molded plastic trays (Watson 1965) and embedded in Permount or clear fiberglass resin (Fig. 3). Permount may in time react with the molded plastic, producing air bubbles, "yellowing," or crystallization. Application of a few drops of solvent (xylene) after several months of storage removes air bubbles and stabilizes the resin for permanent storage in a sealed bag. Fiberglass casting resin will not adhere to molded plastic, but may be used with other materials such as plexiglass (a high-density acrylic). Other resins ("Eukitt," used in Europe, and Canadian balsam) are either prohibitively expensive or difficult to procure.

Currently, only Atlantic cod (*Gadus morhua*) otoliths are baked, a process that takes from 3 to 6 minutes in a scientific radiant heat oven at about 275°C (525°F). Small otoliths tend to require more baking than large otoliths, possibly as a result of their more rapid growth and greater diffusion of protein. Properly baked otoliths are a caramel color; a grey or ashy color is an indication of overbaking, which may cause the otolith to crumble when broken in half at the nucleus. Visibility of the annuli is enhanced by baking, since the hyaline zones turn brown in contrast to the white opaque zones. Burned otoliths may fade with time, but baked otoliths remain unchanged even after storage for several years.

Dry otoliths are thin-sectioned on an Isomet low-speed saw (Nichy 1977) using a pair of fine-grit diamond-impregnated blades separated by a spacer approximating the desired thickness of the section (Fig. 4). Carborundum blades are an alternative, but they tend to break quite easily. Sectioning is accomplished by mounting the otolith on a small cardboard tag bearing crosslines that facilitate proper alignment. Otoliths of most species are positioned to obtain a transverse cross-section across the collum of the sulcus (Fig. 5). A small piece of double-sided tape covers the crosslines at the center of the tag and secures the otolith in the proper alignment for sectioning. Some species have fragile otoliths and the sections break easily. For these, a bed of molten wax is flooded onto the tag and the otolith is positioned on the wax bed before it hardens completely. The wax is heated in a double boiler or egg poacher, with glycerin in the base, on an adjustable-temperature hot plate at approximately 115°C. The otolith is then completely embedded in a mixture of four parts molten paraffin wax, one part decolorizing carbon (enough to just turn the wax black), and three parts calcium oxide powder (enough bulk to prevent the wax from running and to provide additional abrasive action during sectioning). Only a thin layer of wax covering the otolith is required. After the wax has hardened, the tag is inserted in a custom-machined slotted holder on the saw which aligns the otolith for cross-sectioning by the blades (Fig. 6). The saw's micrometer adjustment may be used for final alignment to produce a precision cut. Two 7.6-cm (3-inch) diameter blades separated by a 6.35-cm (2.5-inch) plastic or metal spacer are mounted on the saw unit to produce a thin-section with one cut. Spacer thickness varies from 0.015 to 0.030 mm (0.006 to 0.012 inch) depending on the viewing requirements for age marks in the section. The diameter of the spacer is normally the same, or slightly smaller, than the flanges supporting the two blades.

The saw is operated at maximum rpm (300) and the otolith is gently lowered onto the spinning blades with their rims immersed in a lubricating/cooling solution of 15 parts cold water to 1 part clear, liquid dishwashing detergent. (If foaming is excessive, the amount of detergent may be decreased.) The detergent solution also washes away particles of wax from the blade surfaces. A balancing weight is used on the saw arm that is light enough to keep sectioning time between 1 and 2 minutes. This also avoids warping the blades. The automatic shut-off on the saw is adjusted to allow the blades to just begin cutting through the double-stick tape, but not into the tag itself. After completing a section, the tag is removed from the slotted holder and bent along the cuts (Fig. 7). This exposes the section for removal and placement on a small square of black construction paper. It is folded inside a protective piece of paper, and returned to the specimen envelope with the cut otolith remaining on the tag. Preparation and sectioning of an otolith generally takes about 2 to 3 minutes.

Bivalves

Age determinations of surf clams are made from thin sections of chondrophores (Ropes and O'Brien 1979). For large surf clams, a portion of the chondrophore is first excised using a pair of 25.4 cm (10 inch) diameter diamond-impregnated sawblades spaced 4 mm apart and mounted in a high-speed (1725 rpm) saw unit (Fig. 8); those of small surf clams are excised using 10-13 cm (4-5 inch) diameter blades and a low-speed (300 rpm) saw to minimize breakage. The excised portion is broken away from the valve by finger pressure. Accurate age determination depends upon careful orientation of the valve during excision of the chondrophore so that the excised portion contains the earliest formed portion of the valve, termed the umbo (Fig. 9).

After minor polishing on carbide paper to flatten the surface and remove saw marks, the excised portion is glued to a glass slide using epoxy cement and thin-sectioned by a single blade on the low-speed saw (Fig. 10). An acceptable section is about 0.25 mm thick and takes less than 15 minutes to cut. Surf clams have a chondrophore in each valve of a pair, so the second valve may be processed if the first does not produce a suitable thin-section. Photographically enlarged prints may also be obtained by using the section as a negative in a photographic enlarger. At maximum lens aperture opening of a photographic enlarger, a 5-second exposure of the image onto photographic paper usually produces a suitable print.

Age determinations for ocean quahogs are made from acetate peels (Ropes 1987). The left valve is used, since it has a single prominent tooth in the hinge containing annuli which can be exposed by sectioning. The valve is marked on the ventral margin at a point from the posterior end equal to one-third of the valve length (Fig. 11). This orients sectioning through the umbo and parallel to the broadest tooth surface. The valve is fastened to the adjustable arm holder of an Isomet low-speed saw with its concave, inner surface toward a diamond-impregnated sawblade. The valve is oriented so that the cut is made from the ventral margin through the middle of the tooth, or beside the posterior edge of the tooth for small valves (Fig. 12). The anterior end of the valve is saved for subsequent processing. The cut surfaces are immersed in full-strength household bleach for removal of the periostracum.

An epoxy resin with a colored pigment is used to support the valves during subsequent polishing. The mixed epoxy is poured into molds to a depth of about ½ cm and the sectioned valve is lowered cut-surface-down into a mold (Fig. 13). After an overnight hardening period, three successively finer grits (240, 400, and 600) of carbide paper are used for grinding to expose the cut valve surface, followed by polishing with a vibrating lap machine, to obtain a blemish-free high-gloss surface. Treatment with a 1% hydrochloric acid solution for 1 minute etches the valve surface. Acetate peels are then made by applying an acetate sheet (0.013 mm (0.005 inch) thick) over the etched block surface after flooding it with acetone. After a 1-hour drying period, the acetate is peeled off and sandwiched between glass slides for examination under a light compound microscope.

Methods of examination

Appendix B contains detailed information on equipment mentioned in this section and possible commercial suppliers.

Scale impressions are viewed using microprojectors (Figs. 14 and 15) and microfiche readers (Fig. 16) with transmitted light at magnifications of 20× to 52×, depending on the size of the scales.

Otoliths are examined under binocular stereomicroscopes with a reflected light illuminator inclined at 45-60 degrees at magnifications of 10× to 65× (Fig. 17). Under reflected light, winter (hyaline) zones appear dark and summer (opaque) zones are white. Polarizing filters may be used to reduce glare and enhance contrast. Growth patterns of otoliths or sections are also enhanced by applying wetting agents, such as Kodak Photo-Flo 200, alcohol, glycerin, or clove oil. Whole, individual otoliths are placed in black plastic or clear glass holders with a black base or background and then immersed in the wetting agent for viewing. Embedded otoliths are viewed in the trays with no additional preparation. Broken otolith halves are either hand-held under the microscope or are temporarily mounted in a piece of soft, black plasticene. Otolith thin-sections may be placed on small squares of black construction paper for examination. A wetting agent flooded onto the surface soaks into the paper providing the necessary background contrast.

Thin-sectioned chondrophores from surf clams are examined microscopically using transmitted light. The annuli are translucent and in sharp contrast to opaque growth increments. Photographically enlarged prints of the sections show annuli appearing as dark zones alternating with white growth zones (the opposite of the image seen under transmitted light).

Acetate peels of ocean quahogs are sandwiched between glass slides for examination under a light compound microscope with transmitted light. Annuli appear as dark lines curving down from exit locations at the valve surface toward the umbo; growth increments, which are most evident in the early ontogeny of a specimen, have a lightly textured, granular and homogeneous appearance.

A TV camera monitor connected to a binocular stereomicroscope or dual-viewing heads on binocular stereomicroscopes may be used for viewing various age structures. Such systems are very useful for training personnel and resolving age determination of difficult specimens.

Interpretation and conventions

In temperate-zone waters, both fish and shellfish species exhibit seasonal growth patterns indicative of age. Generally, growth is fast during warm "summer" months, and slow during cold "winter" months. One year of growth consists of one summer zone plus one winter zone. The annulus is usually defined as the winter zone. Summer and winter growth zones differ in appearance, thus providing the basis for age determinations. Increase in length is proportional to the growth of the age structure being used and is a basis for empirical relationships.

By convention, a birthdate of 1 January is assigned for almost all species in the Northern Hemisphere (exceptions to this rule are given in the individual species descriptions). This means that a winter growth zone forming on the edge of the age structure is designated as an annulus on 1 January, even though the zone is not complete.

Growth patterns on age structures and growth rates often vary geographically. Growth is generally faster in more southern areas and slower in more northern areas. Sedentary species tend to show greater geographical variations than migratory species. Geographical patterns are discussed in greater detail under the individual species descriptions.

Certain areas on age structures are preferred for interpretation because the annuli are more distinct or have fewer visible checks. The best area is dependent on the species being examined. Regenerated scales (Fig. 18) or crystallized otoliths (Fig. 19) cannot be used for age determination.

Problems in age determination occur because of deviations in growth. These may result from checks or split annuli occurring in the age structure. Such accessory zones must be recognized as anomalies when assigning an age to a specimen. A knowledge of typical growth patterns helps in distinguishing checks from annuli. Checks tend to be discontinuous, weak or diffuse, and inconsistent with the general growth pattern of true annular zones.

Checks most often occur during periods of rapid growth and are especially common at younger ages. Some may be due to changes in food habits (e.g., a "settling check" forms when some young fish settle to the bottom and begin feeding). Maturation, migration, summer aestivation, or spawning may also cause checks. Some species exhibit distinctive checks typical for certain geographical locations.

Another common problem occurs because of atypical edge growth. For a specimen showing winter edge type in August, a determination must be made as to whether summer growth is retarded (in which case the winter edge zone would be counted as an annulus) or winter growth is advanced (in which case the zone would not be counted), or a check is being formed.

A major problem in assigning age is the determination of the first annulus. Knowledge of the geographic spawning times of a given species helps in determining if the first annulus is expected to be very small. Such annuli consist of minimal growth around the nucleus of an otolith or focus of a scale. For other fish, the first winter zone is expected to be some distance from the nucleus or focus.

Whenever possible, each specimen is examined independently by two age readers, to prevent long-term deviations in results. In general, percent agreement between the two readings has been maintained at better than 85% and exceeds 90% for many species. Comparisons of summaries of age-length data from one season to the next also help maintain consistency in age determinations.

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Watson, J.E.
1965. A technique for mounting and storing herring otoliths. Trans. Am. Fish. Soc. 94(3):267-268.

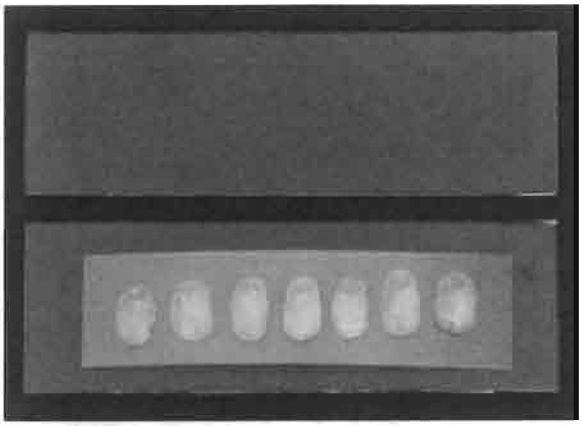


Figure 1
Scales arranged on a base of heavy plastic, sculptured-side-up, and a slide of laminated plastic, soft-side-down, placed over the scales.

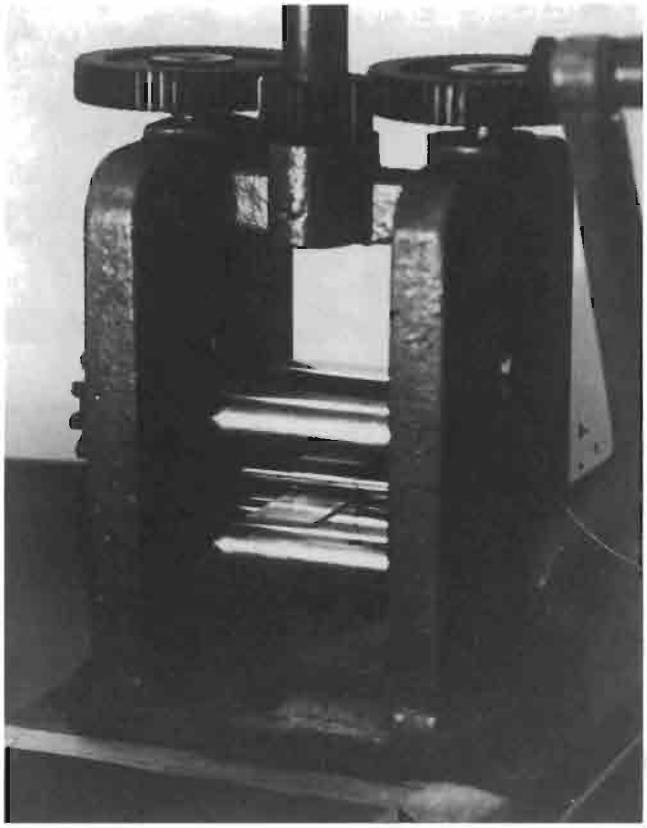


Figure 2
Scales "sandwiched" between plastic slides are passed through a jeweler's press.

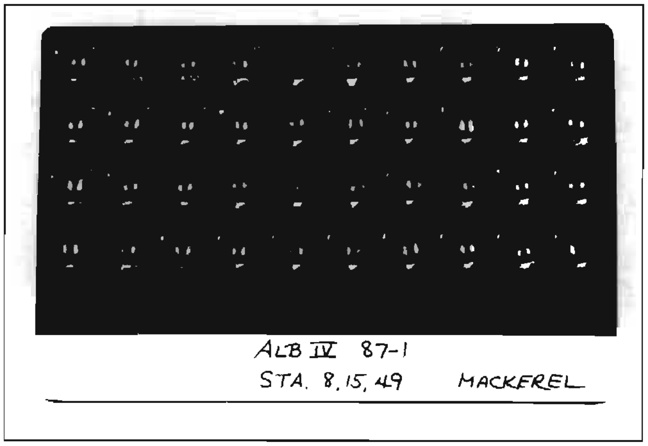


Figure 3
Tray of embedded Atlantic mackerel otoliths.

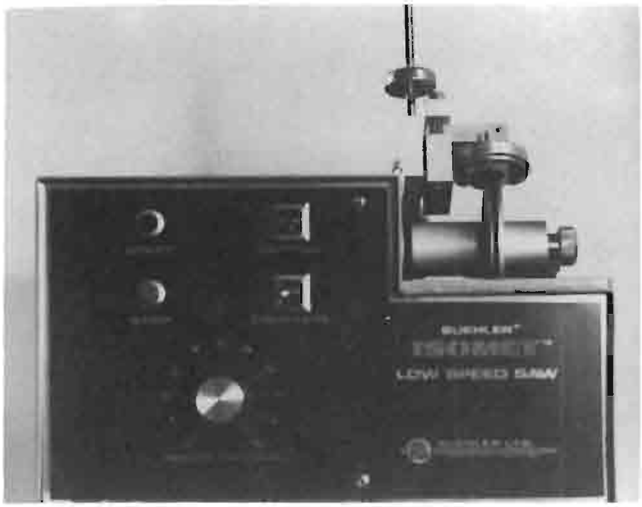


Figure 4
Otolith being sectioned on an Isomet low-speed saw.

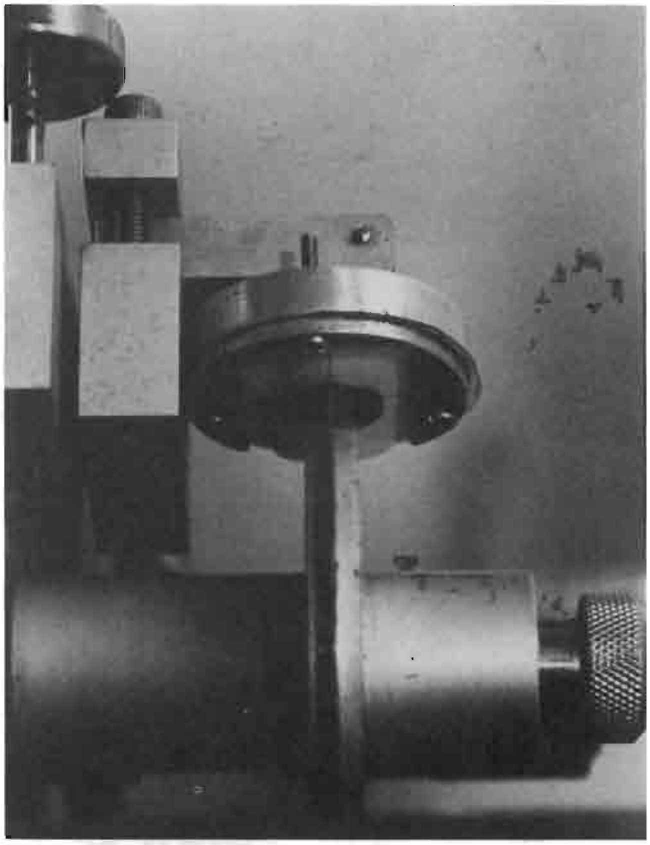
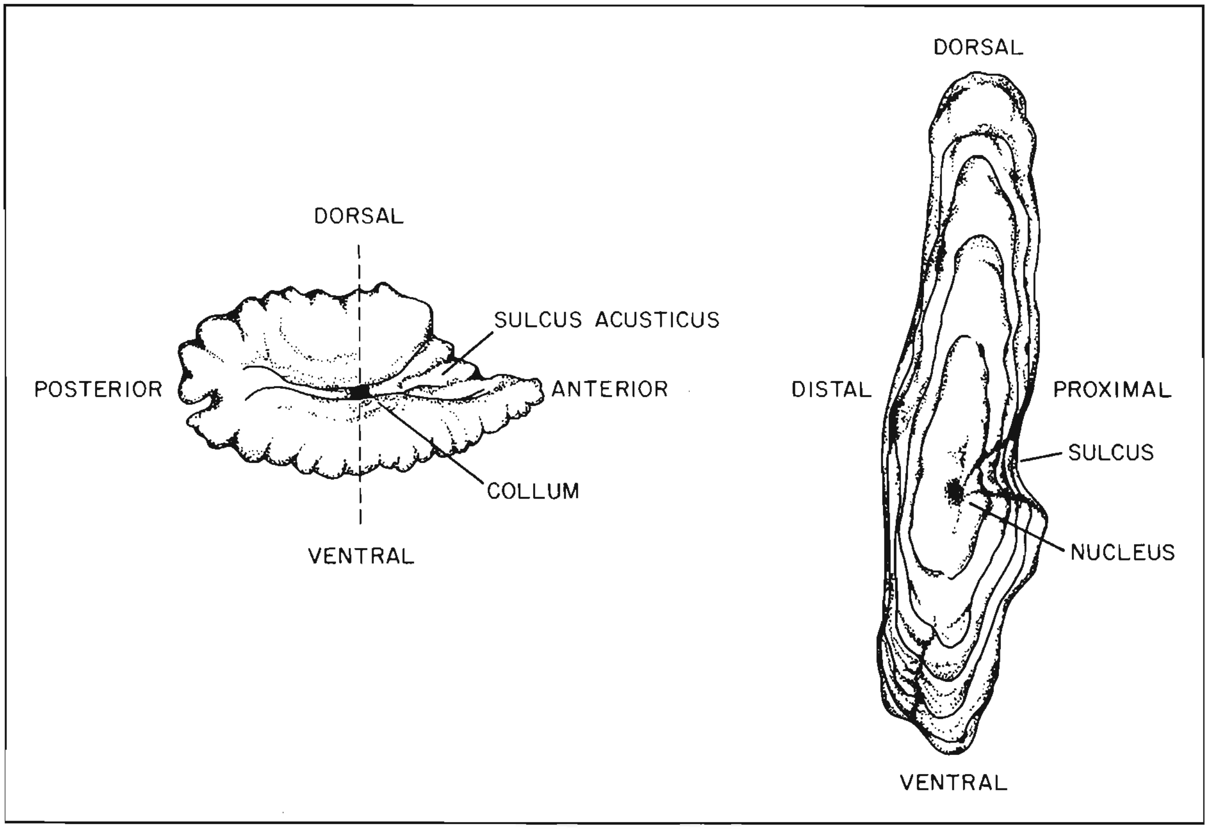


Figure 6
Close-up view of an otolith mounted in the chuck of the low-speed saw being sectioned by two diamond blades.

Figure 5
Sketch of an otolith and cross-section from a demersal species with descriptive terms and direction of cut (dashed line) for removing a thin-section. Proximal side of the whole otolith, with sulcus acusticus and collum, is shown.



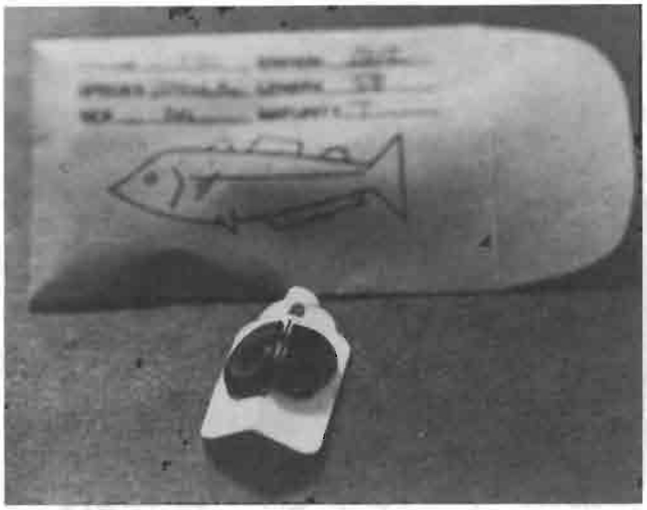


Figure 7
Tag bent open to reveal the thin-section. A printed coin envelope used for specimen storage is in the background.

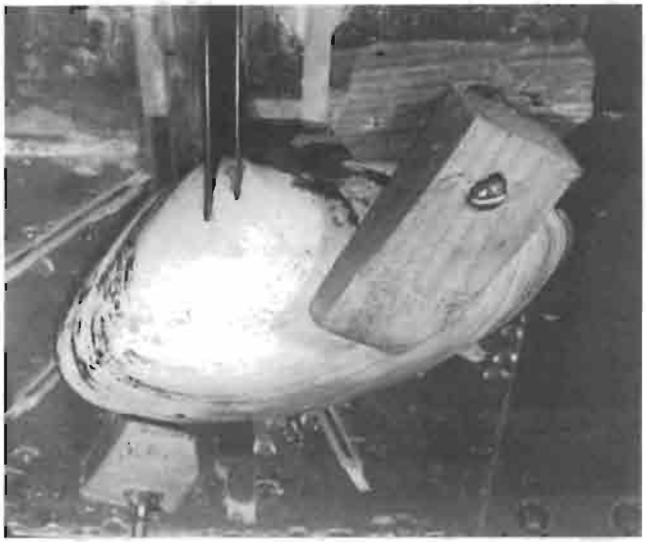


Figure 8
High-speed saw excising a portion of a chondrophore from a large surf clam.

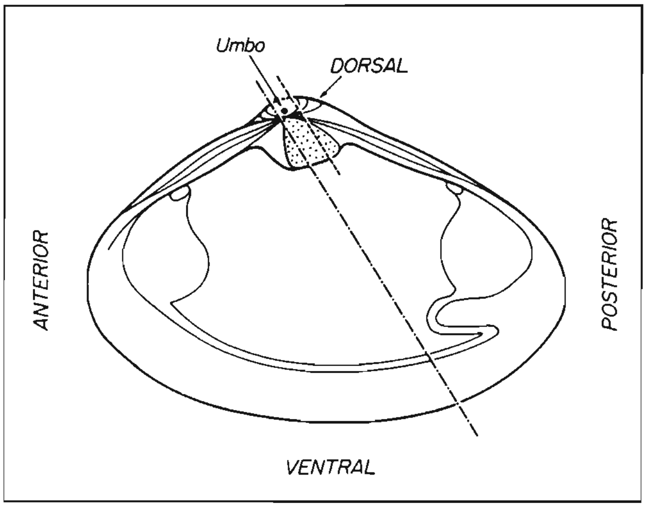


Figure 9
Internal valve features and direction of cuts (dashed line) required to excise the chondrophore from a surf clam.

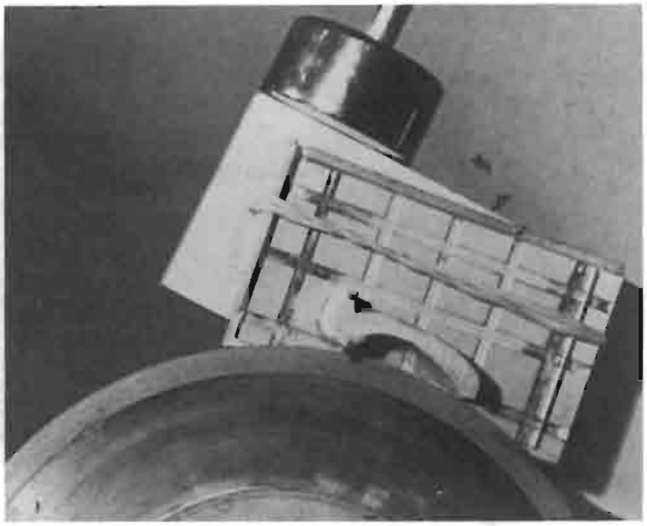


Figure 10
Excised chondrophore portion being thin-sectioned by a single blade on a low-speed saw.

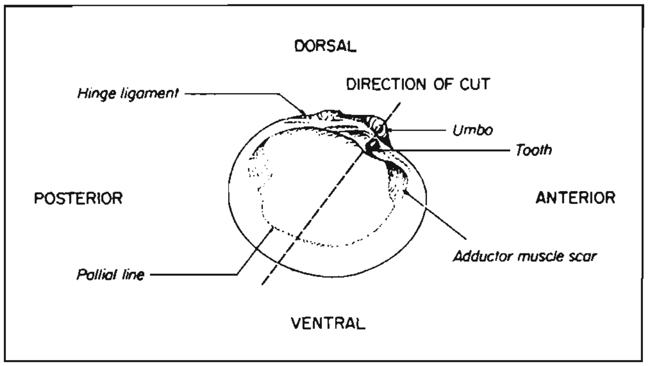


Figure 11
Internal valve features and direction of cut (dashed line) required to completely section the left valve of an ocean quahog.

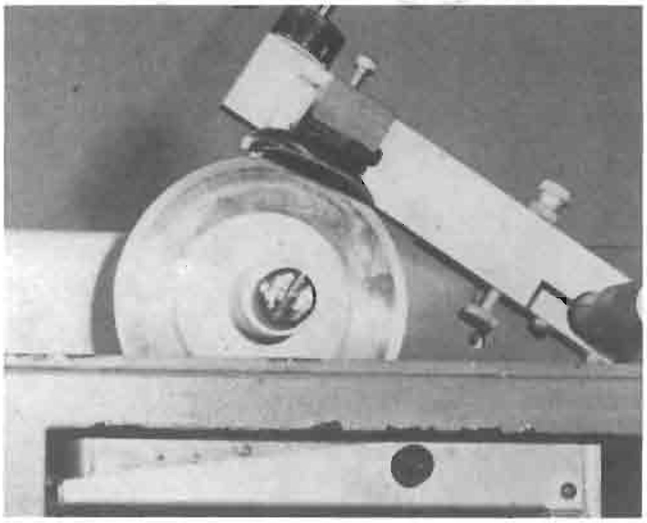


Figure 12
Close-up view of left valve of an ocean quahog fastened to the adjustable arm of a saw unit and oriented with the tooth beside the diamond blade.

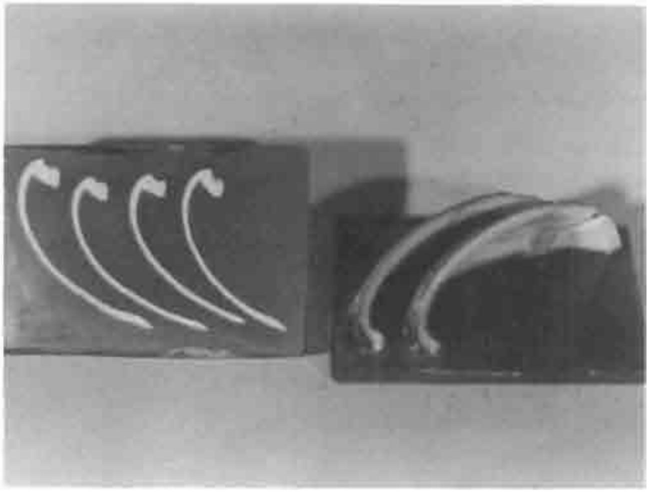


Figure 13
Ocean quahog valves embedded in epoxy molds.

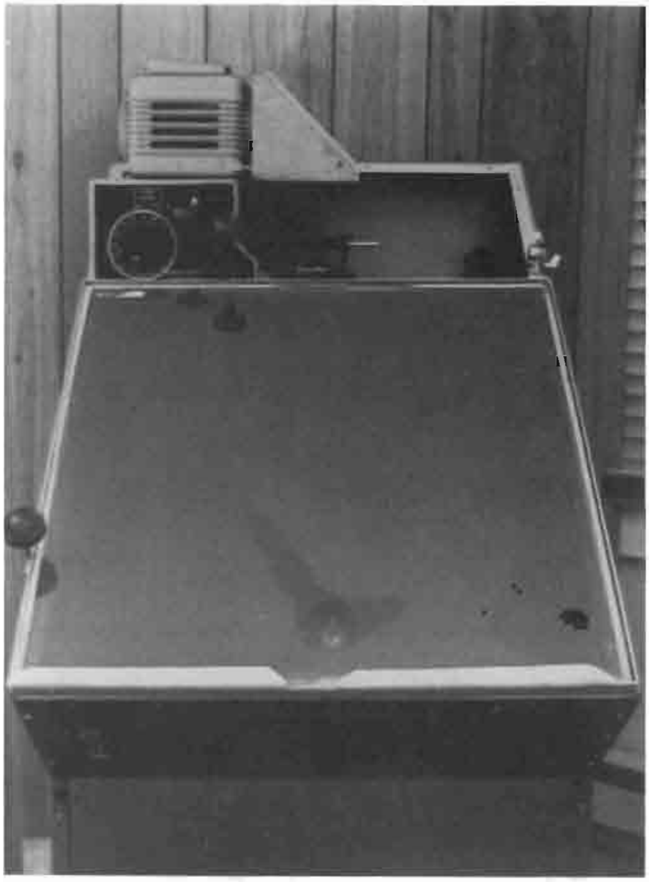


Figure 15
Older type of microprojector used for viewing scale impressions.

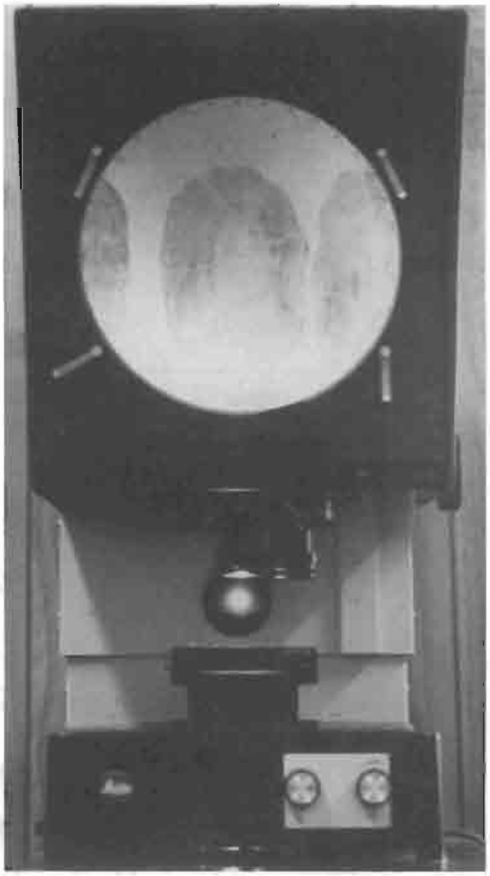


Figure 14
Microprojector (contour bench projector) used for viewing scale impressions.

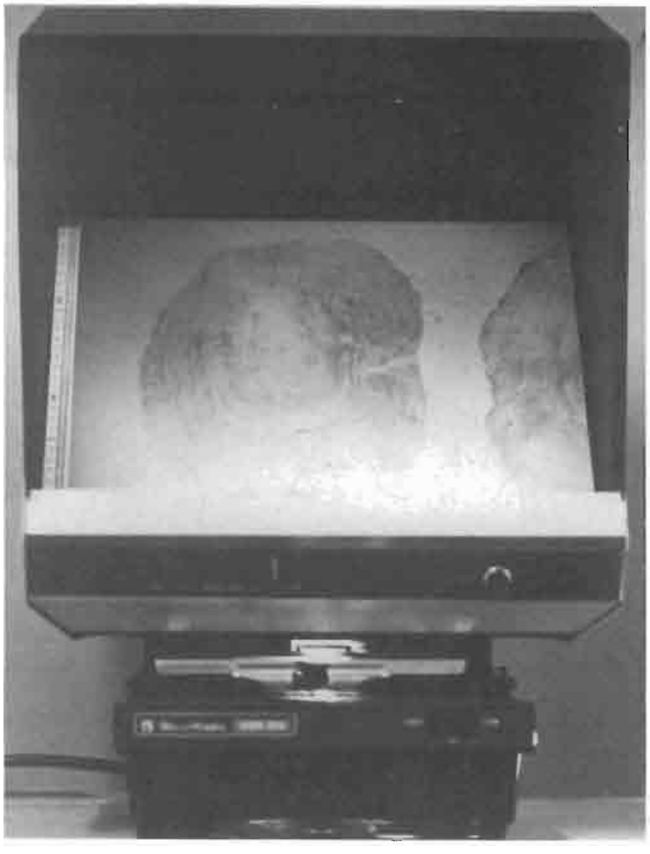


Figure 16
Microfiche reader used for viewing scale impressions.

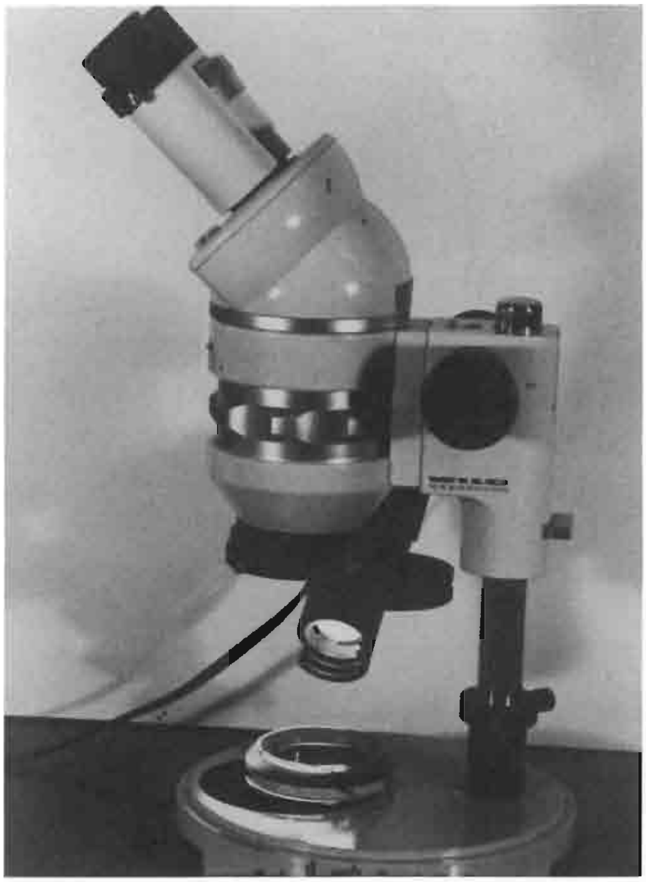


Figure 17
Binocular microscope used for viewing thin-sections.

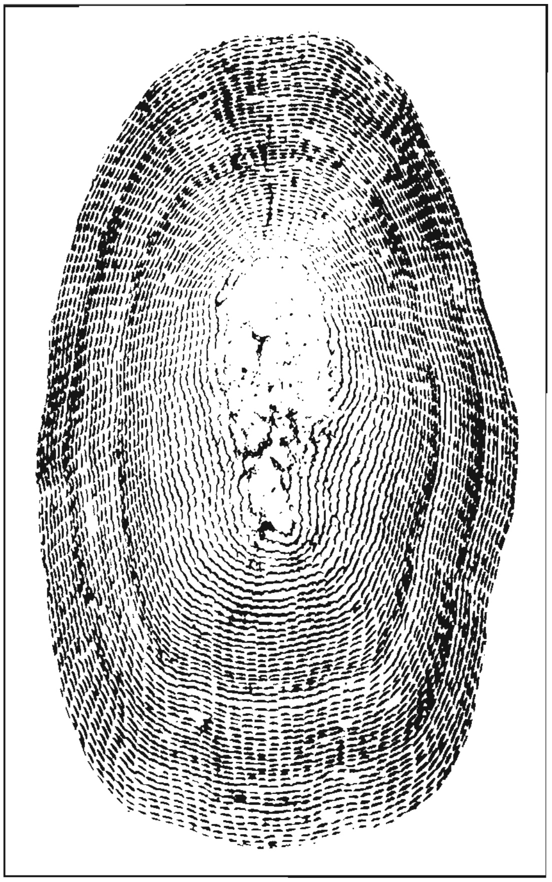


Figure 18
Regenerated scale from a haddock.

Figure 19
Crystallized Atlantic cod otolith broken in half at the nucleus.

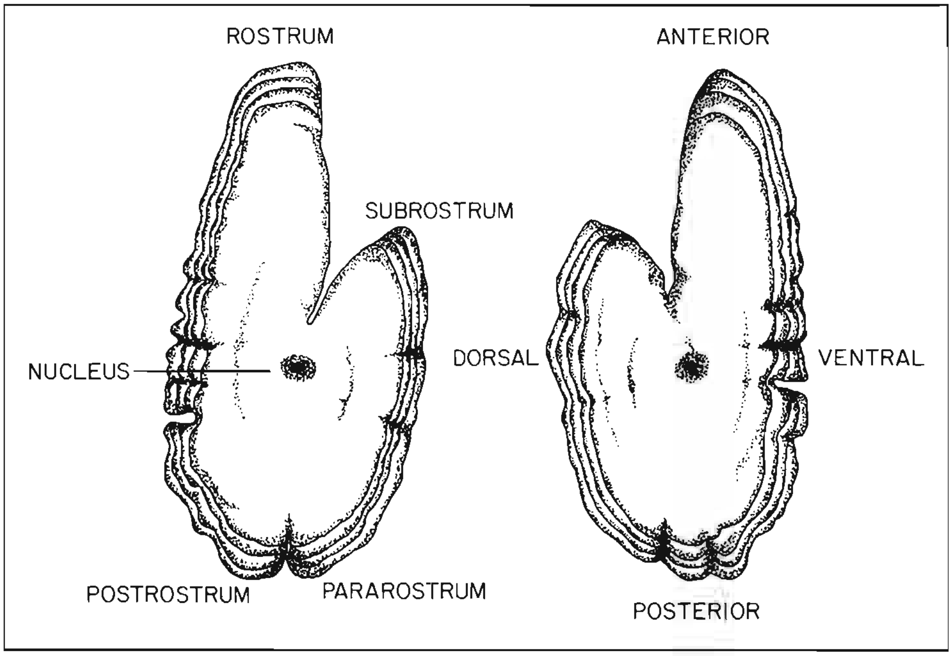


Figure 20
Sketch of a pair of otoliths from a pelagic species with descriptive terms. Distal side of the otoliths is shown.

4

Atlantic herring
Clupea harengus

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The Atlantic herring is a pelagic schooling clupeid ranging in the Northwest Atlantic from Greenland and Labrador south to Cape Hatteras (Bigelow and Schroeder 1953). In U.S. waters herring attain an overall maximum length and age of about 35 cm TL (14 inches) and 15 years, respectively (Anthony 1972). Male and female herring grow at about the same rate and become sexually mature by age 4 or 5.

Herring stock structure and migration patterns remain poorly understood (Anthony and Waring 1980, Graham et al. 1984, Sinclair and Iles 1985). Off the U.S. coast, large feeding and prespawning concentrations have been observed from May through August along the fringes of Georges Bank (prior to the late 70's) and off southwest Nova Scotia (Sinclair and Iles 1985). Spawning occurs between August and October from Nantucket Shoals and Jeffreys Ledge northward to coastal Maine (Anthony and Waring 1980, Graham et al. 1984, Sinclair and Iles 1985). In November, herring migrate to overwintering areas from the New York Bight to Cape Hatteras (Anthony and Waring 1980, Sinclair and Iles 1985).

Scales were originally used to determine the age of herring. During the 1960's, however, many researchers began using otoliths routinely due to problems with scale loss during handling (Hunt et al. 1973) and difficulty with interpretation of herring scales from waters south of Cape Breton (Huntsman 1919, Lea 1919, International Passamaquoddy Fisheries Board 1960). Otolith nucleii have been found to be quite useful in discriminating between races that spawn at different times during the year (spring vs. autumn spawners) (Einarsson 1951).

Although use of otoliths to age young herring from the Gulf of Maine was validated by Watson (1964), difficulties with the ageing of older fish persisted through the mid 1970's, with poor levels of age agreement between scales and otoliths (Messieh and Tibbo 1970), and between otolith age readings by different age readers (e.g., Parsons and Winters 1972). More recently, however, improvements in techniques, informal otolith exchanges, documentation of anomalous zones on otoliths (Dery and Chenoweth 1979) and indirect validation through the tracking of prominent year-classes through the fishery have resulted in substantial improvement in the accuracy and consistency of age interpretations.

Herring otoliths have been prepared for ageing in several ways. Otoliths have been stored in small vials prior to examination (Lissner 1925); or an adhesive, such as ethylene dichloride, has been used to bond each pair at the bottom of small depressions in black plastic trays (Watson 1965). Otoliths were then covered by distilled water or ethyl alcohol for examination. Another method, currently preferred by most age readers, is to embed the otoliths in a plastic resin after the method of Raitt (1961). At the Woods Hole Laboratory, herring otoliths are embedded in Permount resin (distal otolith surface up) on black molded plastic trays, and viewed under reflected light at a magnification of 15-20×. Since hyaline zones are not strongly developed on herring otoliths, the use of a plastic resin affords a much higher level of opaque/hyaline zone contrast than is possible by simple immersion in ethyl alcohol. It is especially useful in interpreting the outer zones on the otoliths close to the margin, which may be thin, split, or poorly defined. Difficulty with interpretation of the otolith margin was cited by Messieh and Tibbo (1970) as a major impediment to the use of otoliths for ageing Atlantic herring. At that time, most researchers used ethyl alcohol rather than plastic resin as a viewing medium (Hunt et al. 1973).

Herring otoliths from the Georges Bank and Gulf of Maine region exhibit a prominent hyaline core area immediately surrounding the nucleus, the outer edge of which is interpreted as the first annulus if hyaline zones are counted as annuli (Hunt et al. 1973) (Fig. 1). This zone is typically overgrown with calcium on the otoliths of older fish. Backcalculation results in an average fish size of 4-5 cm when this zone is completely formed (Dery, unpub. data). Bigelow and Schroeder (1953) and Boyar et al. (1973) found that herring in this size range occur in the Gulf of Maine during the spring months. Therefore, the hyaline core area is formed over a period of approximately 6 months, assuming that herring are spawned during the early autumn months. Few, if any, otoliths of herring from the Gulf of Maine or Georges Bank exhibit a tiny hyaline core area surrounded by opaque material that would indicate that they were spring-spawned fish. Scattergood (1952) and Bigelow and Schroeder (1953) reported no evidence of spring spawners in the Gulf of Maine area in the early 1950's; subsequent reports and observations tend to support the continuance of this pattern (Watson 1964, Anthony and Waring 1980, Kornfield et al. 1982).

The hyaline annulus on the otoliths of age-2 fish and older generally begins to form in October and is completed by the following March or April (Messieh 1974, Dery and Chenoweth 1979) (Fig. 2). However, opaque edge may persist on otoliths of age-5 fish or older in the late autumn months (Fig. 3) and may resume later in the spring relative to younger fish (Fig. 4). By convention, a birthdate of 1 January is used; the hyaline zone forming on the edge of the otolith is interpreted as an annulus whether or not it is complete. Time of annulus formation can vary in some years, however. The formation of opaque edge during winter has been characteristic of several year-classes (Dery and Chenoweth 1979). Checks can also cause confusion with edge interpretation. Therefore, correct evaluation of the type of edge formed on the otolith, and its meaning in terms of the growth of the fish, is an important aspect of herring age interpretation.

False annuli, formed during the first summer of the first full year of growth, are typical anomalies encountered on herring otoliths and may be especially characteristic of particular year-classes. Normally, false annuli are weak or incompletely formed hyaline zones, and because of their proximity to the nucleus, are not readily confused with the second annulus (Fig. 5). However, false annuli may occasionally appear as strong, continuous zones often associated with a wide summer growth increment (Fig. 1). These zones can be readily confused with annuli, and therefore the resulting age interpretation should be compared with that of fish of similar sizes.

The second annulus, which is sometimes a split, diffuse zone (Figs. 6 and 7) tends to be somewhat overgrown with calcium on otoliths of older fish, especially on the rostrum (Figs. 3 and 4). Subsequent to the second annulus, checks are normally formed during the second, third, and fourth summers corresponding to the prespawning period. These checks are especially prominent on the posterior and lateral part of the otoliths and are weak or not evident on the rostrum (Figs. 2, 4, and 8). Therefore, the rostrum is very useful in discriminating between checks and annuli.

The rostrum is also critical for an accurate identification of the fifth and subsequent annuli. With increasing age, herring otoliths accrete relatively little calcium on the posterior edge, although adequate deposition for annulus formation continues on the rostrum, which is the longest axis of the otolith (Fig. 9). Therefore, older herring otoliths may exhibit fewer annuli on the posterior edge relative to the rostrum; or annuli may be so closely spaced on this part of the otolith that it is difficult to distinguish them (Fig. 10). Occasionally, clustering of annuli on the posterior edge may result in underinterpretation of age relative to the rostrum (Fig. 11).

Annuli on the rostrum, however, are sometimes split into two (rarely more) hyaline zones (Fig. 12). Overinterpretation of age can be avoided by tracing each zone around the ventral edge of the rostrum. On this part of the otolith, the split zones reliably resolve into annuli. The second, third, and fourth annuli, however, must be carefully traced from the posterior edge of the otolith up to the rostrum, because the overgrowth of calcium on the rostrum may obscure these rings.

Growth patterns and/or morphology of herring otoliths do not appear to vary substantially in the Gulf of Maine area south to Cape Hatteras, except that adult herring otoliths from Georges Bank and south of Cape Cod exhibit somewhat more complex patterns. This greater complexity involves split, diffuse and somewhat less coherent annular zones and more prominent checks.

In summary, although current methodology has made the age determination of Atlantic herring relatively routine and reliable, misinterpretations can occur because of the formation of false annuli, checks, and split zones. Older herring otoliths should be interpreted with caution, using the rostrum and not the posterior edge of the otolith.

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Figure 1
Whole embedded otolith of a 22-cm age-3 Atlantic herring collected in April showing a prominent first annulus and false annulus.

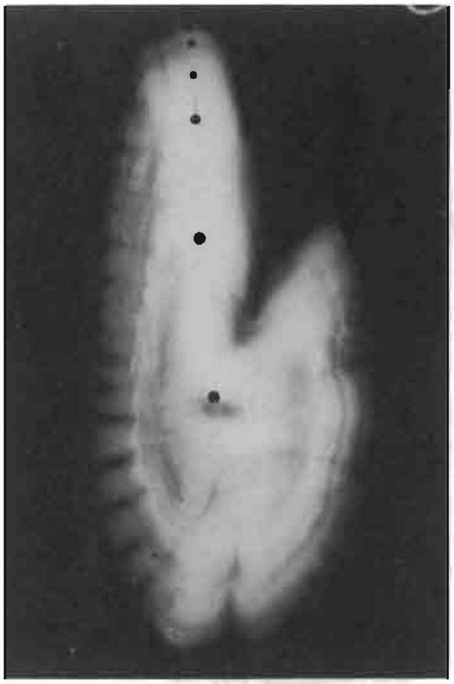

Figure 2
Whole embedded otolith of a 32-cm age-6 Atlantic herring collected in February showing hyaline edge. Second, third, and fourth annuli are weak on the rostrum; prominent checks on the postrostrum are difficult to distinguish from annuli.

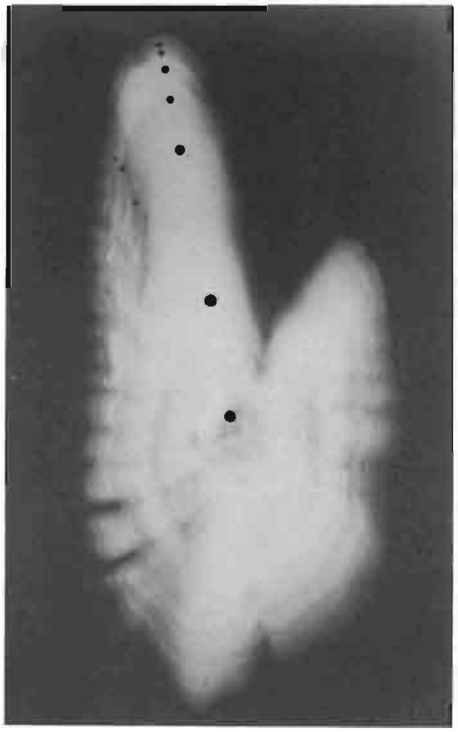

Figure 3
Whole embedded otolith of a 33-cm age 7+ Atlantic herring collected in October and showing hyaline edge. Annuli 2-5 are weak on the rostrum.

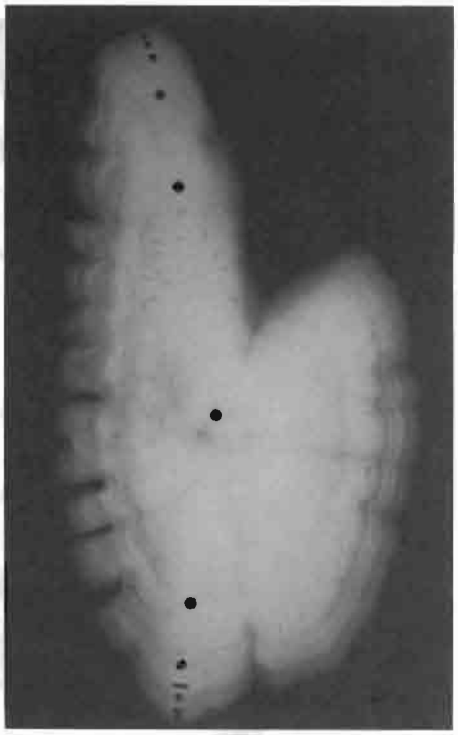

Figure 4
Whole embedded otolith of a 30-cm age-5 Atlantic herring collected in July and showing hyaline to narrow opaque edge. Strong checks are evident between the third and fourth, and fourth and fifth, annuli.

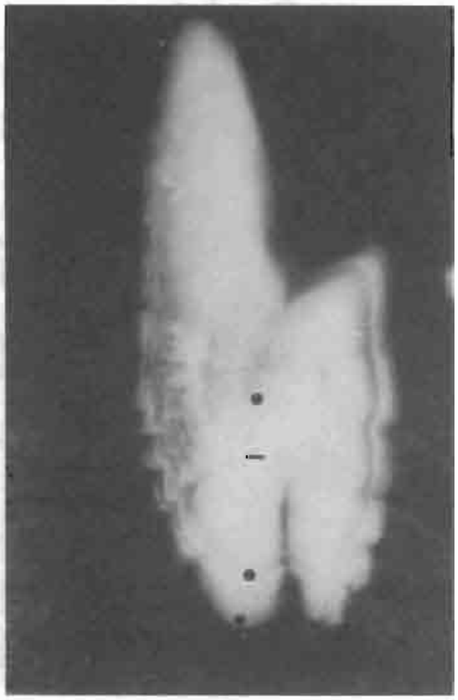

Figure 5
Whole embedded otolith of a 23-cm age-3 Atlantic herring collected in April showing a weak false annulus and large second annulus.

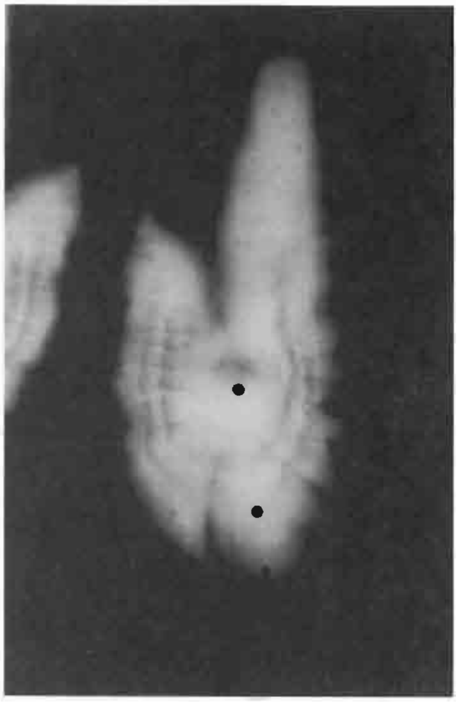

Figure 6
Whole embedded otolith of a 25-cm age-3 Atlantic herring collected in April showing a split second annulus.

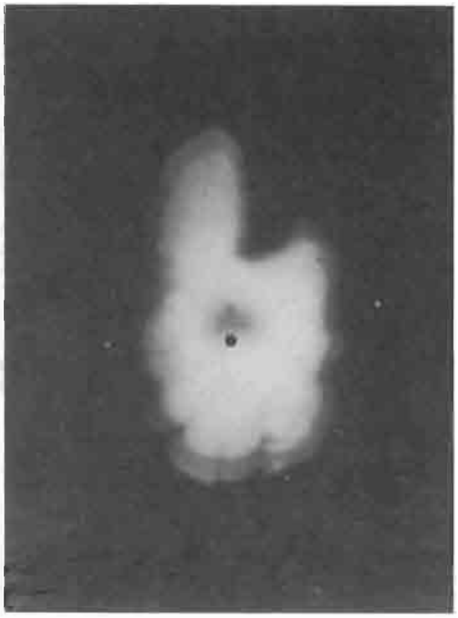

Figure 7
Whole embedded otolith of a 16-cm age 1+ Atlantic herring collected in September showing a split second annulus.

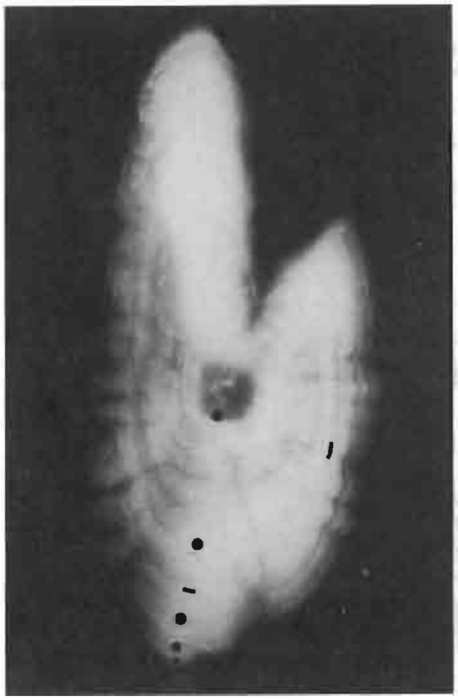

Figure 8
Whole embedded otolith of a 31-cm age 5+ Atlantic herring collected in October showing a weak check between the second and third annuli.

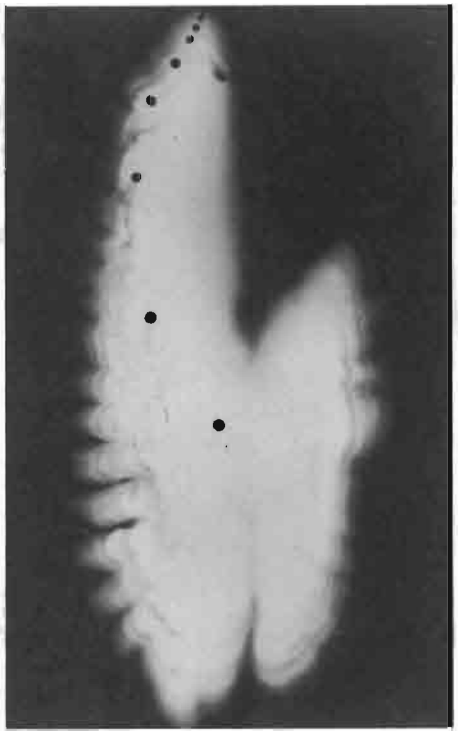

Figure 10
Whole embedded otolith of a 34-cm age-9 Atlantic herring collected in February. Weak second and third annuli on the rostrum; annuli compacted on the postrostrum.

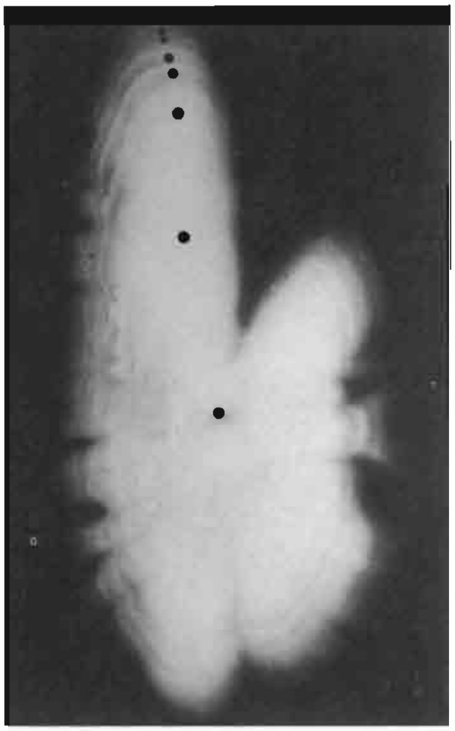

Figure 9
Whole embedded otolith of a 36-cm age-11 Atlantic herring collected in March showing clear annuli on the rostrum.

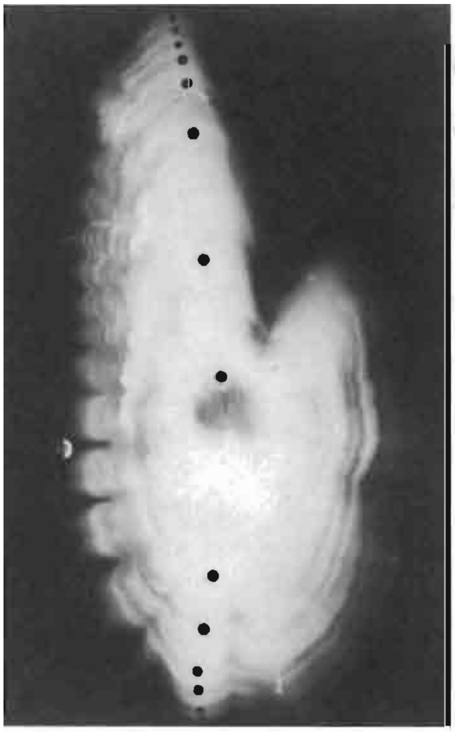

Figure 11
Whole embedded otolith of a 34-cm age-9 Atlantic herring collected in February showing nine annuli on the rostrum while only seven annuli are evident on the postrostrum.

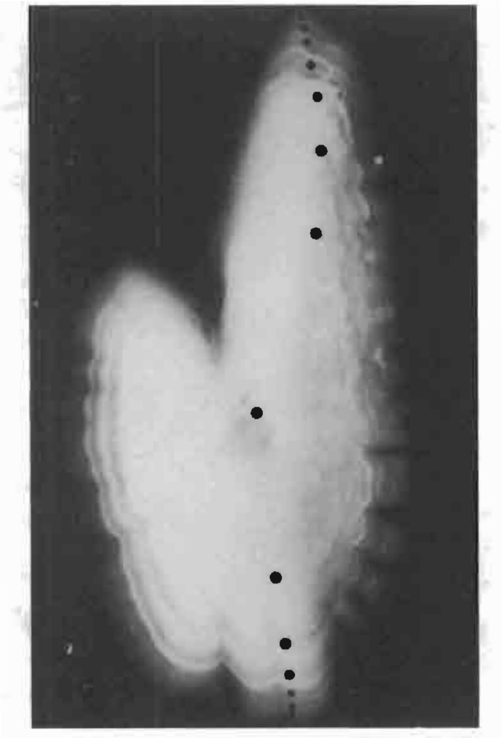

Figure 12
Whole embedded otolith of a 34-cm age-9 Atlantic herring collected in February showing split annuli on the rostrum and postrostrum.

5

Haddock
Melanogrammus aeglefinus

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The haddock is a demersal gadoid found on both sides of the North Atlantic; in the Northwest Atlantic, they occur from West Greenland to Cape Hatteras, and are most common in water temperatures of 2-10°C (36-50°F) and at depths of 45-135 m (Bigelow and Schroeder 1953). In U.S. waters, two stocks from the Gulf of Maine and on Georges Bank have been recognized (Clark et al. 1982). Although Georges Bank haddock are relatively sedentary, seasonal coastal migrations are known to occur in the western Gulf of Maine.

Growth rates for males and females are similar. Haddock become sexually mature at age 2 or 3, with individual females producing up to 3 million eggs. Spawning occurs from January through June, with peak activity in late March and April. A maximum age of 18 years has been documented for Georges Bank, although fish greater than 9 years of age are uncommon. Haddock attain lengths of 75-80 cm (30-32 inches) and weights to 5 kg (11 pounds).

Scales have been used for age determinations for haddock taken off North America since the early 1900's. Kohler and Clark (1958) compared age determinations based on scales and otoliths and reported no significant differences up to about 7 years; but thereafter, scale readings were consistently lower than otolith readings. Jensen and Wise (1962) subsequently validated the use of scales for haddock, particularly during their first 5 years of life. Scales are currently used at the Woods Hole Laboratory, because they are easier to work with, but age determinations can be readily made from both scales and otoliths over the range of ages normally present.

Scales are removed from the lateral line region anterior to the caudal peduncle for dry storage. About 5 or 6 scales from each fish are impressed on a laminated plastic slide by a roller press and viewed on a microprojector using transmitted light at a magnification of about 40×. Regenerated scales are discarded.

The scales are cycloid, oval or elliptical in shape, with no radii or transverse grooves (Figs. 1 and 2). The outer surface is sculptured with concentric rings of circuli, comprised of individual platelets, but the inner surface is smooth. The focus is generally anterior to the center of the scale, and growth zones are most clearly defined on the posterior portion of the scale. Generally, a pie-shaped sector from the longest portion of the scale, starting at the focus and extending to 15° on either side of the center of the posterior edge, is the area preferred for age determination. The spacing of circuli and shape of the platelets indicate periods of rapid and slow growth. Rapid summer-type growth is characterized by circuli which are spaced relatively far apart and are composed of platelets with curved edges. Slow winter-type growth is characterized by closely spaced circuli which are composed of platelets with straight edges (Fig. 3).

The annulus is defined as a zone of close winter circuli marking the end of a year of growth, i.e., the winter growth zone. Jensen and Wise (1962) report the following characteristics: 1) the annulus is concentric with the margin of the scale; 2) it can be traced, by careful scrutiny if necessary, entirely around the scale; 3) it is clearly separated from other such zones and does not ordinarily meet them at any point; and 4) if present, it is on all the normal scales of an individual.

By convention, a 1 January birthdate is used; therefore, a winter growth zone forming on the edge of the scale is designated as an annulus on 1 January, even though the zone is not complete.

Summer-type edge generally forms during May-July (Fig. 4), with winter-type growth predominating during August-April (Figs. 5, 6, 7, 8). Older fish begin forming summer-type edge growth later than younger fish and start winter-type growth earlier.

A major pattern feature in the differentiation of an annulus is the number of circuli per unit area. The number increases during slow winter growth and decreases during rapid summer growth. Measurement of the distance between circuli shows relatively wide interspaces during rapid summer growth, and as the interspace decreases, compaction of the circuli forms the broad, dark-appearing rings that represent the period of slow winter growth (Fig. 5). The end of an annulus (i.e., the last true winter circulus in the winter growth zone) is generally followed by a rapid transition from narrow to widening interspaces, signifying the start of the next period of rapid summer growth. This usually occurs in the spring of the year, but can vary in different geographical areas. Number of circuli per unit area and circuli interspaces are useful in determining the first few annuli, but after the fifth or sixth annulus, there is a gradual reduction in the number of the circuli formed during a year's growth. This diminishes the usefulness of these two methods.

Changes in the shape of the individual platelets forming the circuli are also useful in recognizing an annulus, especially after the fifth or sixth year. The outline of platelets formed during summer growth are frequently curved or crescent-shaped, while winter platelets tend to be straight. Scanning electron microscopy (SEM) reveals this to be caused by greater protrusion of the summer platelets from the basal plate. They also have a rounded upper edge, while winter platelets have a relatively straight upper edge (Fig. 3).

Checks may be distinguished from annuli by relative width, location, and platelet shape (true winter platelets may not be formed) (Figs. 2, 4, 6, 9). Although checks generally begin abruptly, annuli usually have a transition zone showing a relative decreasing of the interspace between circuli before the true winter zone is reached (Figs. 4 and 6). Absence of a rapid transition to summer growth after the check may also help to distinguish it from an annulus (Fig. 9). Checks may also be distinguished by following them around towards the sides of the scale to determine if they merge with an annulus to form one zone. Checks may be stronger on some scales and weaker, or even absent, on others, while annuli are present on all scales from a particular fish. This is one reason why several scales are examined from each fish in order to verify the assigned age. It is sometimes necessary to make two or three impressions, of 5 or 6 scales each, before a clear "composite" picture of the fish's growth can be determined.

Spacing of the annuli relative to each other and to the focus of the scale may be used to differentiate between annuli and checks. For example, if two winter growth zones are found relatively close together on a scale from a younger fish, but all the other winter zones on the scale are relatively far apart, then one of the two close zones will probably be a check and not an annulus. This type of annulus construction (i.e., two close zones) is generally described as a split annulus due to difficulty in differentiating between the check and the annulus (Figs. 2, 4, 7, 8). On scales from older fish, annuli formed after the fifth or sixth year are expected to be fairly close together (Figs. 5, 9, 10).

Characteristic patterns based on geographic origin are useful for identifying annuli. Fish from Georges Bank often have a characteristic check, called a false annulus, before the first annulus. This may be distinguished from a true annulus by the number of circuli contained between the focus of the scale and the end of the false annulus (generally 10-16 circuli) (Figs. 2, 4, 8). These fish also grow more rapidly during their first and second years than do fish from other areas, so that larger first and second annuli are expected on these scales compared with scales from Gulf of Maine or Browns Bank fish. Georges Bank haddock scales also show more distinctly formed annuli than do fish from the Gulf of Maine, which often exhibit annuli lacking winter platelets with straight edges (Figs. 1 and 2). This presents some difficulty in ageing Gulf of Maine fish as often only circuli spacing and relative annuli spacing can be used as age determination criteria. Moving back from the microprojector screen for a better perspective of the overall pattern of growth often permits an easier age interpretation.

Rapid first, second, and third-year growth followed by gradually slower growth in later years by Georges Bank haddock is in contrast to Browns Bank fish (Schuck and Arnold 1951, Wise 1957). They exhibit comparatively slow growth during the first three years, followed by more rapid growth in the fourth, fifth and sixth years, and then a gradual slow down in growth in subsequent years (Figs. 10 and 11).

Recognition of checks caused by damage or injury to haddock is a problem in ageing fish from all areas. In these cases, the scale is physically shifted in the scale pocket, resulting in subsequent circuli that are not quite in line with previous circuli, "lost" circuli, and irregular spaces (Fig. 7). Circuli in the damaged area may disappear when an attempt is made to trace them around the scale. These marks on the scale correspond to the area of regeneration after the scale was lost. The effect is similar to that known as "cutting over," caused by erosion of the scale edge, which is commonly seen with flounder and certain other species' scales.

The relative location of annuli is the most reliable general criterion for discriminating between checks and annuli for haddock scales. An initial procedure that is useful during scale examinations is to mentally superimpose a regular growth pattern, based on prior knowledge of typical patterns for the geographic origin of the fish. Any zone not fitting the pattern is closely scrutinized to determine if it is a check.

On occasion, particular year-classes may exhibit peculiar growth characteristics which assist in determining age. The 1960 year-class on Georges Bank exhibited very regular growth patterns with no checks (Fig. 5). Other year-classes may form a certain split annulus, or a strong check between two particular annuli, or perhaps two close annuli. Such characteristic growth patterns may be very useful in assigning the most probable age, particularly for difficult specimens.

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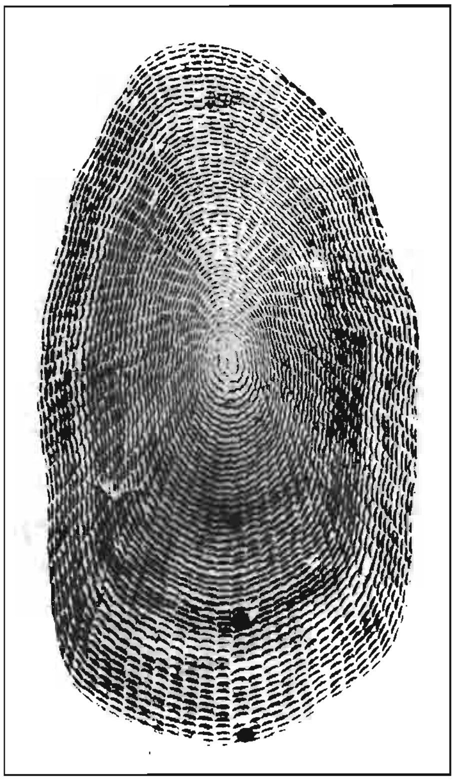

Figure 1
Scale impression of a 41-cm age-3 haddock collected in January from the Gulf of Maine showing the poorly differentiated annuli typical for this area.

Figure 2
Scale impression of a 47-cm age-3 haddock collected in January from Georges Bank showing a checky first year (with a false annulus) and a split second annulus.

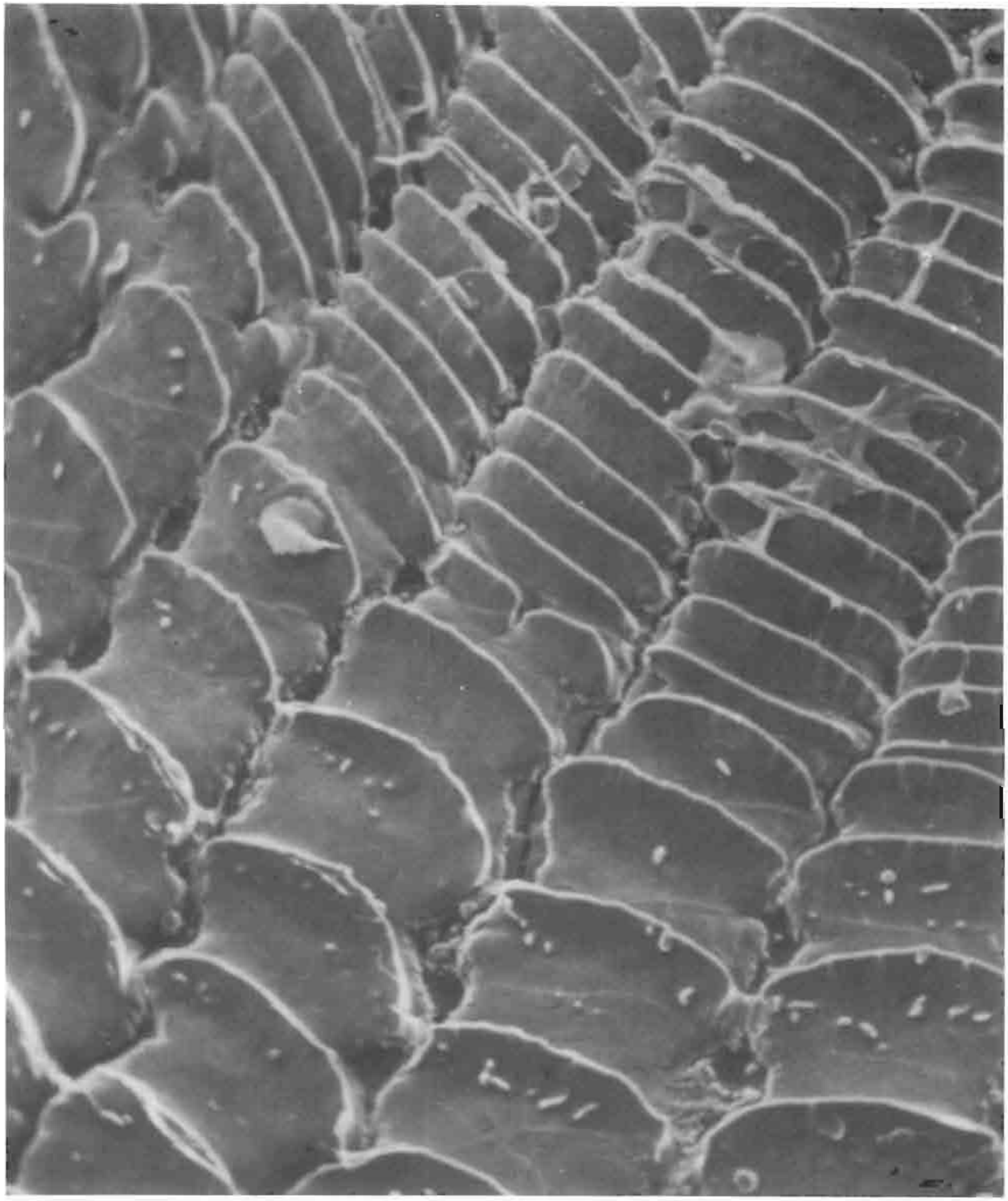

Figure 3
SEM photograph of an actual haddock scale showing summer platelets with curved edges and winter platelets with straight edges.

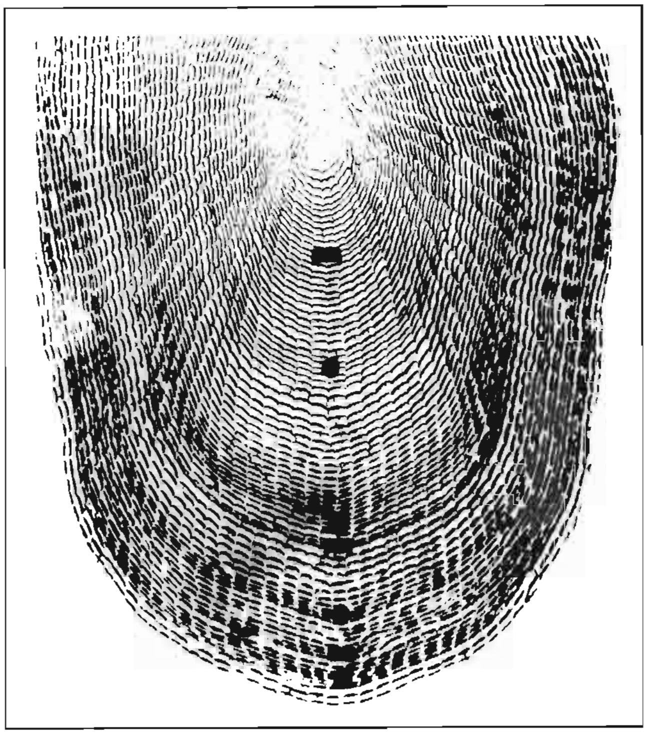

Figure 4
Scale impression of a 52-cm age-4 haddock collected in June from Georges Bank showing a false annulus, a split second annulus, and a strong check before the fourth annulus, with summer edge forming.

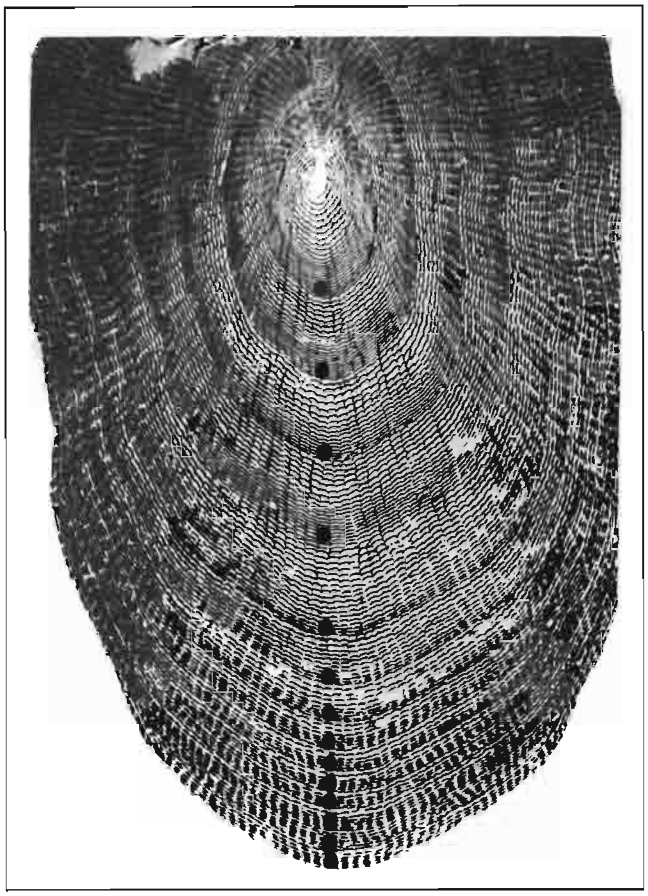

Figure 5
Scale impression of a 74-cm age-14 haddock collected in April from Georges Bank showing a "textbook" pattern of extremely regular annuli formed by alternating zones of rapid summer and slow winter growth.

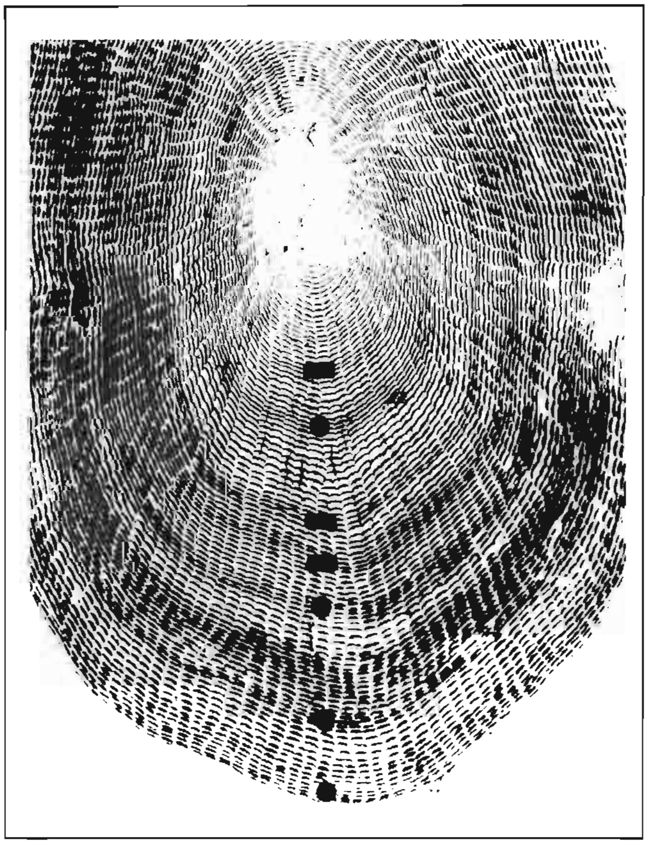

Figure 6
Scale impression of a 64-cm age-4 haddock collected in January from Georges Bank showing very checky first and second years, with winter edge forming.

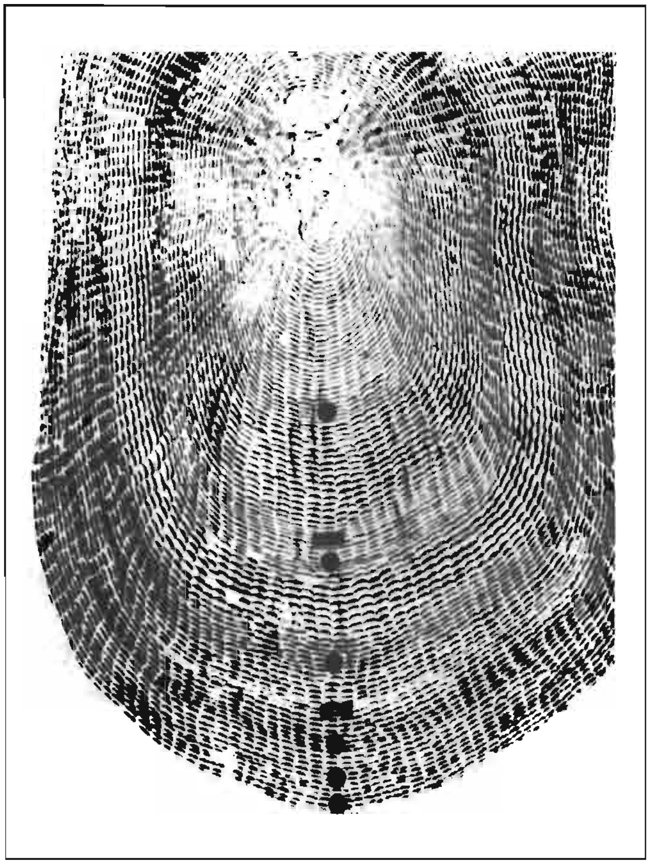

Figure 7

Scale impression of a 60-cm age-6(5) haddock collected in February from Georges Bank showing a split second annulus, a check (caused by damage or injury to the fish) between the third and fourth annuli, and a weak fifth annulus, with the sixth annulus forming on the edge.

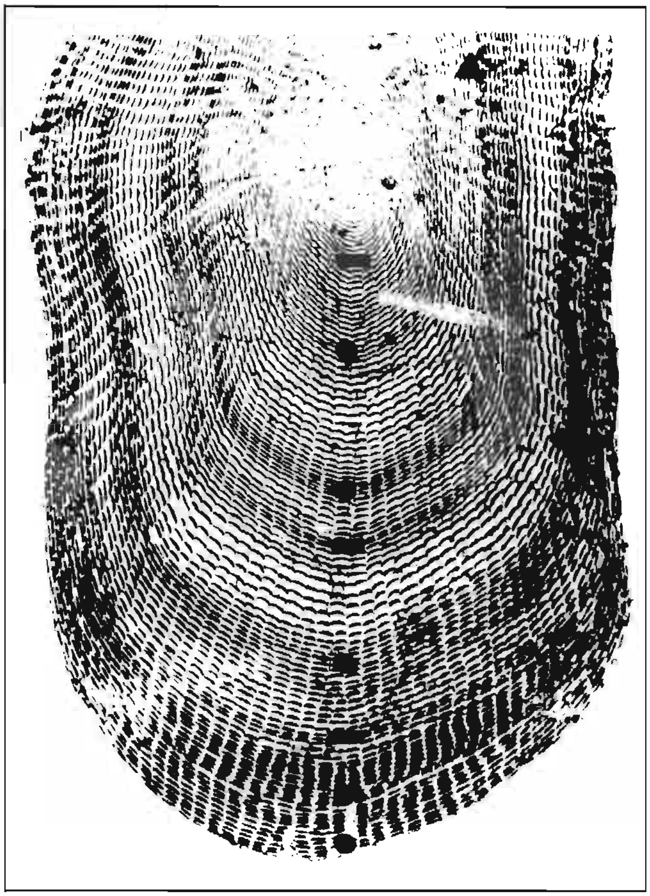

Figure 8

Scale impression of a 64-cm age-5 haddock collected in February from Georges Bank showing a false annulus, split second annulus, weak third annulus, split fourth annulus, with the fifth annulus forming on the edge.

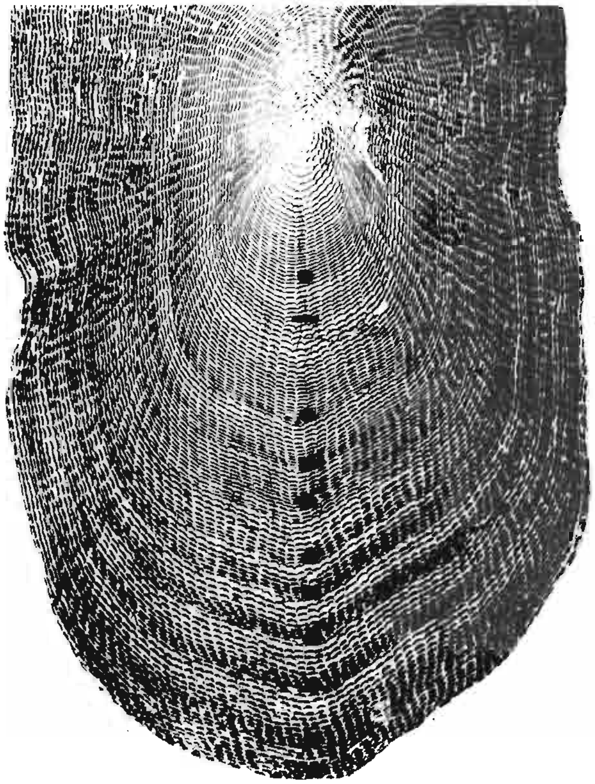

Figure 9
Scale impression of a 68-cm age-10 haddock collected in February from Georges Bank showing a check after the first annulus and close third and fourth annuli.

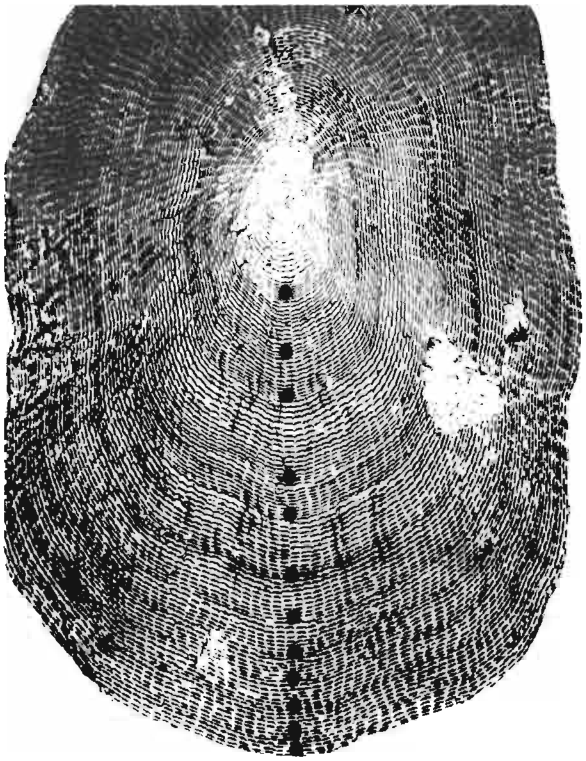

Figure 10
Scale impression of a 66-cm age-12 haddock collected in January from Browns Bank showing the close first, second, and third annuli typical for this area.

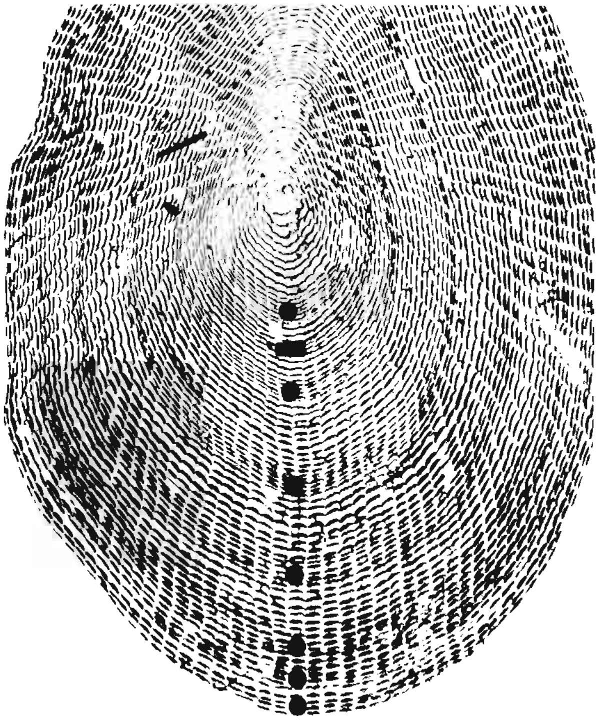

Figure 11
Scale impression of a 54-cm age-7 haddock collected in February from Browns Bank showing the close first, second, and third annuli typical for this area.

6

Atlantic cod
Gadus morhua

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The Atlantic cod is a cold-temperate, demersal gadoid distributed in the Northwest Atlantic from Greenland to North Carolina; commercially important concentrations occur southward from Greenland to Cape Cod. Wise (1963) proposed the existence of four separate groups of cod in the New England area: Gulf of Maine, Georges Bank, Southern New England, and Mid-Atlantic coastal cod. The latter group spends the summer in Southern New England. Age data from the Southern New England and Mid-Atlantic groups are usually combined because of the difficulties experienced in separating cod from the two groups collected during the autumn (Penttila and Gifford 1976). They migrate towards the south and west (shoaler waters) in winter and early spring and towards the north and east (deeper waters) in late spring and summer (Schroeder 1930, Wise 1963). Growth rates for males and females are similar and sexual maturity is typically attained at age 2 or 3. Spawning occurs from December through April. Maximum age is in excess of 20 years, and a record size of a 96 kg (211 pounds) fish >180 cm in length is reported (Bigelow and Schroeder 1953). Cod commonly attain lengths of 120-130 cm, and 12-15 year old fish are often landed.

Early studies depended on scales for age determination, but Schroeder (1930) concluded that they were not reliable for individuals older than 6 or 7 years. Studies at the Woods Hole Laboratory during the early 1970's confirmed that scales are not generally reliable even for younger specimens, and, accordingly, age determinations for Atlantic cod are based on otoliths. An improved method of otolith preparation involves baking to produce brown hyaline zones in contrast to white opaque zones. This enhances visibility of the annuli during examination with reflected light. Otolith thin-sections may also be prepared, but require much more time than baking and do not appreciably improve the distinction of annuli. Ageing criteria and techniques based on baking have been informally validated by comparing cod length frequencies from 1969-74 research vessel surveys with corresponding age-length frequencies. Jensen (1970) validated the use of otoliths for Gulf of Maine cod, and Kohler (1964) validated otoliths for western Gulf of St. Lawrence cod.

Otoliths are stored dry, and one otolith from each fish is baked at 275°C (525°F) for 3-6 minutes, or until it turns a caramel brown color. Smaller otoliths require a longer baking time than do larger otoliths, perhaps because their hyaline zones are not so well defined. The otolith is then broken in half at the nucleus and examined under a binocular microscope at a magnification of about 15× using reflected light. Wetting the broken surface of the otolith with undiluted Kodak Photo-Flo 200 solution enhances visibility of the rings. Rings formed during periods of slow winter-type growth appear as brown, hyaline zones, while growth increments formed during periods of rapid summer-type growth appear as white, opaque zones. Shifted or crystallized otoliths should not be aged (Fig. 1).

The annulus is defined as a hyaline zone marking the end of a year of growth, i.e., the winter growth zone. It has the following characteristics (following Jensen and Wise 1962): 1) it is concentric with the margin of the otolith; 2) it can be traced entirely around the otolith; and 3) it is separated from other such zones and does not ordinarily meet them at any point. Age determinations for cod are usually made by counting from the nucleus out to the distal edge of the otolith. On older specimens, the dorsal end of the otolith may be best for distinguishing annuli close to the edge. Also, the proximal edge on either side of the sulcus groove often displays clear annuli.

By convention, the birthday of all fish in the northern hemisphere is 1 January, therefore, a winter hyaline zone forming on the edge of the otolith is counted as an annulus on 1 January, even though the zone is not complete. Opaque edge material generally forms on the otolith during the late spring, summer, and early fall (Fig. 2); hyaline edge forms during the late fall, winter, and early spring (Fig. 3). Older fish begin depositing opaque edge material later than younger fish, and the hyaline edge also tends to form earlier.

A check may be distinguished by its width and location relative to true annuli. If two hyaline zones are found relatively close together, but all the other hyaline winter zones on the otolith are relatively far apart, one of these zones is considered a check, or the annulus is termed split (Fig. 4). For a split annulus, it is often difficult to determine which zone is the check. Checks and splits may also be distinguished by following them around the otolith to the proximal side near the nucleus to determine if they merge to form one annular zone. On otoliths from older fish, annuli formed after the fifth or sixth year are often split and these annuli are expected to be fairly close together (Figs. 2, 3, 5, 6, 7, 8).

General patterns based on the geographic origin of the fish may also be used as an aid in identifying annuli. Fish from Georges Bank grow more rapidly than do fish from other areas and tend to have more checks on their otoliths. They generally have a large, checky first year with the first annulus situated a good distance out from the nucleus, and may often show a strong check before the second annulus (Figs. 3 and 5). The second annulus is generally the strongest (i.e., the darkest or widest hyaline zone) on the otolith (Fig. 9). Annual growth increments are usually largest during the first and second years but good growth still occurs through the third year, followed by a gradual decrease in growth thereafter (Figs. 6 and 7).

Cod from Southern New England show a small first-year growth increment and rapid growth in the second year, followed by a gradual decrease in growth in subsequent years (Fig. 10). Their growth rate is slower than that of Georges Bank fish for the first two years, but after the third year, growth is comparable (Penttila and Gifford 1976).

Fish from the Gulf of Maine have a slower growth rate and a fairly small first-year annulus. Comparatively good growth in the second year is followed by a decrease thereafter (Figs. 2, 8, 11).

Browns Bank and Scotian Shelf cod exhibit the slowest growth of any of the areas. Browns Bank cod otoliths often exhibit a growth pattern characteristic of the area with comparatively slow growth during the first three years, followed by better growth in the fourth through sixth years, and then a gradual decrease in subsequent years (Fig. 12).

A major problem in age determinations of cod is distinguishing between the settling check and the first annulus. The settling check is usually a thin, single, or split hyaline zone immediately surrounding an amorphous, translucent nucleus. This check tends to be more nearly circular in shape than annuli, and a definite opaque summer zone found inside the first hyaline annulus is not apparent inside a settling check (Figs. 5, 7, 8).

Difficulties may arise in finding the first annulus on otoliths that have not been broken precisely at the collum of the sulcus. Inability to find the settling check or the appearance of an abnormally small or irregularly shaped first annulus usually indicates that the otolith was not properly broken at the nucleus and that the remaining stored otolith for the fish should be processed. Figure 13 shows serial sections cut from a single cod otolith, from the anterior to the posterior area of the first annulus. This figure illustrates the change in appearance and shape of the settling check and first annulus according to how far off the collum the break or cut is made.

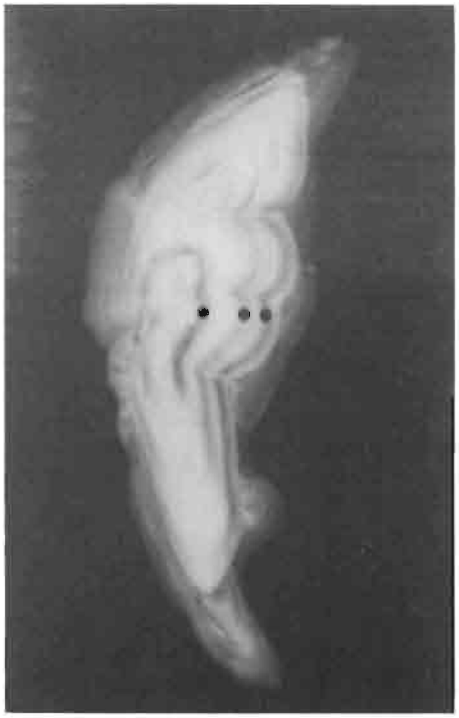

Figure 1
Otolith section cut from a typical shifted otolith of an 88-cm, age ?? cod collected in July from Georges Bank.

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Figure 2
Baked otolith half from a 79-cm age-4 cod collected in September from the Gulf of Maine showing split second and fourth annuli, with opaque edge still forming.

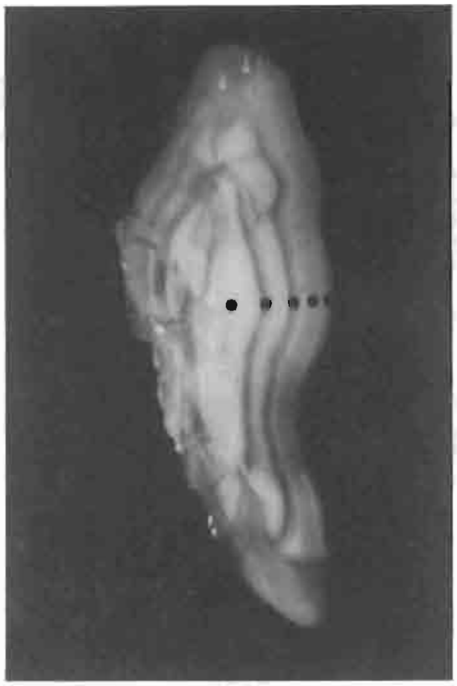

Figure 4
Baked otolith half from an 87-cm age-5 cod collected in January from Georges Bank showing a split fourth annulus.

Figure 3
Otolith section from a 59-cm age-4 cod collected in the spring from Georges Bank showing a checky first annulus with hyaline edge still forming.

Figure 5
Baked otolith half from a 70-cm age-5 cod collected in March from Georges Bank showing a strong settling check, large and checky first annulus, and a strong check before the second annulus.

Figure 6
Baked otolith half from a 122-cm age-7 cod collected in July from Georges Bank showing a split third annulus.

Figure 8
Baked otolith half from a 90-cm age-7 cod collected in September from the Gulf of Maine showing a strong settling check and a split third annulus.

Figure 7
Baked otolith half from a 107-cm age-9 cod collected in August from Georges Bank showing a clear settling check.

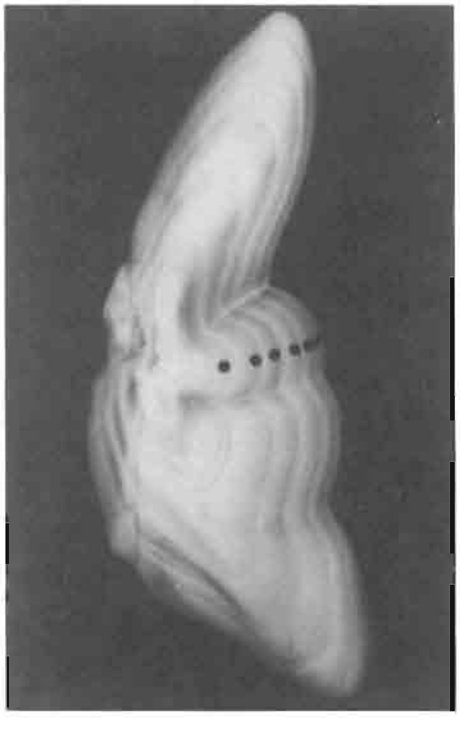

Figure 9
Otolith section from an 88-cm age-6(5) cod collected in the spring from Georges Bank showing a strong second annulus and a possible weak fifth annulus (?) close to the sixth annulus forming on the edge.

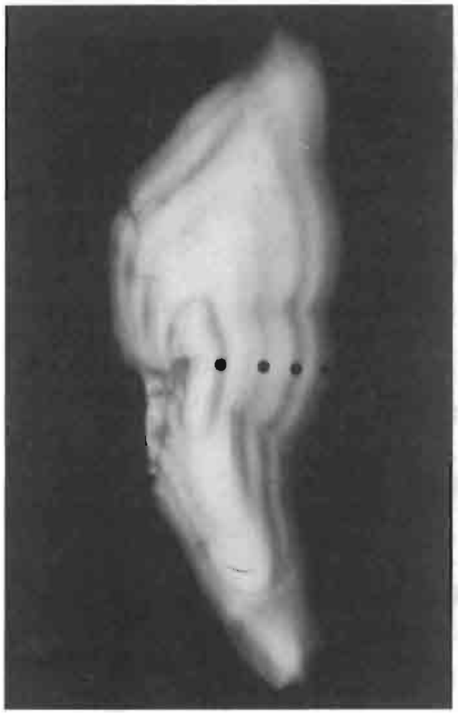

Figure 10

Otolith section from a 69-cm age-4 cod collected in the spring showing the small first annulus typical of Southern New England cod.

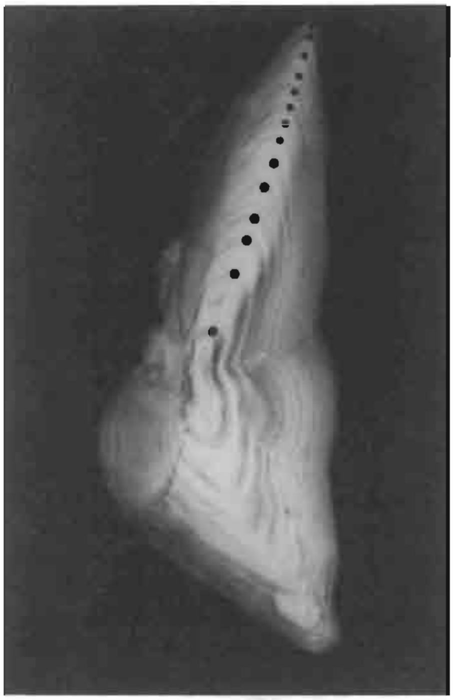

Figure 11

Otolith section from a 130-cm age-14? cod collected in the spring from the Gulf of Maine showing the small first annulus typical for this area.

Figure 12

Otolith section from a 97-cm age-7 cod collected in the spring from Browns Bank showing the close first, second, and third annuli typical of this area.

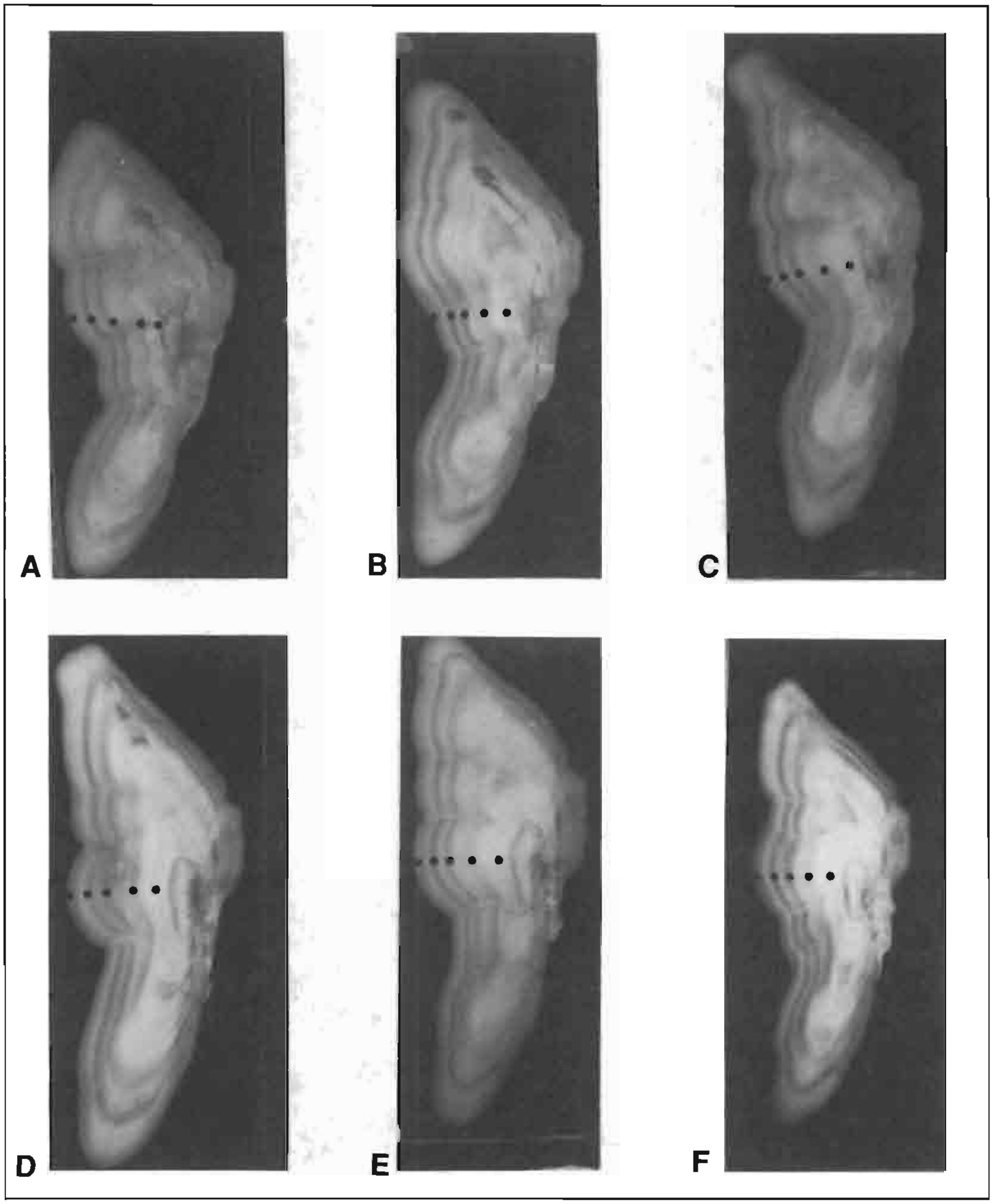

Figure 13

Series of sections cut from a single cod otolith (84-cm age-5 cod collected in the spring from Southern New England), from the anterior (13A) to the posterior (13F) of the first annulus. Section cut at the center of the nucleus is 13C.

7

Pollock
Pollachius virens

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Pollock is an amphiboreal gadoid found on both sides of the North Atlantic. In the western North Atlantic it ranges from Labrador to Cape Hatteras, but is most abundant on the southwestern Scotian Shelf and in the Gulf of Maine. Pollock are highly migratory and extensive movements have been documented (Steele 1963). Stock structure has not been elucidated in the Northwest Atlantic.

Pollock spawn in winter; most individuals attain sexual maturity in 3 or 4 years, although some pollock may not mature before age 6 or so. Pollock are comparatively long-lived, attaining a maximum age of 23 years. They may reach lengths of 110 cm (43 inches) and weights of 16 kg (35 pounds). No significant differences have been observed in growth rates between sexes (Bigelow and Schroeder 1953, Hoberman and Jensen 1962, Steele 1963, Clark et al. 1978).

Scales and otoliths have been used for age determinations of pollock, but otoliths are preferred for older individuals because of the difficulty in distinguishing outer annuli on scales. Otoliths are stored dry.

Otolith processing involves making transverse thin-sections of from 0.17 mm to 0.28 mm in thickness cut exactly at the nucleus along the dorsoventral axis. For large otoliths (taken from individuals greater than 75 cm in length), it is preferable to produce a 0.17-mm thick section. Section thickness from otoliths of smaller fish is not as crucial.

Age determinations are made by placing the section on a square of black paper and applying Kodak Photo-Flo 200 solution to the surface, allowing for some spillage onto the paper. It is then viewed under a binocular microscope at 20 to 30× magnification using reflected light.

Annual zones on a pollock section are composed of a white opaque zone representing fast summer growth and a dark hyaline zone representing slow winter growth. The annulus is defined as the hyaline zone marking the end of a year of growth, i.e., the winter growth zone. These zones are easily distinguished in fish up to 6-8 years of age. Deposition of hyaline material begins as early as October in some fish and is clearly evident in almost all specimens collected by December and January (Fig. 1). Opaque material may be deposited as early as December, but is typically found May-September (Steele 1963) (Fig. 2).

By convention, a 1 January birthdate is used; therefore, a hyaline zone forming on the edge of the otolith is counted as an annulus on 1 January, even though the zone is not complete.

Age determinations are usually made by counting hyaline rings from the center to the edge. The first annulus is rarely located close to the nucleus. Spacing between the opaque and hyaline rings is important in locating a settling check. This settling check consists of a series of light hyaline lines more widely spaced than the hyaline lines comprising a true first annulus. The overall shape of the settling check is more elliptical and has fewer, if any, undulations than the true first annulus. The settling check in some otoliths encircles a dark area, but for others, it is barely visible. An opaque space usually marks its location (Figs. 2 and 3).

Age determinations may be made on several areas of the otolith. The proximal side near the nucleus is optimal for an initial reading and the side closer to the dorsal end sometimes is easier to read because the annuli tend to be more spread out in this region. Rings are also well defined on the dorsal end, but an age reader should carefully follow the rings around to the distal and proximal sides to insure that no splitting has occurred. For older fish, the dorsal end can be very helpful, particularly when searching for the most recently formed annulus. Older specimens taken in summer (July-

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September) can be particularly difficult, because for most fish little opaque summer growth is visible. The dorsal end generally magnifies the size of the annuli, a condition that clarifies differentiation. Another consideration when viewing the edge is the angle or bevel of the otolith surface. Occasionally, the section may have been cut through an area of the otolith which had a particularly rough surface. The result can be an optical illusion of a hyaline edge when in reality the age reader is looking at an opaque edge. It appears hyaline because it is thinner at the bevelled plane (Fig. 4). Therefore, it is important to examine the side of the otolith section to be sure a bevelled edge is not present.

The second, third, and fourth annuli are usually broad and frequently paired or split. The fifth and sixth annuli may also be split. Split rings must be carefully followed around the otolith. Split hyaline rings commonly occur for one or two successive years and may continue for up to four years (Figs. 5 and 6).

Several criteria must be considered when evaluating split hyaline rings. Spacing is most important. Usually, the opaque increment between the split rings is narrower than the opaque increment between the outside split ring and the next hyaline ring towards the edge. This next annulus, if not split, is usually darker with more clearly defined boundaries than the split ring. If two closely spaced hyaline rings, which appear suspect, merge as the reader follows them around to the ventral/proximal side, and/or if they merge at the sulcus, they should be recognized as a split ring and should be counted as a single annulus (Fig. 7).

Annuli become thinner and more crowded towards the edge of the otolith and are occasionally difficult to read. In such cases, an age reader should count back toward the center to locate a strong hyaline ring. It may be followed down toward the dorsal end until an area is located that has more distinct hyaline rings. Repeated counts on each otolith are necessary, especially for older fish. Otoliths from specimens >12 years of age should be examined several times until a consistent determination is reached (Fig. 8).

Although pollock otoliths cause the age reader many problems because of settling checks and split annuli, there is a close correspondence between age and length, especially in small and medium-sized fish.

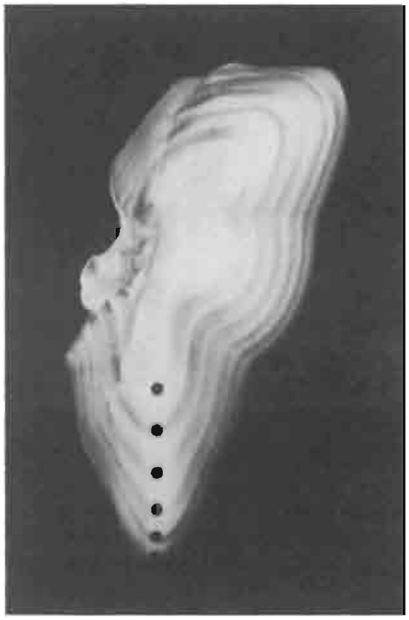

Figure 1
Otolith section from a 69-cm age-6 pollock collected in January showing clearly defined annuli with hyaline edge (sixth annulus) forming.

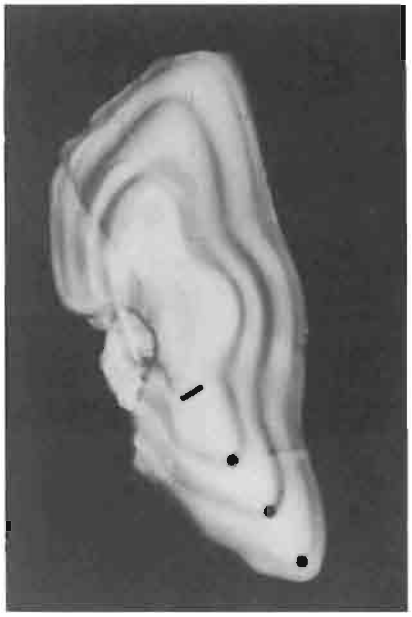

Figure 2
Otolith section from a 50-cm age-3 pollock collected in July with opaque edge forming. A fairly strong settling check is evident.

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Figure 3
Otolith section from a 59-cm age-5 pollock collected in March showing a strong settling check.

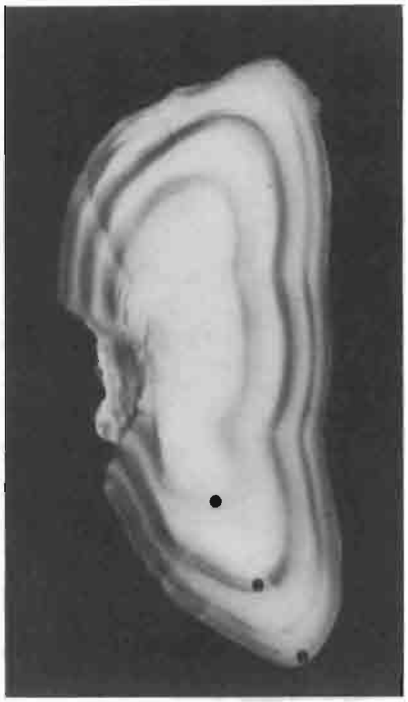

Figure 5
Otolith section from a 46-cm age-3 pollock collected in April showing a strongly split third annulus.

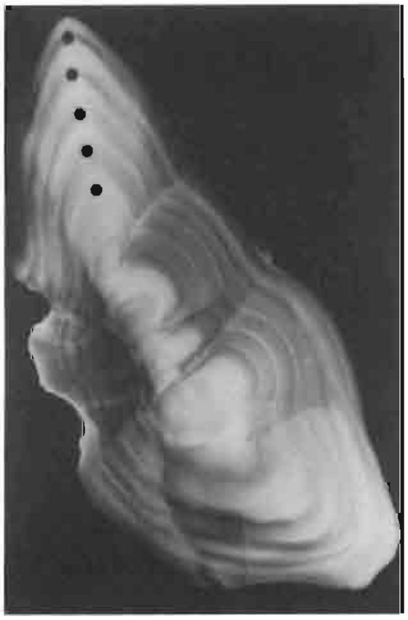

Figure 4
Otolith section from a 54-cm age-5? pollock collected in July with opaque edge forming. This otolith has not been sectioned exactly at the nucleus.

Figure 6
Otolith section from a 57-cm age-4 pollock collected in April showing a strongly split fourth annulus.

Figure 7
Otolith section from a 50-cm age-3? pollock collected in September showing close second and third annuli with hyaline edge forming.

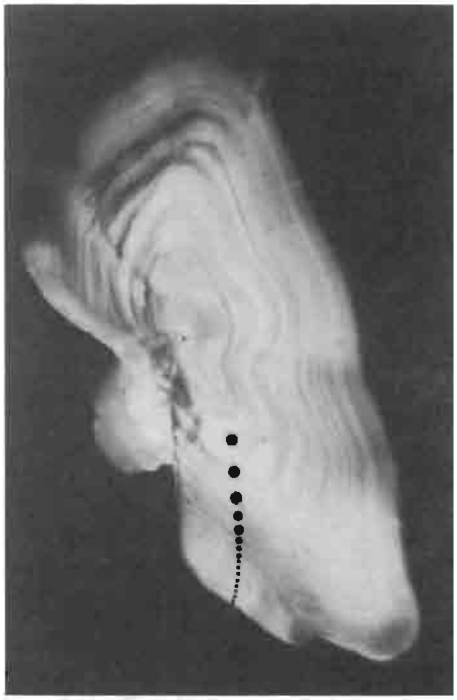

Figure 8
Otolith section from a 94-cm age-17? pollock collected in April. Clearest area on this otolith for ageing is on the dorsal/proximal side.

8

Silver hake
Merluccius bilinearis

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Silver hake is an important gadoid ranging from Newfoundland to South Carolina and is most abundant from Nova Scotia to New Jersey (Bigelow and Schroeder 1953). Silver hake are found over a wide range of depths, from shallow waters to depths greater than 400 m (Almeida 1984).

Two genetically distinct stocks have been defined south of Nova Scotia: a ''northern stock'' occupying the Gulf of Maine-northern Georges Bank region and a ''southern stock'' occurring from southern Georges Bank to Cape Hatteras (Anderson 1974, Schenk 1981, Almeida 1984). Some mixing of the two stocks occurs throughout all or most of the year, perhaps facilitated by the wide temperature tolerance of this species.

Silver hake of the southern stock overwinter primarily along the outer continental shelf from Georges Bank to Cape Hatteras. During spring and summer, these fish move northward and inshore onto the southern and southeast parts of Georges Bank (Almeida 1984). Spawning occurs on the southern slopes of Georges Bank from May to November, reaching a peak in Southern New England and mid-Atlantic waters by May and June. Silver hake of the northern stock overwinter in deep basin areas of the Gulf of Maine, moving into shallower waters in late spring-early summer. Spawning occurs in inshore waters from Cape Cod to Grand Manan Island from June through November, peaking in July and August (Bigelow and Schroeder 1953, Colton and St. Onge 1974, Fahay 1974). Secondary spawning occurs on the north to northwestern slopes of Georges Bank (Sauskan 1964, Sauskan and Serebryakov 1968).

Female silver hake grow faster and live longer than males. Males attain a maximum length and age of about 42 cm (17 inches) and 10 years, respectively, contrasting with 67 cm (26 inches) and 12 years for females (Dery unpubl. data). Most silver hake are sexually mature by age 2.

Ageing methods for silver hake, based on the otoliths, remain somewhat controversial despite intensive research (Nichy 1969; Anderson and Nichy 1975; ICNAF 1976, 1977, 1978; Hunt 1980a). All investigators have counted hyaline zones as annuli. The prolonged spawning season of this species, and the variability of growth patterns due to genetic and environmental factors have made accurate identification of the first annulus, and discrimination between checks and annuli, difficult. In addition, age interpretation using whole otoliths may differ significantly from interpretations based on thin otolith sections and or sectioned halves. The edge type on some otoliths, either hyaline or opaque, has appeared to some investigators to be inconsistent with the season of the year, causing confusion with regard to edge interpretation. Age validation studies for silver hake of the Scotian Shelf have been conducted by Hunt (1978, 1979, 1980b). Although relatively few otoliths of these fish have been examined at this laboratory in recent years, some of the growth patterns appear similar to that observed on Gulf of Maine otoliths, and others resemble those of the southern stock.

The methods used at Woods Hole Laboratory have evolved from early studies by Nichy (1969) on the growth of young silver hake, from participation in age and growth workshops (ICNAF 1976, 1977, 1978), and from research on types of silver hake growth patterns and their distribution in the study area (Dery unpubl. data). Particular attention has been focused on otolith growth patterns of young fish (age 0+ to 1) to facilitate accurate interpretation of the first annulus, and to avoid assigning earlier hatched fish of the southern stock, and later hatched northern Georges Bank-Gulf of Maine fish, to different year classes. Hunt (1980a) summarized previous research in the literature concerning ageing methods used for this species. He described some aspects of otolith growth patterns as characteristic of given geographic areas but did not present an integrated description of these patterns for different stocks of silver hake. The method of presentation of Hunt's interpretations, in addition to his use of whole otoliths rather than thin-sections, makes difficult direct comparisons of his criteria with those used at Woods Hole. Validation of methods at Woods Hole has involved comparisons of modal groups in fish length-at-age data with the modal groups in length frequencies, and monitoring modal progression of prominent year-classes in the fishery on a seasonal and annual basis.

Methods of preparing otoliths have been described by Nichy (1969, 1977), Anderson and Nichy (1975), ICNAF (1976, 1977, 1978) and by Hunt (1980a). Such methods have involved storage of whole otoliths in glycerin or some other medium to "clear" the otolith and enhance the hyaline zones. Other methods include dry storage and soaking for a short period in ethyl alcohol before viewing. At the Woods Hole Laboratory, otoliths are stored dry in coin envelopes. A thin transverse section 0.20-0.23 mm thick is removed at the nucleus and examined under reflected light against a dark background, using a method developed by Nichy (1977). The cut surfaces of the sectioned otolith may be used in addition to, or instead of, the thin-section, depending on the degree of complexity of the growth pattern.

For some fish, whole otoliths, examined in ethyl alcohol, are used to verify age from thin-sections, but are not considered completely reliable. This is because the pattern of early growth on the otolith, which is often difficult to interpret, tends to be obscured by subsequent calcification, despite the use of strong clearing media such as glycerin (Anderson and Nichy 1975). In general, silver hake otoliths become thicker with increasing age relative to an increase in width. Therefore, misinterpretation of early growth, especially of older fish, is more likely. Nevertheless, some whole otoliths exhibit a clearer pattern of annulus formation than do thin-sections, especially if the annular zones are weak or diffuse.

Growth patterns observed on silver hake otoliths tend to support Almeida's (1984) definition of two separate stocks from the Gulf of Maine to Cape Hatteras. Variations in the growth patterns on otoliths with geographic location include size and formation of the first annulus, relative growth increment widths between annuli due to differences in growth rate, formation of checks and split zones, and time of annulus formation. Although characteristic growth patterns can be identified for each stock, some patterns are difficult to classify (due in part to individual variability). Other patterns are intermediate in type, with aspects characteristic of both Gulf of Maine fish and those further south. This may reflect stock intermixture as suggested by Almeida (1984). Seasonal shifts in the distribution of growth patterns also appear to be consistent from year to year and seem to reflect observed migratory movements (Dery unpubl. data).

The otoliths of silver hake from the southern stock tend to exhibit moderate to large amounts of opaque edge as early as March or April, indicating that annulus formation is complete by the end of the winter and probably earlier (Fig. 1). By convention, a birthdate of 1 January is used; the hyaline zone evident on the edge of the otolith is interpreted as an annulus whether or not it is complete. As is typical for many fish species, seasonal growth resumption is quite advanced for young fish relative to older individuals (Fig. 2) and age-1 fish otoliths show considerable amounts of "+" growth as early as April (Fig. 3). The timing of annulus initiation in the autumn is somewhat variable. Opaque edge may persist on the otoliths of age 0+ or 1+ and older individuals into autumn (September-October) (Fig. 4); however, most otoliths collected during autumn tend to exhibit a narrow hyaline edge which is not included in the age determination (Figs. 5 and 6).

In spite of variability in size and timing of formation of the first annulus, the characteristically large growth increment (wide opaque zone) between the first and second annuli provides a means of distinguishing between the two zones (Figs. 1 and 6). The differences in mean length at age-1 and age-2 in the spring months do not fully reflect the magnitude of this growth increment, because of the early growth resumption of age-1 fish (growth beyond the first annulus). The first annulus frequently appears as a small, dense, but split zone of hyaline material surrounding the nucleus of the otolith (Fig. 2), not evident on the otoliths of 0+ fish (Fig. 5). The annulus may also be a large and/or complex zone, with a significant amount of opaque material formed between the nucleus and the first annulus (Fig. 2). Occasionally, however, there is minimal evidence of this annulus, probably due to later hatching (Fig. 4).

The "pelagic" zone, or settling check, traditionally noted as important in age determination (Nichy 1969; ICNAF 1976, 1977, 1978; Hunt 1980a), was initially described by Nichy (1969) in his study of the growth of small silver hake as a small weak zone of hyaline material surrounding the nucleus that appears to form between the pelagic and demersal stages in the life history of this species. Some investigators including Hunt (1980a) have interpreted the "pelagic" ring as an occasionally large and strong zone of hyaline material that may be formed as late as 5 months of age. According to Fahay (1974), however, the length of the pelagic phase following hatching is about 2 months. Therefore, it is possible that a small first annulus formed close to the nucleus of later hatched or slower-growing fish is occasionally mistaken for the pelagic zone by some age readers. At the Woods Hole Laboratory, the pelagic zone has been conservatively interpreted as a small, usually weak zone, following the criteria of Nichy (1969) and verified by the appearance of this zone on age 0+ otoliths (Figs. 4 and 7). It should be noted, however, that accurate differentiation of the pelagic zone from the first annulus is difficult and remains a significant source of error.

Checks formed between the first and second annuli on otoliths collected from southern stock individuals may confuse interpretation of annular zones. The formation of a spring check on age-1 otoliths has been documented by Nichy (1969) (Fig. 6) and similar checks may also be formed later in the season. A check formed in late summer to early autumn, close to the time at which the annulus begins to form, is characteristic of most silver hake otoliths of the southern stock. Such checks are usually weak and/or discontinuous zones that are not as prominent as annuli in the sulcus area (Figs. 6 and 8). By comparison, the second and subsequent annuli are strong and consistent around the periphery of the otolith, particularly on the proximal (sulcus) and distal sides of the section (Figs. 2, 6, and 8). Typically, annular zones on otoliths of silver hake of this stock are evident as thick dense bands of hyaline material layered on the proximoventral part of the otolith (Figs. 6 and 8). Occasionally, however, these bands are split into several rings that must be traced around the periphery of the section in order to resolve the annular zones (Figs. 9 and 10). Whole otoliths can be especially useful in helping to resolve anomalous zones such as checks and split zones on thin otolith sections.

Subsequent to the second annulus, growth increments tend to be quite narrow on otolith sections due to a decrease in growth rate, so that annuli are layered rather closely together. This is particularly characteristic of male silver hake whose growth rate is slower than females after age-2 (Fig. 6). Because of these narrow growth increments, older fish may be difficult to age, especially if there are strong checks in between the annuli. Even where growth increments are relatively wide, annuli may be weak or diffuse (Fig. 10). If growth is "shifted," that is, if there is an unusual amount of growth on one part of the section in contrast to the normal pattern of deposition, interpretation should be focused in the direction of the shift in growth in order to avoid underinterpretation of age. The large amount of accreted material in each growth zone as a result of this shift enhances definition between the annuli and therefore facilitates age interpretation.

Time of annulus formation for the northern stock is later relative to hake further south, and follows a more seasonal pattern, as would be expected for more northerly latitudes (Williams and Bedford 1974). Annuli of these fish are completed in the late-winter to early-spring months with the exception of age-1 fish, some of whom resume growth during the winter months. Therefore, the otoliths of most fish in March and April may continue to exhibit hyaline edge or a small amount of opaque edge, particularly on the thin-sections (Fig. 11), while age-1 fish may exhibit a larger amount of opaque edge (Fig. 12). By October-November, some hyaline edge is usually evident on otoliths of age 2+ and older fish (Fig. 13), while opaque edge is likely to persist somewhat longer on age 0+ and 1+ fish.

The first annulus on otoliths of Gulf of Maine fish is somewhat variable in size, reflecting a tendency for some year-classes (e.g., 1982, 1984) to evidence a bimodal distribution of length at age-1. The first annulus on small one-year-old fish (e.g., 5 cm) may appear as a relatively weak hyaline zone and coincident with the pelagic zone (Fig. 14). It may be difficult to distinguish from the pelagic zone, spring check, and second annulus because the growth increment between the first and second annulus in the Gulf of Maine is generally smaller than further south, and because more of these fish are hatched later in the year. In some cases, the first annulus may not be evident but is assumed near the nucleus because of the large growth increment between the nucleus and the first strong hyaline zone that can be interpreted as an annulus (Fig. 15). While this interpretation may not appear justifiable biologically, it is necessary in order to avoid assigning these fish, and earlier hatched individuals, to different year-classes. Figure 16, for example, shows a section from a 13-cm fish sampled in April with no evidence of an annulus. This fish is interpreted as age-1 (late hatched) and not 0+, because spawning in the Gulf of Maine does not begin until the summer months.

Otoliths from larger age-1 fish may exhibit a well-defined hyaline zone (first annulus) formed some distance from the pelagic zone, which may be more prominent on these otoliths (Figs. 13 and 17). No marked discontinuity appears to exist between the growth patterns of these large age-1 fish and age-2 fish with a tiny first annulus. One technique for differentiating large age-1 fish from small age-2 fish with similar growth patterns involves measurement of the first annulus on otoliths collected from fish identified as age-1 using length-frequency data. Such measurements can provide an estimate of average and maximum first-annulus width for adult fish so that overinterpretation of age can be avoided in cases where the pelagic zone is prominent.

The weak zone formed around the pelagic zone on some silver hake otoliths can be difficult to identify as either a spring check (typical for silver hake of the southern stock) or a weak first annulus (more characteristic of the northern stock), since these zones form at the same time (April-May) (compare Figures 6, 13, and 14). Some silver hake from the Gulf of Maine exhibit a large first annulus with a very weak or nonexistent pelagic zone (Fig. 18). This type of pattern is more commonly observed on silver hake from the Scotian Shelf. Because of the variations in first-year growth patterns observed in the Gulf of Maine, the detail available on the otolith section seems necessary in order to make the most accurate interpretation possible.

The otoliths of Gulf of Maine silver hake are narrower and thicker in cross-section in contrast to those from more southern areas. Subsequent to the second annulus, accurate annulus interpretations of these fish are facilitated by this increased thickness and relatively wide increment widths between annuli resulting from faster growth (Figs. 11 and 14). In addition, the annular zones are quite easy to interpret because of relatively few anomalies (checks and split zones) and because the annular zones are strong and well defined (Figs. 14 and 19). Prominent checks are evident on some Gulf of Maine otoliths, but they are easily recognized by weak formation in the sulcus area in contrast to the annular zones (Fig. 11).

Some fish collected in the Southern New England and southern Georges Bank area exhibit growth patterns that appear to be hybrids of the two basic growth patterns described above. For example, the growth increment between the first and second annulus is intermediate in width, or the growth pattern will exhibit larger numbers of checks than is characteristic for the Gulf of Maine but fewer than typically seen further south (Fig. 20). Other otoliths, especially from southern Georges Bank fish, exhibit numerous strong checks and split zones that make annulus identification difficult (Fig. 21). In general, growth patterns observed among fish collected in the spring from the southern New England-southern Georges Bank area are rather heterogeneous compared with the greater consistency observed in the mid-Atlantic or northern Georges Bank-Gulf of Maine areas.

In summary, systematic study of the types of otolith growth patterns exhibited by silver hake of various stocks may facilitate consistency of age interpretation of these fish because of their prolonged spawning season and the variability of their growth patterns. Although bias may be created in anticipating an interpretation based on the geographic location of the sample, errors due to inconsistent interpretations could be more serious. Age readers at the Woods Hole Laboratory, having noted the variability of growth patterns on silver hake otoliths, attempt to apply standard criteria for the identification of annuli and checks that are agreed upon as valid by other age readers.

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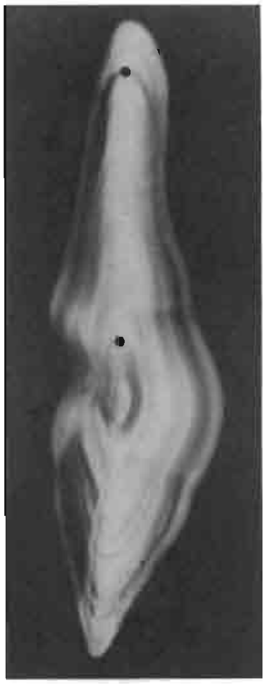

Figure 1

Otolith section of a 31-cm age 2+ female silver hake (southern stock) in April showing strong first and second annuli and wide opaque edge.

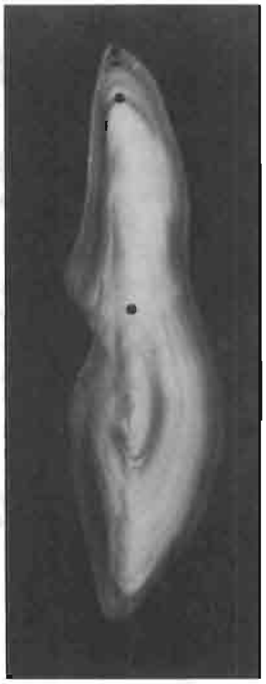

Figure 2

Otolith section of a 28-cm age-3 male silver hake (southern stock) collected in April showing strong annuli and a hyaline edge. Checks are evident between the second and third annuli.

Figure 3

Otolith section of an 18-cm age 1+ silver hake (southern stock) collected in April showing a weak settling check, large and complex first annulus, and opaque edge.

Figure 4

Otolith section of a 24-cm age 1+ silver hake (southern stock) collected in October showing no evidence of a first annulus, and opaque edge.

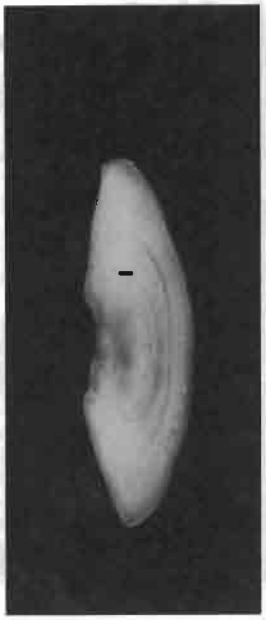

Figure 5
Otolith section of a 10-cm age 0+ silver hake (southern stock) collected in October showing a weak settling check and narrow hyaline edge.

Figure 6
Otolith section of a 33-cm age 5+ male silver hake (southern stock) collected in October showing spring, summer, and autumn checks between the first and second annuli and a narrow hyaline edge. Annuli 2-5 are closely spaced.

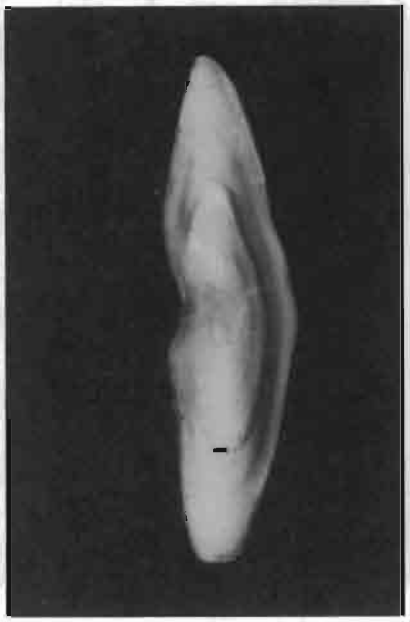

Figure 7
Otolith section of a 13-cm age 0+ silver hake (southern stock) collected in September showing a strong settling check.

Figure 8
Otolith section of a 36-cm age-5 male silver hake (southern stock) collected in April showing strong annuli with spring and autumn checks between the first and second annuli.

Figure 9
Otolith section of a 42-cm age-4? female silver hake (southern stock) collected in April showing split diffuse annuli and numerous checks between the second and third annuli.

Figure 10
Otolith section of a 39-cm age-4? female silver hake (southern stock) collected in April showing vague diffuse annuli and spring and autumn checks formed between the first and second annuli.

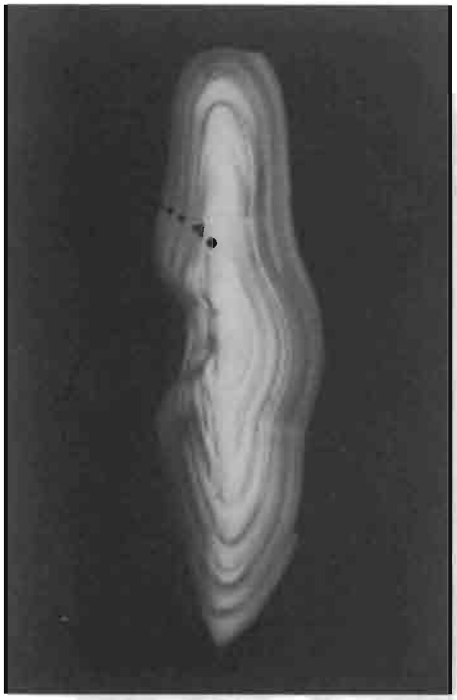

Figure 11

Otolith section of a 48-cm age-5 female silver hake (northern stock) collected in May showing strong widely spaced annuli, a check formed between the first and second annuli, and hyaline edge.

Figure 12

Otolith section of a 10-cm age-1 silver hake (northern stock) collected in May showing a prominent settling check and opaque edge.

Figure 13

Otolith section of a 30-cm age 2+ female silver hake (northern stock) collected in October showing a prominent settling check, large first annulus, and narrow hyaline edge.

Figure 14

Otolith section of a 48-cm age-5 female silver hake (northern stock) collected in November showing a small weak first annulus followed by a spring check and strong, widely spaced annuli 2-5.

Figure 15

Otolith section of a 39-cm age-4 female silver hake (northern stock) collected in November showing a large second annulus and no evidence of a first annulus.

Figure 16

Otolith section of a 13-cm age-1 silver hake (northern stock) collected in April showing a weak settling check but no first annulus, possibly due to a late hatch date.

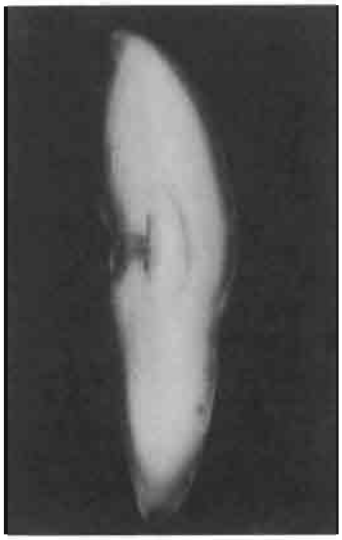

Figure 17
Otolith section of a 14-cm age-1 silver hake (northern stock) collected in April showing a weak settling check and strong first annulus formed on the edge.

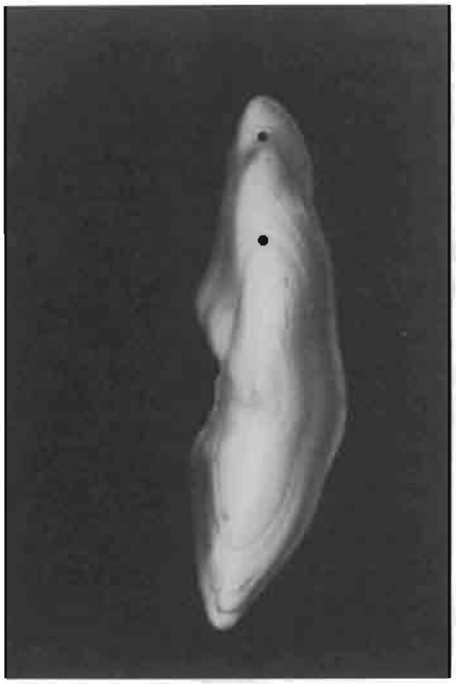

Figure 18
Otolith section of a 31-cm age 2+ female silver hake collected in November showing a Scotian Shelf type with a large first annulus and very weak or non-evident settling check.

Figure 19
Otolith section of a 41-cm age 4+ female silver hake (northern stock) showing a large first annulus and strong, widely spaced annuli on the ventral tip due to shifting of otolith growth.

Figure 20
Otolith section of a 35-cm age-9 male silver hake collected in April from Southern New England waters showing a large complex first annulus and closely spaced annuli 3-9.

Figure 21
Otolith section of a 44-cm age-5? female silver hake collected in April from the southern edge of Georges Bank. Numerous checks and split zones are evident on this section.

9

Red hake
Urophycis chuss

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Red hake is a demersal gadoid inhabiting the continental shelf waters from Nova Scotia to North Carolina. North of Nova Scotia this species is thought to be rare; its sibling species, the white hake (*Urophycis tenuis*) is much more common in that region (Musick 1974). However, according to Markle et al. (1980) and Svetovidov (1982), confusion still exists as to the identity of *Urophycis* species in Canadian waters, at least on the eastern part of the Scotian shelf. *Urophycis chuss* is most abundant from southwest Georges Bank to the New York Bight (Anderson 1982).

Two stocks of red hake are currently recognized: one inhabiting the Gulf of Maine-northern Georges Bank region and a second stock inhabiting the area from southern Georges Bank to Cape Hatteras (Conservation and Utilization Division 1985). The migratory movements of this species are largely inshore-offshore in response to seasonal changes in water temperatures. Spawning occurs May through November from the New York Bight to Georges Bank. It may not begin until June in the Gulf of Maine based on the absence of red hake eggs and larvae through May in those waters (Marak and Colton 1961, Colton and St. Onge 1974).

Growth of this species is sexually dimorphic, with females generally larger and longer lived than males. Males attain a maximum length and age of about 53 cm (21 inches) and 11 years, respectively, contrasting with 63 cm (25 inches) and 12 years for females. Most red hake are sexually mature by age 2 (McBride and Brown 1980).

Rikhter (1968) presented the results of age and growth studies of red hake using otoliths but did not describe methods other than his technique for preparing samples. At the Woods Hole Laboratory, age determinations have been conducted for a number of years. Validation studies (unpublished) indicate that hyaline zones traceable in the sulcus acusticus area of the transverse section (the collum) are valid annuli.

The otoliths of red hake from southwest Georges Bank to Cape Hatteras generally exhibit well-defined annular zones, including the first annulus. Accurate age determinations of northern Georges Bank and Gulf of Maine fish are considerably more challenging. The pattern of first annulus formation is more variable, and subsequent annuli are often weak, split, or difficult to trace around the periphery of the otoliths. Prominent second-summer checks also occur which are difficult to distinguish from annular zones. Time of annulus formation is variable and difficult to assess because of the formation of split zones. Thus far, seasonal progression of age groups (based on age-length keys) in the length-frequencies indicate that the methods are valid, but the possibility for error appears to be much greater for northern Georges Bank and Gulf of Maine red hake than for more southern areas.

Rikhter (1968) prepared red hake otoliths by cutting them in half and then polishing and burning the cut surfaces. The method developed at the Woods Hole Laboratory and now in current use involves removing a thin transverse section (0.20-0.23 mm) from the thickest part of the otolith through the nucleus. As a rule the otolith is not heated, although in some cases those from Gulf of Maine fish with weak growth patterns are baked at 450°F for 2-3 minutes before sectioning.

All thin-sections are viewed in ethyl alcohol under reflected light against a black background at 15-20×. The orientation of the thin-section from unbaked otoliths in relation to the light source may be critical to accurate evaluation of annuli in the sulcus acusticus area. This part of the section is very important for age determination of this species. The sulcus is more translucent than other parts of the otolith; therefore, the angle of incident reflected light affects the appearance of the annuli in the sulcus. They are most clearly evident if oriented perpendicular to incident light so that they are reflecting light rather than channeling light rays through them.

Observed differences in otolith growth patterns between the southern and northern stocks reflect differing times of annulus formation, growth rates, and environmental/genetic factors. These differences are stable from year to year and tend to support current stock definition. Silver hake from southwestern Georges Bank to Cape Hatteras exhibit clear growth patterns with strong distinct annuli. Red hake of the northern Georges Bank and Gulf of Maine region are faster growing after age 2 than more southern fish based on age-length keys, resulting in more widely spaced annuli, and their typically anomalous growth patterns often make analysis difficult. Some fish from this area do exhibit clear growth patterns, but the relatively wide growth increments between annuli often distinguish them as Gulf of Maine fish.

Annulus formation (completion of hyaline zone) of most red hake from the southern group is complete by April or May. By convention, a birthdate of 1 January is used; from that date until seasonal growth resumption, the hyaline zone evident on the edge of the otolith is interpreted as an annulus whether or not it is complete. The type (opaque or hyaline) and amount of edge is somewhat variable during most seasons of the year. The observed edge during the spring months may be narrow-to-wide hyaline, depending, in part, on the width of the annulus being formed (Fig. 1), or growth resumption may be indicated by the presence of narrow opaque edge (Fig. 2). Age-1 fish often show wide opaque edge (Fig. 3). In general, the edge evaluation can best be made on the dorsal tip of the transverse section, which is the longest radius of the section.

By September or October, seasonal growth is largely complete, especially for young fish. While opaque edge will persist on some otoliths, others exhibit large amounts of hyaline material that indicate that the beginning of annulus formation is well under way (Fig. 4). An advanced cycle of seasonal growth for young relative to older fish is not as predictable as it tends to be for other fish species. For that reason, the otolith edge should be interpreted with caution.

The pattern and rate of growth reflected on otoliths of red hake of the southern group is in many respects similar to what is observed for silver hake (*Merluccius bilinearis*) of the same geographic area. However, the growth patterns are less complex. The first annulus of red hake otoliths is usually a well defined hyaline zone in the central part of the otolith (Fig. 3). This zone, however, is variable in strength, size, and complexity due to individual variations and differences in spawning time. But, as with silver hake, the generally large growth increment between the first and second annulus facilitates interpretation of this zone (Fig. 2). On some otoliths the first annulus is tiny and/or coincident with the larval (pelagic) zone, or not evident at all, especially if the otolith was not sectioned precisely across the nucleus (Fig. 5). The large increment to the second annulus, however, indicates that the otolith is from a late-hatched fish with no apparent first annulus.

A small "settling" check surrounding the nucleus, which is representative of a shift from the larval pelagic habitat to the demersal habitat, is evident on the otoliths of both red and silver hake. This zone is fully described in the accompanying article concerning the latter species. The settling check on red hake otoliths is usually a weak zone and not easily confused with the first annulus (Fig. 3). In some cases where the first annulus is weak, however, the settling check may appear relatively prominent (Figs. 1, 2, and 4).

Check formation is characteristic of the second season of growth following the first annulus. However, since annuli on red hake otoliths from the southern area are normally distinct, they can usually be readily differentiated from spring, summer, or autumn checks. Figure 3 shows an otolith section from a 13-cm, age-1 fish sampled in April which shows a very narrow hyaline edge indicative of the formation of a spring check. Figure 6 shows an otolith taken from a 40-cm, age 3+ fish with complex check formation. A check formed just before the second annulus on some otoliths may be confused with that zone, except that the check is not strongly evident in the sulcus and is a relatively thin or weak ring. Figure 4 shows an otolith section from a 45-cm, age 4(3)+ fish with a weak second hyaline zone interpreted as the second annulus because it was rather distinct in the sulcus. In general, checks are not prominent in the sulcus area. Figure 7 is an otolith section from a 47-cm, age-5 female red hake with a small second annulus not interpreted as a check because of the width and strength of the zone.

The second and subsequent annuli on these otoliths tend to be wide, prominent hyaline zones, and closely parallel to one another for slow-growing fish, particularly males (Figs. 1 and 8). Annuli on some otoliths, although strong, are somewhat split or diffuse, and narrow growth increments between the annuli can make it difficult to distinguish one annulus from another. In addition, the increments between annular zones often become increasingly translucent after age 2 or 3. Examination of these zones in the sulcus area tends to minimize these problems, because annular zones develop resolution into clear separate bands in the sulcus (Figs. 4 and 8), due to the nature of its crystalline structure and its discontinuity with the rest of the otolith. With increasing age, red hake of the southern group do not usually become more difficult to interpret. In fact, many of them show unusually clear growth patterns (Fig. 9). The annuli, however, are spaced increasingly close together out to the edge of the section.

The rounded ventral part of the section can be useful in identifying the first few annuli because of the prominence of hyaline zones in this area. Nevertheless, it should be used with caution when interpreting subsequent annuli, because hyaline zones tend to fuse together on this part of the otolith. Also, the crowding of zones near the edge may result in underinterpretation of age (Figs. 1 and 8).

Time of annulus formation for red hake of the northern group is as variable as observed for the southern group. Correct edge evaluations are often quite difficult due to the extensive splitting of hyaline zones on many of the otoliths. With the exception of some young fish, growth resumption in the Gulf of Maine is not as advanced as observed further south. By April or May, annuli may not be completely formed judging by the presence of only tiny amounts of hyaline edge on some of the otoliths (Fig. 10). Relatively few of these otoliths exhibit opaque edge indicating seasonal growth resumption.

During the late summer (late July and August), most otoliths of all age groups exhibit at least some opaque edge (Fig. 11). In October or November, many otoliths continue to exhibit small to large amounts of opaque edge indicating that seasonal growth is not complete (Fig. 12).

It may be inferred from these observations that annulus formation for some Gulf of Maine red hake may not be complete until the early summer months. The presence of split hyaline zones and the shift in time of annulus formation from north to south may confuse attempts to decide whether or not the edge of the otolith should be included in the age. In addition, the type of edge observed may vary on different parts of the same otolith. It is helpful in this situation to locate the last fully formed annulus in the sulcus, which shows the annuli more clearly. However, the amount of newly formed material will be somewhat underestimated because the short radius from the nucleus to the edge of the sulcus results, overall, in less accreted material on this part of the otolith (Fig. 12).

The pattern of growth observed on many red hake otoliths from the northern area appears quite anomalous when compared with the southern group or to the closely related white hake (*U. tenuis*). The latter species often exhibits weak growth patterns on its otoliths, but few other anomalies complicate age determination. As will be shown, some red hake otoliths from the Gulf of Maine tend to resemble those of white hake both in terms of external morphology and the internal growth pattern. It is sometimes difficult to distinguish the two species using otoliths alone in this geographic area.

Similar to the southern group of red hake, the first annulus on northern group otoliths is variable in size and complexity. However, the integrity of hyaline zones in the center of the otolith representative of the first annulus can be difficult to establish because of the formation of numerous and sometimes prominent checks, and because the growth increment to the second annulus is sometimes rather small (Fig. 13). In addition, the first and second annuli are sometimes not very strong hyaline zones (Figs. 13 and 14). All of these factors result in the blurring of distinctions between annular zones.

One pattern especially difficult to interpret involves a relatively simple first hyaline zone formed some distance away from the nucleus that, because of its size, could represent a large first annulus or a small second annulus in otoliths of older fish (Fig. 15). The interpretation of this pattern is a persistent problem. However, by first establishing reference measurements of the second annulus on otoliths of known age-2 fish (via length-frequencies), it is possible to measure the questionable annulus and interpret it based on a comparison with reference measurements (for each year-class).

Subsequent to the first annulus, age interpretation of anomalous otoliths encounters further problems. Some otoliths generally form distinct annular zones, but the second and possibly third annuli are very weak. However, the structure of these wide (although weak) hyaline zones, especially in the sulcus, and their relative spacing, tends to confirm their identity as annuli (Figs. 13, 14, and 15).

Some otoliths exhibit a very weak growth pattern. Enhancement by baking may be necessary to identify any of the annuli (Fig. 16). Usually, traces of these zones are evident in the sulcus or on other parts of the otolith. If not, these otoliths cannot be interpreted.

Hyaline zones may be prominent in some otoliths, but each annulus is split into two or more hyaline zones. Strong checks are often associated with this pattern to further confuse age interpretations (Fig. 17). Figure 18 shows an otolith section from a 42-cm, age 5(4) female sampled in May where the pattern of annulus formation is so obscure that the sulcus is required to identify the annular zones.

Occasionally, the sulcus is of no use in distinguishing annuli because growth zone formation is discontinuous around the periphery of the otolith. Annuli on these otoliths cannot be traced with any confidence through the sulcus, since discrete hyaline zones that are recognizable as annuli may not be exhibited. Fortunately, the pointed dorsal area of the otolith section often shows enough evidence of discrete zones to estimate age with some accuracy (Fig. 15).

Red hake otoliths with anomalous patterns are typical in the Gulf of Maine. The reasons for these patterns are not understood. Some otoliths, however, exhibit a clearer pattern that resembles that of red hake of more southern waters, although relatively wider growth increments between annuli are characteristic because of a faster growth rate in the Gulf of Maine. The growth patterns on these otoliths are usually characterized by 1) a generally prominent first annulus; 2) relatively discrete annular zones, and 3) few anomalies such as splits and checks. Age interpretation of Gulf of Maine otoliths may not be more difficult with increasing age, despite anomalous growth patterns. Deposition of subsequent annuli may actually elucidate the pattern of earlier growth on the otolith (Fig. 19).

Some otoliths collected in the Gulf of Maine, particularly from the northwestern and eastern part near the Bay of Fundy, vary morphologically from red hake otoliths sampled elsewhere. Removed from fish taxonomically identified as *U. chuss*, these otoliths show characteristics that appear to be intermediate between what is normally observed for *U. chuss* and for *U. tenuis*. Red hake otoliths are characterized by a smooth surface and curvature, are rounded in cross-sectional dimension, and have smooth, reduced rostra (Fig. 20, left). In contrast, white hake otoliths are more angular, exhibit numerous surface ridges and dentations, are somewhat flattened in cross-sectional dimension, and have more prominent rostra (Fig. 20, right). ''Mixed'' otoliths are more angular than those of red hake, may show more surface irregularities, are more flattened, and have larger rostra (Fig. 20, center).

Since otolith shape and size are sensitive to genetic variations and are often used to trace the evolutionary patterns of fishes (e.g., Gaemers 1976), it is interesting to speculate as to whether the observed variation in otolith morphology is characteristic of red hake of the Gulf of Maine, or whether the existence of a hybrid *Urophycis* is indicated. Musick (1973) noted significant meristic-morphometric differences between white hake of Nova Scotian and southern New England waters and red hake from the Gulf of Maine and southern New England waters. However, his samples from the Gulf of Maine were collected from the southwest and southeastern parts of the Gulf and *not* from the northwestern-northeastern area where the ''mixed'' otolith types are most frequently observed. The ambiguity represented by the mixed otolith types is a problem for age determination because the growth patterns also reflect a mixed pattern. Since the spawning season and, therefore, the interpretation of the first annulus, differs for red hake and white hake, some uncertainty exists as to ageing methods for these otoliths. Thus far, the approach has been to assume these fish to be red hake until other evidence is available. Fortunately, white hake otoliths normally exhibit weaker growth patterns and wider growth increments than red hake otoliths with mixed patterns (compare Figures 21 and 22).

In summary, age determinations of red hake from southern Georges Bank to the Mid-Atlantic are as straightforward and reliable as the same procedure is difficult and relatively unreliable for many red hake of northern Georges Bank and the Gulf of Maine. Aspects of the growth patterns of red hake of the southern group are similar to what is observed for silver hake from the same area. Otolith growth patterns of the more northern group are often anomalous. In addition, some otoliths from the northwestern and eastern parts of the Gulf of Maine show characteristics that are intermediate in type between what is normally observed for red hake and for white hake.

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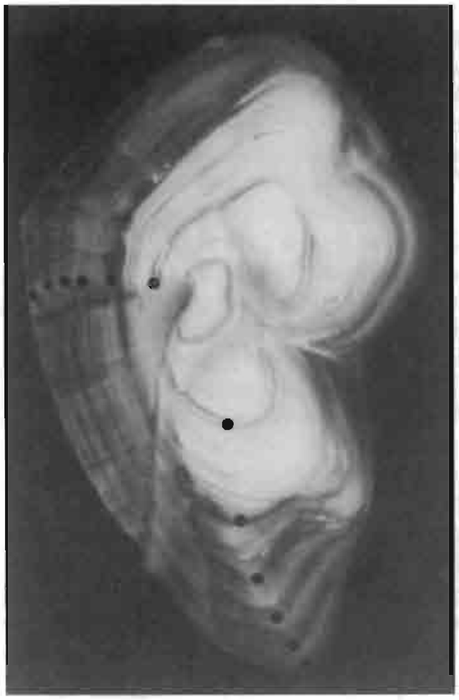

Figure 1
Otolith section of a 37-cm age-6 male red hake (southern stock) collected in April showing strong annuli and a hyaline edge.

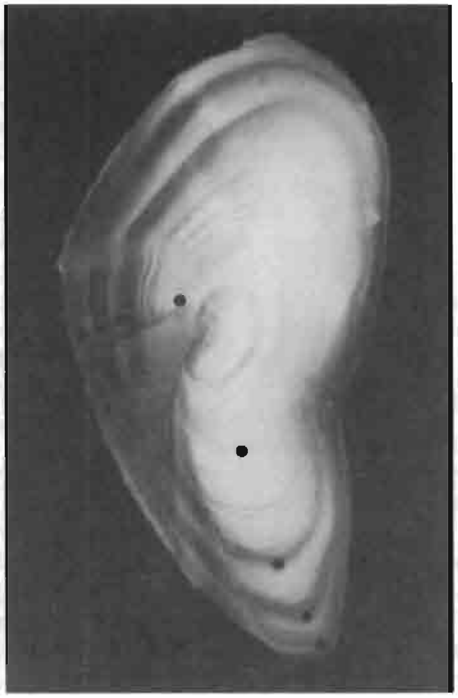

Figure 2
Otolith section of a 36-cm age-4 female red hake (southern stock) collected in April showing a strong settling check, weak first annulus, and narrow/opaque edge.

Figure 3
Otolith section of a 13-cm age-1 red hake (southern stock) collected in April showing a spring check forming on the edge.

Figure 4
Otolith section of a 45-cm age 4(3)+ female red hake (southern stock) collected in October showing a weak diffuse second annulus and narrow hyaline edge.

Figure 6
Otolith section of a 40-cm age 3+ female red hake (southern stock) in October showing a diffuse first annulus and summer and autumn checks between the first and second annuli.

Figure 5
Otolith section of a 34-cm age-4 male red hake (southern stock) collected in April with no first annulus evident.

Figure 7
Otolith section of a 47-cm age-5(4) female red hake (southern stock) collected in April showing a large first annulus and small second annulus.

Figure 8
Otolith section of a 34-cm age-7 male red hake (southern stock) collected in April showing closely spaced annuli.

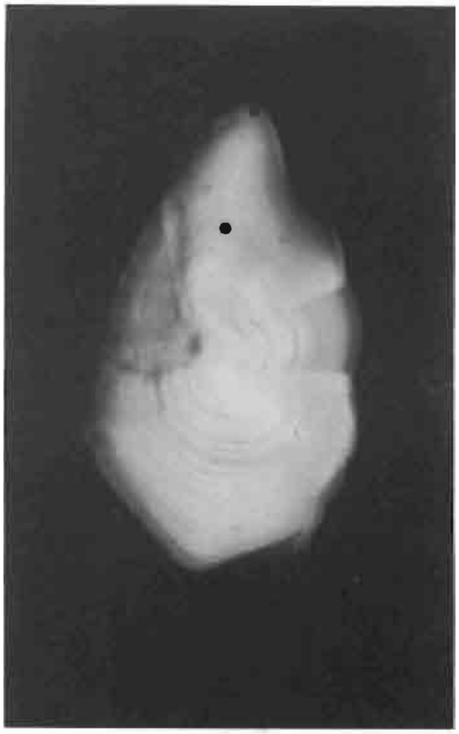

Figure 10
Otolith section of a 26-cm age-2 male red hake (northern stock) collected in April showing a weak first annulus and narrow hyaline edge.

Figure 9
Otolith section of a 45-cm age-8 female red hake (southern stock) collected in April showing clear annuli.

Figure 11
Otolith section of a 40-cm age 3+ female red hake (northern stock) collected in August showing split annuli and opaque edge.

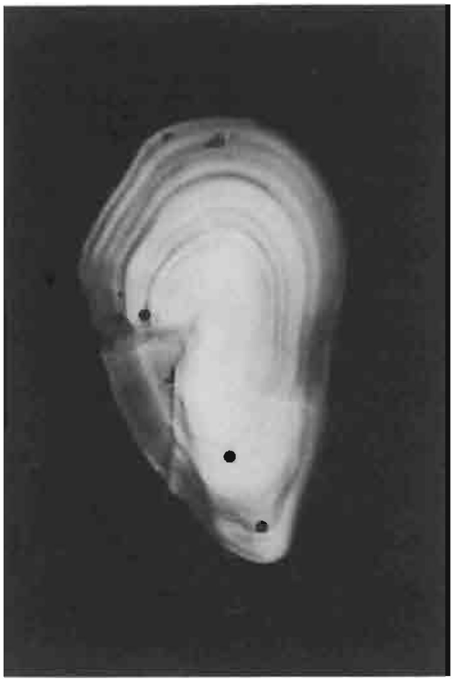

Figure 12
Otolith section of a 28-cm age 2+ male red hake (northern stock) collected in October showing a split second annulus in the ventral area and opaque edge.

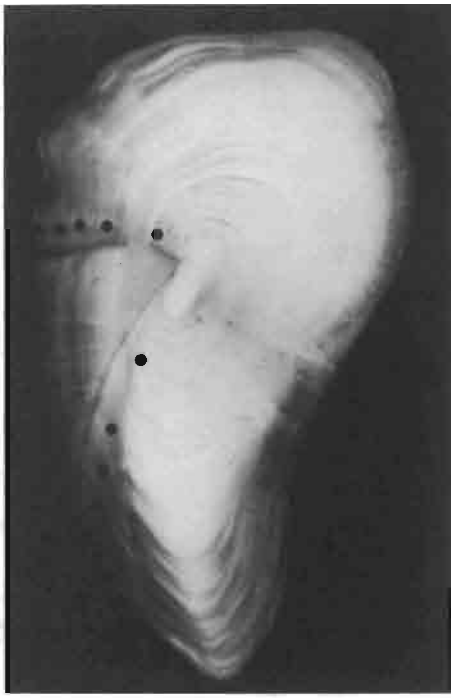

Figure 14
Otolith section of a 42-cm age-5(4) female red hake (northern stock) collected in May showing a weak second annulus that may be a check.

Figure 13
Otolith section of a 40-cm, age-6(5) male red hake (northern stock) collected in May showing a small weak second annulus.

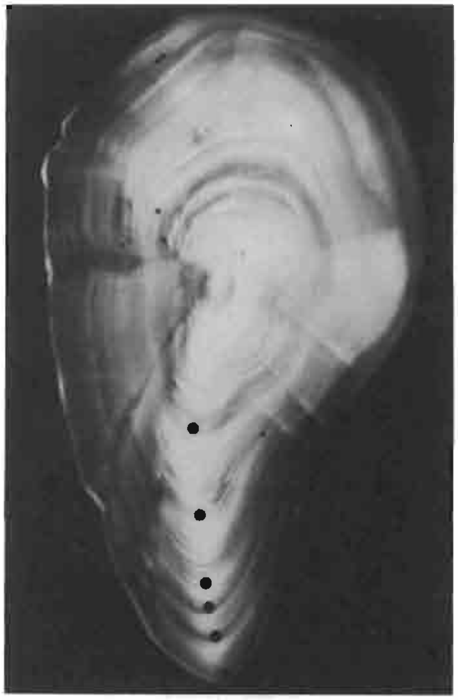

Figure 15
Otolith section of a 40-cm age-6 male red hake (northern stock) collected in April showing a large first annulus and weakly formed second and third annuli.

Figure 16
Otolith section of a 40-cm age 5? female red hake (northern stock) collected in April showing very weak annuli interpretable mainly in the dorsal area near the sulcus.

Figure 18
Otolith section of a 42-cm age-5(4) female red hake (northern stock) collected in April showing an obscure pattern of annulus formation requiring use of the sulcus to identify annuli.

Figure 17
Otolith section of a 46-cm age-4 female red hake (northern stock) collected in May showing checks and split annuli.

Figure 19
Otolith section of a 59-cm age-12(11) female red hake (northern stock) collected in May showing numerous annuli interpretable ventral to the sulcus.

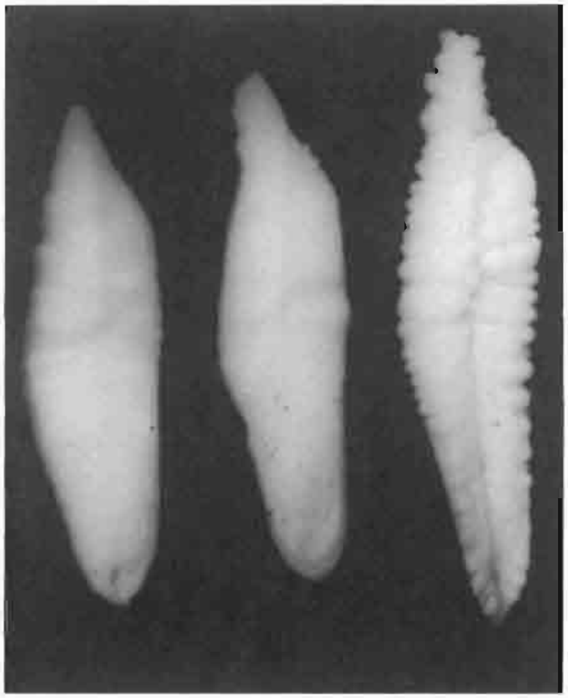

Figure 20
(Left) Whole otolith of a 46-cm red hake; (center) Whole otolith of a 48-cm red hake with "intermediate" characteristics; (right) Whole otolith of a 54-cm white hake.

Figure 21
Otolith section of a 48-cm age 5+ female red hake collected in May showing an "intermediate" type growth pattern.

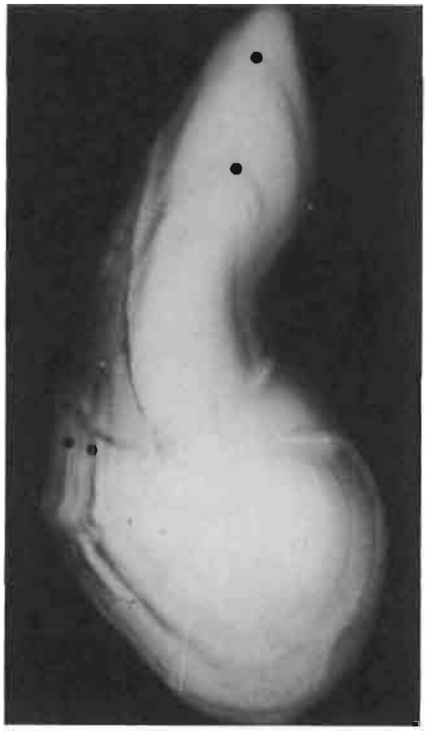

Figure 22
Otolith section of a 54-cm age 2+ female white hake collected in November showing weak annuli separated by wide growth increments.

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Black sea bass *Centropristis striata*

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Black sea bass is an economically important serranid ranging from New England to Florida (Kendall 1977). A protogynous hermaphrodite, sex reversal from female to male occurs for at least half of the population, usually between the ages of 2 and 5 (Mercer 1978). Males are faster growing than females, attaining a maximum length and age of over 60 cm TL (24 inches) and 20 years, respectively. Females reach a maximum length and age of 38 cm and 8 years (Lavenda 1949). Female black sea bass are sexually mature by age 2; males may not mature until age 4 (Mercer 1978).

Two stocks of black sea bass have been recognized north and south of Cape Hatteras (Cupka et al. 1973). The northern stock migrates seasonally in response to temperature changes. Most of these fish overwinter along the edge of the continental shelf in the southern part of the Mid-Atlantic Bight. In the spring they move inshore and northward to depths less than 40 meters for spawning and feeding on live bottom areas during the summer months (Musick and Mercer 1977, Kendall and Mercer 1982). Spawning extends from June through October, reaching a peak progressively later further north (Mercer 1978, Kendall and Mercer 1982). The southern stock does not appear to be seasonally migratory, frequenting the live bottom areas south of Cape Hatteras (Kendall and Mercer 1982). Spawning for these fish commences in February, reaching a peak in April or May (Mercer 1978).

Several hard structures have been used for age determination of black sea bass. Lavenda (1949) and Briggs (1978) used cellulose acetate impressions of the scales to age black sea bass of New York and New Jersey waters. They identified zones of closely spaced circuli as annuli on these structures. This technique, however, was not validated and has since been questioned by other investigators (Cupka et al. 1973, Mercer 1978, Link 1984). In studies of Virginia-South Carolina fish, otoliths were preferred over scales and were found to have valid age marks. Although investigators did not find operculae or vertebrae to be useful, pelvic spine sections and impressions of scales from behind the pectoral fin have recently been cross-validated with otoliths and found to be acceptable alternate ageing structures, although not as reliable as otoliths (Dery unpubl.). In this study, "cutting-over" marks on scales and hyaline zones on spines were validated as annuli. These marks were found to form at approximately the same time as the deposition of opaque material on otoliths is completed. The outer edge of the opaque zone has been interpreted as the annulus by other investigators (Cupka et al. 1973, Mercer 1978, Link 1984, Wenner et al. 1986). In general, otoliths are preferred at Woods Hole for routine age determinations, but scales are also collected for verification purposes since checks and split zones can cause difficulties with age interpretation.

Glycerin has been used as a storage medium to enhance the clarity of hyaline zones on otoliths (Cupka et al. 1973). Mercer (1978) reported that glycerin tended to overclear the otolith's edge, and therefore used glycerin to clear only those otoliths where annuli were obscured by the overgrowth of calcium. Wenner et al. (1986) stored otoliths dry, and viewed them in water. At Woods Hole, otoliths are stored dry and examined in ethyl alcohol to avoid the overenhancement of hyaline zones. Thin transverse sections (0.20-0.23 mm thick) are removed at the nucleus and are examined instead of whole otoliths if the annuli are obscured by later calcification. Otoliths are examined distal-surface-up against a black background at 10-15× using reflected light.

Five or six scales from behind the pectoral fin are impressed in laminated plastic (Dery 1983) and viewed under a microprojector at 40×. Pelvic spines require more preparation time. The outer tissue covering the spine can usually be readily peeled off prior to sectioning, but may first require soaking in water or bleach. A thin-section, about 0.20 mm thick, is removed just above the base of the spine. This thinness is required to clearly define the annuli on the spine section. Subsequently, the spine section must be soaked in clove oil for several minutes to whiten "opaque" zones and provide the necessary contrast with hyaline zones.

Annulus formation on otoliths, pelvic spines, and scales occurs in May or June. The outer edge of the opaque zone is interpreted as the annulus on otoliths, the outer edge of the hyaline zone as the annulus on spines, and the cutting-over mark as the annulus on scales. By convention, a birthdate of 1 January is used; the annulus forming on the edge of these structures is included in the age whether or not it is completely formed. Formation of opaque material may persist on some otoliths into the early autumn and should be taken into consideration when backcalculating otoliths. The formation of hyaline zone on otoliths, which normally occurs from June through the following January (Mercer 1978), is unusual because hyaline material, indicating slow growth, generally forms during the colder months of the year. It is possible that the lack of opaque material deposited during the warmest months may be due to shifts in calcium metabolism during onshore movements into very warm coastal water in the summer. The summer flounder (*Paralichthys dentatus*) otolith shows similar seasonal calcification patterns, a species that has a migration and distribution pattern similar to that observed for black sea bass.

Otoliths show the clearest record of first-year growth. Figure 1 shows the age structures from a 22-cm, age 1+ fish collected in November. A weak hyaline core area formed after hatching occurs close to the center of the otolith (Fig. 1A). Opaque material is deposited around this central core. The deposition of this material is complete by the following spring, forming the first annulus. A wide hyaline zone then forms during the summer and autumn of the second year. This otolith shows an unusual amount of opaque edge for November. Hyaline edge would persist on most adult black sea bass otoliths until January.

The spine section (Fig. 1B) shows minimal evidence of the first annulus and first-year growth, which is generally characteristic of black sea bass spines. This annulus is located on the inner edge of the lumen and appears as a thin band of hyaline material. All the opaque material formed after the first annulus represents growth in summer and autumn of the second year. A tiny amount of hyaline material is evident on the edge. Hyaline edge normally begins to form on the spine during the winter months.

A poorly defined first annulus on the corresponding scale is typical for most black sea bass (Fig. 1C). If present it will usually appear as a zone of closely spaced circuli without a cutting-over mark. The first cutting-over or erosion mark representing the second annulus is not yet evident.

Figure 2 shows the three types of age structures for a 40-cm, age-4 fish collected from Nantucket Sound in June, the time of annulus formation. All show rapid growth typical of more northern areas. The otolith (Fig. 2A) shows prominent hyaline zones separated by wide growth increments. Four opaque zones, including the edge, are formed on this otolith. Figure 2B shows the corresponding spine section with four clearly formed annuli (hyaline zones), including the edge of the lumen and the outer edge of the spine. A check is also evident between the first and second annuli. Such checks formed during the second summer of growth are typical and can be confused with the second annulus, especially on the spine sections of age 1+ or 2 fish. However, such checks are not continuous around the lumen or visible in the indented area of the section marking the spinal groove. The scale of this 4-year-old fish (Fig. 2C) has two clear cutting-over marks representing the second and third annuli. The first annulus is vaguely indicated by the zone of compacted circuli near the focus of the scale. The outer edge of the scale is the fourth annulus.

Clear growth patterns are also characteristic of slower growing black sea bass from more southerly ranges of the northern stock. Figure 3 shows the growth patterns of a 35-cm, age-6 fish collected from off Virginia in February. Growth increments on these structures are relatively narrow. The last (sixth) annulus on the outer edge of the otolith, spine, and scale is not complete because of the February collection date. Nevertheless, we include the edge in the age of the fish because of the 1 January birthdate convention.

Although opaque zones are usually well defined on the otoliths, they may sometimes be bordered by such thin hyaline zones that annuli could be missed in the age interpretation. Figure 4 shows the age structures of a 31-cm, age-3 black sea bass sampled from Nantucket Sound in June. The second hyaline zone bordering the second annulus (opaque zone) is weakly defined on the otolith (Fig. 4A). The second annulus, however, is very strong on the spine section (Fig. 4B) and the scale (Fig. 4C).

Weak annuli are also characteristic of the central region of the otoliths for some older fish, prior to a sharp increase in growth rate after four or five years of slow growth. This pattern occurs in the age structures of a 46-cm, age-8 fish collected from Virginia waters (Fig. 5). The second, third, and fourth annuli are clearly formed, although they are closely spaced on the spine section (Fig. 5B) and scale (Fig. 5C). The third annulus is split into two rings on the spine section. On the otolith (Fig. 5A), however, these annuli (2-4), are very difficult to distinguish without referring to one of the other two structures. The change in growth rate reflected by these structures may be the result of sex reversal or migration.

The formation of strong checks and split hyaline zones (or split cutting over marks on the scale) may make annulus interpretation difficult. Figure 6 shows the age structures of a 30-cm, age 2+ fish sampled from New Jersey waters in November. Both the first and second annuli (opaque zones) (Fig. 6A) are split into two rings, but the relative spacing between them does not identify these rings as "split" zones without reference to the other two structures (Figs. 6B and 6C). Therefore, based on examination of the otolith alone, an age of 3+ or 4+ could be interpreted. It should be noted that the first hyaline zone is split into two or more rings on many otoliths. Identification of this anomaly is difficult only if there are narrow growth increments between the first several annuli.

Figure 7 shows the difficult-to-interpret age structures of a 34-cm, age 4(3)+ black sea bass sampled from New Jersey waters in November. If the second annulus is bordered by a weak hyaline zone (Fig. 7A), the age would be interpreted as 4+, otherwise the age would be 3+. The spine section (Fig. 7B) indicates an age of 4+, although the hyaline zones are somewhat close together. The most likely interpretation of the scale impression, however, would be age 3+, recognizing a false cutting-over mark formed between the first and second annuli (Fig. 7C). For such fish the final age must be determined using the strongest evidence for a particular age.

Annuli may remain relatively easy to interpret at older ages, although increasingly narrow growth increments may cause some confusion. The age structures of Figure 8 from a 57-cm, age-10 fish sampled from Virginia waters in February, show the clear annuli typical of most older fish. Annuli on the otoliths may be somewhat obscured by overgrowth of calcium, and erosion of the scale may obliterate the annuli close to the central anterior edge of the scale. Nevertheless, these structures can still be accurately aged, especially if the otolith is sectioned and the anterior corners of a scale are carefully studied.

In summary, some geographic variation in growth patterns appears to exist. For example, the growth patterns on the structures of some New Jersey fish are especially difficult to interpret because of the formation of strong checks or split zones (Figs. 6 and 7). Characteristic differences between the northern and southern stocks have not been documented, however.

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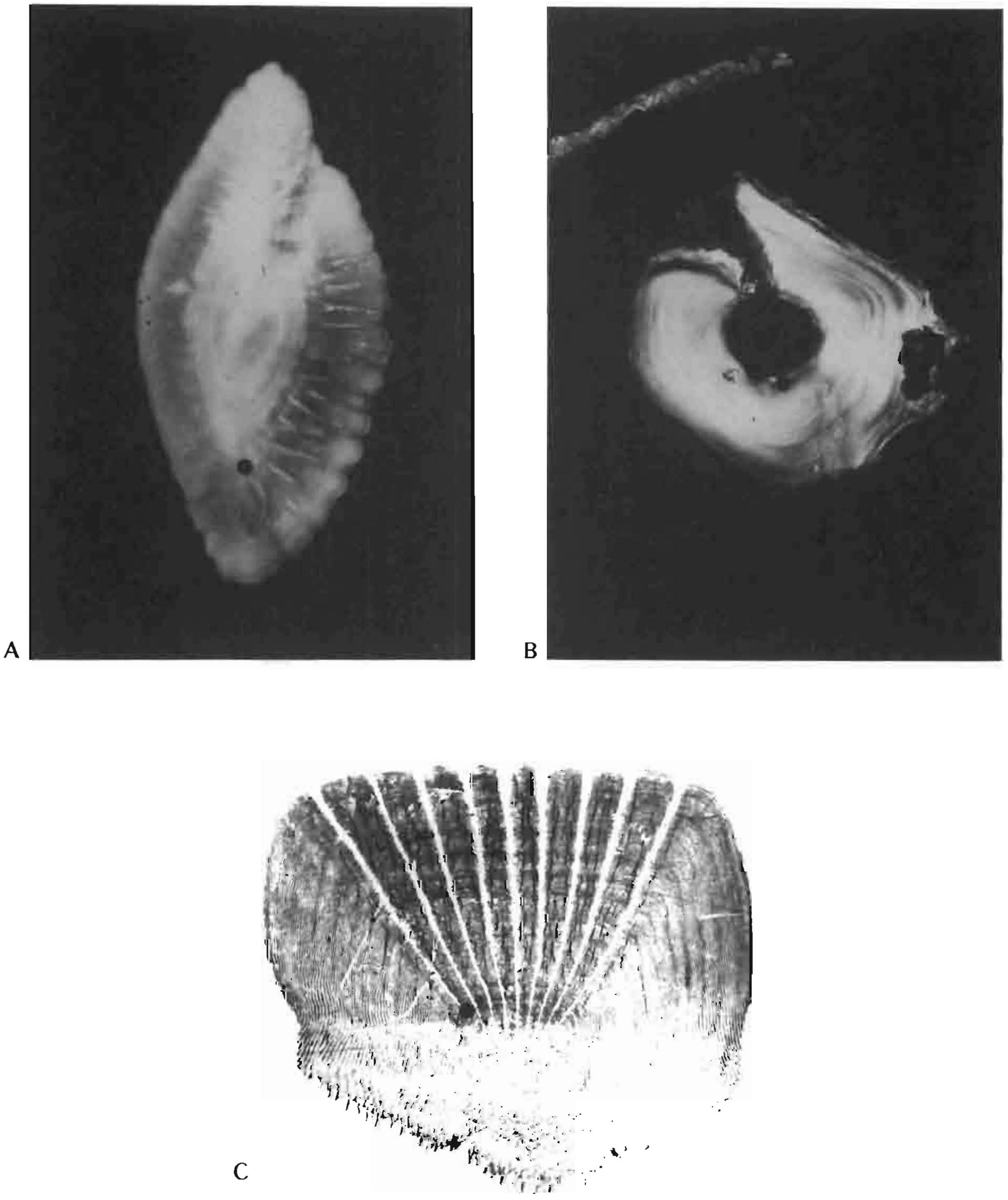

Figure 1

(**A**) Whole otolith of a 22-cm age 1+ black sea bass collected in November showing wide opaque edge. First annulus is bordered by a wide hyaline zone. (**B**) Pelvic spine section showing a thin first annulus (hyaline zone) bordering the lumen and the beginnings of hyaline edge. (**C**) Pectoral scale impression (expanded view) showing a zone of compacted circuli near the focus which may represent the first annulus.

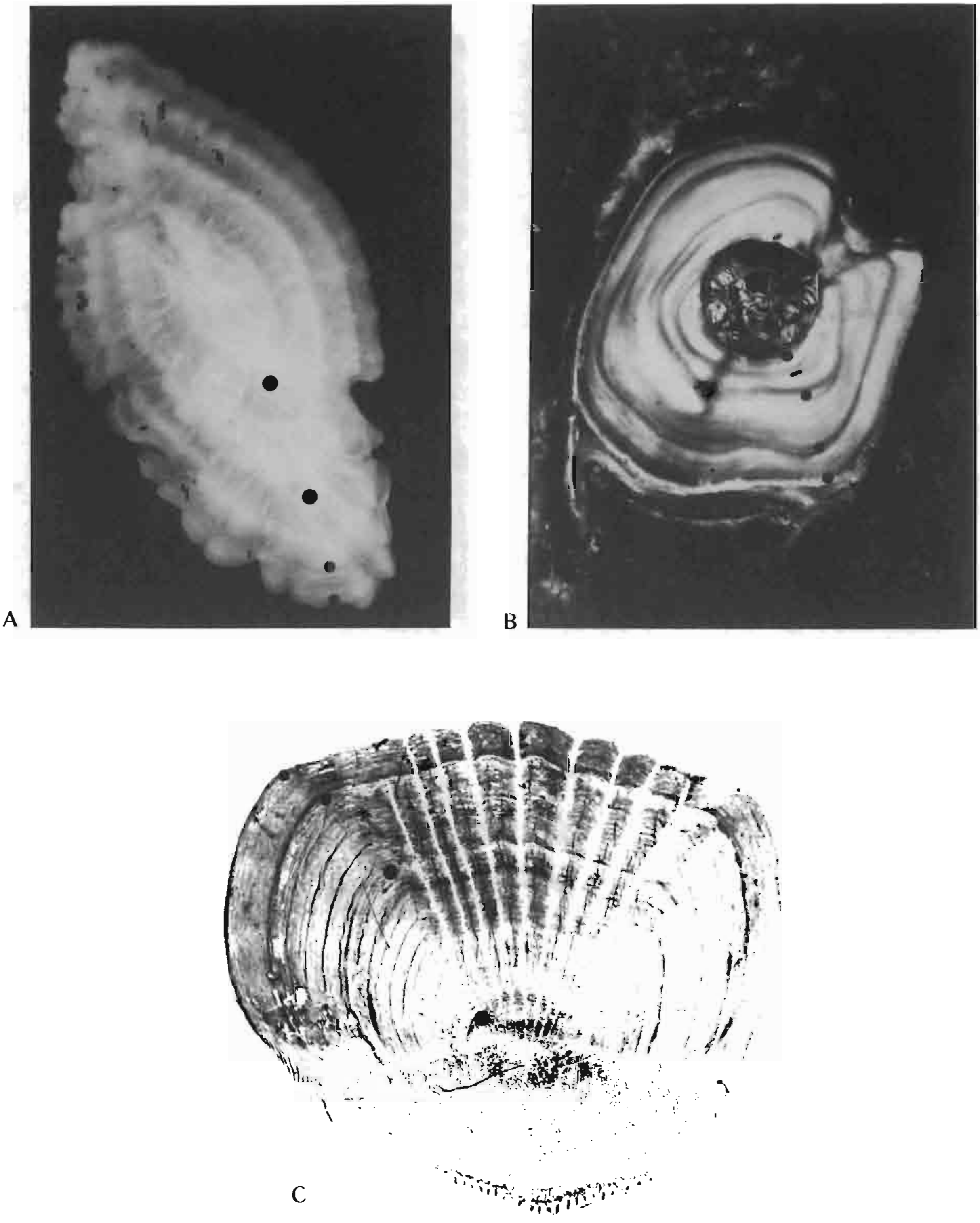

Figure 2

(A) Whole otolith of a 40-cm age-4 female black sea bass collected in June from Massachusetts waters, showing narrow hyaline edge. Clear annuli with wide growth increments are evident. (B) Pelvic spine section showing clear annuli and a hyaline to narrow opaque edge. Check between the first and second annuli is not continuous around the lumen or separate from the first annulus. (C) Pectoral scale impression from the black sea bass of Figure 2A showing two clear "cutting-over" marks at the second and third annuli and a "cutting-over" mark (annulus) at the edge of the scale included in the age.

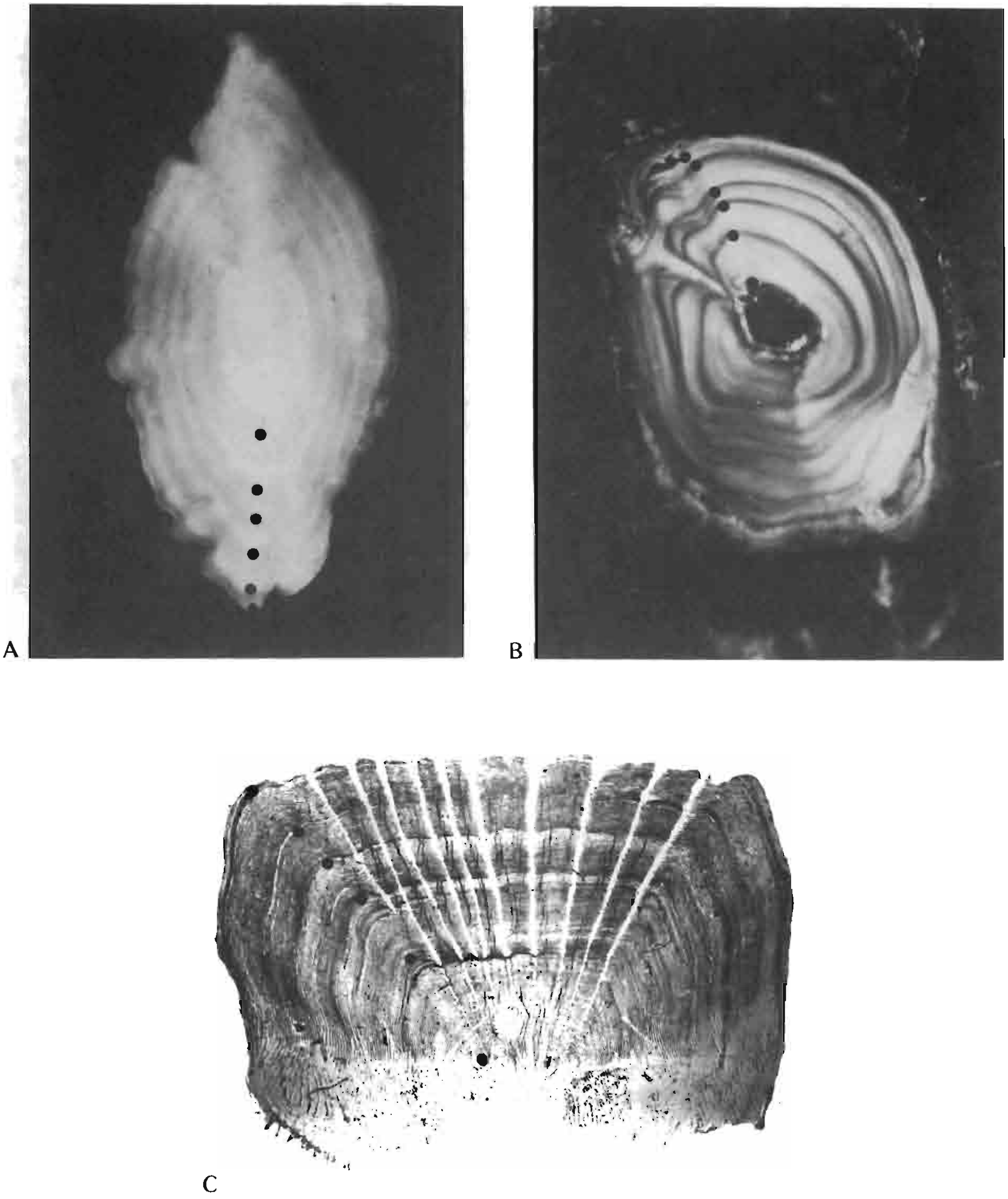

Figure 3

(A) Whole otolith of a 35-cm age 6? male black sea bass collected in February from Virginia waters. Clear annuli (opaque zones) are evident with an incomplete sixth annulus on the edge. (B) Pelvic spine section showing a sixth annulus (hyaline zone) barely evident on the edge of the section. (C) Pectoral scale impression showing split second, third, and sixth annuli.

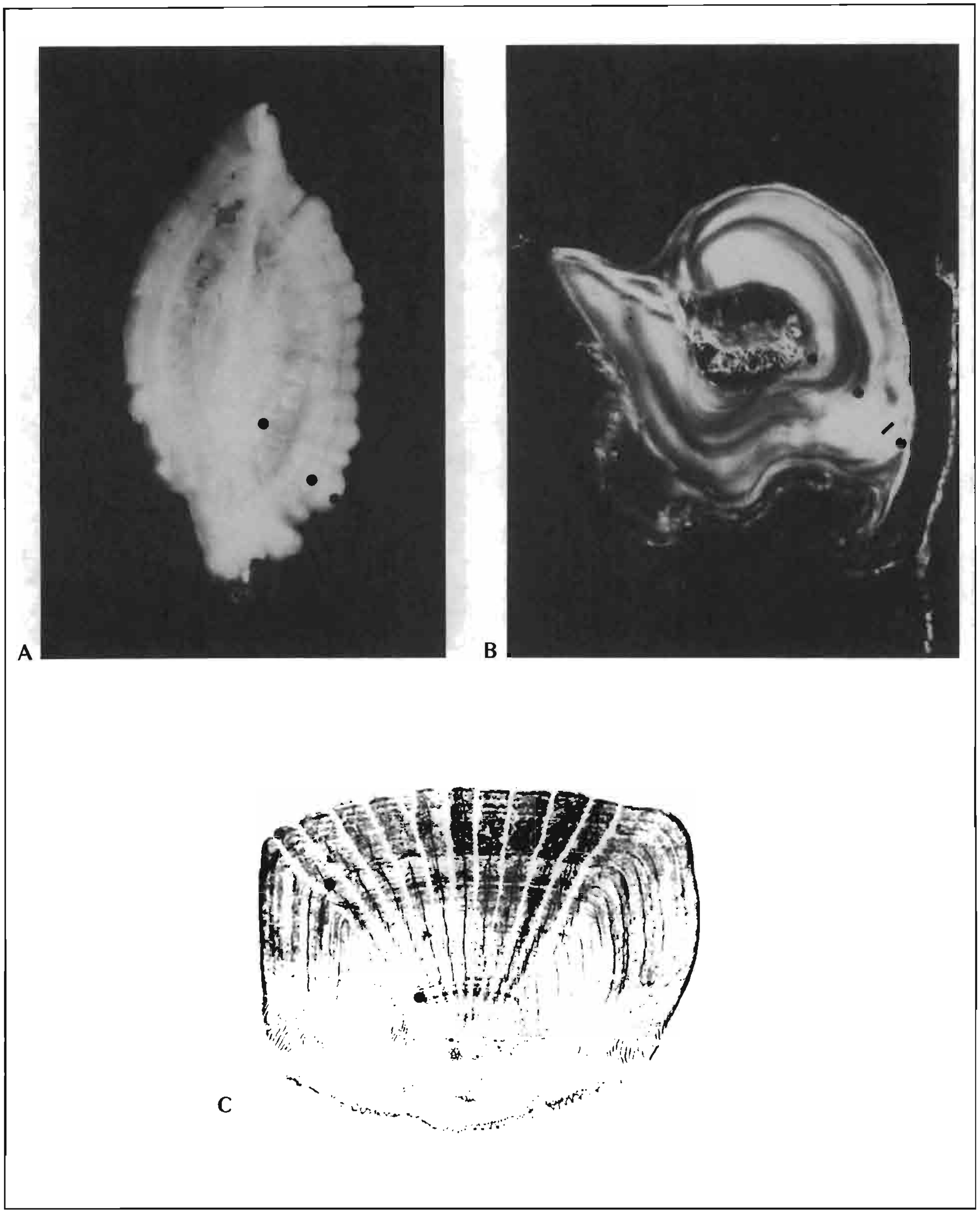

Figure 4

(A) Whole otolith of a 31-cm age-3 female black sea bass collected in June showing a narrow hyaline edge. A very thin second hyaline zone borders the second annulus. (B) Pelvic spine section showing three clear annuli (hyaline zones) including the edge of the section. Also evident is a split second annulus and weak check between the second and third annuli. (C) Pectoral scale impression showing a split second annulus and checks between the second and third annuli.

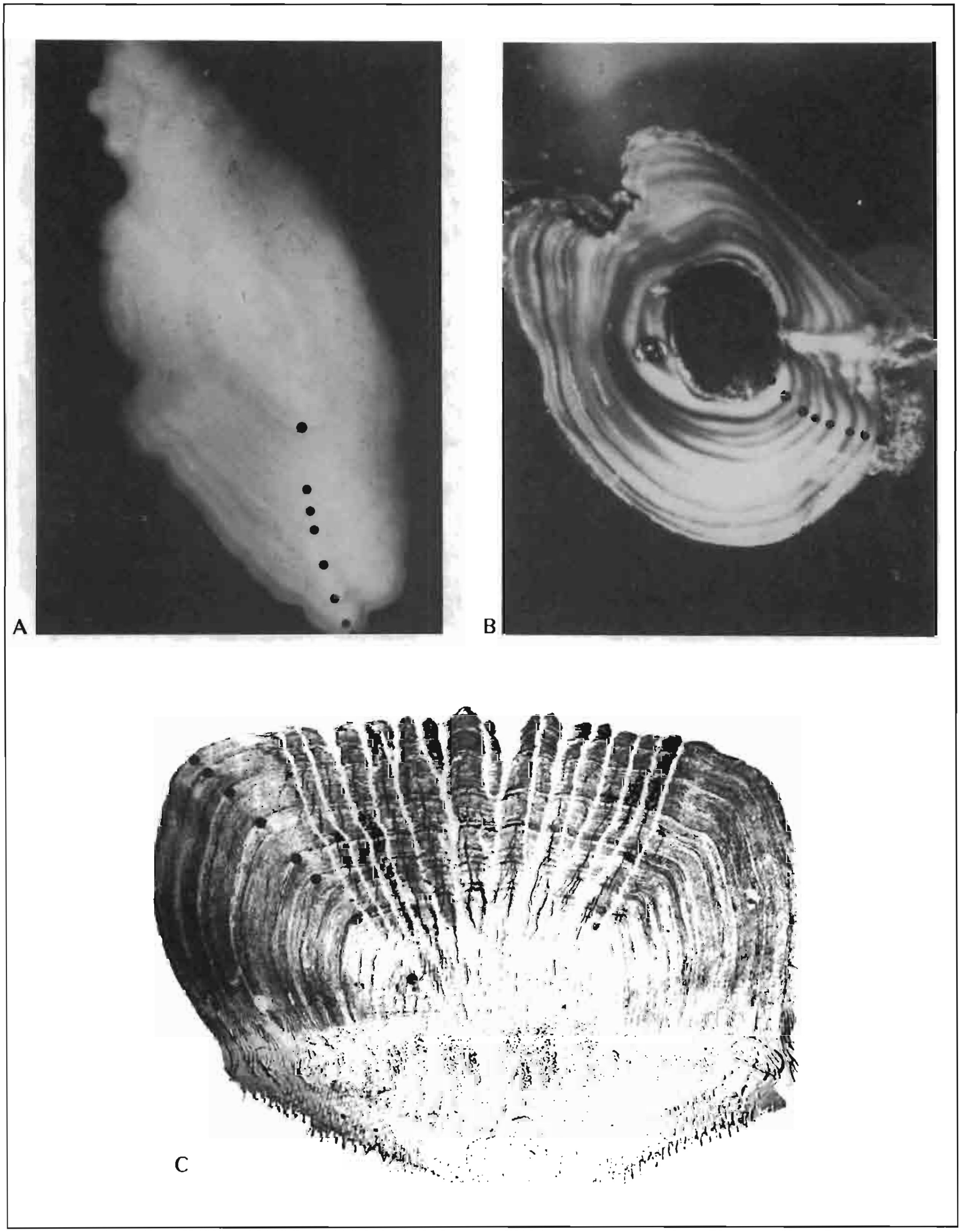

Figure 5

(A) Whole otolith of a 46-cm age-8 male black sea bass collected in February from Virginia waters showing poorly defined annuli (opaque zones). (B) Pelvic spine section showing closely spaced second, third and fourth annuli (hyaline zones). In the area beneath the lumen, the third annulus is split. (C) Pectoral scale impression showing eight clear annuli including the edge of the scale.

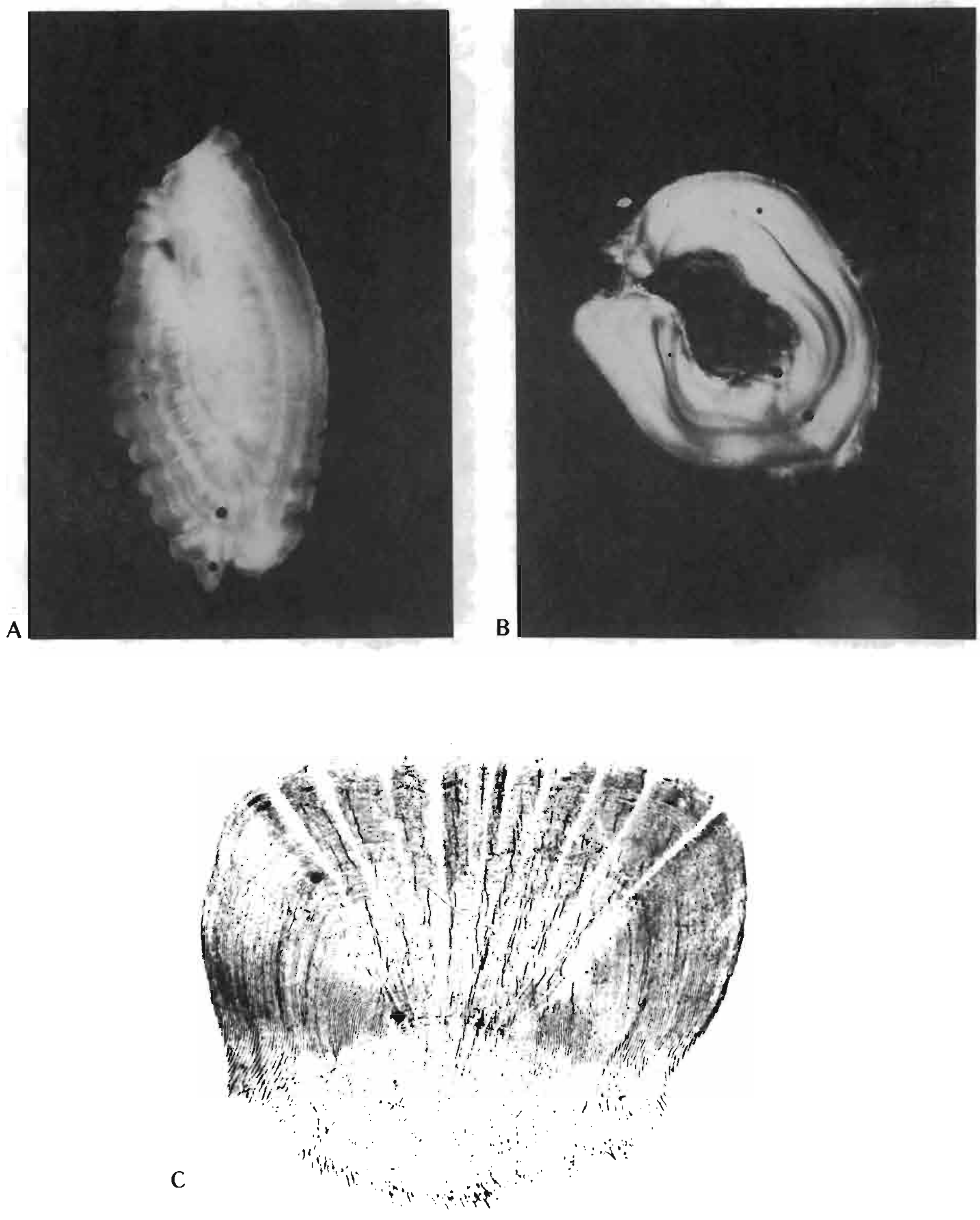

Figure 6

(A) Whole otolith of a 30-cm age 2+ female black sea bass collected in November from New Jersey waters showing a split first annulus. **(B)** Pelvic spine section showing two annuli (hyaline zones) not including the hyaline zone beginning to form on the edge. **(C)** Pectoral scale showing a discontinuous "cutting-over" mark interpreted as a check formed close to the edge of the scale.

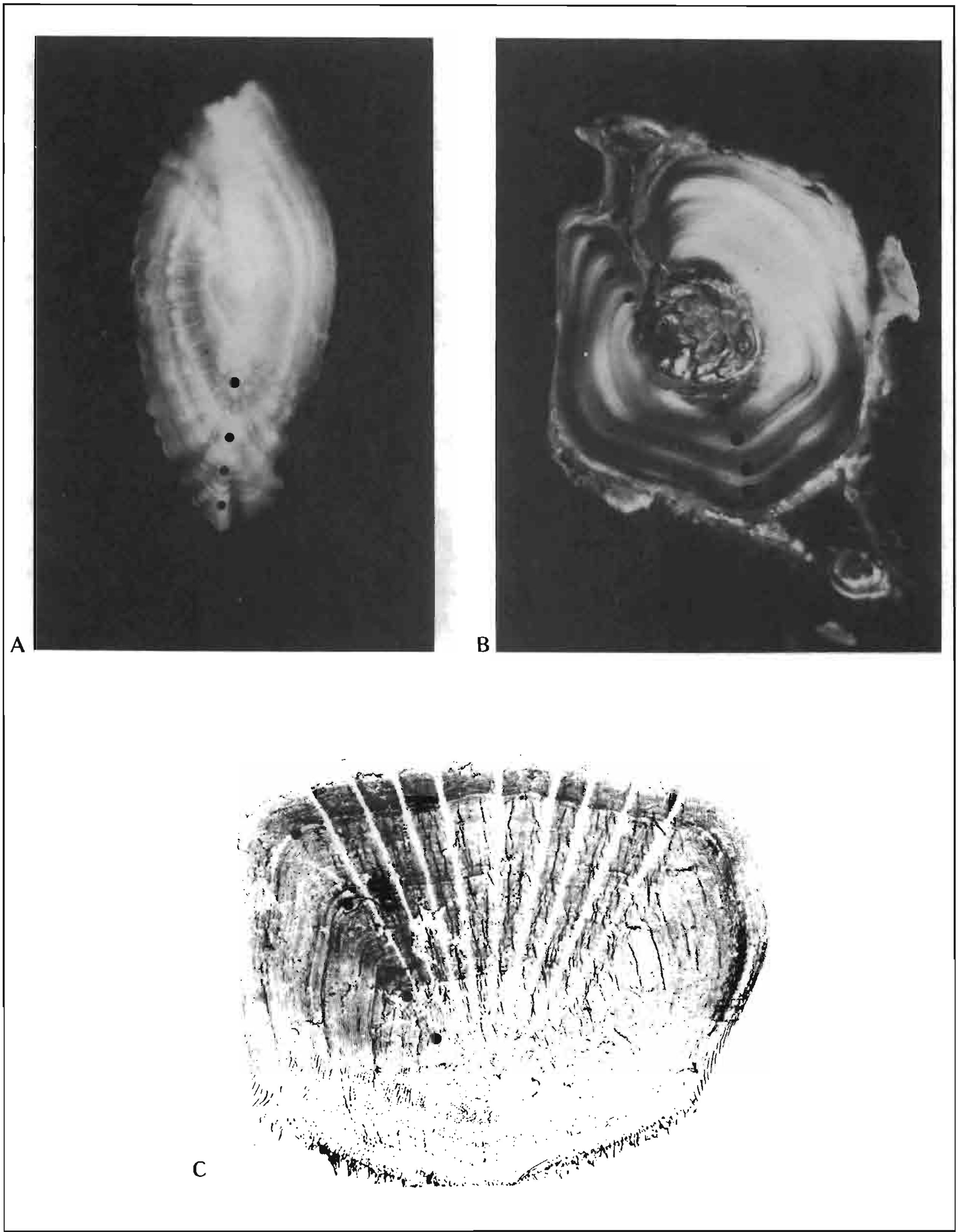

Figure 7

(A) Whole otolith of a 35-cm age 4(3)+ female black sea bass collected in November from New Jersey waters. If the (second) thin hyaline zone borders the second annulus, an interpretation of "age 4" would result. (B) Pelvic spine section showing four complete, although somewhat diffuse, annuli not including the hyaline zone near the edge. (C) Pectoral scale impression showing four annuli (not including the edge) if the first weak "cutting-over" mark is the second annulus.

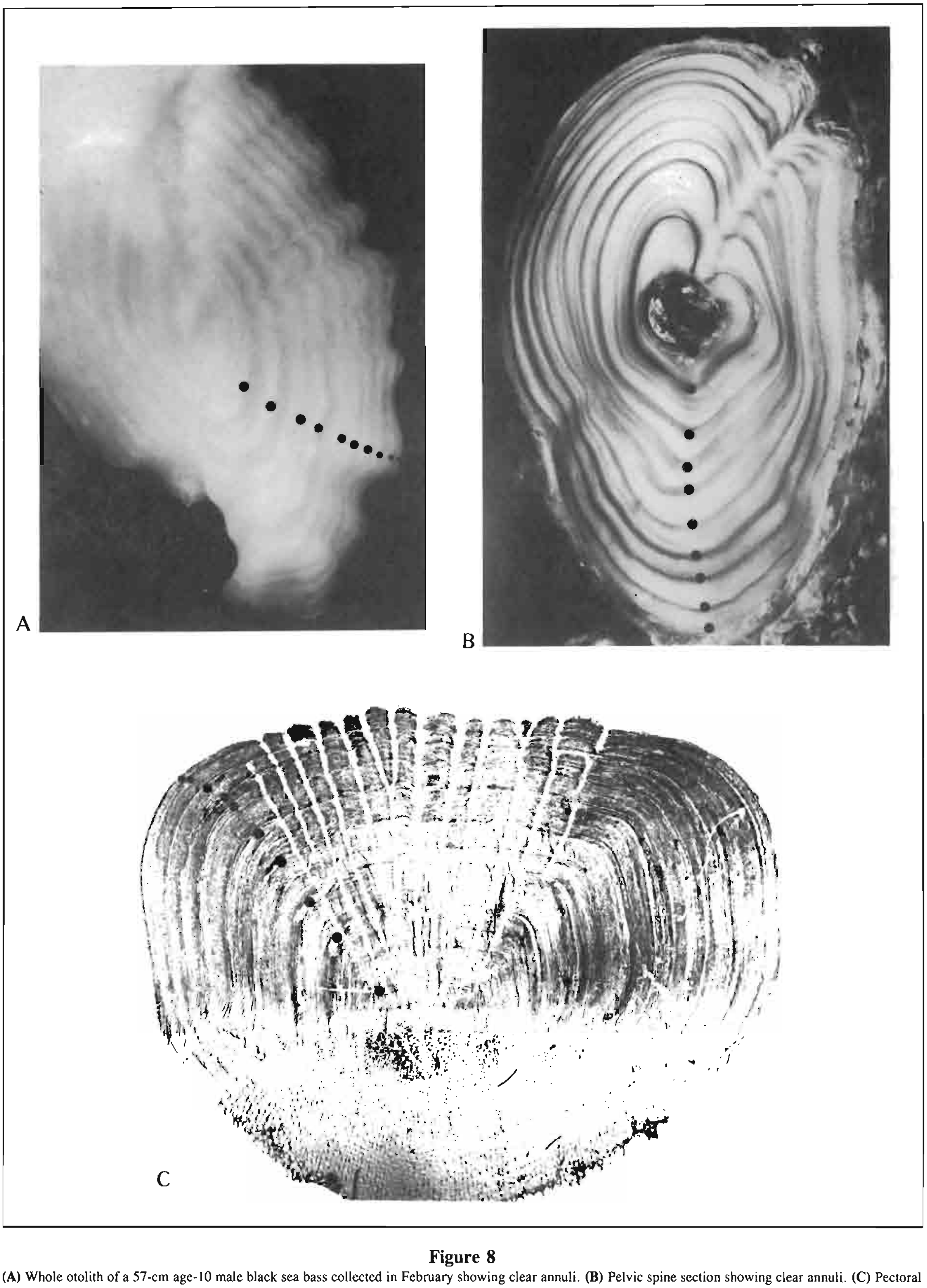

Figure 8
(A) Whole otolith of a 57-cm age-10 male black sea bass collected in February showing clear annuli. **(B)** Pelvic spine section showing clear annuli. **(C)** Pectoral scale impression showing clear annuli.

11

Weakfish
Cynoscion regalis

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Weakfish, or gray seatrout, is a sciaenid species indigenous to eastern United States coastal waters, where it ranges from Cape Cod to Florida. Seasonal migrations consist of northward movement along the coast during the spring followed by a return migration in autumn to overwinter in warmer, southern waters (Wilk 1979). Spawning takes place primarily from April to July (Mercer 1983). Ages of up to 12 years and maximum sizes up to 95 cm have been reported (Shepherd and Grimes 1983), with females generally larger at age than males.

Weakfish growth is variable, depending in part on the location sampled and the growth peculiar to a particular year-class. Pronounced variation in growth over the last 60 years has been observed. Size at age 3 from the same general locality has varied between studies by 15 cm (Mercer 1983). Differences may be due to density-dependent growth, mixing of different groups of fish, or differences in ageing techniques. It is important to keep such differences in mind to avoid the pitfalls of bias due to previously determined expectations of age-at-length.

Age studies of weakfish were initiated in 1901 when Eigenmann (1901) examined the natural history of young weakfish in southern New England. Since then, investigations have been conducted for weakfish throughout the geographic range and during different levels of population abundance (Welsh and Breder 1923, Perlmutter et al. 1956, Thomas 1971, Merriner 1973, Seagraves 1981, Shepherd and Grimes 1983). In each case, the primary ageing structure has been scales. Otoliths and vertebrae have also been used (Merriner 1973) but provided no increased information or clarity. Scales are usually more accessible from commercial and recreational landings and therefore are the age structure traditionally used for weakfish.

Age information generated from scales has been validated through comparison with modal progressions in length-frequencies (Taylor 1916, Perlmutter et al. 1956) and examination of seasonal changes in the marginal increments (Taylor 1916, Massmann 1963, Shepherd and Grimes 1983). Marginal increment analyses have shown that annulus formation occurs once a year from April to September, with the principle period between May and June (Fig. 1). Time of annulus formation coincides with migration and spawning activity, but a cause-and-effect relationship has never been verified. Information from tagging experiments has not been adequate to validate annulus formation.

Scales are traditionally removed from one of two places on weakfish. The primary location is an area between the middle of the second dorsal fin and the lateral line (Fig. 2). Perlmutter et al. (1956) chose these scales because they contain the greatest number of circuli. An alternative area for scale samples is posterior to the pectoral fin. Merriner (1973) used these scales because they are the first formed during weakfish ontogeny. Weakfish have a high number of regenerated scales and lose scales easily during the sampling process. Therefore, it may not always be possible to collect scales from the preferred location and the alternate area must be used. Both areas provide valid ages, although scales from the primary location may be easier to read. After removing scales, they are best stored dry prior to use.

Preparation of scales for age reading involves impressing the scales on laminated plastic slides. Clean, nonregenerated scales should be chosen. The thickness of the laminate should be enough to accommodate the relatively thick scales of large weakfish. If too thin a plastic is used, information on the thinner anterior edge may be lost. Scale impressions are generally examined on a standard microprojector at a magnification of 32×, although this can be modified depending on the scale size.

Weakfish scales are of the ctenoid type, and have a rather short and wide configuration (Fig. 3). Scales are characterized by distinct radii emanating from the focus to the anterior edge, and prominent circuli in the lateral fields. Annual marks appear as thin, opaque, broken lines and are most distinct in the radii zone. These annuli are sometimes referred to as "cutting-over" zones. One distinctive feature of the annuli is the shape or lack of circuli in the thin band. Unlike some fish scales, such as haddock, the annuli appear as an abrupt stoppage or change in growth. This is followed by immediate resumption of regular growth, as opposed to a gradual change. Consequently, identifying an annulus becomes more of a "yes or no" decision rather than a "possibly."

In assigning an age to an individual fish, the researcher should be aware of advantages in using a standardized 1 January birthdate. Weakfish are somewhat unusual since the annulus is formed in late spring, at the same time that spawning occurs. If the age reader uses the mean spawning date for a birthdate, the same year-class may be assigned to 0 and 1-year-old fish. A 1 January birthdate eliminates this problem, especially if the reader assigns the correct age for samples between January and the time of annulus formation.

Checks may create some confusion for an age reader. Checks are distinguished from annuli by their appearance in the lateral field and by the relative spacing since the last annulus (Fig. 4). An incomplete annulus in the lateral field is probably a check. Also, circuli in the lateral field intersect the annulus at oblique angles, whereas the circuli are parallel to checks. A check may also create a false annulus near the focus (Fig. 5). In older fish greater than age 6 or 7, annuli may be difficult or impossible to follow into the lateral fields because of crowding. In such specimens, the appearance of a check in the anterior field will be the sole source for a decision (Fig. 6).

The focus of a weakfish scale is usually a large area lacking any clearly defined circuli. Often there is a small degree of regeneration that occurs near the focus which can be ignored. If the scales are collected too close to the lateral line, the scale may have a hole in the area of the focus. The first year of weakfish growth is generally quite rapid and total length reaches 15-25 cm. Consequently, the first annulus on the scale is relatively far from the center and usually quite distinct (Fig. 7). Scale growth during the first year is fairly constant for fish throughout the geographic range.

The second annulus may be quite close to the first, indicating a growth-rate decrease of 10-15 cm per year, but may vary somewhat depending on the origin of the sample. Fish in the northern end of the range tend to have slower annual growth than fish from southern waters and may have closely spaced first and second annuli (Fig. 8). This phenomena, which has also been noted by other researchers (R. Seagraves, Del. Div. Fish Wildl., P.O. Box 1401, Dover, DE 19903, pers. commun. April 1980) may not be consistent for each year. Nevertheless, close annuli are possible and age readers should be aware of this possible source of error. Growth between the second and third annuli varies, with the third annulus often relatively close to the second (Fig. 9). The growth to the fourth annulus may be as great, or greater, than the second to third increment (Fig. 10). At ages of 6, 7, and older, annuli are harder to identify and may only be visible as a line of distorted circuli in the anterior field (Fig. 6). The frequency of older fish tends to be greatest at the northern end of the weakfish range.

With adequate preparation techniques, weakfish scales can be relatively easy to age. Annulus interpretation problems can be minimized if the sampling time and location are considered.

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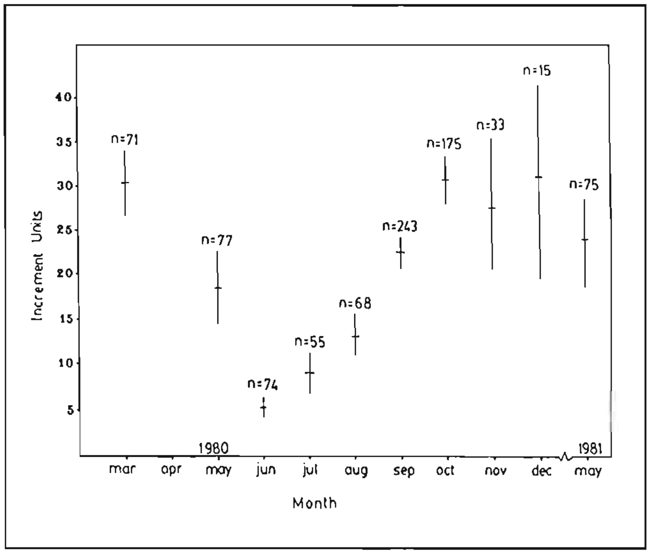

Figure 1
Mean marginal scale increments with 95% confidence intervals of weakfish for all ages combined. Sample size given for each month.

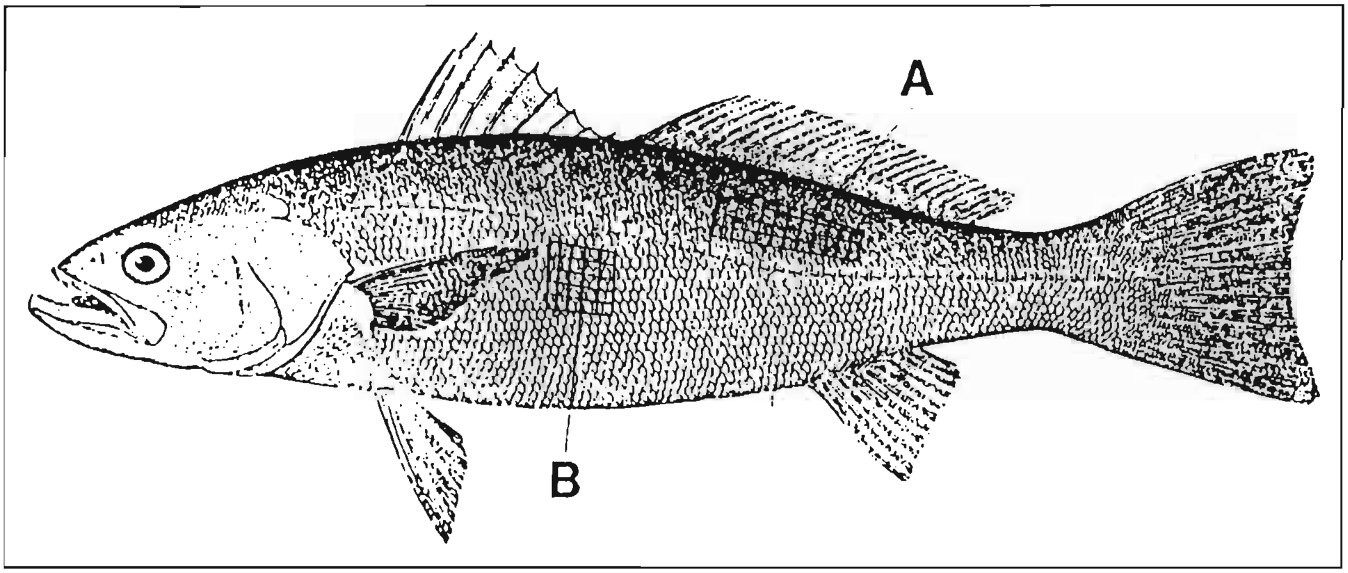

Figure 2
Weakfish, *Cynoscion regalis*, showing primary (A) and secondary (B) locations for collecting scales.

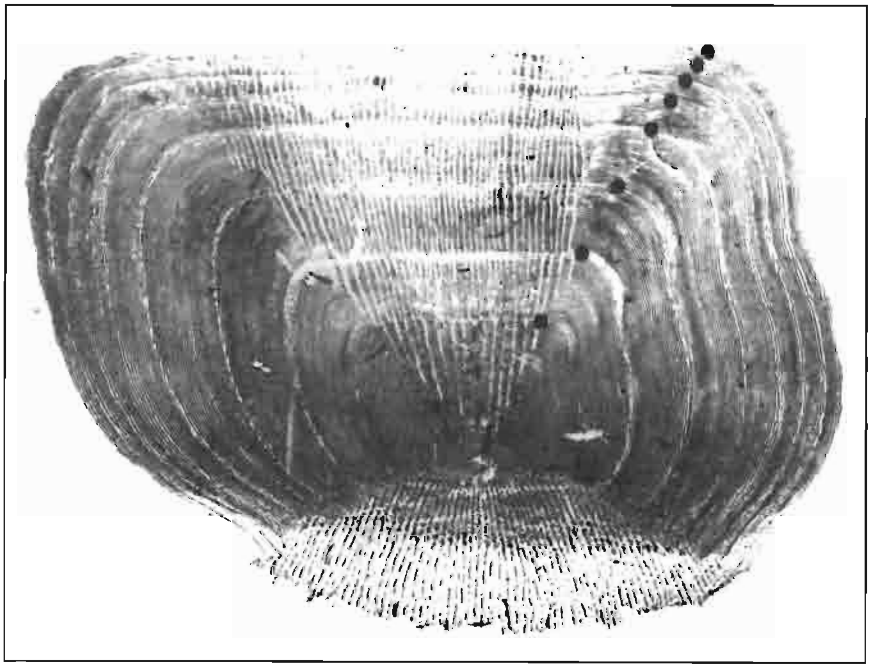

Figure 3
Scale impression from a 75-cm age-8 female weakfish showing common configuration of annuli with cutting edge.

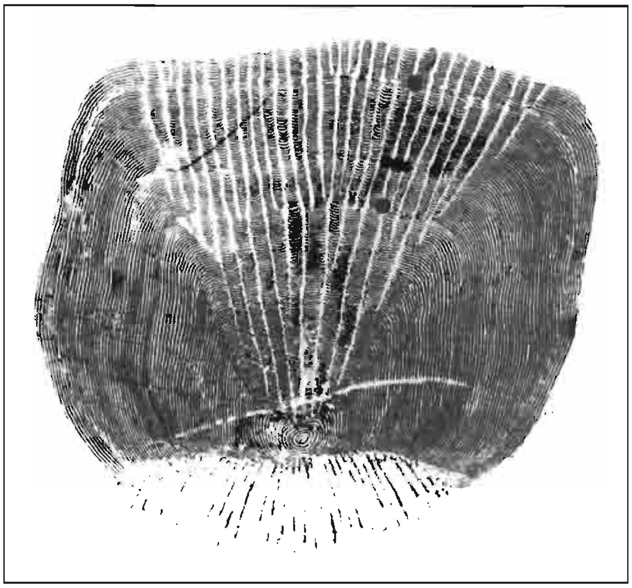

Figure 4
Scale impression from a 35-cm age-2 female weakfish showing 2 annuli and a check after first annulus.

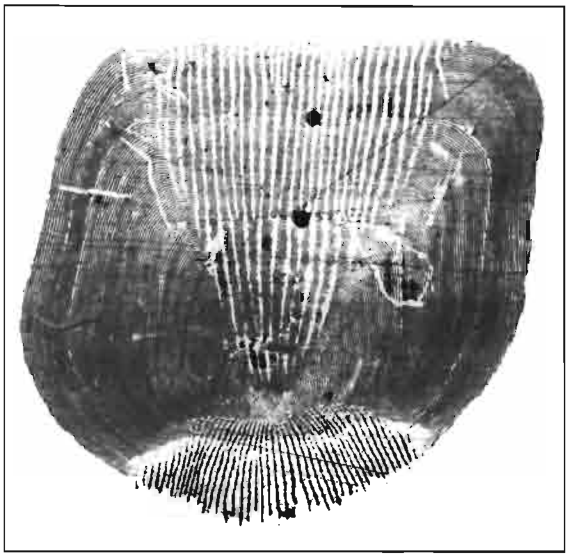

Figure 5
Scale impression from a 47-cm age-2 male weakfish showing a false annulus near the focus.

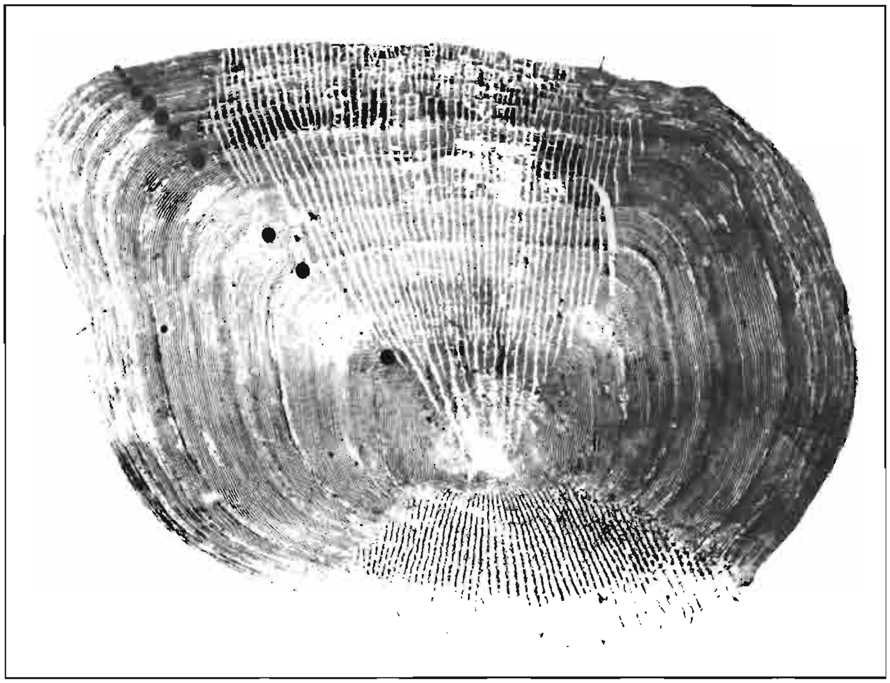

Figure 6
Scale impression from a 79-cm age-11 female weakfish showing crowding of recent annuli near the anterior edge of the scale. Last annulus on edge not yet formed.

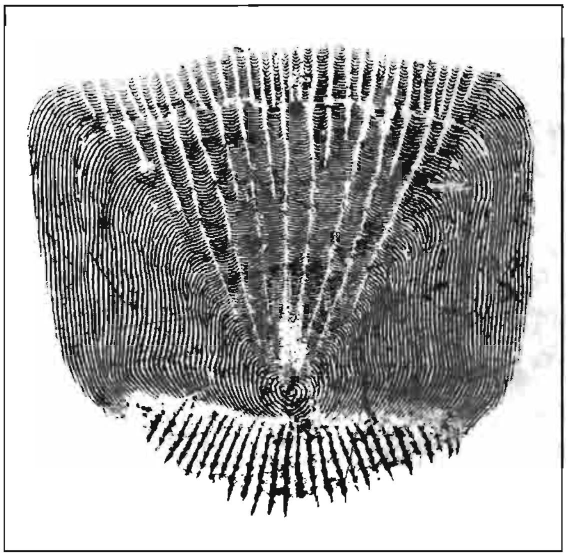

Figure 7
Scale impression from a 24-cm age-1 male weakfish showing typical configuration of the first annulus.

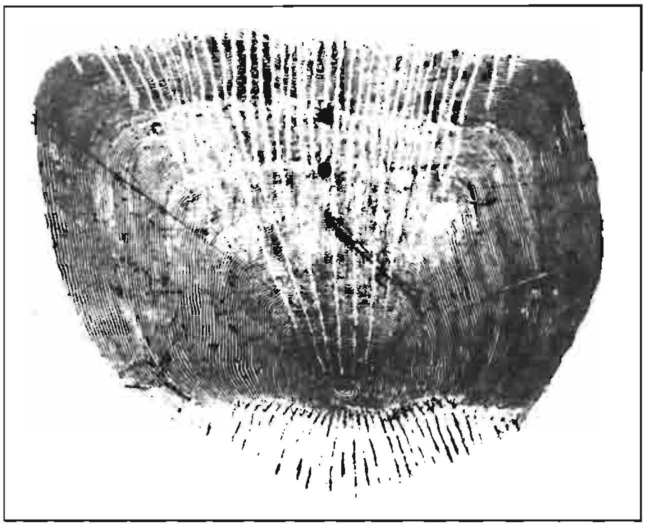

Figure 8
Scale impression from a 34-cm age-2 female weakfish showing close first and second annuli.

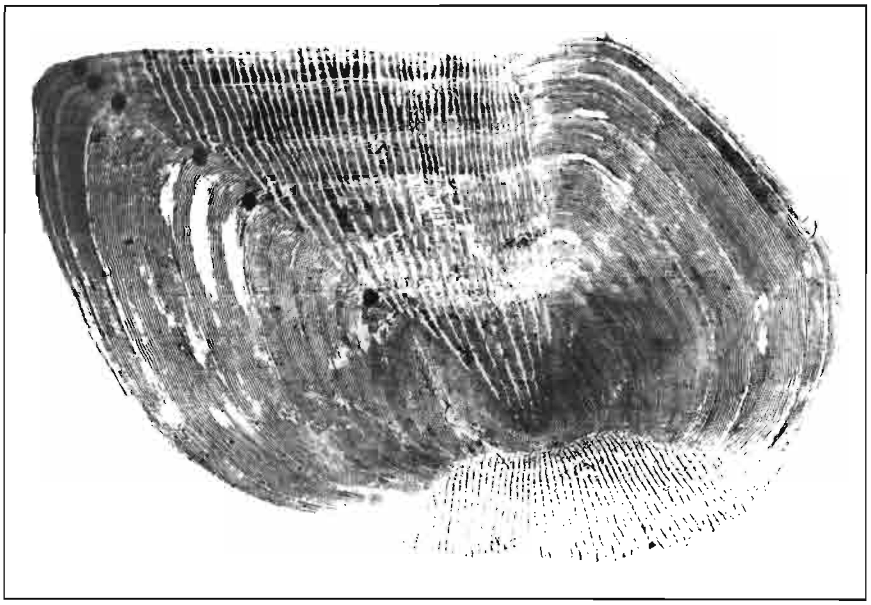

Figure 9
Scale impression from a 73-cm age-6 female weakfish showing close second and third annuli.

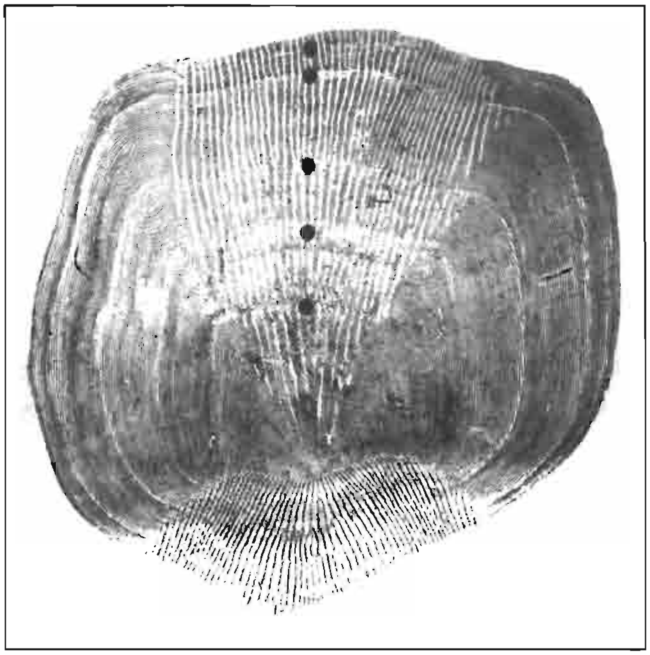

Figure 10

Scale impression from a 68-cm age-5 female weakfish showing amount of growth between the third and fourth annuli relative to growth between the second and third.

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Atlantic mackerel *Scomber scombrus*

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Atlantic mackerel is a pelagic schooling species found on both sides of the North Atlantic; in the Northwest Atlantic it occurs from Labrador to North Carolina. Males and females grow at about the same rate, reaching a maximum age of about 20 years and a maximum fork length of about 47 cm (19 inches). Most mackerel are sexually mature by age 3.

The Northwest Atlantic population of mackerel consists of two major components which follow different migratory patterns but do not appear to be genetically distinct. Mackerel of both components overwinter along the edge of the continental shelf from Cape Sable, Nova Scotia, to Cape Hatteras, North Carolina. The southern component moves inshore and northward to spawn in the Middle Atlantic Bight in spring and then move further northward into the Gulf of Maine in summertime. The northern component reaches Southern New England waters in late May and then moves northward to the Gulf of St. Lawrence where spawning occurs in June and July. Fish of both groups move southward en route to overwintering areas during the autumn (Anderson and Paciorkowski 1980).

Ageing methods for this species were first described by Steven (1952) for mackerel of the English Channel and Celtic Sea. Growth patterns on mackerel otoliths are not as complex as some other fish species, but can be difficult to interpret if viewed in water or alcohol. Several types of growth pattern anomalies, such as checks, may cause difficulty with age interpretation. In addition, older mackerel (>10 years) can be difficult to age because annuli are extremely thin and closely spaced near the edge of the otolith.

The whole otolith, mounted in clear Permount resin, is used to age this species. The resin enhances definition of the finely detailed growth patterns and provides a protective, long-term storage medium. Pairs of otoliths are mounted in circular depressions on black plastic trays distal-surface-up with the rostra of both otoliths aligned together. This enables a detailed comparison of zone formation and ring counts on each otolith. Magnification of as much as 60X under reflected light is required to distinguish annuli formed near the edge of otoliths from older mackerel.

Young-of-the-year mackerel are fast growing, usually completing about 20 cm (8 inches) of growth by the first autumn after hatching (Anderson and Paciorkowski 1980). The first annulus begins to form as early as August (Dery and Anderson 1983) and is deposited after a large amount of opaque material has formed around the nucleus. For that reason, the first strong hyaline zone after the nucleus is interpreted as the first annulus. Subsequent hyaline zones are counted as annuli. By convention, a birthdate of 1 January is used. As of this date, the hyaline zone forming on the edge of the otolith is included as an annulus until seasonal growth resumes.

Annulus formation is completed by March or April for a few young fish (ages 1-3), but for most individuals it may not be complete until May or June (Dery and Anderson 1983). Seasonal growth resumption (opaque edge) may not be apparent on older fish otoliths until late August or September. This is partly an artifact of the relatively narrow growth increment formed, which is not easily detectable when first deposited. On most otoliths, opaque edge is first evident on the tip of the rostrum rather than on the posterior edge (Fig. 1). This is because the rostrum is usually part of the longest axis of growth on the otolith, and forms wider growth increments relative to other parts of the structure.

Age interpretations should be based upon the examination of more than one part of the otolith. It is possible to interpret different numbers of annuli on the rostrum, subrostrum, and postrostrum, due to close spacing or weak formation of zones on various parts of the otolith. The rostrum usually affords the widest separation of hyaline zones unless it is truncated, with most otolith growth shifted to the posterior end of the otolith. In Figure 2, hyaline zones are well spaced and clearly defined on both the rostrum and the posterior edge of an otolith from a 41-cm, age-11 fish. Figures 3A, 3B, and 3C show different parts of an otolith from a 39-cm, age 16+ fish with a truncated rostrum. No age can be determined using the rostrum (Fig. 3A), but 16 hyaline zones can readily be interpreted on the subrostrum (Fig. 3B) and the postrostrum (Fig. 3C).

As previously mentioned, the tip of the rostrum may be more reliable for edge interpretation, and checks usually appear weaker on the rostrum than on other parts of the otolith. Generally, if the number of annuli interpreted on various axes of the otolith differ, and the cause of the difference cannot be identified as a check, split, or weakly formed zone, the "best" age may be assigned on the basis of the highest ring count. This is because ageing error for older individuals has been found to be biased toward underinterpretation of age.

On most mackerel otoliths, the first hyaline zone, representative of the first annulus, is generally well defined. A large first annulus is evident on an otolith from a 16-cm, age 1+ fish sampled in July (Fig. 4). The first hyaline zone is clear and distinct around the entire periphery of the otolith. In contrast, Figure 5 shows an otolith from a (young-of-year) age 0+ mackerel sampled in October that could be interpreted as age 1+. However, the weak incomplete hyaline zone was interpreted as a check and not an annulus.

During the second year of growth following the first annulus, a check can form on the otolith during the summer months which could lead to overinterpretation of age, especially if strongly formed on the postrostrum (Dery and Anderson 1983). If such otoliths are collected in late summer or early autumn after the check has formed they could be interpreted as age 2+ rather than 1+ (Fig. 6). Normally, the check is not continuous around the otolith, and is faint or absent on the rostrum (Fig. 7). Frequently, it appears as a diffuse stippling of hyaline material and does not form a discrete hyaline zone. In Figure 8, showing an otolith from a 26-cm, age 2+ fish sampled in July, the second hyaline zone, although weak and diffuse, is the second annulus. This zone is very strong on the rostrum, confirming it as an annulus.

On otoliths from older fish, the identity of the hyaline zone as a check is more obvious because of the relative spacing of annuli and the contrast of the check with the more strongly formed second, third, or fourth annuli. Frequently, such checks are also formed during the third or fourth summers (Fig. 9).

After formation of the third annulus, age interpretation may be complicated by an irregular spacing of annuli. In Figure 10, showing an otolith from a 42-cm age-9 fish, the third, fourth, fifth, and sixth annuli are separated by very narrow growth increments, but the increment between the sixth and seventh annuli is wide. Anomalous spacing of annuli tends to be quite typical of mackerel otoliths. On some otoliths, annuli may be spaced very close together and may seem to constitute one split annulus. If the "split" zone is traced along the pararostrum or examined on the rostrum, it may be resolved into two separate annuli. The pararostral area is therefore very important in distinguishing annuli. An otolith from an age 5+ fish has very closely spaced third and fourth annuli on the posterior edge of the otolith, but the same annuli are spaced more widely apart on the rostrum (Fig. 11).

For age-10 or older mackerel, thickening of the otolith may partially obscure the first several annuli. Outer annuli are difficult to interpret because the hyaline zones are often thin, weak, split, or closely spaced. Figures 12A and 12B show the rostrum and postrostrum of an otolith from a 42-cm mackerel which was aged as a possible 16+ after multiple age readings. Although the rostrum is well developed, the annuli are poorly formed and diffuse (Fig. 12A). On the postrostrum (Fig. 12B), several annuli are so weak and obscured by calcium that they can easily be overlooked in the age interpretation.

Contrast between hyaline and opaque zones tends to deteriorate toward the outer edge of otoliths of older mackerel. This results from a decrease in the relative amount of calcium aragonite deposited during the summer months, causing the "opaque" zones to appear more translucent. Figure 13 shows an otolith from a 40-cm age 9+ fish collected in December. All the annuli (not including the edge) can be readily interpreted on both the rostrum and postrostrum, but after the sixth annulus there is less contrast between the hyaline and opaque zones. Figures 14A and 14B are more extreme examples, showing the rostrum and postrostrum of an otolith from a 42-cm mackerel. It could not be aged because of the increasing translucence of the otolith toward the edge, and because of poorly formed hyaline zones.

No significant differences in growth patterns between mackerel otoliths of the northern and southern components have thus far been established. Individual variation in the shape of otoliths and relative length and thickness of the rostrum relative to the postrostrum are considerable.

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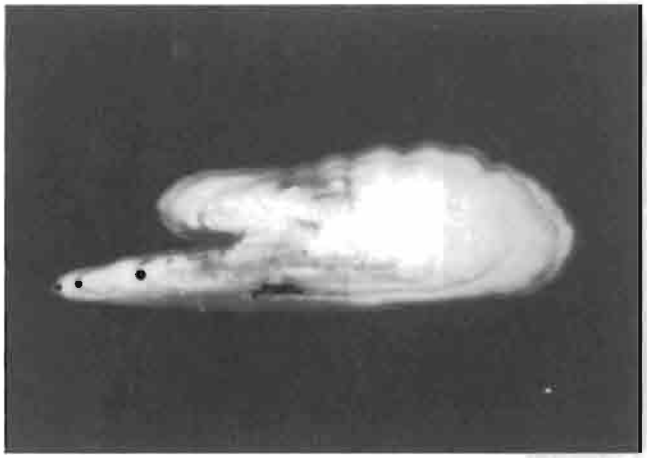

Figure 1
Whole embedded otolith of a 34-cm age 3+ Atlantic mackerel collected in August showing a tiny amount of opaque material visible on the tip of the rostrum.

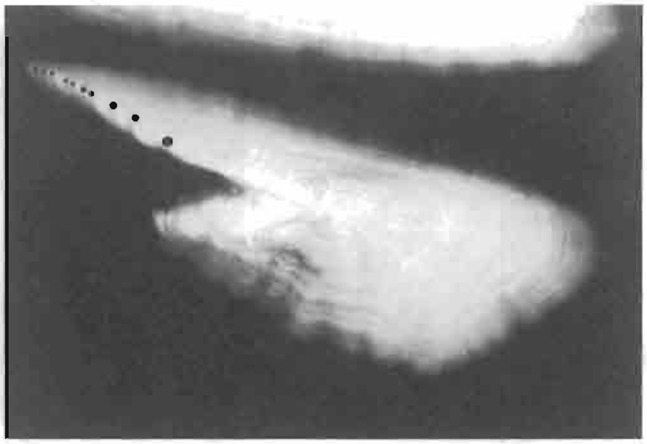

Figure 2
Whole embedded otolith of a 41-cm age-11 Atlantic mackerel collected in July showing clearly defined annuli and persistent hyaline edge.

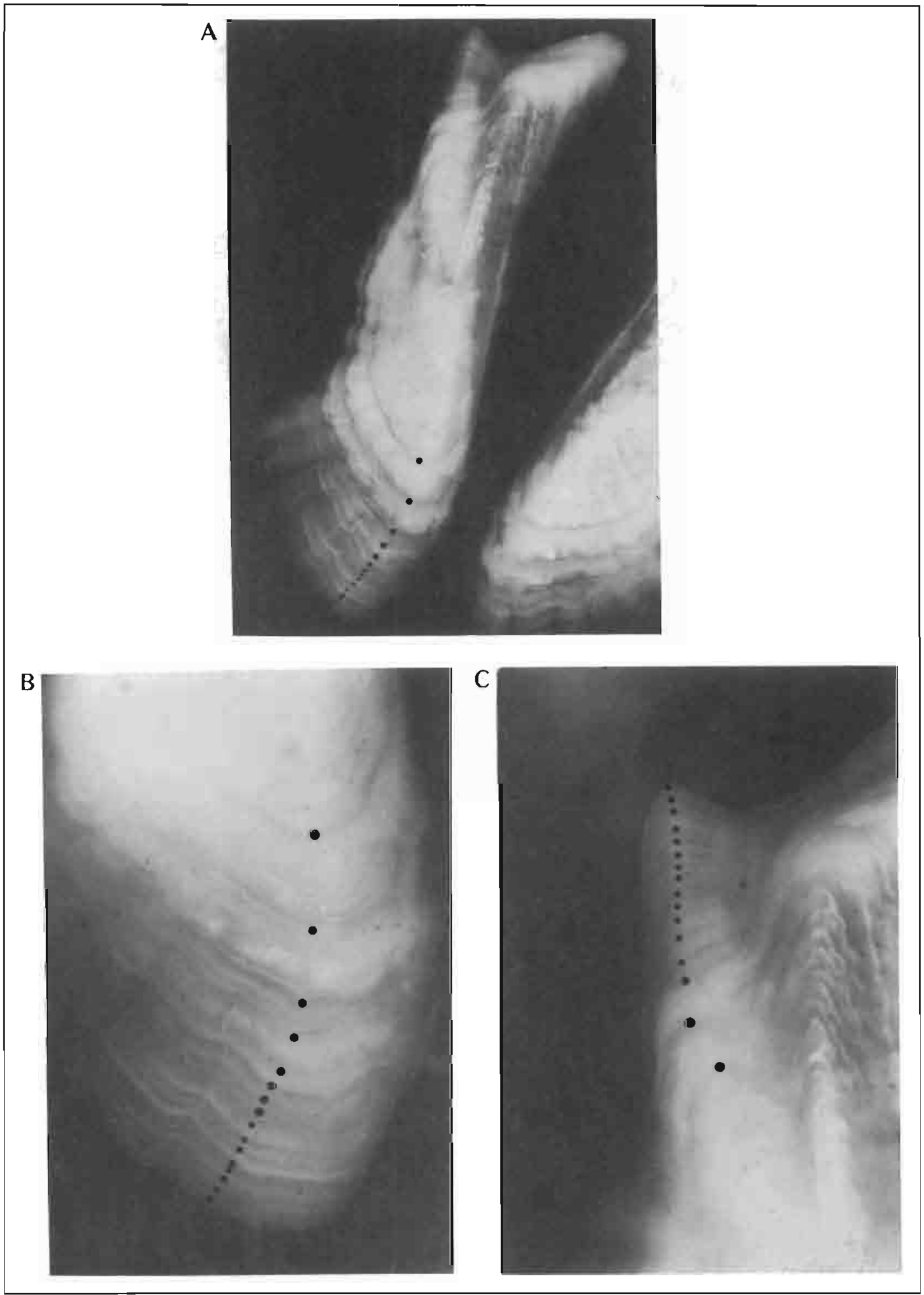

Figure 3

(A) Whole embedded otolith of a 39-cm age 16+ Atlantic mackerel collected in August with a truncated rostrum, unsuitable for age interpretation. **(B)** Subrostrum showing 16 clear annuli and narrow opaque edge. **(C)** Posterior part of the otolith showing 16 clear annuli and narrow opaque edge.

Figure 4
Whole embedded otolith of a 26-cm age 1+ Atlantic mackerel collected in July showing a large first annulus.

Figure 5
Whole embedded otolith of a 20-cm age 0+ Atlantic mackerel collected in October showing a first summer check formed close to the edge of the otolith.

Figure 6
Whole embedded otolith of a 25-cm age 1+ Atlantic mackerel collected in August showing a thin summer check formed just inside the posterior edge.

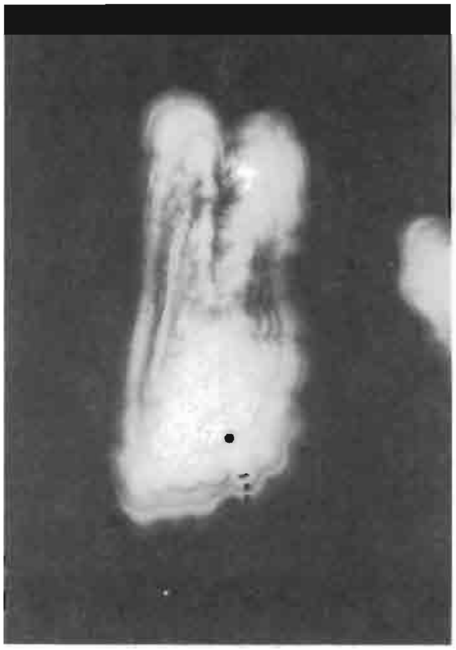

Figure 7
Whole embedded otolith of a 39-cm age 4+ Atlantic mackerel collected in December showing a weak check formed between the first and second annuli.

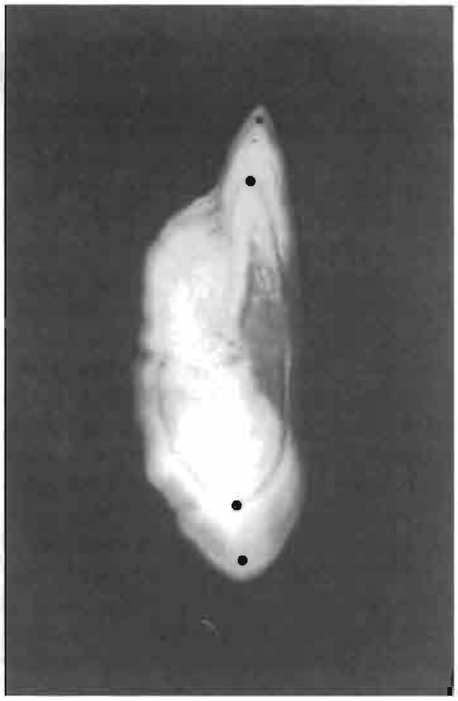

Figure 8
Whole embedded otolith of a 26-cm, age 2+ Atlantic mackerel collected in July showing a weak second annulus strongest on the rostrum.

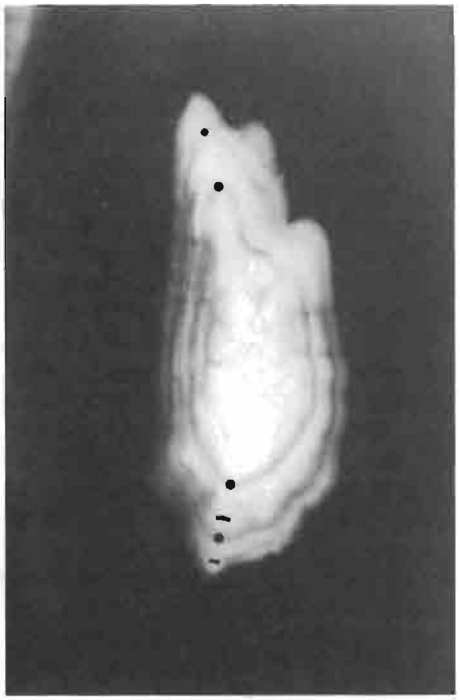

Figure 9
Whole embedded otolith of a 32-cm age 2+ Atlantic mackerel collected in November showing a small first annulus and second and third summer checks.

Figure 10
Whole embedded otolith of a 42-cm age-9 Atlantic mackerel collected in August showing persistent hyaline edge. Annuli 3-6 are spaced closely together.

Figure 11
Whole embedded otolith of a 41-cm age 5+ Atlantic mackerel collected in December showing the third and fourth annuli spaced closely together on the posterior part of the otolith, but spaced more widely apart on the rostrum.

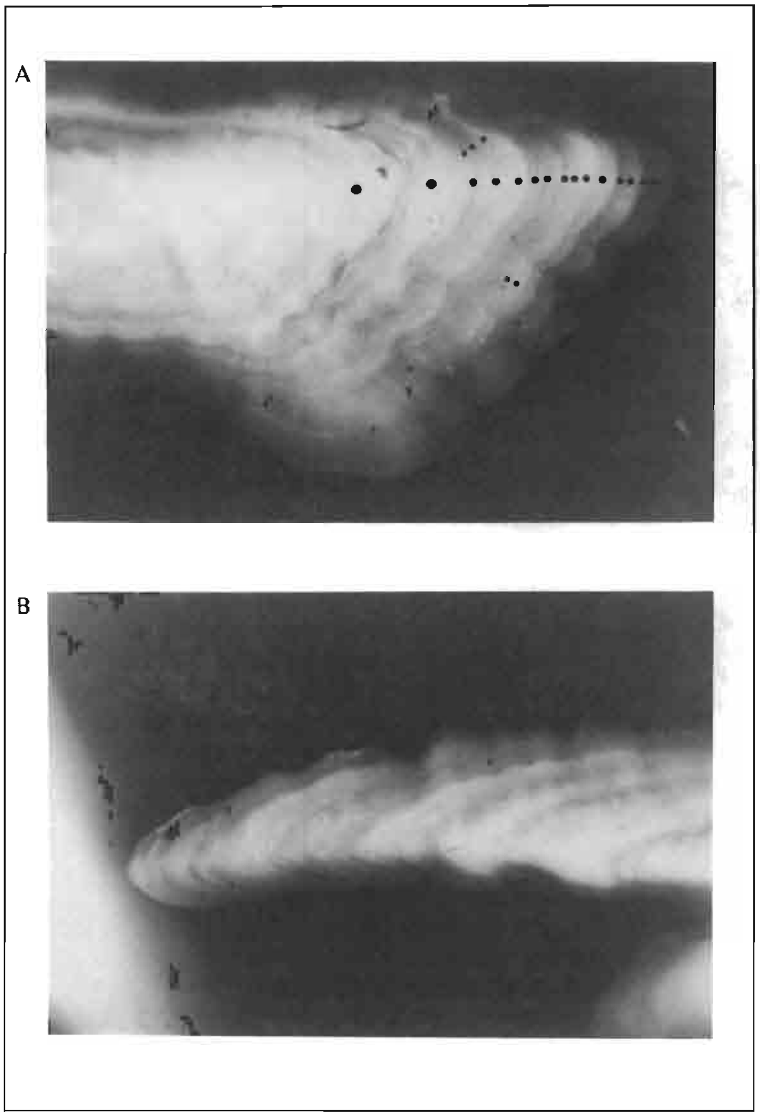

Figure 12

(A) Whole embedded otolith rostrum of a 42-cm age 16+ Atlantic mackerel collected in August showing weak, diffuse annuli. **(B)** Postrostrum showing a second summer check and split twelfth annulus. Calcium overgrowth obscures annuli 3-5, 9, 10 and 13.

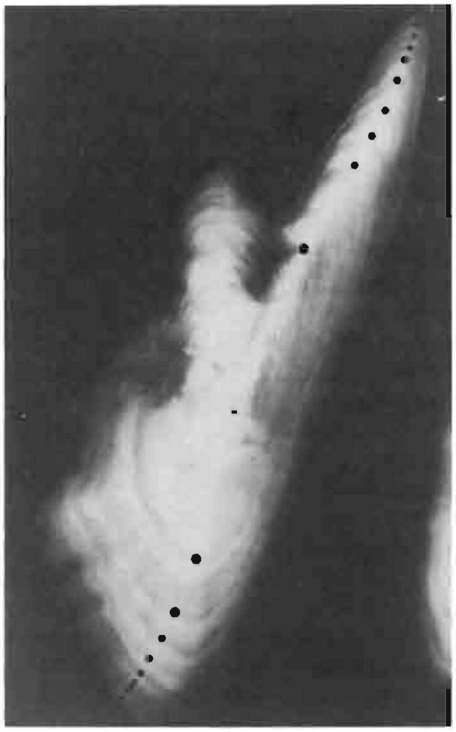

Figure 13

Whole embedded otolith of a 40-cm age 9+ Atlantic mackerel collected in December showing clear annuli, but decreased opaque/hyaline zone contrast after the sixth annulus.

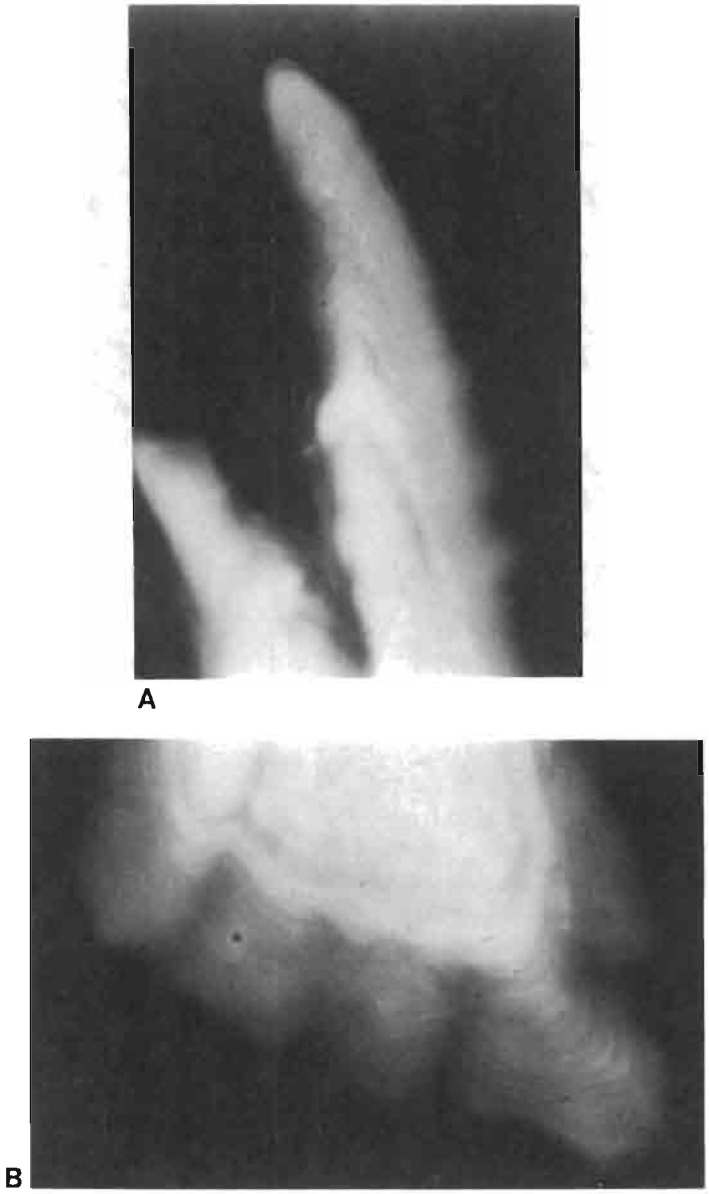

Figure 14

(A) Whole embedded otolith rostrum of a 42-cm age ? Atlantic mackerel collected in January showing very poor annulus definition due to poor calcification. (B) Postrostrum of the otolith showing poor annulus definition due to poor calcification.

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Butterfish
Peprilus triacanthus

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Butterfish is a small, semipelagic schooling species of commercial importance from Southern New England to Cape Hatteras, although it has been reported from Nova Scotia south to deep waters off Florida (Nichols and Breder 1927, Bigelow and Schroeder 1953). Butterfish overwinter along the 183-m (100-fathom) contour of the continental shelf from late autumn through early spring. North of Cape Hatteras, these fish begin to disperse over the shelf in April or May, moving inshore and northward with increasing water temperatures (Horn 1970, Waring 1975). South of Cape Hatteras seasonal inshore/offshore migrations are not thought to be significant (Caldwell 1961, Horn 1970). Spawning occurs from May through October, reaching a peak in July and August (Colton et al. 1979).

Butterfish are fast-growing and short-lived, attaining lengths of up to 24-25 cm (9-10 inches) and a maximum age of 4 although a few fish may reach age 5 or 6. Females are somewhat faster growing than males. Many butterfish are sexually mature by age 1; all are mature by age 2 (DuPaul and McEachran 1973).

Previous investigators (DuPaul and McEachran 1973, Kawahara 1978) have validated hyaline zones as annuli using whole otoliths, but did not describe growth patterns in detail. Although this species is short-lived, growth patterns on some butterfish otoliths are quite complex. Annuli may be very difficult to identify due to formation of checks within opaque zones and split or diffuse hyaline zones.

Whole otoliths are used at the Woods Hole Laboratory and are stored dry because storage in alcohol or glycerin tends to weaken contrast between the hyaline and opaque zones. Otoliths are examined by viewing the distal surface in ethyl alcohol against a dark background using reflected light at a magnification of 15×.

Most butterfish, except those that hatch late, complete at least half of their growth by age 1 (Waring 1975). Therefore, the first annulus, completely formed by the end of the first spring after hatching, is some distance away from the nucleus of the otolith. By convention, a birthdate of 1 January is used. As of this date, the hyaline zone evident on the edge of the otolith until spring growth resumption is interpreted as an annulus. Due to an overgrowth of calcium, the nucleus is seldom visible on the otolith after the fish has attained 4 or 5 cm in length. Larger young-of-the-year (YOY) fish, age 0+, complete seasonal growth by early autumn, judging by the appearance of hyaline material on the edge of the otolith. Smaller late-hatched fish appear to continue growing through autumn and perhaps winter, because opaque edge is still evident by early spring. In general, the time of annulus formation for butterfish is mid-autumn through late spring. This may vary for different age groups as older fish tend to begin and end seasonal growth slightly later in the season.

For butterfish sampled from the waters of the Gulf of Maine to Cape Fear, two types of otolith growth patterns may be identified, though not clearly differentiated. The "offshore" pattern is characteristic of butterfish sampled in waters deeper than 27 m, although such otoliths are also found in specimens taken inshore in summer and autumn. The "inshore" type of otolith growth pattern is characteristic of specimens collected at depths of less than 27 m, especially from the New York Bight south to Cape Fear. Infrequently, otoliths with the inshore pattern will be noted among offshore samples collected during the overwintering period. This distribution of inshore and offshore growth patterns has been stable from year to year among NEFC research and commercial samples.

Otoliths with the offshore growth pattern are predominant in survey and commercial catches, and tend to exhibit clearly defined annular zones. Checks may be prominent on these otoliths but can easily be distinguished from annular zones, and do not normally complicate age interpretation. The otoliths are usually well calcified, with good contrast between the hyaline and opaque zones. They are somewhat elongate in shape and the posterior edge is squared in outline.

Figures 1 to 12 illustrate variations in the offshore pattern. During the summer months, otoliths of young-of-the-year fish exhibit opaque edge, indicating vigorous growth (Fig. 1). By September or October, hyaline edge begins to form, especially on otoliths of age 0+ individuals. The initial deposition of hyaline material often appears as a closely spaced series of thin hyaline rings (Fig. 2); continuous hyaline edge may form during the winter months. If this "split" zone (intermittent deposition of hyaline material) is composed of two apparently distinct but closely spaced hyaline zones, the first zone may be misidentified as a separate annulus, resulting in overestimation of age (Fig. 3).

Checks formed before the first annulus are characteristic of offshore otoliths, but because they contrast with more prominent annular zones they are usually not difficult to differentiate from annuli (Compare Figures 2, 3, and 4). Figure 5 shows an otolith from a 13-cm, age-1 fish with three checks formed before the first annulus on the edge. These are thin, superficial, and/or discontinuous. In Figure 6, however, the hyaline zone near the center of the otolith of a 10-cm fish may represent the first annulus of a late-hatched fish. The zone is narrow but deeply formed and continuous around the periphery of the otolith.

Subsequent to the first annulus, growth increments (opaque zones) narrow considerably in width. If the first annulus is small, however, growth compensation may result in relatively wide increments (Figs. 6 and 7). In general, growth increments subsequent to the first annulus are larger for more northerly sampled butterfish with faster growth after age 1. Otoliths with very narrow increments between annuli may be difficult to distinguish from otoliths with split annuli (Fig. 3). In addition, individual differences in timing of increment/annulus formation can cause considerable indecision in age interpretation. For example, Figures 8 and 9 show otoliths from fish of similar sizes that were sampled in October. The second summer-growth increment including winter (hyaline) edge is easily recognizable in Figure 8; in Figure 9, however, the growth increment after the first annulus is barely distinguishable, probably due to retarded seasonal growth. In general, a "split" annulus may be distinguished from an annulus close to the edge (due to retarded seasonal growth) by the strength and width of the hyaline zone near the edge. If this zone is thin and/or weak, a split annulus is indicated; if strong and/or wide, the zone may be interpreted as an annulus followed by a narrow growth increment.

Growth patterns become easier to recognize after several annuli have formed. Distinct patterns of check or annulus formation may be repeated on otoliths of individual fish and relative spacing of hyaline zones becomes easier to evaluate. Figure 10, for example, shows an otolith from an 18-cm, age 2+ fish. Both the first and second annuli are weak diffuse zones, but the first annulus can be distinguished laterally on the otolith where the hyaline rings comprising the zone are compacted together.

Figures 4, 7, 11, and 12 show growth patterns characteristic of adult offshore butterfish otoliths. Note especially the otolith in Figure 11. Here, a strong check or split is formed after the first annulus. This is a frequently occurring anomaly and could be confused with an annulus, but the zone is relatively weak in the rostral and subrostral area and closely spaced with the first annulus.

Otoliths exhibiting an inshore growth pattern are typically difficult to age. This pattern involves numerous checks and diffuse annuli. Many of these otoliths have a generally rounder outline than is characteristic of the offshore type, and are often poorly calcified. Figure 13 shows an age 1+ otolith from a 12-cm fish where the increment after the first annulus contains very little opaque material. Otoliths are sometimes so poorly calcified that they are impossible to age, the amount of calcium being insufficient to define annuli (see Figure 14). It is possible that calcification may have been disrupted as these structures were formed, or later resorption removed opaque material. The lack of adequate calcium reduces hyaline/opaque zone contrast making it difficult to distinguish checks from annuli. Otoliths sampled from the shoal waters off Maryland south to Cape Fear are most problematic in this respect.

Figure 15 shows an otolith from a 4-cm YOY fish which has formed a check but is still growing actively in October, judging from the presence of opaque edge. Many larger age 0+ fish are difficult to distinguish from small age 1+ fish if strong checks have formed on the otolith. In Figure 16, numerous checks are present on the otolith, but the obvious contrast of these checks with the stronger first annulus indicates an age-2 individual, since the edge is included in the age. In Figure 17, however, it is difficult to interpret any one of the hyaline zones as an annulus; this 10-cm fish could be age interpreted as 0+ or age 1+.

In addition to problems with checks, annuli of inshore otoliths are frequently split into multiple rings and are not well defined. Figure 18, showing a 9-cm, age 1+ fish, is an extreme example of this type, but the zone of split rings is strong enough to be identified as an annulus.

Figures 19 through 22 are examples of adult inshore butterfish otoliths with complex growth patterns. On such otoliths, it is necessary to search for areas where annular zones are strongest and most condensed, such as the rostrum and lateral edges.

The growth pattern phenomena described for inshore otoliths, involving numerous checks and diffuse annuli, may be correlated with environmental factors. Pannella (1974) has observed that for tropical species the incidence of check formation due to environmental influences tends to increase for fish of shoaler water habitats. These fish are exposed to greater variation and extremes of water temperature, anaerobic conditions, and tidal influences. Pannella has also observed that annular zones may be indistinct or missing due to lack of marked seasonal changes in the environment. Inshore butterfish otoliths, sampled south of the New York Bight, are undoubtedly subject to such influences.

Regarding problems with poor mineralization of inshore butterfish otoliths south from New York Bight, mechanisms controlling otolith calcification in fishes are still poorly understood, although water temperature has been cited as an important factor (Pannella 1980). Poor mineralization of otoliths occurs for a number of tropical and semitropical species. It is possible that high seasonal water temperatures in southern inshore areas are partly responsible for poor calcification. Because poor calcification of fish otoliths can cause serious difficulties with age interpretation, more research is necessary, especially concerning how otolith calcification relates to environmental variables.

Although observations of differences in growth patterns are useful in describing ageing methods used for butterfish, a more systematic study is in order before inferences can be made about their significance, especially for stock separation. Thus far, existing meristic and morphometric studies by Caldwell (1961) and Horn (1970) indicate a separate stock of butterfish, possibly a *P. triacanthus*/*P. burti* hybrid, distributed in shallow water (to 20 m) from Cape Hatteras south to Florida. No comment can be made concerning the appearance of these otoliths, since no such fish have been identified during the Cape Hatteras to Cape Fear component of our bottom trawl surveys. Waring (1986) identified five subregions of butterfish distribution based on length-frequency data and trends in abundance. Two inshore groups, north and south of Delaware Bay, were differentiated from offshore groups. However, no inferences were drawn concerning the existence of separate subpopulations in those regions.

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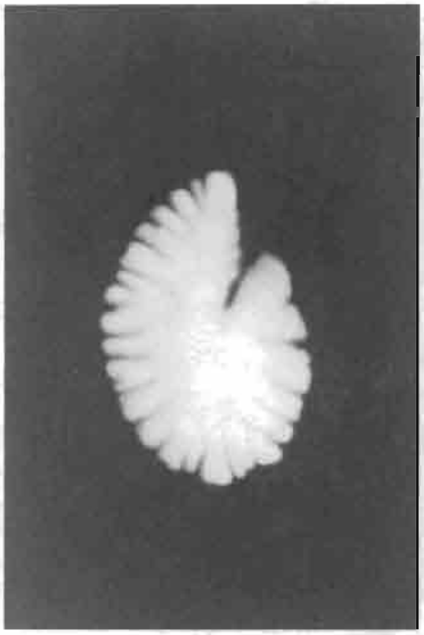

Figure 1
Whole otolith of a 6-cm age 0+ butterfish collected offshore in August showing opaque edge.

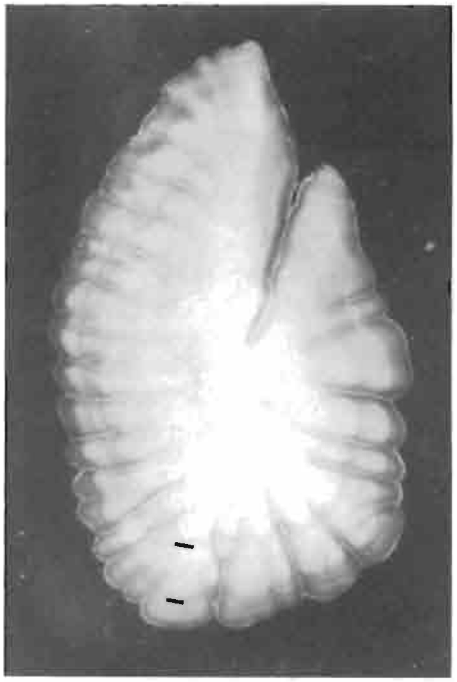

Figure 2
Whole otolith of a 10-cm age 0+ butterfish collected offshore in October showing a first summer check and split hyaline edge forming.

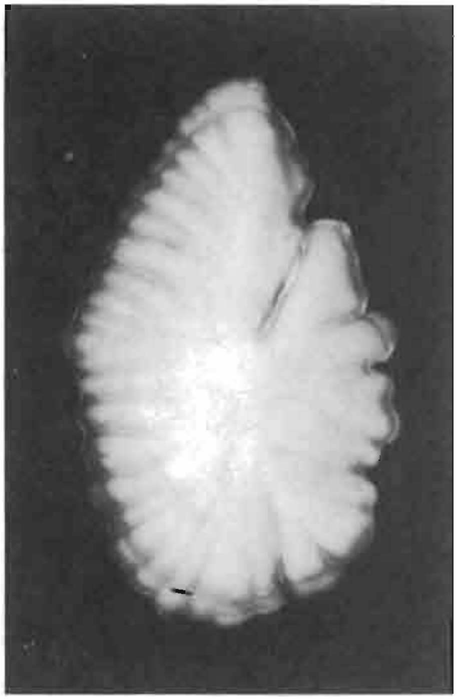

Figure 3
Whole otolith of an 8-cm age 0+ butterfish collected offshore in October showing a check or split formed just inside the (hyaline) edge.

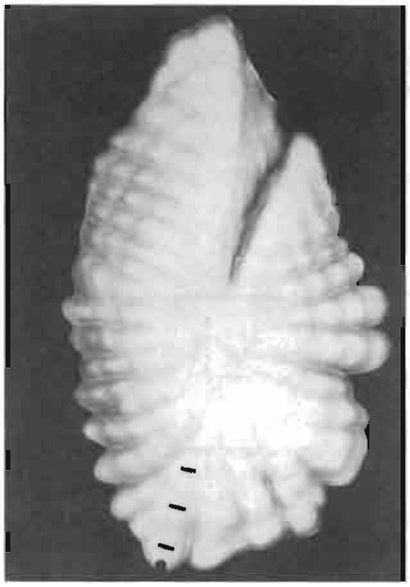

Figure 5
Whole otolith of a 13-cm age-1 butterfish collected offshore in April showing three weak superficial checks formed before the first annulus (hyaline zone) on the edge.

Figure 4
Whole otolith of a 14-cm age 1+ butterfish collected offshore in September showing strong wide annuli and a thin check between the first and second annuli.

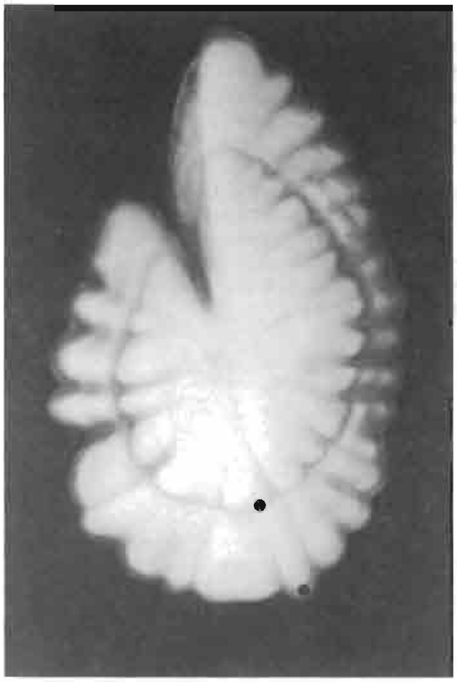

Figure 6
Whole otolith of a 10-cm age-2? butterfish collected offshore in May showing a possible small first annulus.

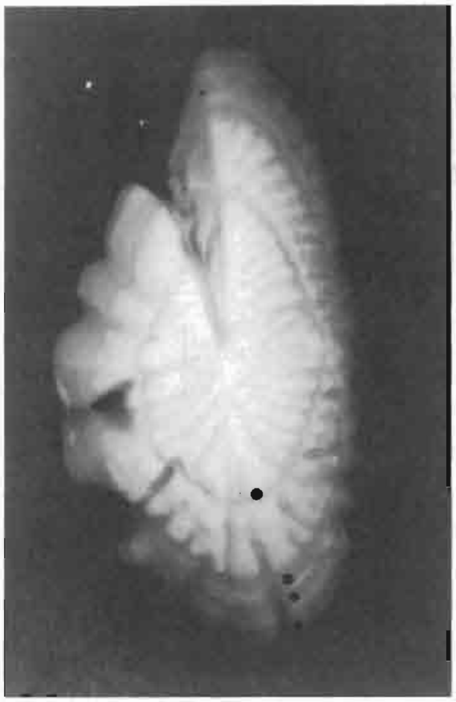

Figure 7
Whole otolith of a 21-cm age-4 butterfish collected offshore in April showing a small first annulus and weak checks between successive annuli.

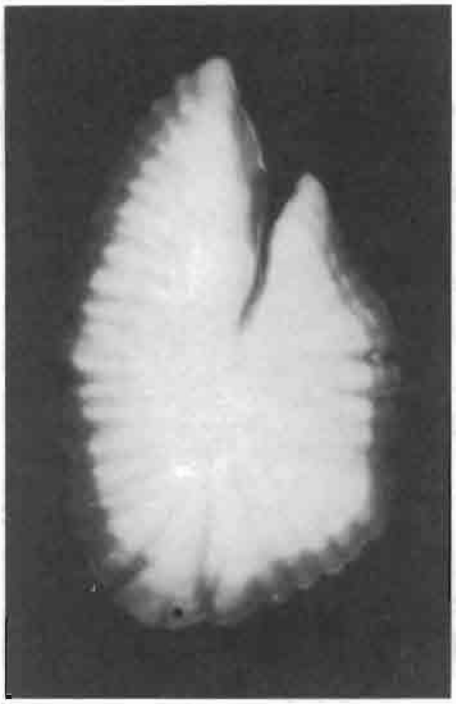

Figure 9
Whole otolith of a 14-cm age 1+ butterfish collected offshore in October showing a diffuse first annulus and unusually narrow seasonal growth increment (opaque zone) on the edge.

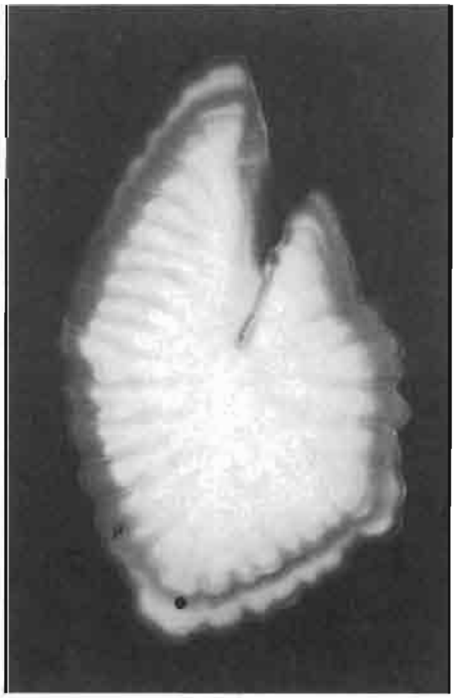

Figure 8
Whole otolith of a 15-cm age 1+ butterfish collected offshore in October showing a strong first annulus and hyaline edge.

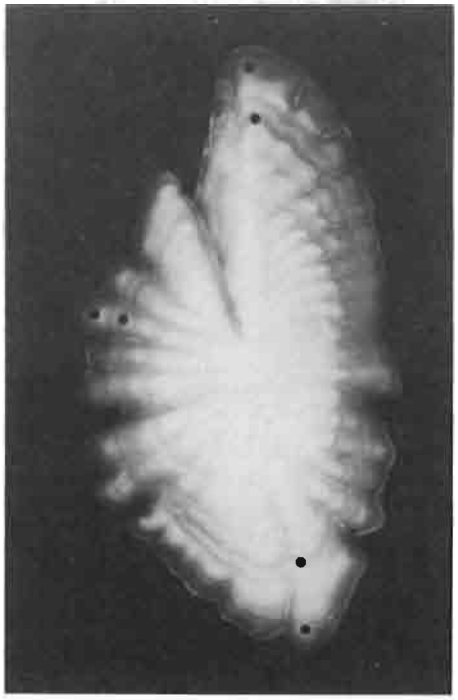

Figure 10
Whole otolith of an 18-cm age 2+ butterfish collected offshore in November showing thin first summer checks and weak, split first and second annuli.

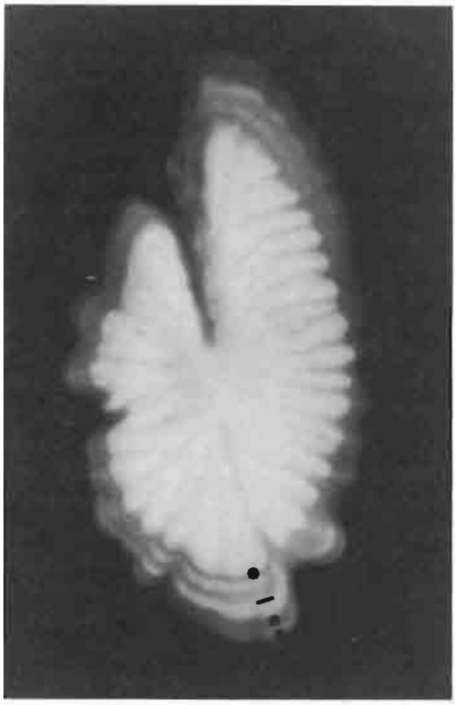

Figure 11
Whole otolith of a 19-cm age-3 butterfish collected offshore in April showing a strong check (or split) formed after the first annulus.

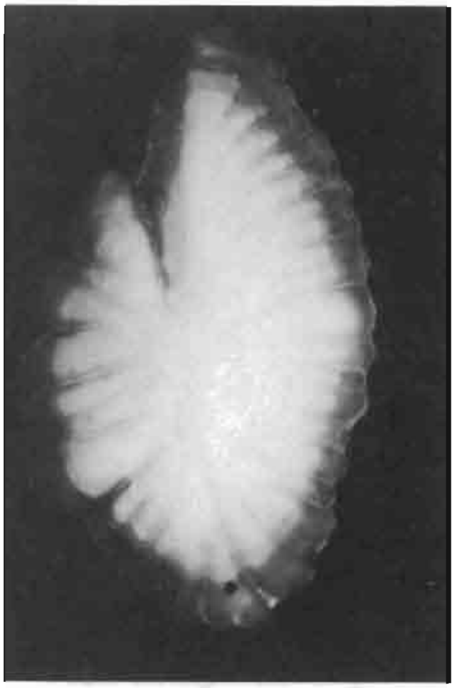

Figure 13
Whole otolith of a 12-cm age 1+ butterfish collected inshore in October showing poor calcification subsequent to the first annulus.

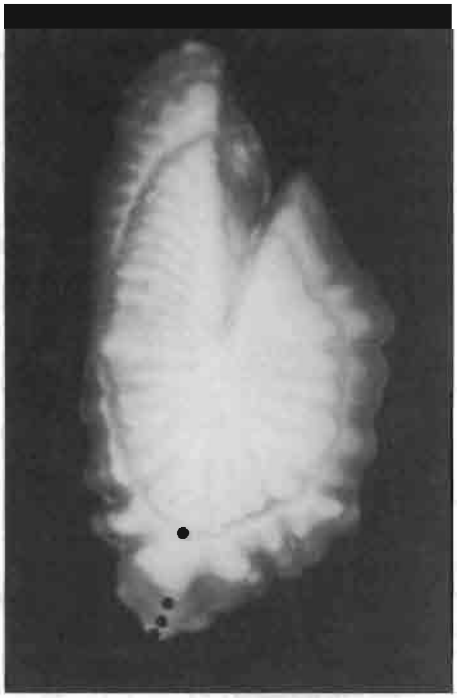

Figure 12
Whole otolith of an 18-cm age-4 butterfish collected offshore in March showing clear annuli.

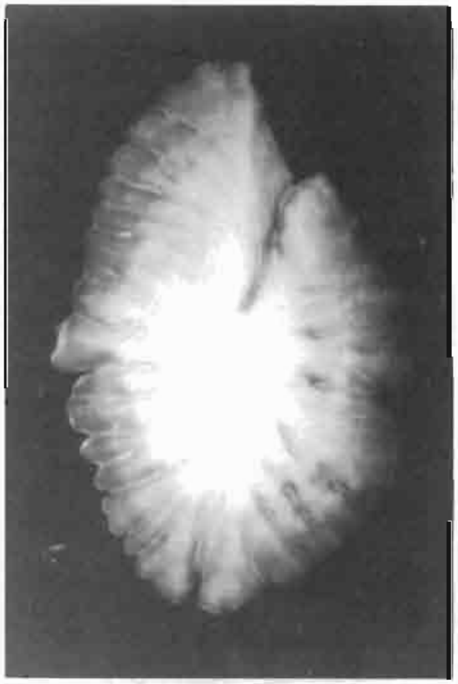

Figure 14
Whole otolith of a 16-cm age? butterfish collected inshore in October showing indistinct zones due to poor calcification.

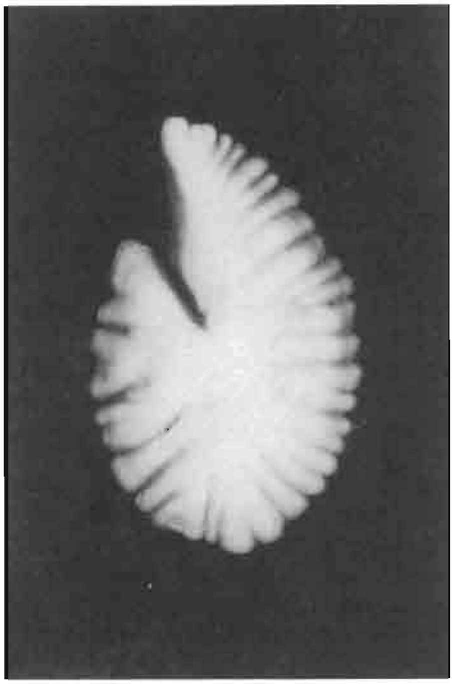

Figure 15
Whole otolith of a 7-cm age 0+ butterfish collected inshore in October showing a thin check formed inside opaque edge.

Figure 17
Whole otolith of a 10-cm age 0(1)+ butterfish collected inshore in October showing a complex growth pattern with a possible small first annulus.

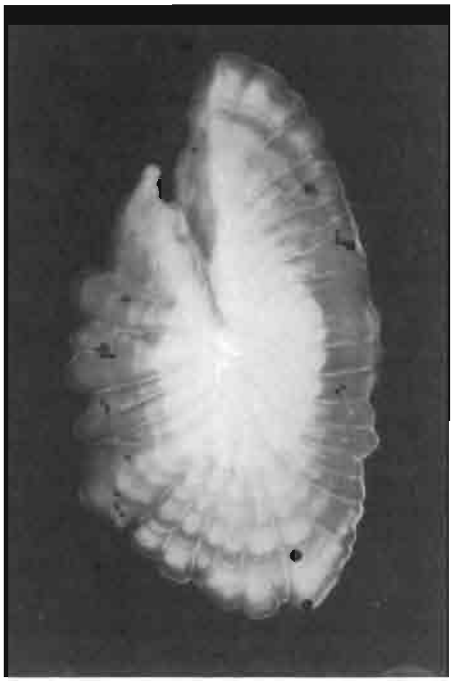

Figure 16
Whole otolith of a 15-cm age-2 butterfish collected inshore in April showing numerous first summer checks and hyaline edge.

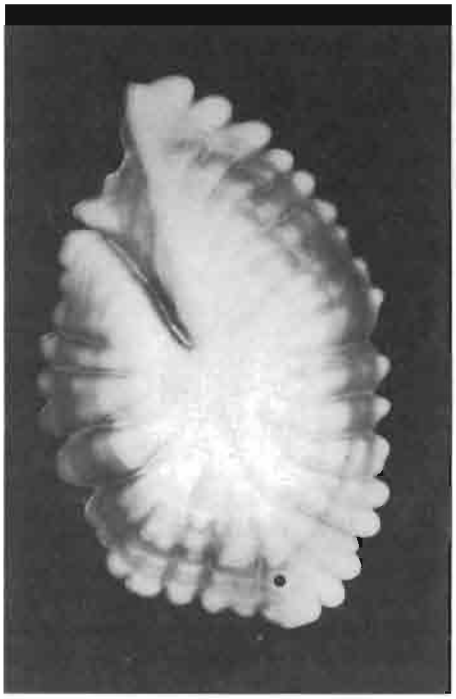

Figure 18
Whole otolith of a 9-cm age 1+ butterfish collected inshore in October showing a split first annulus.

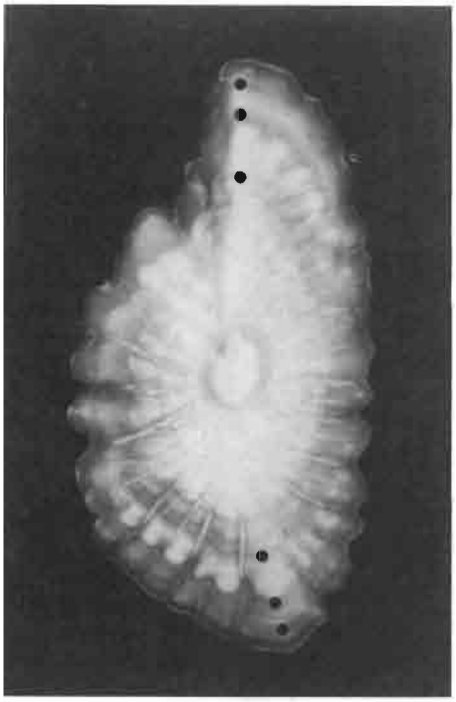

Figure 19

Whole otolith of a 17-cm age 3+(?) butterfish collected inshore in November showing a tiny check close to the nucleus and split, diffuse hyaline zones.

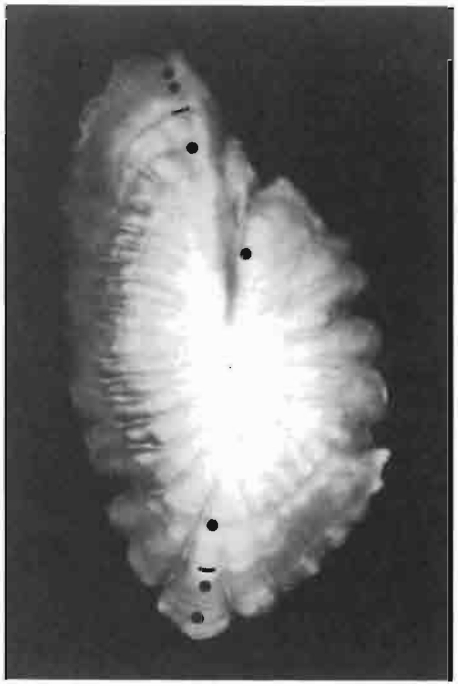

Figure 21

Whole otolith of an 18-cm age 3(4)+ butterfish collected inshore in October showing poorly differentiated annuli.

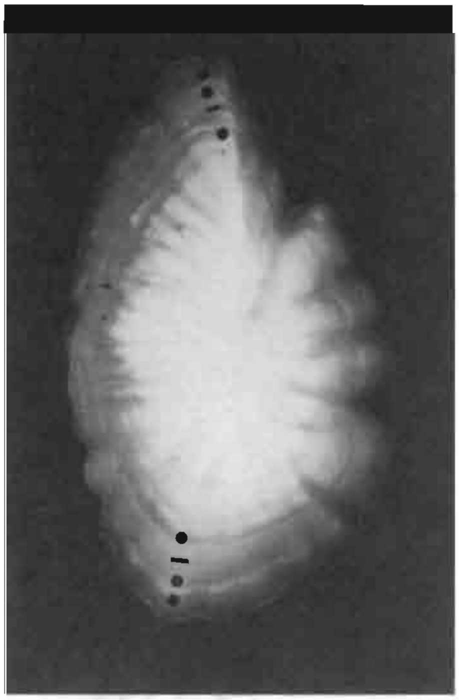

Figure 20

Whole otolith of a 15-cm age 3+ butterfish collected inshore in October showing strong checks and split zones.

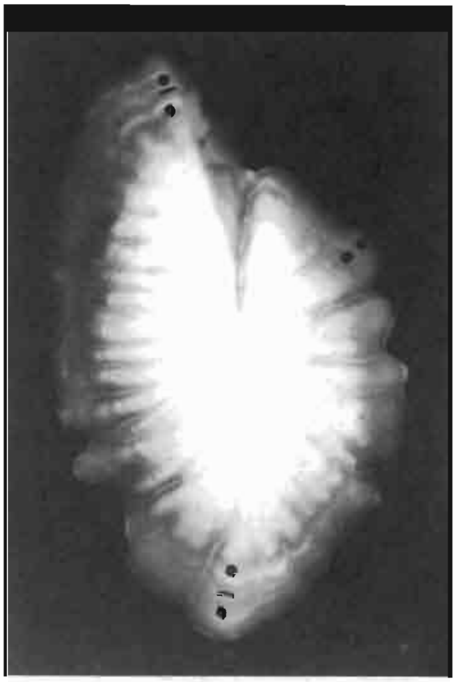

Figure 22

Whole otolith of a 16-cm age 2+ butterfish collected in October showing a complex pattern with two annuli apparent inside the dorsal (lateral) edge.

14

Redfish
Sebastes fasciatus

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Redfish is a slow-growing long-lived scorpaenid. In the Northwest Atlantic it is found from Davis Strait southward to the Gulf of Maine and Georges Bank at water depths up to 300 meters (Bigelow and Schroeder 1953). Growth rates attained to age 6 are the same for both sexes, but thereafter females grow faster than males. Both sexes mature sexually at age 8 or 9. Mating occurs during the fall, and gravid females can be found the following spring with larvae appearing in the water column from April to September (Kelly and Wolf 1959). A maximum age of 50 years has been documented and a size of 45-50 cm (18-20 inches) (Mayo et al. 1983).

Both scales and otoliths have been used to determine the age of redfish. Scales have been used by European investigators and are prepared by impregnating the scales with silver nitrate and viewing under polarized light (Kosswig 1971, 1980). In earliest investigations of redfish at the Woods Hole Laboratory, scales were cleaned in an acidic solution, impressed on a plastic slide, and observed using a microprojector. This method proved unsatisfactory because the annuli on older fish are very compact along the edge of the scale. Attempts were also made to stain otoliths; use of a silver-diammino solution provided a greater contrast between annuli (H. Foster and F. Nichy, unpubl. data, Woods Hole Lab.), but proved to be time-consuming.

Otoliths are preferred because they can be readily processed and annuli on older fish are more distinct than on scales. The otoliths are stored dry and prepared by the following method (Nichy 1977). An otolith is placed on a cardboard tag and covered with wax. A low-speed macrotome saw (fitted with two diamond blades, separated by a spacer) is then used to thin-section through the nucleus along the dorsoventral axis. The result is a transverse section approximately 0.178 mm thick. The section is viewed with a binocular microscope against a dark background using reflected light at a magnification of 25X or 50X. The section is moistened with clove oil, alcohol, or Kodak Photo-Flo 200 solution to enhance the contrast between opaque and hyaline zones.

Along the edge of the otolith, depending upon the time of the year, there may be either an opaque or hyaline zone. The hyaline zone predominates from November to May; the opaque zone is usually formed from March to November. Mayo et al. (1981) were able to validate that growth marks are annual events, based on seasonal formation of hyaline and opaque zones and comparisons of mean lengths-at-age with modes of length-frequencies.

The annual zones on a redfish section consist of a white opaque zone, representing fast summer growth, followed by a dark hyaline zone, representing slow winter growth. An opaque zone succeeded by a hyaline zone constitutes one year of growth. For age determination purposes, the annulus is defined as the hyaline zone marking the end of a year of growth. By convention, a 1 January birthdate is used.

Age determinations may be made by counting annuli from the nucleus to the dorsal edge, with corroborating counts usually made to the proximal and ventral edges (Figs. 1 and 2). The nucleus is centrally located and surrounded by the first annulus, which is a normally distinct, oval-shaped hyaline zone (Figs. 1-5). Figure 1 shows a fairly wide gray zone immediately surrounding the nucleus, inside the first annulus. This zone may be the result of a settling check, but is less apparent on otoliths from older fish (Figs. 2-5). The first annulus may be distinguished from this settling zone because it is separated from the gray zone and extends further out to the edges of the otolith (Fig. 1). If the first annulus appears irregularly shaped, the otolith should be resectioned closer to the nucleus (Fig. 4).

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Annuli from the second to about the tenth year are broad and relatively easy to read. The opaque zone between the first and second annuli is usually a clear white band. After ten years of age, annuli become more compact and less distinct (Figs. 3-5). An age reader can more readily discern split annuli and checks in younger (<12 years) rather than in older fish. Split annuli are recognized because the closely spaced hyaline zones are repeatedly interrupted by narrow opaque bands (Figs. 1-4). Checks are recognized because they are usually quite close to the preceding annulus and become diffuse along the proximal edge (Figs. 1-3).

Difficulties caused by checks can be overcome by following the annuli from the dorsal side of the otolith (where the hyaline zones are relatively broad) to the proximal side (where the hyaline zones narrow) back towards the nucleus. Along the proximal edge, split annuli converge to form a single narrow hyaline zone. Also, the checks become more diffuse and fade away. In specimens >15 years of age, annuli are quite compacted on the dorsal edge. For such specimens, age can be better determined by counting the annuli from the nucleus towards the proximal edge (Fig. 5). Increased thickness relative to width may result in serious underestimation of age if only the dorsal axis is used to interpret age. Age determinations for younger fish (<12) can usually be corroborated by counting the annuli along both the proximal and ventral edges of the otolith (Figs. 1 and 2). Thus, there are three ways of verifying annuli counts on a sectioned redfish otolith. One can begin at the nucleus and count out to the dorsal edge, or to the proximal edge, or count the annuli along the ventral edge.

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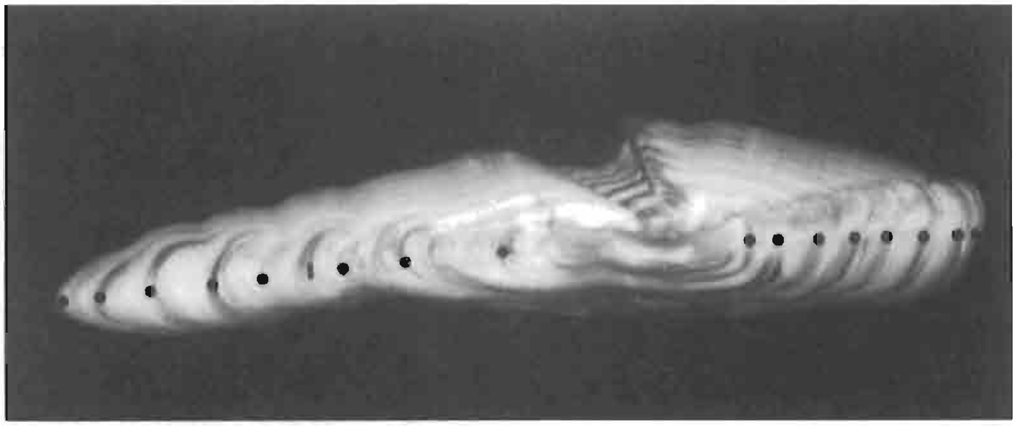

Figure 1

Otolith from a a 27-cm age-8 male redfish. As indicated by the markers, the first annulus separates from the gray settling zone and extends further out to the edges of the otolith. The second and third annuli are split zones which converge to form single annuli along the proximal edge. Note the strong check between the third and fourth annuli. The fourth annulus is fairly weak but discernible, especially along the proximal and ventral edges. The fifth to the eighth annuli are quite distinct and clear.

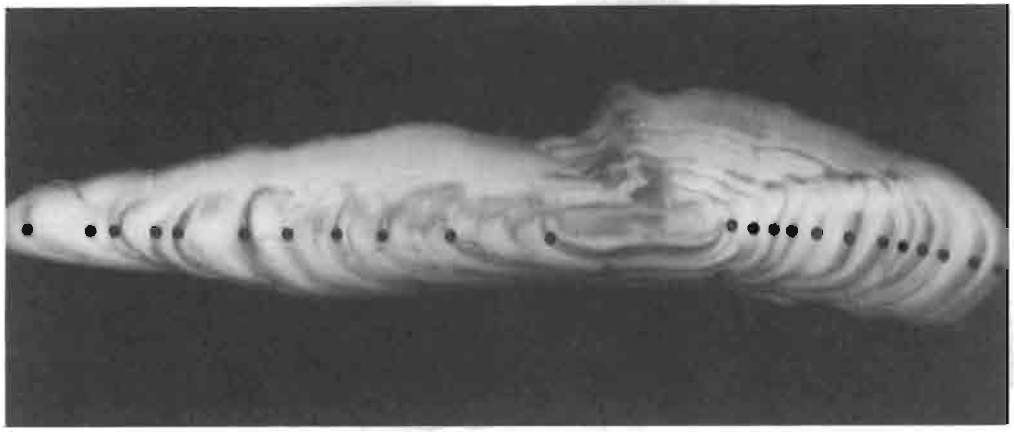

Figure 2

Otolith section from a 28-cm age-12 male redfish. Annuli along the ventral edge are more distinct and corroborate the age found along the dorsal edge. The second to fourth annuli are composed of numerous checks and splits but form clear annuli along the proximal edge. Also, the section shows an irregular growth pattern especially in later years where there are pairs of close annuli (7 and 8, 9 and 10, and 11 and 12) with a large opaque zone evident between the tenth and eleventh annuli.

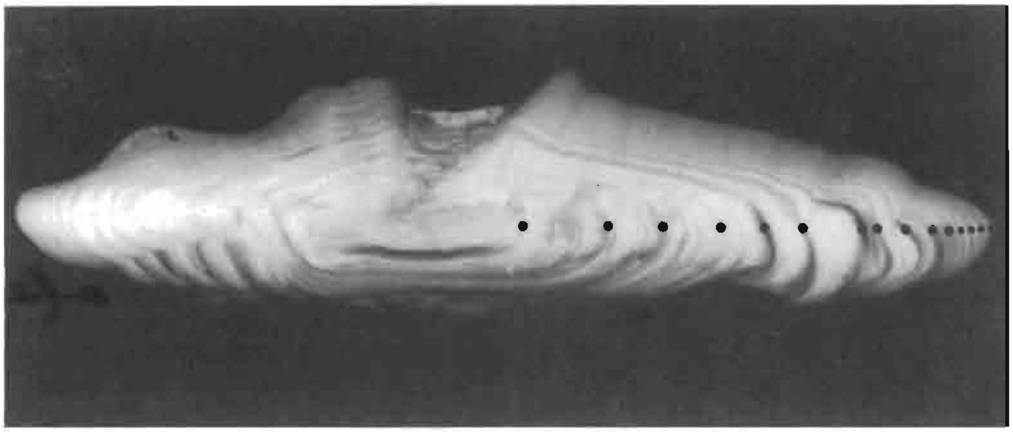

Figure 3

Otolith section from a 34-cm age-15 male redfish. This demonstrates how, with older otoliths, the ages along the dorsal edge are impossible to verify along the ventral edge. Additionally, the annuli are quite compact along the dorsal edge, making the age determination difficult.

Figure 4

Otolith section from a 29-cm age-17 male redfish. This otolith has been sectioned slightly off the nucleus resulting in an irregularly shaped first annulus. Because annuli along the dorsal edge are compact and not very distinct, counting annuli outward towards the proximal edge is the most accurate and precise way of determining age in older fish (>15).

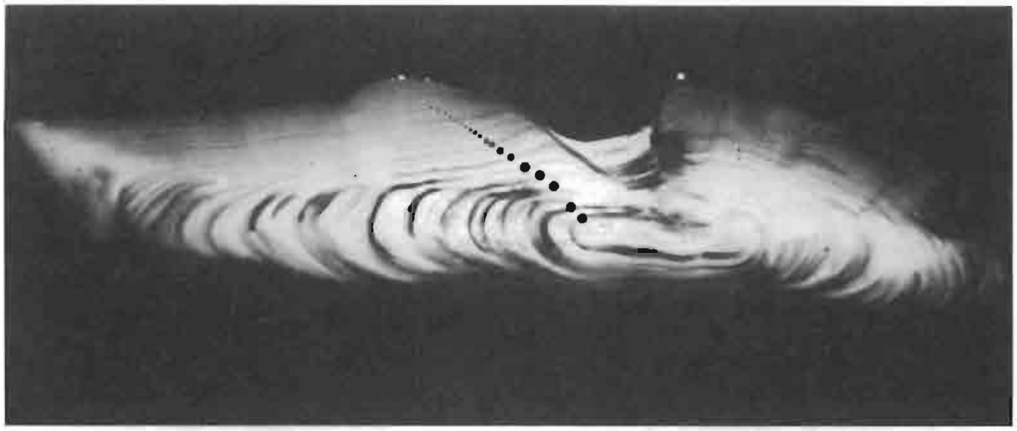

Figure 5
Otolith section from a 33-cm age-31 male redfish. Annuli are clearest on the proximal edge.

15

Summer flounder *Paralichthys dentatus*

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Summer flounder is a relatively active, predaceous, and fast-growing species ranging from Nova Scotia to the northern Gulf of Mexico; it is most common from Cape Cod to South Carolina (Vladykov and McKenzie 1935, Bigelow and Schroeder 1953, Briggs 1958, Leim and Scott 1966). Females grow faster than males; males attain a maximum age and length of about 7 years and 60 cm (24 inches), respectively, as compared with 12 years and 82 cm (32 inches) for females. Summer flounder are sexually mature by age 2 (Morse 1981).

Summer flounder of the U.S. continental shelf appear to be divided into two populations, north and south of Cape Hatteras, although in the vicinity of Cape Hatteras there may be some mixing of the two groups (Wilk et al. 1980, Fogarty et al. 1983). Seasonal inshore/offshore migrations occur in response to changes in water temperature. During winter and early spring, summer flounder are concentrated offshore in depths of 70-155 m along the outer edge of the continental shelf (Byrne and Azarovitz 1982), but in late spring and early summer they move inshore and concentrate in shallow coastal waters and estuaries. An offshore migration begins in August or September (Wilk et al. 1977). Spawning begins in September in southern New England and New Jersey waters coincident with offshore movement, and progresses southward with cooling water temperatures, ending by February off Cape Hatteras (Smith 1973, Morse 1981). Coastal estuarine areas are nurserygrounds for this species.

Many investigators have used otoliths to age summer flounder (Poole 1961; Eldridge 1962; Powell 1974, 1982; Smith and Daiber 1977). The results of some of these studies have been controversial due to uncertainty in locating the first annulus (complicated by north/south differences in spawning times), poor calcification of otoliths, and apparent reversal of the usual seasonal timing of formation of opaque and hyaline zones (Smith et al. 1981). Hyaline edge forms on otoliths during spring and summer months; opaque edge forms during autumn and winter months.

In the late 1970's, investigators at Woods Hole developed ageing methods for summer flounder using laminated plastic impressions of scales; this is now the preferred method at the Woods Hole Laboratory. Shepherd (1980) cross-validated methods in a comparison of zone formation on scales, otoliths, and fin ray sections from individual fish. A State/Federal summer flounder age and growth workshop held at Woods Hole in May 1980 reviewed previous studies and established standard criteria for interpretation of the first annulus on scales, otoliths, and fin ray sections (Smith et al. 1981). Dery (1981) compared growth pattern formation on the scales and otoliths of young fish from various nursery areas and established more accurately the location of the first annulus.

Summer flounder scales are removed from a localized area just above the lateral line anterior to the caudal peduncle and stored dry in coin envelopes. These scales are ctenoid (although the ctenii are greatly reduced) with numerous radial grooves extending from the focus to the anterior edge of the scale. Since the segmented circuli ridges on these scales are finely sculptured, laminated plastic, composed of a thin, soft layer of polyethylene laminated to a harder layer of vinyl chloride (Dery 1983), is used for scale impressions. A minimum of six scales are impressed evenly and quickly under heavy pressure of a roller press. Scale impressions are then examined at 40× using a microprojector or microfilm reader.

"Cutting over" or erosion marks represent annuli on ctenoid summer flounder scales. Such marks are formed from annular erosion of the scale edge, and appear as a sudden break or discontinuity in the formation of one or more (segmented) circuli. In the anterior field of the scale impression, these marks resemble concentric "white" lines, indicating fragmentation or absence of circuli due to erosion (Fig. 1). In the lateral field of the scale, circuli immediately following the mark appear to cross-over or cut across previously formed circuli (Fig. 2). A cutting-over mark should be continuous and intersect the ctena to be interpreted as a true annulus (Fig. 2).

North of Cape Hatteras, annulus formation (scale edge erosion) normally occurs during late spring-early summer. By September, most summer flounder scales exhibit substantial amounts of seasonal growth beyond the last cutting-over mark formed. From late June through July, however, interpretation of the scale edge may be difficult due to individual variation in the timing of annual scale edge erosion. Although cutting-over marks form as late as June, the edge of the scale is included in the assigned age as of 1 January by convention. Scale edge formed subsequent to cutting-over (through December) is interpreted as "+" growth and is not included in the assigned age. Figure 3 shows a scale from an age 5+, 51-cm female summer flounder collected in June with a small amount of scale growth ("+" edge) beyond the last cutting over mark.

Variations in scale growth patterns (north of Cape Hatteras) have not been systematically studied. However, latitudinal differences in rate of growth as reflected by spacing of circuli on the scales, and changes in the size of the first annulus (presumably due in part to north-south shifts in spawning time) have been observed.

Following criteria established at the 1980 summer flounder age and growth workshop (Smith et al. 1981), the first annulus is interpreted as the first continuous cutting-over mark formed on the scale. For summer flounder hatched in October in more northerly waters (e.g., off southern New England and New Jersey), the distance from the focus to the first annulus may be relatively great, reflecting 18 to 21 months of growth from hatching to formation of the first cutting-over mark (second spring following hatching). Relatively wide spacing between circuli up to the formation of the first cutting-over mark is characteristic of the scales of many of these fish (Fig. 4) (see also Dery 1981).

Further south, with a later spawning season (13 to 21 months of growth) and different environmental conditions, circuli in the "0+" region of the scale tend to be spaced more closely together reflecting slower growth (Fig. 5). The distance from the focus to the first annulus decreases; on some scales the first annulus marks a sharp discontinuity between a "0+" zone of closely spaced circuli and a subsequent zone of widely spaced circuli (Fig. 6). Dery (1981) studied scales from small samples of young summer flounder from several coastal nursery areas. Mean backcalculated fish length at the first cutting-over mark was 26 cm for New Jersey samples, 21 cm for Delaware/Maryland samples, and 19 cm for Virginia samples.

Accurate interpretation of the first annulus may be complicated by several factors. Some scales exhibit a small zone of thin, closely spaced circuli near the focus that is not bounded by a cutting-over mark (Fig. 7). This zone may be confused with the small first annulus described above for summer flounder of more southern areas because of close circulus spacing. However, the lack of a cutting-over mark bordering this zone and more frequent association with scales from more northerly areas suggests that this zone may reflect slow growth experienced by autumn-early winter hatched juveniles during the first winter of life.

The presence of a weak, incomplete cutting-over mark formed prior to the first annulus may also result in overinterpretation of age. Such marks are generally evident only in the anterior field of the scale and do not continue into the ctena (Fig. 5). Sudden and marked shifts in circulus spacing can also resemble annuli, especially if the location of such a shift suggests an annulus (Fig. 7). These shifts in spacing do not involve cutting-over; the rows of circuli remain parallel to one another and do not intersect (Fig. 2). Erratic shifts in circulus spacing are generally characteristic of summer flounder scales and will be discussed in more detail. For this reason circulus spacing is not normally used as a criterion for annulus interpretation unless a consistent annual pattern is evident for individual fish.

Subsequent to the first cutting-over mark, age is interpreted by counting the number of complete cutting-over marks (Figs. 1 and 3). In general, scales of summer flounder from more northerly areas are easier to age due to relatively rapid scale growth (indicated by wide circulus spacing) and clear cutting-over marks (Figs. 1 and 3).

Anomalies causing difficulty with age interpretation include checks, split annular zones (double cutting-over marks), and erratic changes in circulus spacing. In addition, scale size may vary significantly for individual fish and for fish of similar lengths, despite attempts to remove scales from a specific location. This makes it difficult to achieve a perspective concerning the size of scales and proportional size of growth zones in relation to fish length.

A check (false cutting-over mark) is characteristic of the second year of growth on some scales and has been a major source of age disagreement. This check may be strongly evident in the anterior field and resemble an annulus since it is often associated with a wide increment between the first and second annuli. However, cutting over of circuli is weak or absent laterally on the scale. Alternatively, an erosion mark may appear weak in the anterior field but more pronounced laterally on the scale (Figs. 6 and 8). In rare cases the check may resemble an annulus, but the anomalous spacing of zones indicates that the mark is more likely to be a check (Fig. 1).

Checks greatly complicate interpretation of some scales from older fish, since annuli become increasingly closely spaced near the edge of the scale. These marks may appear identical to annuli in the anterior field but on close examination are not continuous into the lateral fields. Some scales exhibit many such marks that may reflect scale damage (Fig. 9).

Two cutting-over marks spaced closely together are interpreted as constituting a single annular zone if they fuse together laterally on the scale (Fig. 10). Scales forming such marks ("split" annuli) tend to repeat this pattern from year to year, facilitating interpretation of this anomaly.

Erratic shifts in circulus spacing can mask cutting-over marks. As previously mentioned, sudden brief growth spurts can superficially resemble annuli (Fig. 7). If the same pattern of circulus spacing is repeated on individual scales from year to year, true annular marks, otherwise masked by erratic growth shifts, may be more easily identified (Fig. 11). One type of repeating pattern that actually highlights the annular zones is the tendency for some scales to exhibit a zone of very wide circulus spacing immediately prior to cutting-over (Fig. 12). In general, a helpful approach to interpreting scales exhibiting confusing or erratic patterns is to avoid examining the scale in detail until a general sense of the pattern has been achieved. Figure 13, however, shows a scale from a 71-cm, age 5(6)+ female where the pattern of growth is so erratic that age interpretation is very difficult. An estimated age was assigned based on the number of completed cutting-over marks observed laterally on the scale.

In summary, age interpretation of most summer flounder scales is straightforward if good scale impressions are available, and if cutting-over marks are carefully evaluated for continuity into the ctena and spacing relative to other zones. Familiarity with variation in the pattern of first-year growth with geographic area seems necessary for accurate interpretation of the first annulus.

In terms of general orientation to scale patterns, it is recommended that a sense of the overall pattern of growth be achieved before more detailed features are evaluated. Experience has indicated that overinterpretation of age is otherwise likely to result, since growth patterns on some summer flounder scales are quite complex.

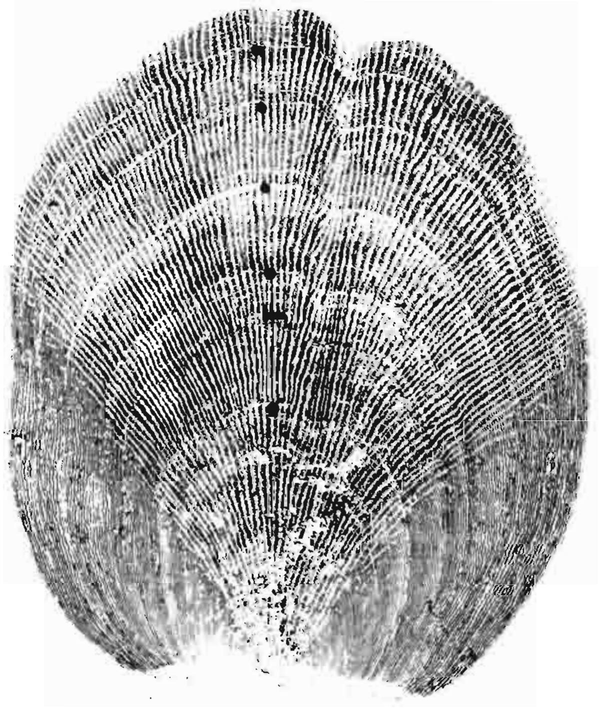

Figure 1

Scale impression of a 64-cm age 5+ summer flounder collected in November showing clear annuli (cutting-over marks) and a strong second summer check.

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Figure 2
Detail of scale impression from the summer flounder of Figure 11 showing a true "cutting-over" mark (annulus) contrasted with a growth shift (indicated by an "x") formed previous to the annulus.

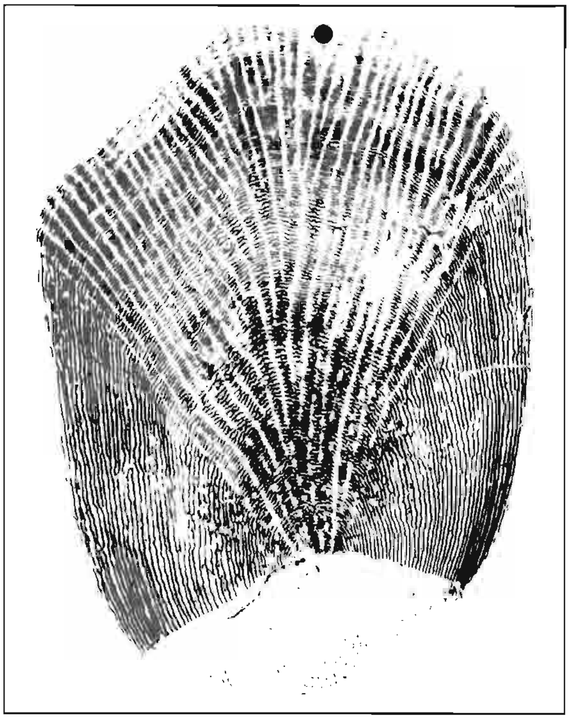

Figure 4
Scale impression of a 26-cm age-1 summer flounder collected in May showing an expanded view of widely spaced circuli with some erratic growth close to the scale edge.

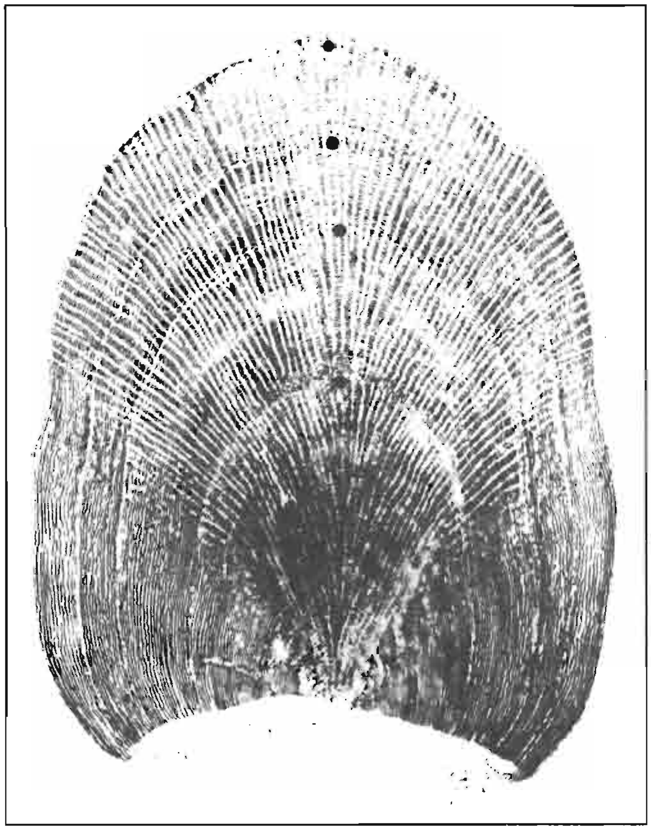

Figure 3
Scale impression of a 51-cm age-4 summer flounder collected in June showing clear annuli and "cutting over" just inside the scale edge.

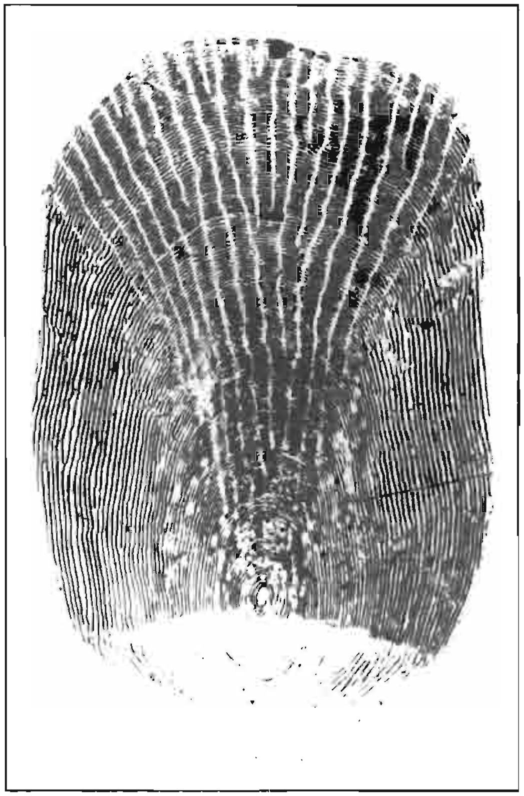

Figure 5
Scale impression of a 29-cm age-1 summer flounder collected in April showing an expanded view of closely spaced circuli and a weak check most evident laterally on the scale.

Figure 6
Scale impression of a 38-cm age-2 summer flounder collected in May showing a strong check between the first and second annuli most evident laterally on the scale.

Figure 8
Scale impression of a 50-cm age 2+ summer flounder collected in November showing a strong check between the first and second annuli most evident laterally on the scale.

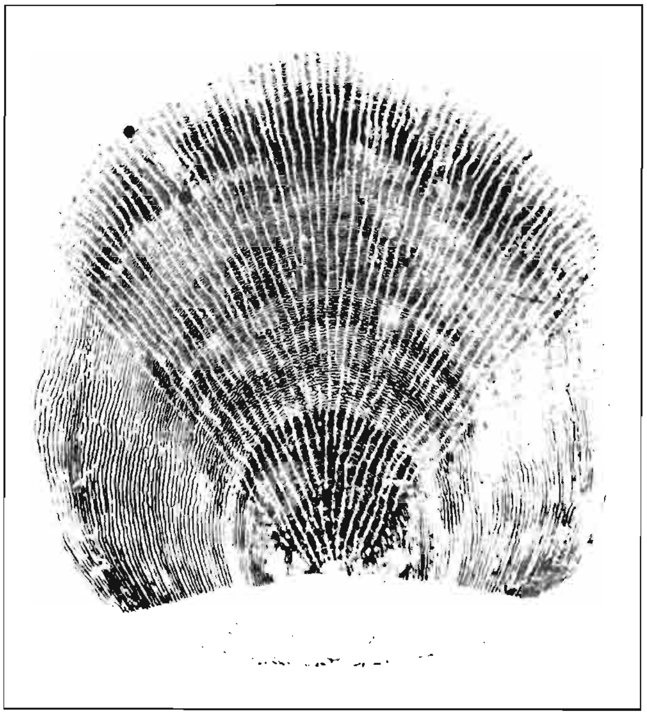

Figure 7
Scale impression of a 31-cm age-2 summer flounder collected in March showing a dense zone of circuli not bounded by "cutting over" close to the focus of the scale. This zone is interpreted as the first winter zone of autumn spawned summer flounder.

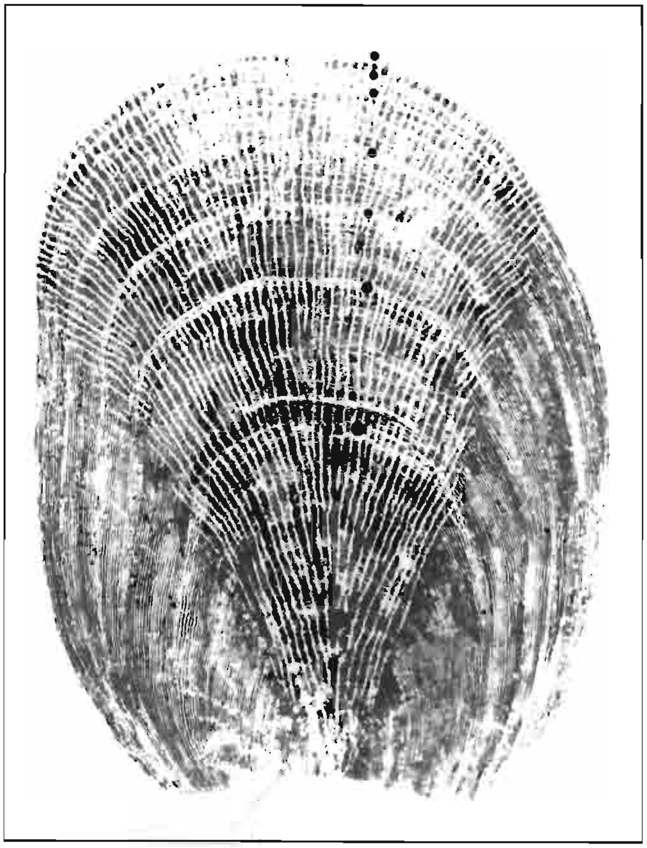

Figure 9
Scale impression of a 62-cm age-8(7) summer flounder collected in March showing numerous checks and scale damage.

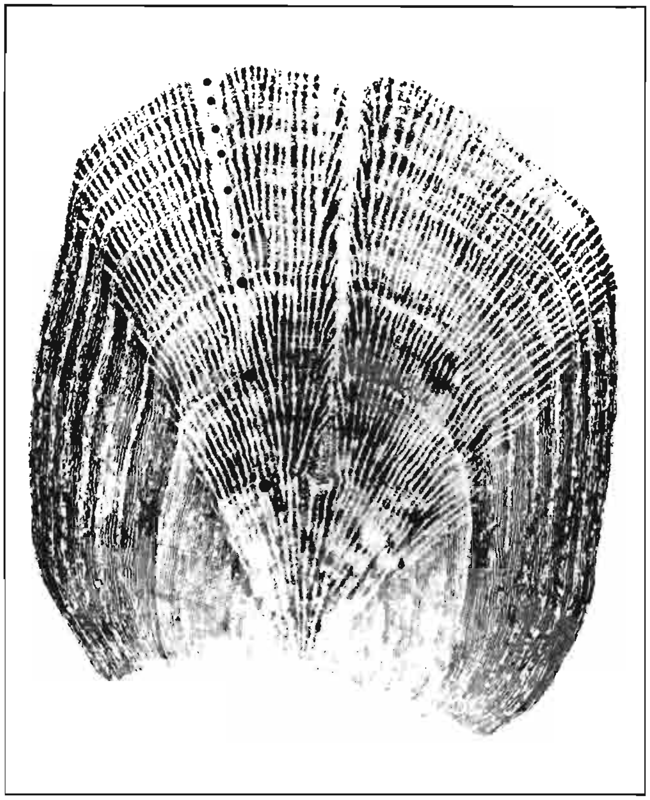

Figure 10
Scale impression of a 54-cm age-9 summer flounder collected in May showing a weak first annulus and split zones (double "cutting-over" marks).

Figure 12
Scale impression of a 61-cm age 4? summer flounder collected in May showing erratic growth and numerous checks. Repeating pattern of wide circulus spacing occurs just prior to "cutting over."

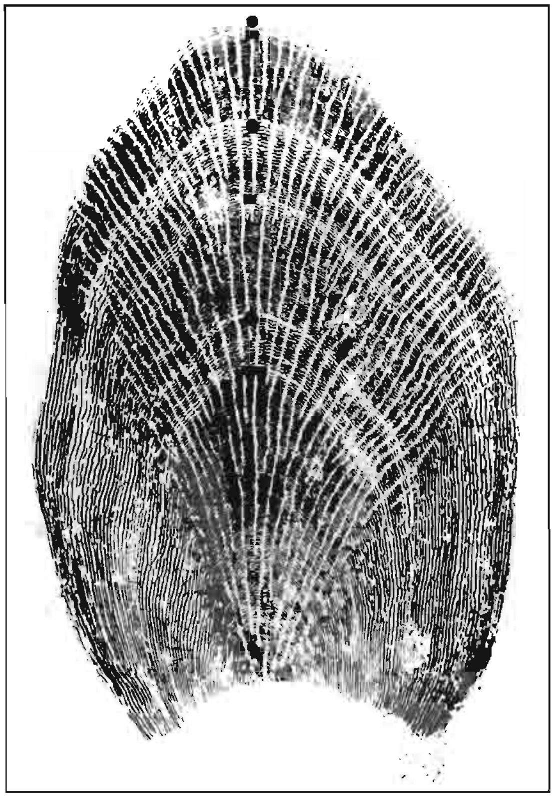

Figure 11
Scale impression of a 49-cm age-3 summer flounder collected in March showing growth shifts formed in a repeating pattern prior to the first and second annuli.

Figure 13
Scale impression of a 71-cm age 5(6)+ summer flounder collected in October showing highly erratic growth and numerous checks.

16

Winter flounder *Pseudopleuronectes americanus*

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Winter flounder is distributed in the Northwest Atlantic from lower Labrador southward to Georgia, and is most abundant from the Gulf of St. Lawrence to Chesapeake Bay. For descriptive purposes, the winter flounder resource and fishery have been divided into four major geographic groups: Gulf of Maine, Georges Bank, Southern New England, and Mid-Atlantic. Migrations of winter flounder are not extensive. The fish appear to be broken up into local subpopulations that are relatively stationary. Movements north of Cape Cod are relatively localized and confined to inshore waters, whereas south of Cape Cod, flounder disperse seasonally in relation to water temperature. Little mixing occurs between Georges Bank and inshore areas (Howe and Coates 1975). Lux (1973) observed that winter flounder on Georges Bank grow faster than fish from inshore areas.

Winter flounder are relatively long-lived, reaching a maximum age of about 15 years and a length of 58 cm. The growth rate up to age 2 is the same for both sexes, but thereafter females grow faster and live longer than males (Lux 1973). Both males and females mature sexually at age 2, although males mature at a smaller size than females. Winter flounder spawn from January through May with spawning beginning earlier in southern portions of its range.

Because interpretation of winter flounder growth in inshore areas has not been validated, this section deals only with age determination for Georges Bank fish. Recent research employing daily growth increments as an age-validation tool for inshore winter flounder has shown promise, but such studies are as yet incomplete.

Early investigators (Lobell 1939, Perlmutter 1940) chose scales as the preferred structure; later investigators based age determinations on otoliths (Berry at al. 1965, Pearcy 1962). At the Woods Hole Laboratory, scales are used as the primary structure, although otoliths are used quite frequently for a corroboration of age determination.

Scales are taken from the lateral line area a few centimeters anterior to the caudal peduncle for dry storage. Impressions of the dried scales are made in laminated plastic using a roller press and viewed on a microprojector at a magnification of 40×. Regenerated scales are discarded.

The scales are ctenoid, with radial grooves extending from the focus to the forward margin of the scale. Otoliths from young-of-the-year fish may be covered with Kodak Photo-Flo 200 solution for viewing whole by a binocular microscope at a magnification of 25×, under reflected light. Otoliths from older fish are thin-sectioned by a low-speed macrotome saw (Nichy 1977). The sections are covered with Photo-Flo, and examined under a binocular microscope against a dark field with transmitted light at a magnification of 25-50×.

By convention, a 1 January birthdate is used. Annular zones on winter flounder scales appear as changes in the circuli pattern. Zones of fast and slow growth are reflected by wide or narrow spacing, respectively, of circuli, made up of individual platelets on the sculptured upper surface of the scale.

When using either whole or sectioned otoliths, a year's growth consists of a white opaque zone, representing fast summer growth, followed by a dark hyaline zone, representing slow winter growth. The annulus, by definition, is the hyaline zone marking the end of a year of growth, i.e., the winter growth zone.

On winter flounder scales and otoliths, the first winter zone representative of the first annulus is well defined for slow-growing fish but not for fast-growing fish. The scale winter zone appears on the edge approximately coincident with the hyaline edge on otoliths. Studies have demonstrated close agreement between scale and otolith readings from the same fish through age 4.

The first annulus on a scale is identified by a dense mass of winter growth (closely spaced circuli) near the focus; the end of the annulus is considered to be the outermost of these circuli. Sometimes, pigmentation on the scale will cover the first annulus almost completely. The first annulus on many scales is barely discernible and is usually estimated by slight changes in the formation of the circuli (Fig. 1). For all succeeding years, spring and summer growth are characterized by widely spaced circuli (rapid length accretion) and fall and winter growth by closely spaced circuli (slow length accretion). The outer edge of the zone of closely spaced circuli is considered to be the end of the annulus. Slight checks in growth consisting of only a few closely spaced circuli on the scale are considered to be checks and may be ignored in assigning age (Fig. 2).

On scales from older fish the identity of checks is more obvious because of the relative spacing of annuli and the contrast of checks with the more strongly formed annuli (Fig. 3 and 4). After formation of the third annulus, age interpretation may be complicated by irregular spacing of annuli (Fig. 5). The growth increment between the second and third annuli is generally wide, with decreasing growth increments between later annuli (Figs. 6 and 7).

Contrast between winter and summer zones tends to deteriorate toward the outer edge of scales of older winter flounder. After the fourth winter zone, summer growth appears to merge with the slow winter growth and the narrow growth increments may make interpretation difficult (Fig. 8).

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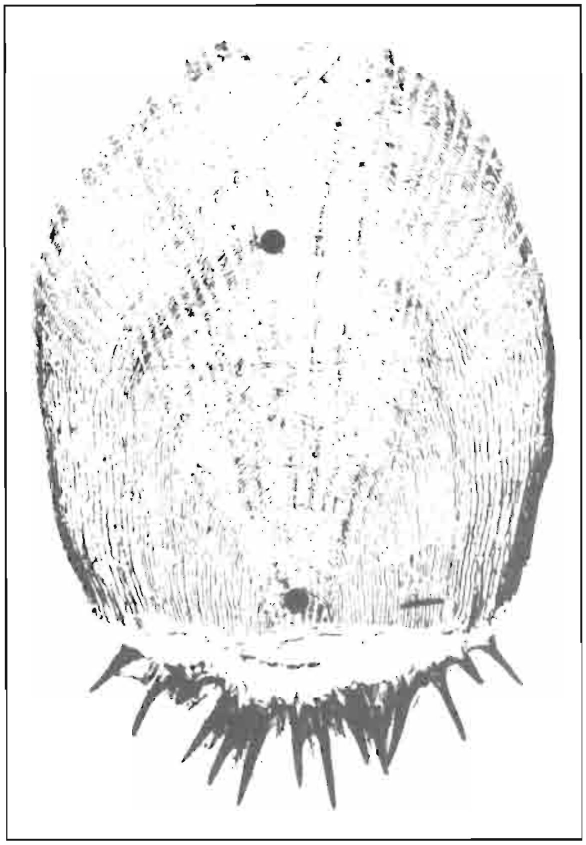

Figure 1

Scale impression of a 36-cm age-2 female winter flounder collected in the fall from Georges Bank showing a small first annulus with good growth in the second year.

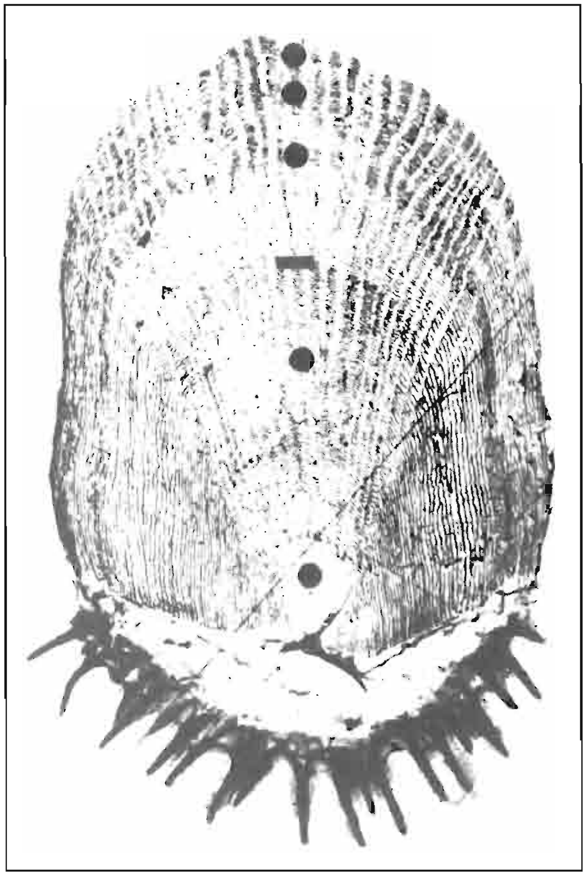

Figure 2

Scale impression of a 46-cm age-5 male winter flounder collected in the spring from Georges Bank showing a fairly strong check after the second annulus.

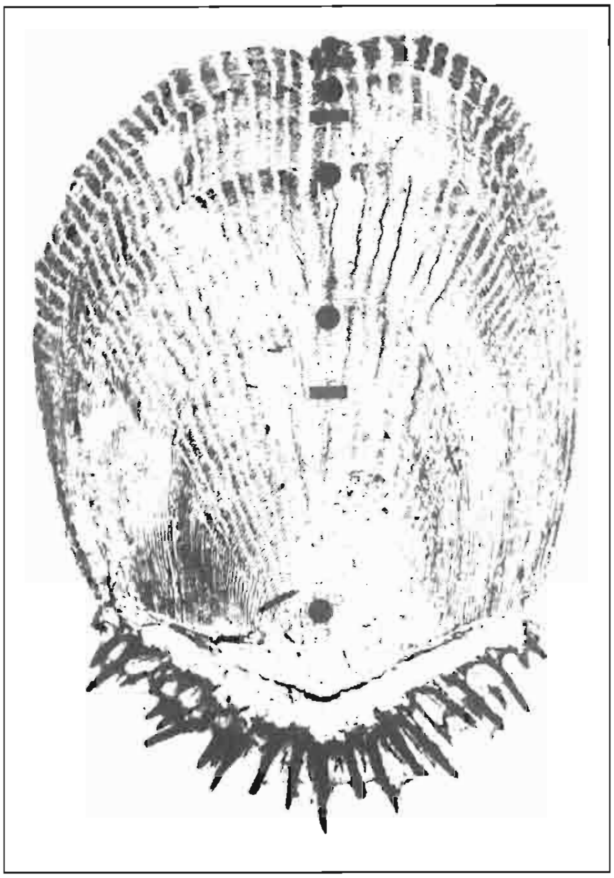

Figure 3

Scale impression of a 54-cm age-5? female winter flounder collected in the spring from Georges Bank showing a check before the second annulus with split fourth and fifth annuli.

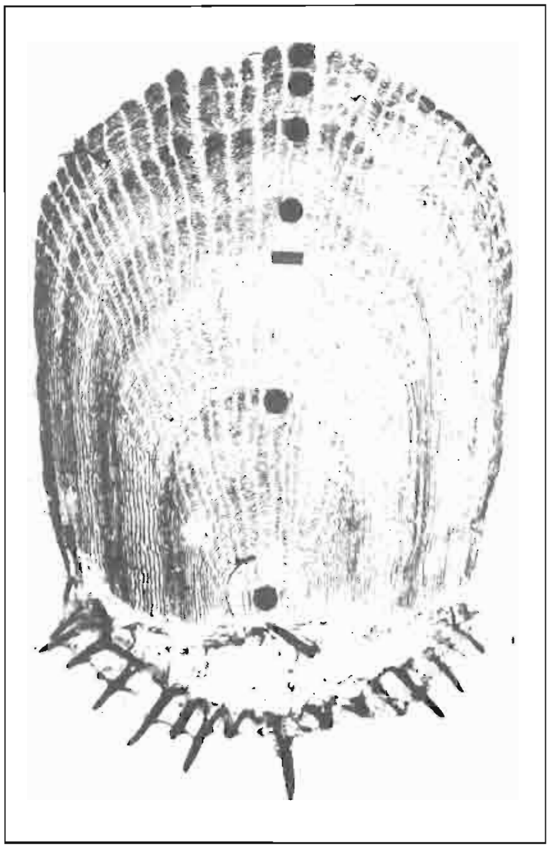

Figure 4

Scale impression of a 47-cm age-6 male winter flounder collected in the spring from Georges Bank showing a split third annulus.

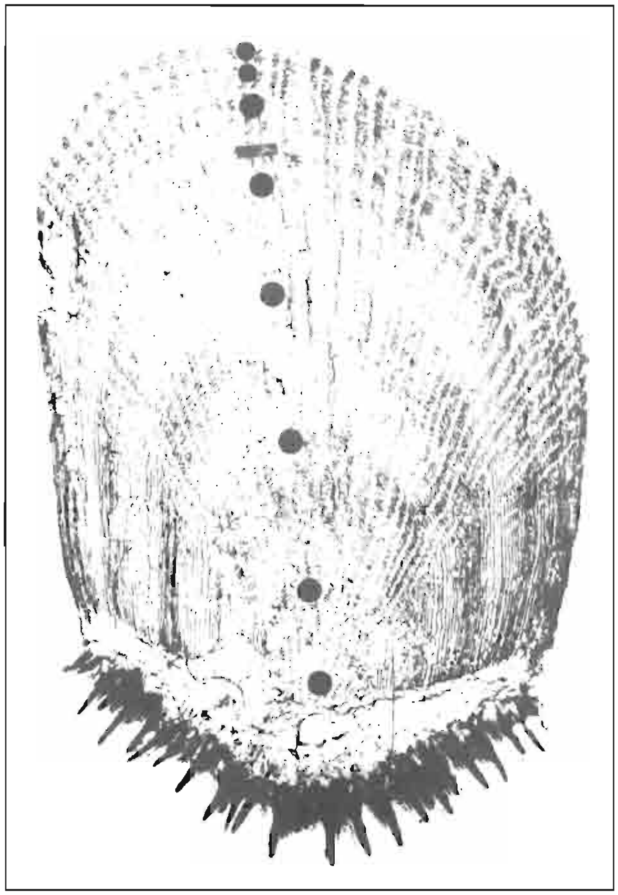

Figure 5
Scale impression of a 49-cm age-8 female winter flounder collected in the spring from Georges Bank showing fairly small first, second, and third annuli. There is also a check or damage evident between the fifth and sixth annuli.

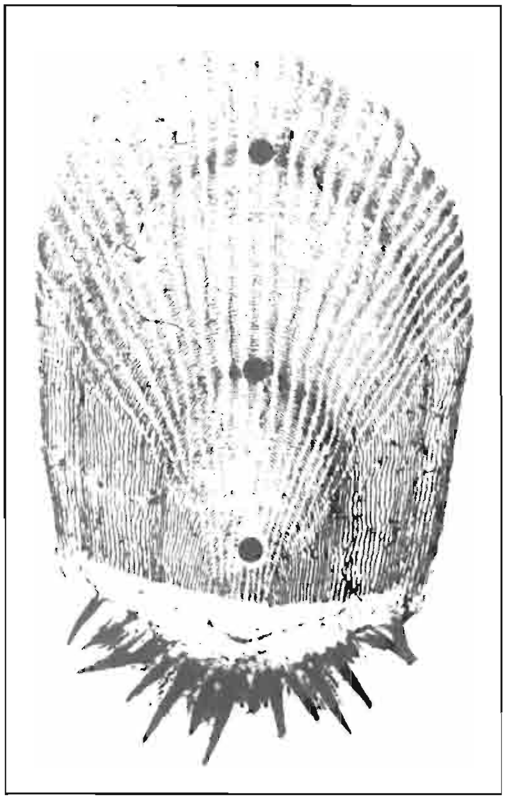

Figure 6
Scale impression of a 46-cm age-3 female winter flounder collected in the fall from Georges Bank showing a fairly small second annulus with good growth in the third year.

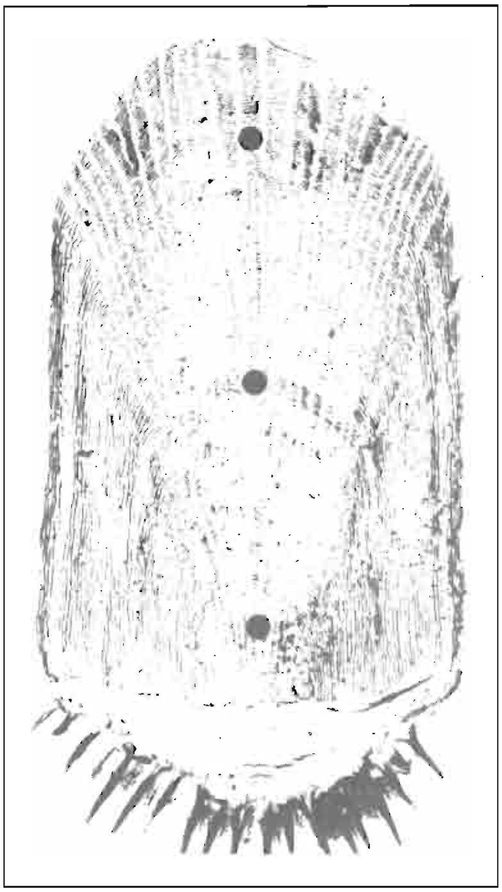

Figure 7
Scale impression of a 40-cm age-3 female winter flounder collected in the fall from Georges Bank showing moderate growth in the second year with good growth in the third.

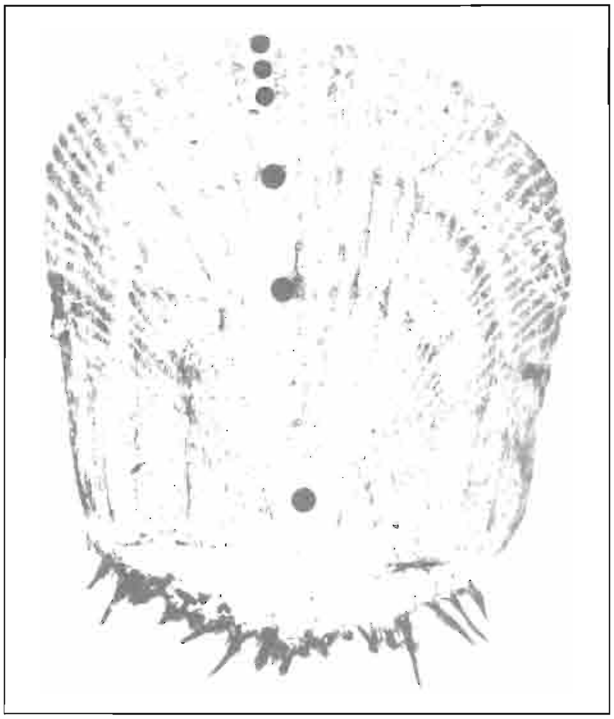

Figure 8
Scale impression of a 51-cm age-6 female winter flounder collected in the spring from Georges Bank showing close fourth, fifth, and sixth annuli.

17

Witch flounder *Glyptocephalus cynoglossus*

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Witch flounder, or grey sole, is a small-mouthed, right-sided pleuronectid distributed in deep, cold waters from Labrador to North Carolina. Although numerous stocks of witch flounder have been delineated in Canadian waters (Fairbairn 1981, Bowering and Misra 1982), no stock-identification studies have been conducted for our region. Witch flounder in the Gulf of Maine-Georges Bank region are considered to be a unit stock for assessment purposes. Witch flounder are sedentary and do not undertake seasonal migrations (Bigelow and Schroeder 1953). Most commercial catches occur at depths of 90-270 m over mud bottom at temperatures ranging from 2°C in winter to 9°C during summer (Burnett and Clark 1983).

Relative to other flatfish in the region, the witch flounder can be characterized as a slow-growing, late-maturing, long-lived species. Maximum observed length and age for the Gulf of Maine-Georges Bank region are 72 cm total length and 30 years, respectively. Median age at sexual maturity for male witch flounder is 4 years; for females, 6.5 years. Spawning occurs over a protracted season with a peak occurring during May and June. The pelagic larval stage is lengthy compared with other flounders, lasting from 4-6 months (Bigelow and Schroeder 1953) to a year (Evseenko and Nevinsky 1973).

The first study of witch flounder age was conducted by Huntsman (1918), who used scales as the ageing structure. Molander (1925) and Bowers (1960) both employed whole otoliths for witch flounder from the eastern Atlantic, but did not validate their methodology. Powles and Kennedy (1967) polished whole otoliths from Scotian Shelf samples, validating their interpretation of hyaline zones as annuli by using modal analysis of back-calculated mean lengths-at-age of younger fish. Burnett (1987) examined thin-sectioned otoliths from the Gulf of Maine-Georges Bank collections. Validation techniques in this study consisted of comparing ages obtained from scales with otolith-based ages of individual fish and examining the seasonal progression of otolith edge type.

At Woods Hole, thin-sectioned otoliths are examined with the following exceptions: 1) Whole otoliths are used when possible for younger fish to save preparation time, and 2) scales are used for commercial samples when dealers do not allow otolith extraction by National Marine Fisheries Service port samplers. (However, scales cannot be aged accurately beyond 10 years-of-age due to compression of annuli on the scale edge). Upon removal from the fish, otoliths are stored dry.

Although either otolith is a suitable structure, the ventral otolith (easily distinguished in larger fish by its greater length and lesser height) generally provides better interpretations in older fish due to minimal dorsolateral compression within the sacculus. A low-speed macrotome saw is used for thin-sectioning otoliths to thicknesses of 0.178±0.051 mm (0.007±0.002 inches); the most successful orientation of the section is transversely through the nucleus along the dorsolateral axis. The resulting section allows tracing of hyaline zones from the sulcus area into the otolith body.

Sections are immersed in ethyl alcohol and viewed against a dark background at magnifications of 25-50× with reflected lighting. Age determinations are based on the number of hyaline zones present. Figure 1 shows a section from an otolith taken from a 54-cm female witch flounder assigned an age of 17 years. Features of interest include: A) poorly defined first annulus; B) broad, well-defined opaque and hyaline zones present through ages 2-9; C) a check between annuli 6 and 7, possibly associated with initial reproductive efforts; D) narrowing of both zones subsequent to age 5; and E) splitting of opaque zones which can be mistaken for annuli in the outer fields. The section from an 11-cm male illustrates both a settling check within the nucleus associated with metamorphosis and settling to a benthic habitat and the lack of a well-defined first annulus (Fig. 2); this fish, captured in July, was assigned an age of 1+. Figure 3 represents a typical intermediate-aged fish, in this instance, a 34-cm female captured in April. Again, the first annulus is poorly defined; however, the settling check and annuli 2-4 are prominent in this age-5 interpretation. For older fish, both lateral fields must be utilized: earlier annuli, more distinct and less subject to zone-splitting in the ventral field, can be traced around to the dorsal field. This generally affords better interpretation of later annuli. Later annuli may also be more accurately evaluated within the sulcus, providing a point of reference has been established in the otolith body. Care must be taken in evaluating the outer annuli of older fish and in categorizing the type and width of edge material; often increasing magnifications and the examination of the otolith halves are necessary in both instances.

An important clue in the age-determination process is also provided by the spacing of opaque and hyaline zones. Annual incremental growth of witch flounder diminishes sharply after age 12 and remains fairly uniform thereafter; often decisions between true annuli and splits within opaque zones can be made by examining the spacing of otolith events.

To summarize, thin-sectioning of otoliths is a reliable method for witch flounder. Sectioning increases the preparation time, but the resulting improvement in accuracy of age determinations justifies the approach. Reliable age determinations beyond age 10 or so will be an important prerequisite for analytical assessments of this species.

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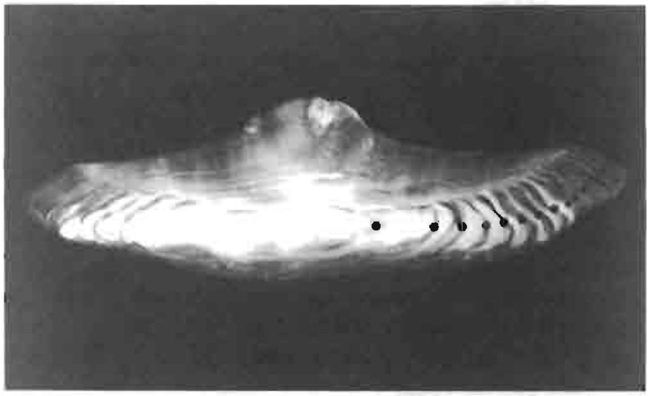

Figure 1

Otolith section from a 54-cm age-17 female witch flounder collected in November showing a poorly defined first annulus; broad, well defined zones present through ages 2-9; a check between annuli 6 and 7, possibly associated with initial reproductive efforts; narrowing of zones subsequent to age 5; and splitting of opaque zones in the outer fields. (This section is not cut exactly at the nucleus.)

Figure 2

Otolith section from an 11-cm age 1+ male witch flounder collected in July showing a settling check and poorly defined first annulus.

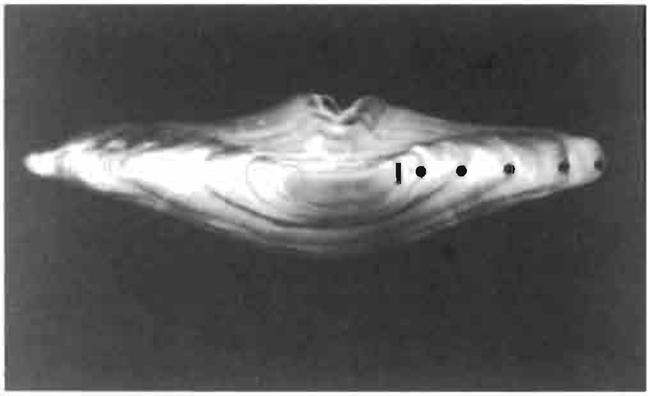

Figure 3

Otolith section from a 34-cm age-5 female witch flounder collected in April showing a prominent settling check, poorly defined first annulus, and well defined annuli 2-4.

18

American plaice
Hippoglossoides platessoides

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American plaice is a sedentary, slow-growing flatfish ranging from southern Labrador to Rhode Island (Bigelow and Schroeder 1953). In the Gulf of Maine and Georges Bank area, individuals attain a maximum length of about 70 cm (28 inches) and ages in excess of 20+ years, with females growing faster than males after age 4 (Sullivan 1982, Dery unpubl.). Most American plaice in these waters are sexually mature by age 3 (Sullivan 1982).

American plaice tend to be distributed in deep water from 90 to 180 m, and do not occur in waters less than 25-35 m. Feeding and spawning migrations appear to be limited (Bigelow and Schroeder 1953, Pitt 1967, Sullivan 1982). Spawning in the Gulf of Maine extends from March through May, with peak activity in April and May (Bigelow and Schroeder 1953, Colton et al. 1979). Coastal waters along the Gulf of Maine are nurserygrounds for this species (Bigelow and Schroeder 1953).

Studies by Powles (1965, 1966) and Pitt (1967) validated hyaline zones on otoliths as annuli for American plaice in Canadian waters. Ageing techniques for the Gulf of Maine-Georges Bank region have not been validated (Lux 1969, 1970, Sullivan 1982). Although the hyaline zones are considered to be valid annuli, a large number of Gulf of Maine and Georges Bank American plaice otoliths are often difficult to interpret, exhibiting weak, diffuse, or split hyaline zones, and, occasionally, strong checks. Little documentation of such problems is available, although Powles (1966) noted the presence of checks on the otoliths of small fish.

Powles (1965) and Lux (1970) examined whole otoliths stored in glycerin; Pitt (1967) broke otoliths in half and examined the broken surfaces. Sullivan (1982) examined thin-sections of otoliths of specimens greater than 35 cm. Smaller otoliths were examined whole in glycerin.

Age determinations have been performed at the Woods Hole Laboratory by examination of the thin-section and cut surfaces of one otolith. Tranverse sections 0.20 mm thick are made precisely at the nucleus of the otolith. The other whole otolith may be used to verify the age from the section and for young fish with clear zone formation. Prior to examination, otoliths are stored dry. For purposes of consistency with terminology applied to otolith sections of other species described in the manual, the terms "dorsal," "ventral," "proximal," and "distal" are used to describe locations on sections as if the fish's left eye had not migrated, resulting in a change of orientation of the otoliths to a vertical position, one above the other. Generally, the right or dorsal otolith provides the best section for age interpretation. This otolith is relatively thick and has a deeper sulcus acusticus. This is important in locating annular zones on thin-sections.

Although glycerin is an effective "clearing" medium for enhancement of hyaline zones, it has not been used at this laboratory because of difficulties with edge interpretation of overly cleared otoliths. Whole otoliths or sections are viewed in ethyl alcohol against a dark background under reflected light. Magnifications of up to 50-60× are used in order to distinguish the closely spaced annuli near the edge of older plaice otoliths.

The size of the first annulus is somewhat variable according to time of hatching and individual growth differences. Annulus formation generally occurs during the winter months and seems to be influenced by temperature (Pitt 1967). American plaice sampled further inshore tend to form opaque edge earlier in the season than deeper-water fish, possibly in response to advanced warming of coastal waters. Younger fish also resume growth earlier than older individuals, with some otoliths exhibiting large amounts of opaque edge as early as April (Fig. 1). By October, most otoliths of young fish have begun to form hyaline edge (Fig. 2), while otoliths of older fish may continue to exhibit opaque edge (Fig. 3). It is important to note that the transverse section will reveal less newly-formed edge than the otolith as a whole. More detailed information on time of annulus formation in the Georges Bank-Gulf of Maine area is not currently available since specimens were available only from spring and autumn survey sampling. By convention, a birthdate of 1 January is used. As of this date, an annulus is interpreted on the edge of the otolith until spring growth resumption.

The dark central kernel or nucleus of the otolith represents the larval-to-juvenile pelagic phase of growth described by Powles (1966) (Fig. 4). Surrounding this central kernel is a thin, weak hyaline ring or "settling check" (Fig. 4) possibly representing the change from pelagic to demersal habitat, and similar to the "pelagic" ring described by Nichy (1969) for the silver hake. This zone is sometimes evident through the surface of the whole otolith and may be confused with the first annulus, which is formed rather close to the nucleus (Powles 1966).

The first annulus is usually a relatively strong hyaline zone and is clearly marked in the sulcus area (Figs. 2 and 5). A few plaice otoliths exhibit an unusually large first annulus, with the settling check surrounding the nucleus (Fig. 1). The first annulus may also be very tiny and close to the nucleus, appearing as thin concentric rings of hyaline material (Fig. 6).

Several factors that appear to influence the clarity of annulus formation on plaice otoliths include depth, temperature, growth rate, and sampling location. Otoliths of plaice from deeper Gulf of Maine waters often have less distinct annuli, probably because seasonal influences on the growth of these fish are muted. Otoliths of faster-growing fish from the western part of the Gulf of Maine and Georges Bank also exhibit less distinct zones than those of the eastern Gulf of Maine and Scotian Shelf areas. These differences in growth rate are apparent from examination of age/length keys (Dery unpubl. data). Since stock structure in the Gulf of Maine is currently unresolved (F.E. Serchuk, Woods Hole Lab., pers. commun.), the significance of these regional differences is unclear.

Figures 1-8 show otolith sections with distinct annulus formation. Although annuli may be clearly evident on all parts of a section (Fig. 2), they are usually most distinct on the proximal side of a section from the right otolith in the area between the sulcus and the dorsal edge (Fig. 7). Annuli tend to be more compacted on the shorter ventral axis which could lead to erroneously low age estimates. Because of the depth of the sulcus on sections shown in Figures 3 and 7, the annuli are especially distinct. Figure 8 provides an example of very slow growth, with the third through eighth annuli formed very close together on the otolith of a fish of only 28 cm. These zones are quite distinct, however, on the proximal (sulcus) side of the section. After age 3, this otolith increased more in thickness than in width or length, resulting in the apparent layering of annuli.

Otoliths with split or diffuse annular zones are more difficult to read, but are nevertheless interpretable in the sulcus area where the hyaline zones are more clearly resolved. Figure 9 provides an example of a split, diffuse second annulus. This section could easily be overaged if interpreted along the transverse axis. However, only one distinct zone (second annulus), in addition to the first annulus and edge annulus, is evident in the sulcus area. Similarly, if numerous checks are formed in-between annuli (Fig. 10), age can be reliably interpreted only in the sulcus, because checks are not normally evident on this part of the section. Figure 11 shows a similar growth pattern for an older fish.

The sulcus area, however, is not always the most reliable part of the otolith section for age interpretation. Although the annuli of Figure 12 are most distinct in the sulcus area, the eleven annuli on the otolith section of Figure 13 are clearest along the dorsoproximal axis. On this section, two groups or clusters of annuli are evident: annuli 2, 3, 4, and 5, 6, 7. In Figure 14, annuli are much more distinct on the dorsal axis than in the sulcus, which is very difficult to interpret. Therefore, each section should be individually evaluated for the best location to interpret the annuli, and alternate locations should be used to verify age.

Individual otoliths of American plaice may exhibit both strongly and weakly defined hyaline zones, unlike individuals of other species which tend to show a consistent pattern of hyaline zone formation from year to year. The first several annuli may be distinct, with those of the outer zones poorly formed (Fig. 15), or the outer annuli may be more distinct and the central or mid zone of the otolith difficult to interpret (Fig. 16). This intra-otolith variability in definition of hyaline zones is typical of many American plaice otoliths.

On some otoliths, the growth patterns are so weak and variable that error in age interpretation is likely. On these otoliths, each hyaline zone must be carefully traced around the periphery of the section to determine whether or not it is continuous and therefore an annulus. The annular zones may appear as indistinct clusters of very thin hyaline rings. In Figure 17, the separation between the annuli is most evident on the distal side (bottom) of the section. A growth pattern such as this may be very difficult to interpret on a section from the thinner, more convex left otolith with a shallow sulcus (zones near the sulcus may be poorly defined). Figure 18 is a left otolith section with a shallow sulcus, which is, however, possible to interpret. The annuli along the dorsoventral axis are quite weak and diffuse, which is characteristic of some fast-growing plaice (Figs. 17 and 18). Some otoliths exhibit such poorly defined growth zones that they cannot be reliably interpreted (Fig. 19).

The otoliths of older American plaice can be quite difficult to age without a clear sulcus area on the section, or without an interpretable whole otolith. Figure 20 shows an otolith section from a 60-cm, age-17(18) fish where the growth pattern is increasingly complex toward the dorsal tip of the section. Annuli can be traced from the sulcus area, which is fairly easy to interpret, around the dorsal edge of the section. Age can also be determined using the whole otolith (Fig. 21), which shows 17 continuous hyaline zones.

In summary, American plaice otoliths often exhibit complex zone formation requiring cross-verification of age using both the thin section and/or whole otolith or sectioned otolith half. Young American plaice can be aged by simply examining the whole otolith in alcohol if the hyaline zones are strong and well defined. However, where the interpretation is not clear, preparation of a thin-sectioned otolith, preferably the left otolith, is necessary.

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Figure 1

Otolith section of a 34-cm age 4+ American plaice collected in April showing a large first annulus and opaque edge.

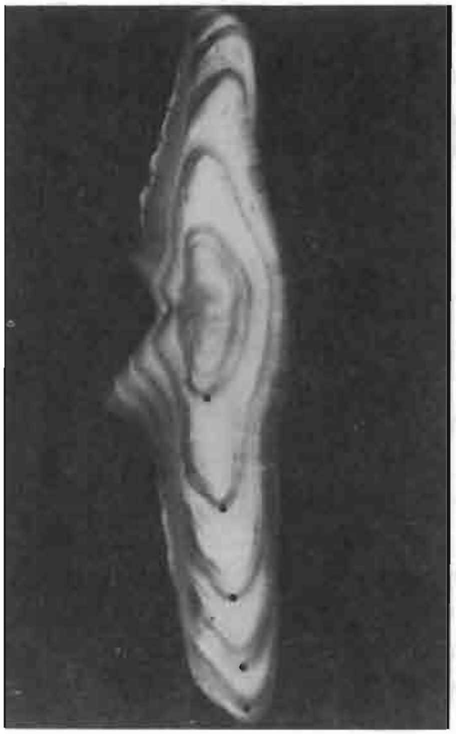

Figure 2

Otolith section of a 35-cm age 5+ American plaice collected in November showing strong clear annuli and a hyaline edge.

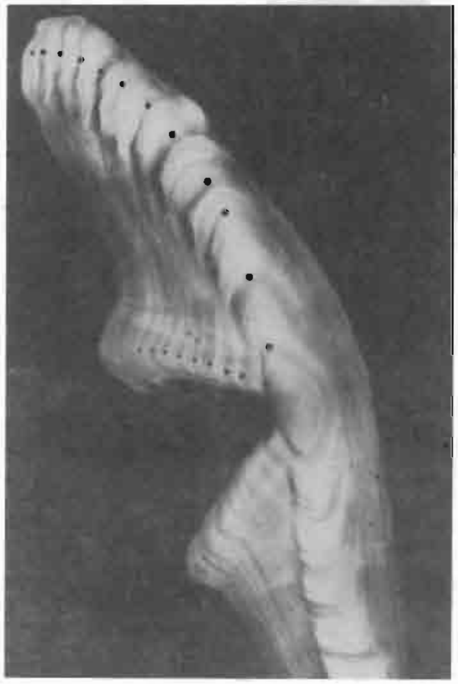

Figure 3

Ventral part of an otolith section from a 54-cm age 12+ American plaice collected in October showing a deep sulcus facilitating interpretation of annuli around that area.

Figure 4
Otolith section of a 9-cm age 0+ American plaice collected in November showing a well defined larval zone and settling check.

Figure 6
Otolith section of a 16-cm age 2+ American plaice collected in October showing a strong, tiny first annulus.

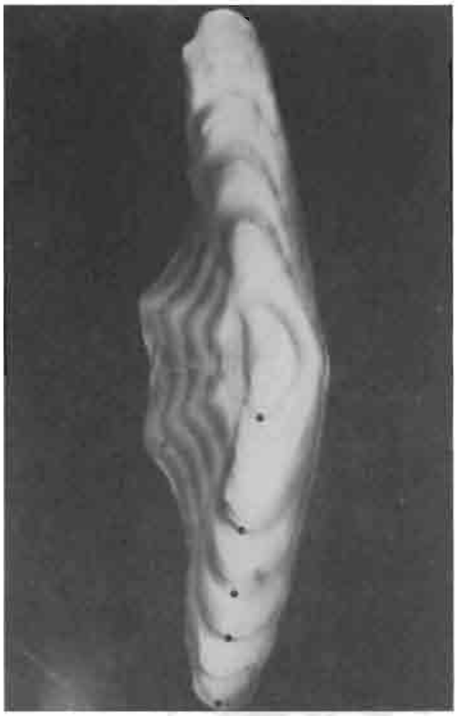

Figure 5
Otolith section of a 33-cm age-5 American plaice collected in March showing strong clear annuli, especially around the sulcus, and split fifth annulus.

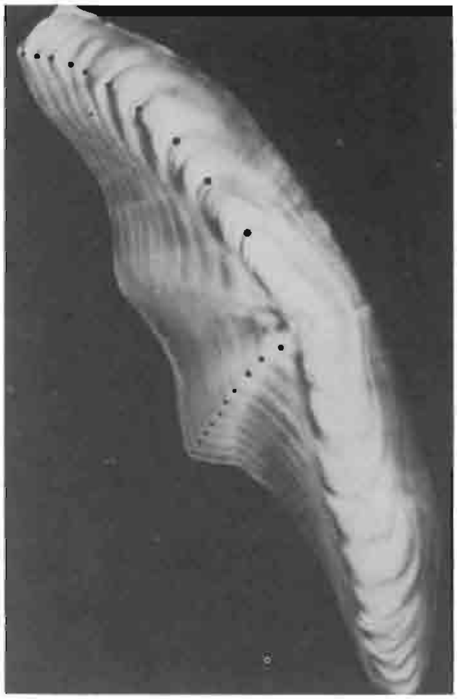

Figure 7
Otolith section of a 48-cm age-10 female American plaice collected in May showing a deep sulcus and strong clear annuli.

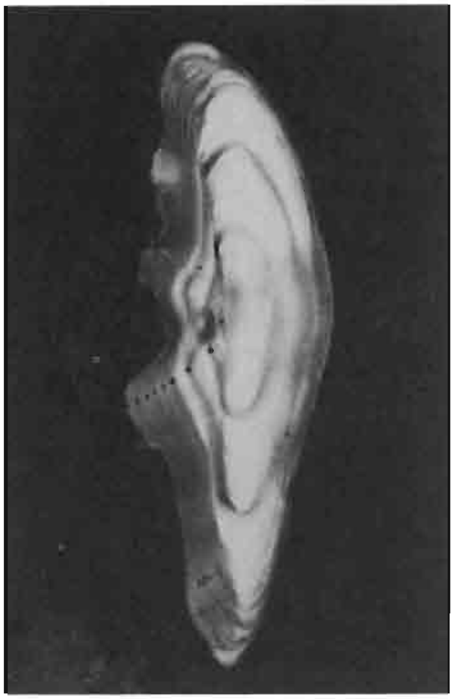

Figure 8

Otolith section of a 28-cm age-8 male American plaice collected in April showing very slow growth, with closely spaced annuli layered on the proximal part of the section.

Figure 10

Otolith section of a 23-cm age 4+ American plaice collected in April showing split zones and checks.

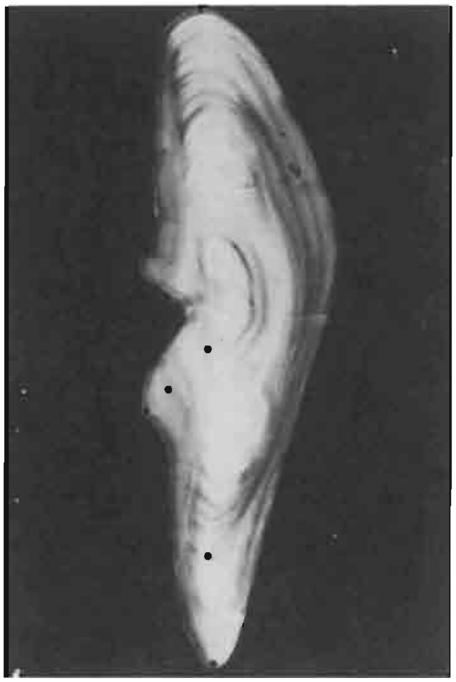

Figure 9

Otolith section of a 25-cm age-3 American plaice collected in April showing a split diffuse second annulus, interpretable near the sulcus.

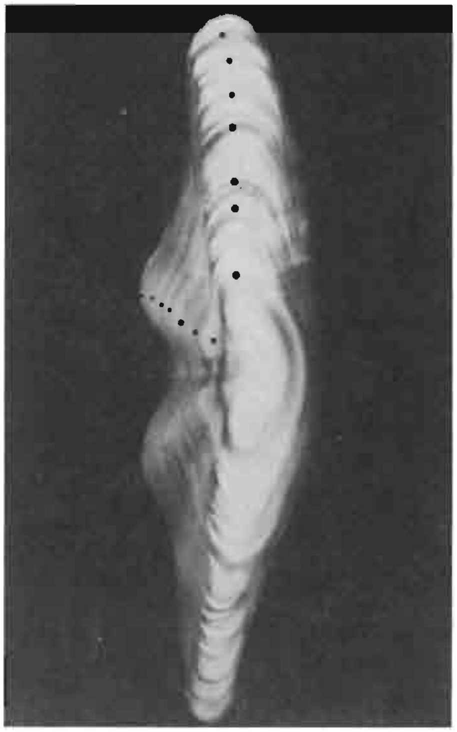

Figure 11

Otolith section of a 34-cm age 7+? American plaice collected in April showing split zones and checks.

Figure 12

Otolith section of a 55-cm age-9 American plaice collected in April showing annuli clearly defined near the sulcus, but becoming more diffuse out to the dorsal edge.

Figure 14

Otolith section of a 63-cm age-13 American plaice collected in May showing annuli more distinct along the dorsal axis than near the sulcus.

Figure 13

Otolith section of a 63-cm age-11? American plaice showing groups of clustered annuli 2-3-4, 5-6-7, and 8-9-10-11.

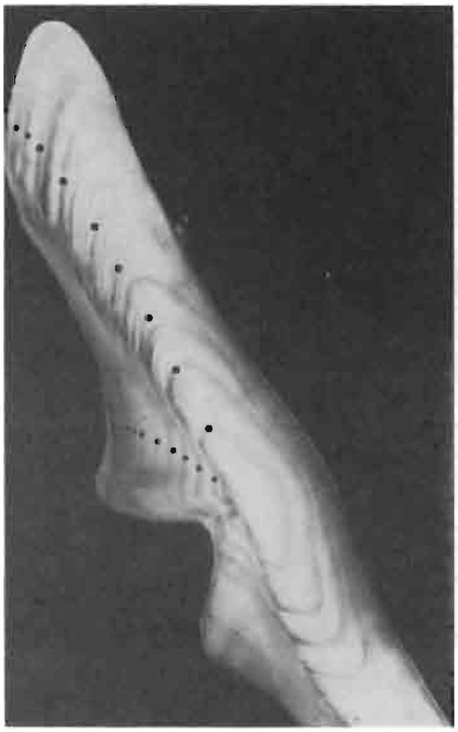

Figure 15

Otolith section of a 56-cm age 9+ or 10+ American plaice collected in October showing weak annuli formed after the fourth annulus.

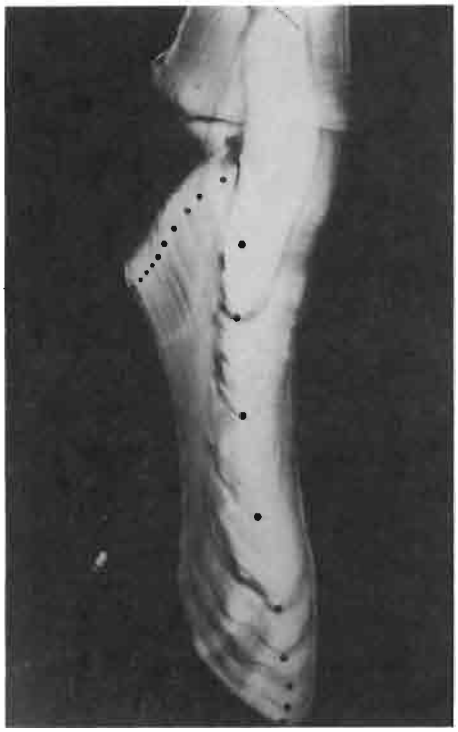

Figure 16
Otolith section of a 56-cm age-10 American plaice collected in April showing weak, diffuse third, fourth, and fifth annuli.

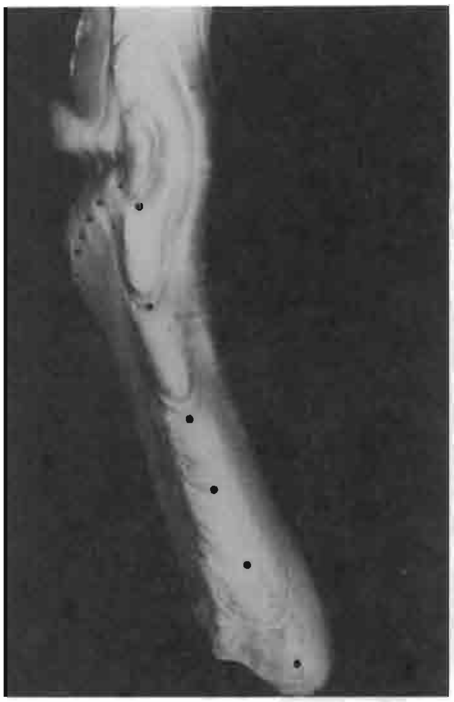

Figure 18
Left otolith section of a 57-cm age-7 American plaice collected in April showing weak, diffuse annuli 4-7.

Figure 17
Otolith section of a 53-cm age 6+? American plaice collected in October showing very indistinct, diffuse annuli, somewhat distinguishable on the distal (bottom) side of the section.

Figure 19
Otolith section of a 33-cm age ? American plaice collected in April showing very weak diffuse annuli.

Figure 20
Dorsal part of an otolith section from a 60-cm age-17(18) American plaice collected in April showing increasingly diffuse annuli out to the dorsal edge.

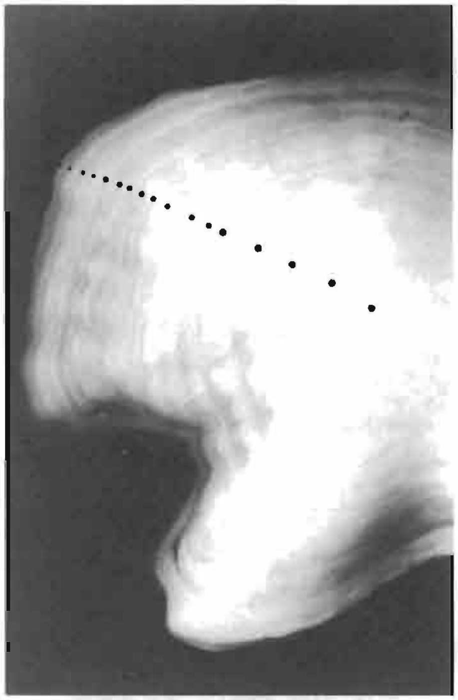

Figure 21
Whole otolith from the American plaice of Figure 20 showing 17 or 18 continuous hyaline zones.

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Yellowtail flounder *Limanda ferruginea*

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Yellowtail flounder are found in the Northwest Atlantic from Labrador to Chesapeake Bay; they prefer sandy bottom at water depths of 37-73 m (20-40 fathoms) (Bigelow and Schroeder 1953). Yellowtail tend to be relatively sedentary although seasonal movements have been documented. In U.S. waters, commercially important concentrations occur on Georges Bank, off Cape Cod, and in the Southern New England-Middle Atlantic region.

The growth rate to age 2 is the same for both sexes, but thereafter females grow faster than males and also live longer (Lux and Nichy 1969). Growth rates differ by geographical area, with fish from Georges Bank generally growing more rapidly than those from other areas. Both males and females become sexually mature at age 2 or 3; males tend to mature at a younger age and at a smaller size than females. Spawning occurs during spring and summer, with peak activity in May. Ages to 17 years have been documented, although individuals more than 7 years old are uncommon. Yellowtail attain lengths of up to 47 cm (18.5 inches) and weights up to 1.0 kg (2.2 pounds).

Historically, scales have been used for age determinations. Royce et al. (1959) and Lux and Nichy (1969) have validated procedures for age determinations based on scales. These procedures give good results over the range of ages normally present in the population (ages 0 to 7).

Scales are removed from the eyed side of the fish along the lateral line immediately anterior to the caudal peduncle for dry storage. About 5 or 6 scales from each fish are impressed on a laminated plastic slide using a roller press and viewed on a microprojector at a magnification of 52× with transmitted light. Regenerated scales are discarded.

The scales are ctenoid and radial grooves extend from the focus to the forward margin of the scale (Fig. 1). Scanning electron microscopy shows the outer surface is sculptured with concentric rings of circuli comprised of individual platelets, while the inner surface is smooth. The spacing of the circuli indicates periods of rapid and slow growth. Rapid summer-type growth is characterized by circuli which are spaced relatively far apart; slow winter-type growth is characterized by circuli spaced relatively close together (Fig. 2).

The annulus is defined as a zone of close winter circuli marking the end of a year of growth, i.e., the winter growth zone. The first annulus is a small, central zone of closely spaced circuli found very close to the focus of the scale (Figs. 1 and 3). The following characteristics, following the criteria of Jensen and Wise (1962) for haddock, also identify annuli on yellowtail scales: an annulus 1) can be traced entirely around the anterior portion of the scale; 2) can be traced, by careful scrutiny if necessary, entirely around the scale; 3) is clearly separated from other such zones, either by a zone of summer-type growth or because of "cutting over" marks on the scale, and does not ordinarily meet other annular zones at any point on the anterior portion of the scale; and 4) if present, is found on all the normal scales from that particular fish. Many of the criteria used for distinguishing annuli on yellowtail scales are also used for scales from other species (e.g., haddock).

By convention, the birthday of all fish in the northern hemisphere is 1 January; therefore, a winter growth zone forming on the edge of the scale is designated as an annulus on 1 January, even though the zone is not complete. Summer-type edge generally forms during spring and summer (Fig. 3) with winter-type growth predominating during fall and winter (Figs. 1 and 4). Older fish begin putting down summer-type edge growth later than younger fish do and start winter-type growth earlier.

A major pattern feature which determines annuli is the number of circuli per unit area. The number increases during slow winter growth and decreases during rapid summer growth. Measurement of the distance between circuli shows relatively wide interspaces during rapid summer growth, and as the interspace decreases, compaction of the circuli forms the broad dark-appearing rings that represent the period of slow winter growth. The end of an annulus (i.e., the last true winter circulus in the winter growth zone) is generally followed by a rapid transition from narrow to widening interspaces, signifying the start of the next period of rapid summer growth. This usually occurs in the spring of the year, but can vary in different geographical areas. Number of circuli per unit area and circuli interspaces are useful in determining the first few annuli, but after the third or fourth annulus, there is a gradual reduction in the number of the circuli formed during a year's growth. This diminishes the usefulness of these criteria. Cutting-over marks are often helpful in determining annuli for older fish.

Checks may be distinguished from annuli by general appearance, relative width, and location. Checks usually begin abruptly, whereas annuli generally have a transition zone showing a relative decreasing of the interspace between circuli before the true winter zone is reached. Absence of a rapid transition to summer growth after the check may also help to distinguish it from an annulus. Checks may also be distinguished by following them towards the sides of the scale to determine if they merge with an annulus to form one zone. This method is generally applicable for only the first few annuli. Later annuli may be too crowded together for easy separation on the sides of the scale. Checks may be stronger on some scales and weaker, or even absent, on others, while annuli are present on all scales from a particular fish. Thus several scales are examined from each fish for a verification of the assigned age. It is sometimes necessary to make two or three impressions, with five or six scales each, before a clear ''composite'' picture of the fish's growth can be determined.

Spacing of the annuli relative to each other and to the focus of the scale may be used to differentiate between annuli and checks. For example, if two winter growth zones are found relatively close together on a scale from a younger fish, and all the other winter zones on the scale are relatively far apart, then one of the two close zones will probably be called a check and not an annulus. This type of annulus construction (i.e., two close zones) is generally described as a split annulus, since it is usually difficult to determine which zone is the check and which is the annulus (Fig. 3). On scales from older fish, annuli formed after the third or fourth year are expected to be fairly close together.

General patterns based on the geographic origin of the fish are also used as an aid in identifying annuli. For example, a characteristic check, called the third summer check, is often apparent in the spring or summer growth zone of the third year of life for fish from Southern New England (Figs. 4 and 5). This check is generally strong on scales from fish from the Southern New England area, but it is usually weak or absent on scales from fish from the eastern part of Georges Bank (Figs. 6 and 7). Southern New England fish also grow more slowly than do fish from other areas so that annuli formed after the third year are composed of few circuli and are very close together. Cutting-over marks are often helpful in determining annuli for older Southern New England fish. A rule of thumb for older fish from this area is to count as annuli all ''possible'' zones delineated by cutting-over after the fourth annulus, especially if the specimen is a male. Females from this area, being faster growing, show slightly better separation of annuli up until the fifth or sixth annulus, after which annuli are closely compacted on the scale.

Fish from the southwestern area of Georges Bank generally show only a weak third summer check, if it is present at all. Because of their more rapid growth rate, their scales show a better separation of annuli which are fairly distinct through the sixth year of life for both sexes. (Note the pronounced alteration of summer and winter growth zones in Figures 6 and 7).

Fish from the northern and eastern parts of Georges Bank generally lack the third summer check and, because of their rapid growth rate, also have distinct, well-separated annuli (Fig. 8). Sometimes, however, growth is so rapid that winter zones do not contain closely spaced circuli and consequently annuli are often indistinct. With these fish it sometimes helps to actually move back from the microprojector screen to gain a better perspective of the overall pattern of growth when assigning an age.

Yellowtail taken on the Cape Cod grounds generally show slower growth in the first two years, followed by more rapid growth in the third and fourth years. There may sometimes be a distinctive check immediately after the second annulus (Fig. 9).

Yellowtail from the Browns Bank region on the Scotian Shelf grow even more slowly in the first three years of life, although growth in the fourth through sixth years is more rapid. A gradual slowdown in growth is evident in subsequent years (Fig. 10).

A consistent problem involves distinguishing checks caused by damage or injury to the fish. In these cases, the scale is physically shifted in the scale pocket so that subsequent circuli are not quite in line with previous circuli and ''lost'' circuli and irregular spaces result. Circuli in the damaged area may disappear when an attempt is made to follow them around the scale. These marks on the scale correspond with the area of regeneration on lost scales. The effect is similar to that of cutting-over, a condition caused by erosion of the scale edge, and can create a good deal of confusion in determining annuli. One way to distinguish between damage and cutting-over marks is to identify marks occurring at the end of a winter zone. If this occurs, then it can usually be assumed to be cutting-over (Fig. 8).

Another major problem is in determining the type of growth present on the edge of the scale. The thinness of the scale edge often results in an impression with a light coloration that may appear to be summer edge (Fig. 5). However, the only true difference between summer and winter edge is seen in the relative spacing between circuli. A simple way of improving impressions to lessen this optical problem is by angling the upper roller of the press. This applies slightly more pressure to the edge of the scale.

The most reliable general criterion for distinguishing checks from annuli for yellowtail scales is the relative location of the annuli. In first looking at a scale, an attempt is made to mentally superimpose a regular growth pattern, based on prior knowledge of typical patterns for the geographic origin of the fish. Any zone not fitting the pattern is closely scrutinized to determine if it is a check or a split (Fig. 7). Particular year-classes may also exhibit peculiar growth characteristics which assist in determining age. Some year-classes may exhibit a certain split annulus, or a strong check between two particular annuli, or perhaps two close annuli. Recognition of a characteristic growth pattern for a difficult specimen may be used to help assign the most probable age for the fish.

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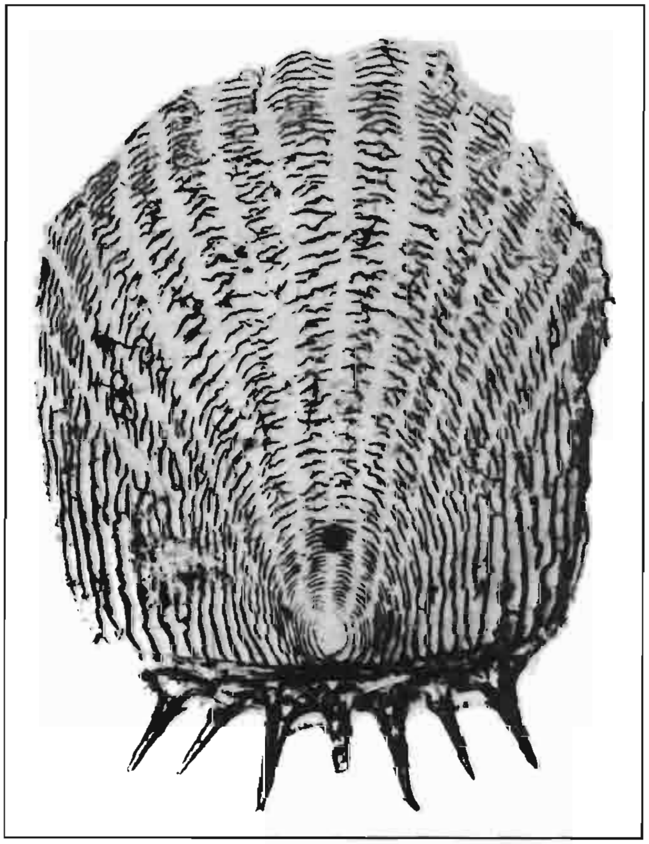

Figure 1
Scale impression from a 20-cm age-1 immature yellowtail flounder collected in the fall from Southern New England, with winter edge just beginning to form.

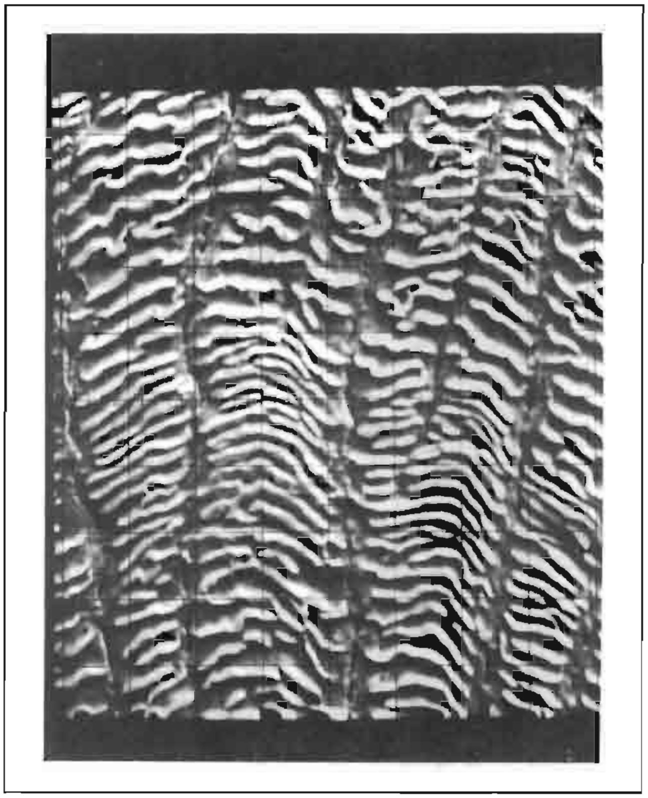

Figure 2
Scanning electron microscope photograph of an actual yellowtail flounder scale (magnification 166×) showing the difference between summer and winter platelets.

Figure 3
Scale impression from a 12-cm age-1 immature yellowtail flounder collected in the spring from Southern New England showing a split first annulus, with summer edge forming.

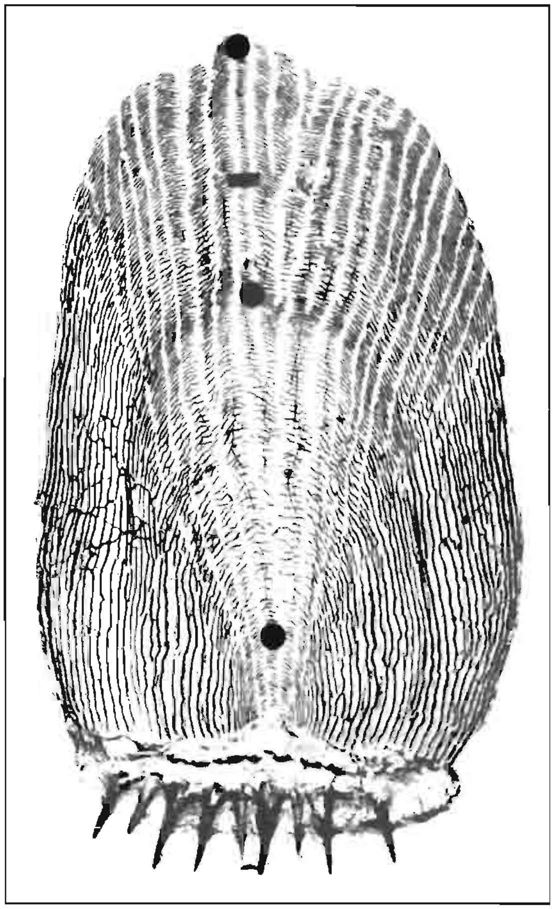

Figure 4
Scale impression from a 30-cm age-3 female yellowtail flounder collected in the spring from Southern New England showing a third summer check.

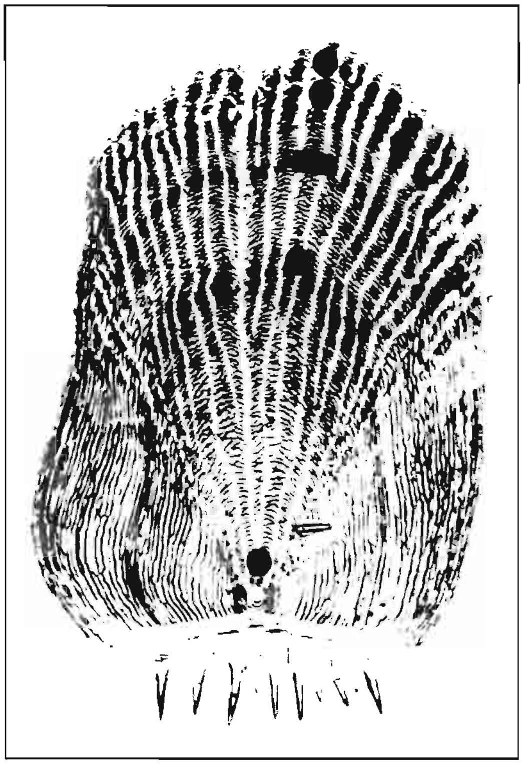

Figure 5
Scale impression from a 32-cm age-4 male yellowtail flounder collected in February from Southern New England showing a strong third summer check, with possible summer edge forming.

Figure 6
Scale impression from a 38-cm age-3 female yellowtail flounder collected in the fall from southwestern Georges Bank showing well-separated and defined annuli, with no third summer check.

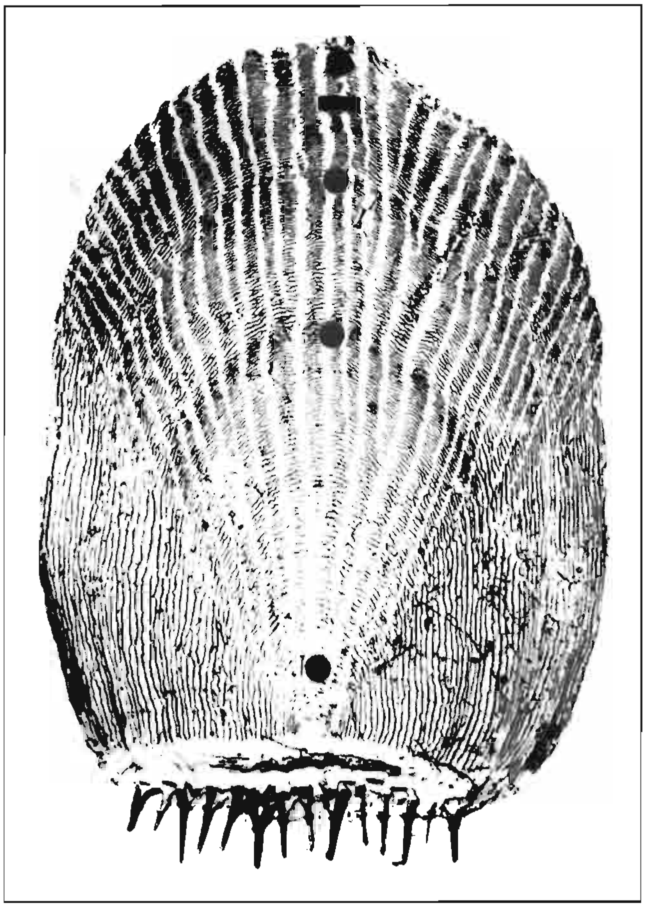

Figure 7
Scale impression from a 38-cm age-4 male yellowtail flounder collected in October from southwestern Georges Bank showing a split fourth annulus, with no third summer check.

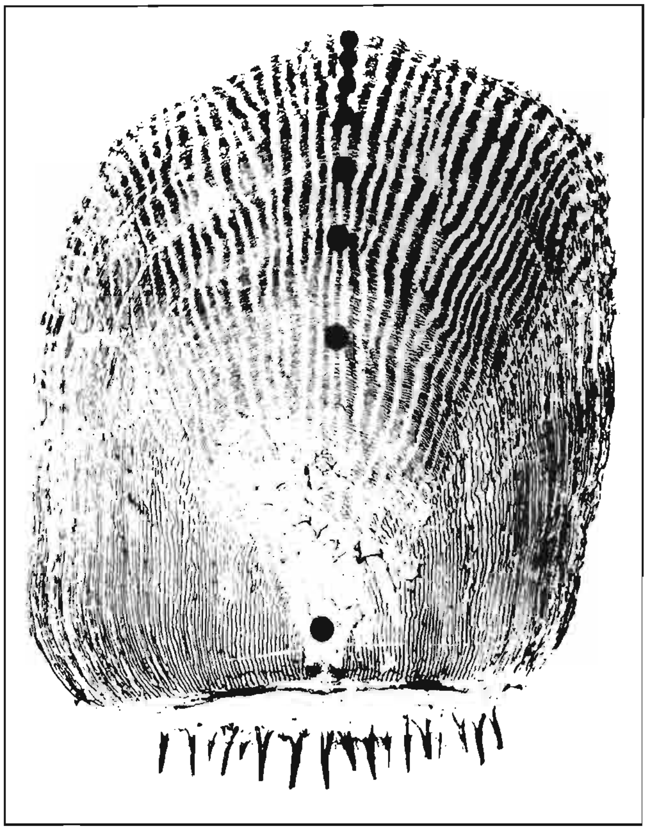

Figure 8
Scale impression from a 54-cm age-8 female yellowtail flounder collected in May from northern Georges Bank showing distinct annuli with strong cutting-over.

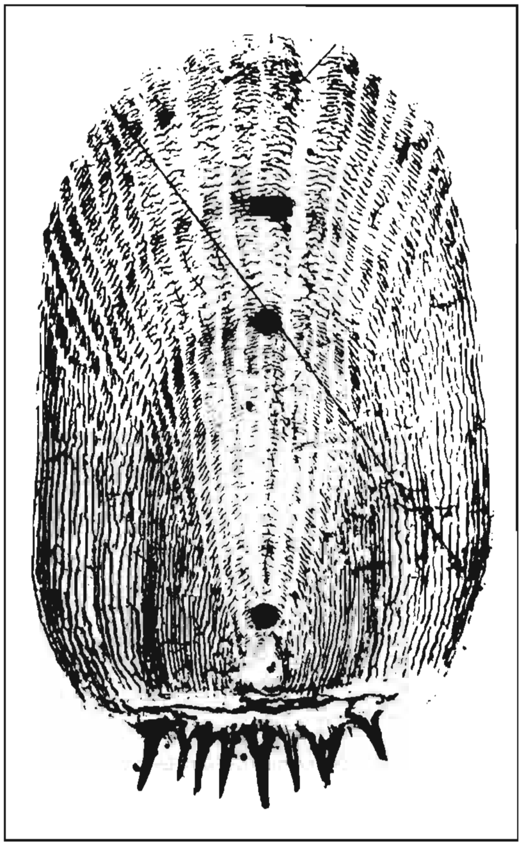

Figure 9
Scale impression from a 31-cm age-2 yellowtail flounder collected in November from the Cape Cod area showing the distinctive check after the second annulus.

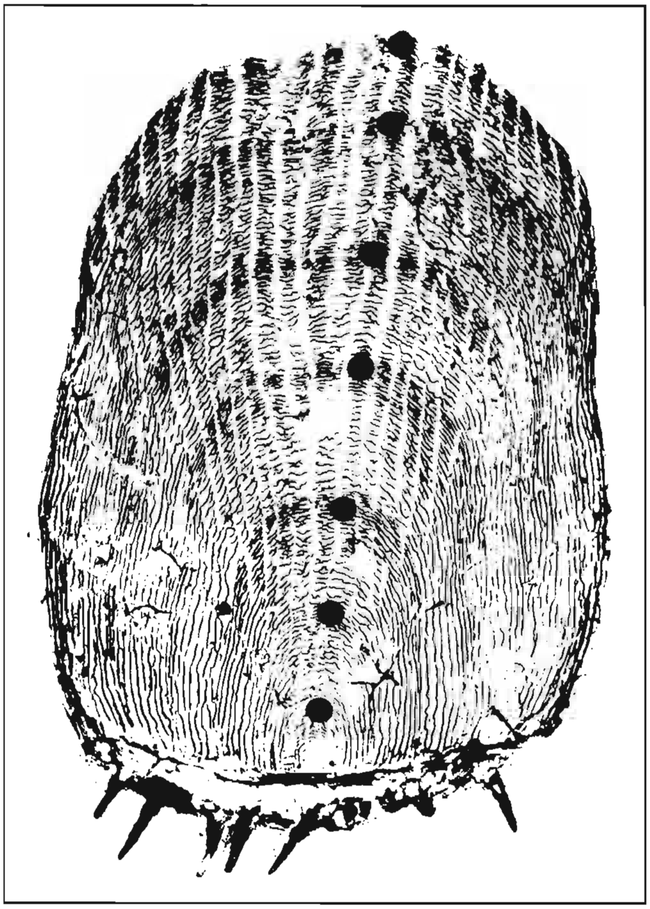

Figure 10
Scale impression from a 36-cm age-7 male yellowtail flounder collected in the spring from Browns Bank showing the close first, second, and third annuli typical for this area.

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Surf clam
Spisula solidissima

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Surf clams are found from the Gulf of St. Lawrence to Cape Hatteras (Ropes 1980). In the Middle Atlantic Bight, where the resource is extensive and an active fishery exists, this mactrid occurs from the coastal beach zone to depths of over 60 m. Off New England, surf clams are found along nearshore ocean beaches, on shoals off Nantucket and Martha's Vineyard, and on Georges Bank.

Surf clams are of separate sexes, although some individuals are hermaphroditic (Ropes 1982). In the Middle Atlantic Bight, spawning occurs primarily during summer, although some activity has also been documented in autumn (Ropes 1968). Sperm and eggs are released into the environment where fertilization and larval development occur. Full sexual maturity is attained in the second year of life at a shell length of 45 to 85 mm (Ropes 1979). Growth is fairly rapid to about age 7, but diminishes thereafter (Fig. 1). A maximum shell length of 226 mm and longevity estimate of 37 years have been reported for surf clams (Ropes and Jearld 1987).

Belding (1910) reported the earliest age information for surf clams based on observations that rings or bands on the external valve surface probably form annually. Mean shell length at age values were reported for surf clams ≤7 years old. The method was extended to studies of surf clams at Prince Edward Island, Canada (Kerswill 1944), off Long Island, New York (Westman and Bidwell 1946), off central New Jersey (W.R. Welch, Maine Dep. Mar. Resour., W. Boothbay Harbor, ME 04575, pers. commun. 1963), off Buctouch, Canada (Caddy and Billard 1976), and off Virginia (Loesch and Ropes 1977). An age of 17 years was the oldest reported in these studies, but none of the shell-length measurements exceeded 163 mm (Ropes 1980). Age was not determined for larger clams, because early rings on the valves were obliterated by erosion, and later rings became too crowded together at the valve margin for definite separation. Since significant numbers of clams >163 mm are found in natural populations (Ropes and Merrill 1976), the method is not generally applicable.

Bivalves have been found to deposit specific internal microstructures annually (Rhoads and Lutz 1980). These are considered to be relatively unaffected by external conditions and can be critically examined by microscopic enlargement. Therefore, methods were developed for exposing and examining such deposits in the shells of surf clams. In 1975, procedures were developed at Oxford, Maryland, for sectioning whole valves from the umbo to valve margin using a diamond-impregnated sawblade. The cut edges were then polished to remove saw marks and enhance the age-growth structures. Distinctive dark lines seen in the cut edges of the valves terminated at external rings. The annual periodicity of these lines was validated by marking experiments (Ropes and Merrill 1970, Jones et al. 1978). Although the method was reliable, age determinations required careful microscopic examination of the cut surface, which, together with the cutting and polishing procedures, proved to be excessively time-consuming.

A more efficient method has been developed based on similarity between the number and relative location of annuli in the valve and chondrophore (Ropes and O'Brien 1979). A linear correspondence has been found between chondrophore and valve growth ($r = 0.97$). The preparation of a chondrophore for examination includes excision of the chondrophore from the valve by a pair of diamond-impregnated blades, gluing the excised chondrophore onto a slide, and production of a thin-section (0.25 mm thick) using an Isomet low-speed saw.

Age determinations are conducted under transmitted light at 50-100×. Wetting agents on the surface of the sections are unnecessary. A television/microscope monitoring unit also provides adequate resolution of most sectioned chondrophores and has the advantage of permitting examinations by several viewers. Such devices are invaluable for training age readers and resolving age determinations of difficult specimens. In these examinations, light is transmitted through the translucent age annuli and blocked by the opaque growth increments, producing alternate zones of white and black, respectively. The exact opposite occurs in a photographic print (Fig. 2).

Bivalves may alter construction of their shells to form annular marks because of extrinsic or intrinsic factors (Lutz and Rhoads 1980). Low winter temperatures have been cited as the cause for annulus formation in several bivalves, including Stimpson's surf clam, *S. polynyma* (Feder et al. 1976). For the surf clam, annuli may form in response to spawning stress (Jones et al. 1978). Anaerobic conditions reportedly contribute to annulus formation in the northern quahog, *Mercenaria mercenaria*, and the ocean quahog, *Arctica islandica* (Lutz and Rhoads 1980).

Jones (1980, 1981a) investigated age-growth phenomena of surf clams and examined shells under a scanning electron microscope. He identified specific microstructural elements constituting the age annuli and growth increments in the outer shell layer and found that very fine layers in the shells of surf clams had no subdaily, daily, or tidal periodicity. Only annual layers were formed with a consistent periodicity.

The aforementioned research provides the basis for making routine age estimates from thin-sectioned surf clam chondrophores. Since age readers customarily use the term "hyaline zones" for age-mark determinations of other animals, this term is used herein to describe and identify the translucent age annulus or portions of an annulus in surf clam chondrophores. Three types of annuli have been recognized: the first annulus formed near the umbo, those formed during the next 9 years or so, and those formed from the 10th year or so onward. This separation is based on the apparent thickness of hyaline zones and a repetitive accretion of hyaline zones in some annuli. Age readers are instructed to count annuli beginning at the distal growing edge of a chondrophore, since it has been found that counts initiated at the umbo often resulted in an underestimation of age.

Environmental conditions at the sample location may influence the time of annulus formation. Jones (1980) found developing annual marks during late summer-early fall for specimens collected off New Jersey. Our observations confirm these data with the exception that clams from off Delmarva Peninsula form the annulus later in the fall, i.e., from October to December. These geographic differences in time of annulus formation may create confusion in age interpretation. A difficulty arises with the designation of 1 January as the standardized birthdate, since all annuli formed in early fall may show that substantial growth had occurred before the January birthdate. Therefore, caution must be exercised assigning an additional year of age due to finding a hyaline zone at the distal edge of chondrophores collected between the time of annulus formation and 1 January. This procedure assumes an annulus is formed in the first few months after larvae settle to the bottom.

The first annulus is usually a single, relatively narrow hyaline zone (Fig. 2a). Distance from the umbo to the most distal edge of the first hyaline zone is often variable. This may occur from improper technique during the cutting or grinding operations. More typically, variation may result from an annual variation in timing of larval production and settlement due to protracted spawning activity. One to three-month periods of peak spawning activities have been reported by Ropes (1968) and Jones (1981b), with some lower levels of spawning before and afterwards. This interpretation, however, does not take into account possible differences in growing conditions at the place of settlement.

The second through about the tenth annuli may be characterized by alternating weak and strong hyaline zones separated by narrow opaque bands (Fig. 3). The terms "strong" and "weak" relate to the relative thickness of the hyaline zones and degree of light transmission through the zones. Double hyaline zones comprising an annulus are delimited from preceding and subsequent annuli by wide opaque bands. A reduction and increase in the growth rate during the formation of an annulus are suggested by this alternative pattern of zones and bands.

The hyaline zones and opaque bands of subsequent annuli are greatly compressed, although variation in growth between annuli may occur (Fig. 3). Each distinct hyaline zone is counted as an annuli. The more compressed pattern of these annuli is suggestive of a reduction in the growth rate.

Occasionally chondrophores have incomplete hyaline zones, particularly in the case of annuli formed after the tenth year of life. These are narrow hyaline zones in the middle of a chondrophore that fail to clearly extend to the lateral edges (Fig. 3). They are categorized as growth checks.

Although patterns of annular growth are similar for most areas that have been sampled, others have unique characteristics. Surf clams from Nantucket Shoals have much more diffuse hyaline bands. The bands are not discrete groups sharply delineated by opaque zones but tend to split more frequently and blend together. The dynamic environment of this area may create conditions which are not conducive to consistent deposition of annular material. Inshore and offshore samples along the Middle Atlantic coast also exhibit different growth patterns (Jones et al. 1978) and consequently the annuli pattern varies. In this area, though, the rings are defined well enough to age.

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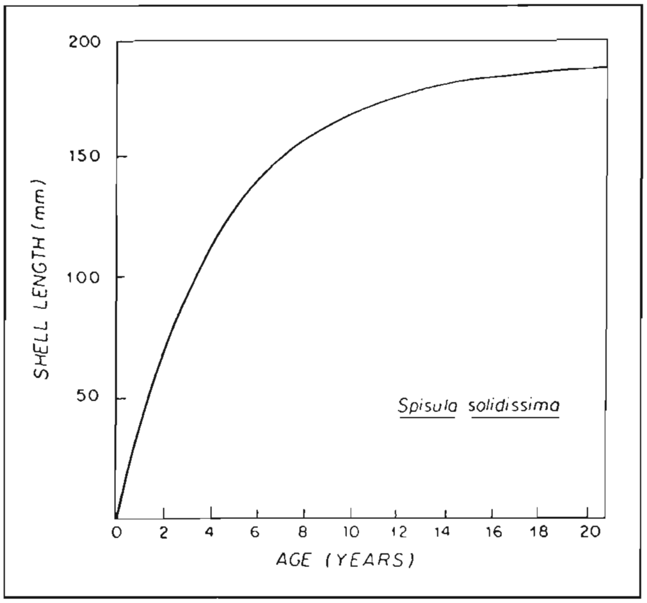

Figure 1

Relationship between age and growth in surf clams.

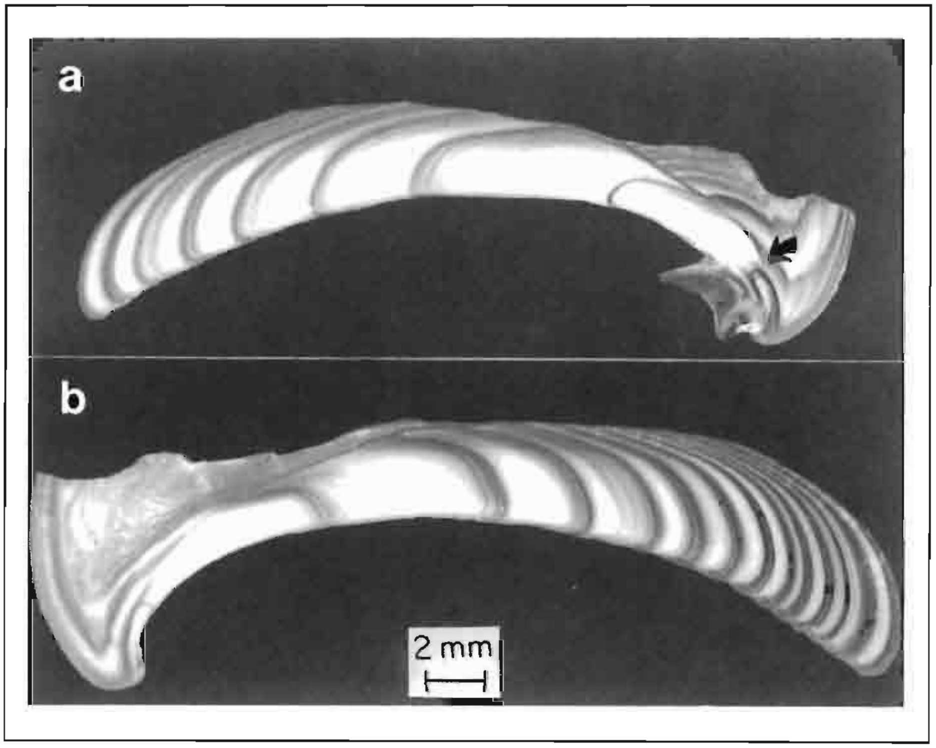

Figure 2

Photographic enlargements of thin-sectioned chondrophores from surf clams: (a) 139 mm (shell length), age 8; and (b) 137 mm (shell length), age 13. The first annulus formed in the life of a surf clam is sometimes faint (arrow indicates a bold annulus in the chondrophore of the upper clam). The most recent annulus at the marginal edge of these chondrophores was not completely formed.

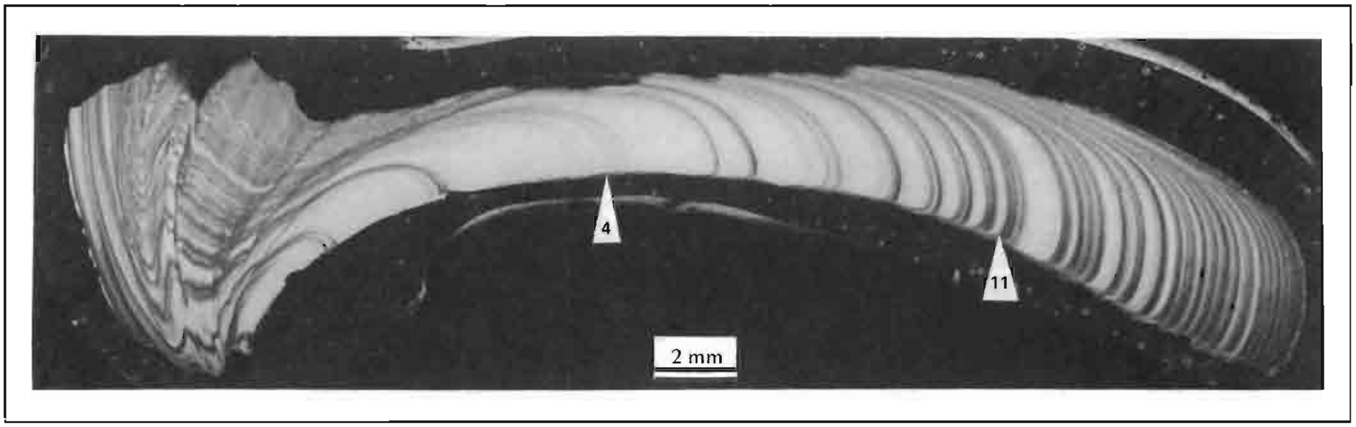

Figure 3
Photographic enlargement of the thin-sectioned chondrophore of a 175-mm (shell length), age-32 surf clam.

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Ocean quahog
Arctica islandica

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The geographic range of the ocean quahog is extensive. This arcticid occurs along the east coast of North America north of Cape Hatteras to St. George's Bay, Newfoundland, Canada, off the southern coast of Iceland, off the Faroe and Shetland Islands, and along the European coast northward from the Bay of Cadiz, Spain, around the British Isles, in the North Sea, and off the Norwegian coast to the White Sea in Russia (Merrill and Ropes 1969, Ropes 1979). Off the Middle Atlantic coast it is common at depths of 35 to 75 m.

The sexes are separate, although hermaphroditism may occur (Mann 1982). A period of intense spawning from August into November has been found for ocean quahogs, although minor spawning activity has been observed in earlier and later months (Loosanoff 1953, Jones 1981, Mann 1982). Sperm and eggs are released into the environment where fertilization and larval development occur. In samples from off Long Island, NY, the youngest ocean quahogs that had attained sexual maturity were an age-5 male and an age-6 female (Ropes et al. 1984b). Growth of ocean quahogs is fairly rapid during the first 20 years of life but lessens greatly thereafter (Murawski et al. 1982, see Figure 1). Ocean quahogs of about 100 years and older are common; a maximum shell length of 140 mm (5.5 inches) and a maximum longevity estimate of 225 years have been reported (Ropes 1985).

Murawski et al. (1982) reviewed early studies that presented largely unsubstantiated age and growth observations for ocean quahogs. Earlier investigators interpreted dark concentric rings or bands found on the external valve surface of small quahogs ($\leqslant$60 mm shell length) as annual marks. Larger, older quahogs were not aged because the rings crowded together at the valve margin and became obscured by the thick, black periostracum.

Recent age determinations at the Woods Hole Laboratory have been based on enumeration of annuli in acetate peel preparations (Thompson et al. 1980a,b; Jones 1980; Ropes et al. 1984a,b). In light microscope examinations of these acetate peels, outer and inner layers of the three-layered aragonitic ocean quahog shell are quite obvious, unlike the very thin prismatic pallial myostracum, which separates the outer from inner layers. Annuli occur in the relatively thick outer valve layer, curve toward the umbo from exit locations at valve surface bands, and seem to merge with the prismatic pallial myostracal layer. Annuli in peels appear as dark lines; growth increments form a lighter, textured background (Figs. 2 and 3). Definite prismatic microstructures, considered to be annuli, were found by investigators at Princeton University (Thompson et al. 1980a, Jones 1980) that separated growth increments from predominantly homogeneous microstructures. Although the microstructures are only visible by scanning electron microscopy (Ropes et al. 1984a), light microscope examinations of acetate peels clearly revealed the periodicity of annuli in small and large marked quahogs (Figs. 2 and 3).

The left valves of ocean quahogs are prepared, since they have a single tooth that contains age marks, and correspondence in the number of marks in the tooth and valve adds confirmation to an age estimate for a specimen. The valves are sectioned by a diamond-impregnated blade on an Isomet slow-speed saw machine. A valve is oriented on the machine to make a cut through the umbo and to the ventral margin such that the broadest surface of the tooth remains in the anterior valve portion. This portion is immersed in bleach (sodium hypochlorite ~ 5.25%) to remove the periostracum, rinsed in tapwater, and allowed to dry before embedding it in Epon 815 resin. After hardening, the embedded valve cut surface is exposed by grinding off excess resin and polished to a high luster on a vibrating lap machine. Etching the cut surface of the valve for 1 minute with 1% HCl preceeds application of sheet acetate and acetone. The sheet is peeled off after the acetone evaporates. The image produced in the peel is a necessary procedure, since the thin-age annuli are microscopically indistinct on the external valve surface, in the cut surface, or thin-sections of ocean quahog shells. Although age annuli and growth increments are reproduced clearly in the peels, they must be examined microscopically. Optimal contrast between annuli and growth increments in examinations of peel preparations is possible under a compound microscope at low (40×) magnifications, low transmitted light intensity, and with the iris/diaphragm of the substage condenser closed down.

Various experimental evidence, including radiometric analyses (Turekian et al. 1982, Bennett et al. 1982), suggests that annual age marks are formed in the valves of ocean quahogs. Validation of an annual periodicity for these marks has been supported by a marking experiment (Murawski et al. 1982). Recovered individuals show the expected number of annuli formed during the period between marking and recapture (Figs. 2 and 3).

Problems in determining an age for an ocean quahog relate to the loss of the earliest-formed annuli in the valve from erosion of the outer valve layer, a condition not uncommon in old individuals. Annuli formed during the first 10-15 years in the life of an ocean quahog may split into multiple lines at the valve-surface exit locations. Careful observation will usually reveal that they merge at the pallial myostracum. These conditions can result in deviations in agreement between annuli counts of the valve and hinge tooth, and individuals have been found to have a confusing pattern of growth lines suggestive of aberrant growth (Ropes et al. 1984b). The labor-intensive preparation of acetate peels and ages approaching or exceeding 100 years for many ocean quahogs are additional problems.

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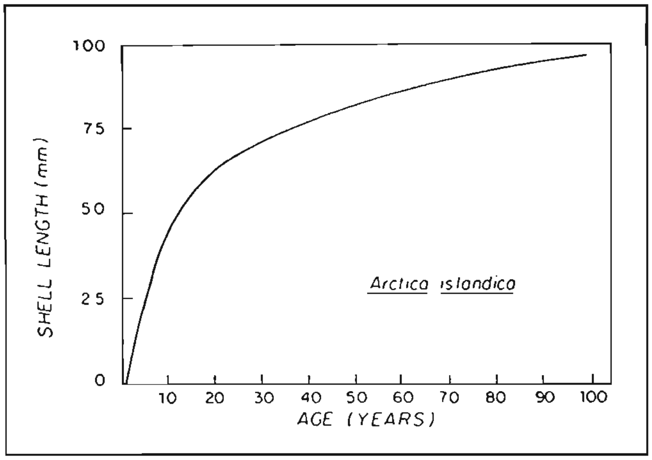

Figure 1
Relationship between age and growth in ocean quahogs.

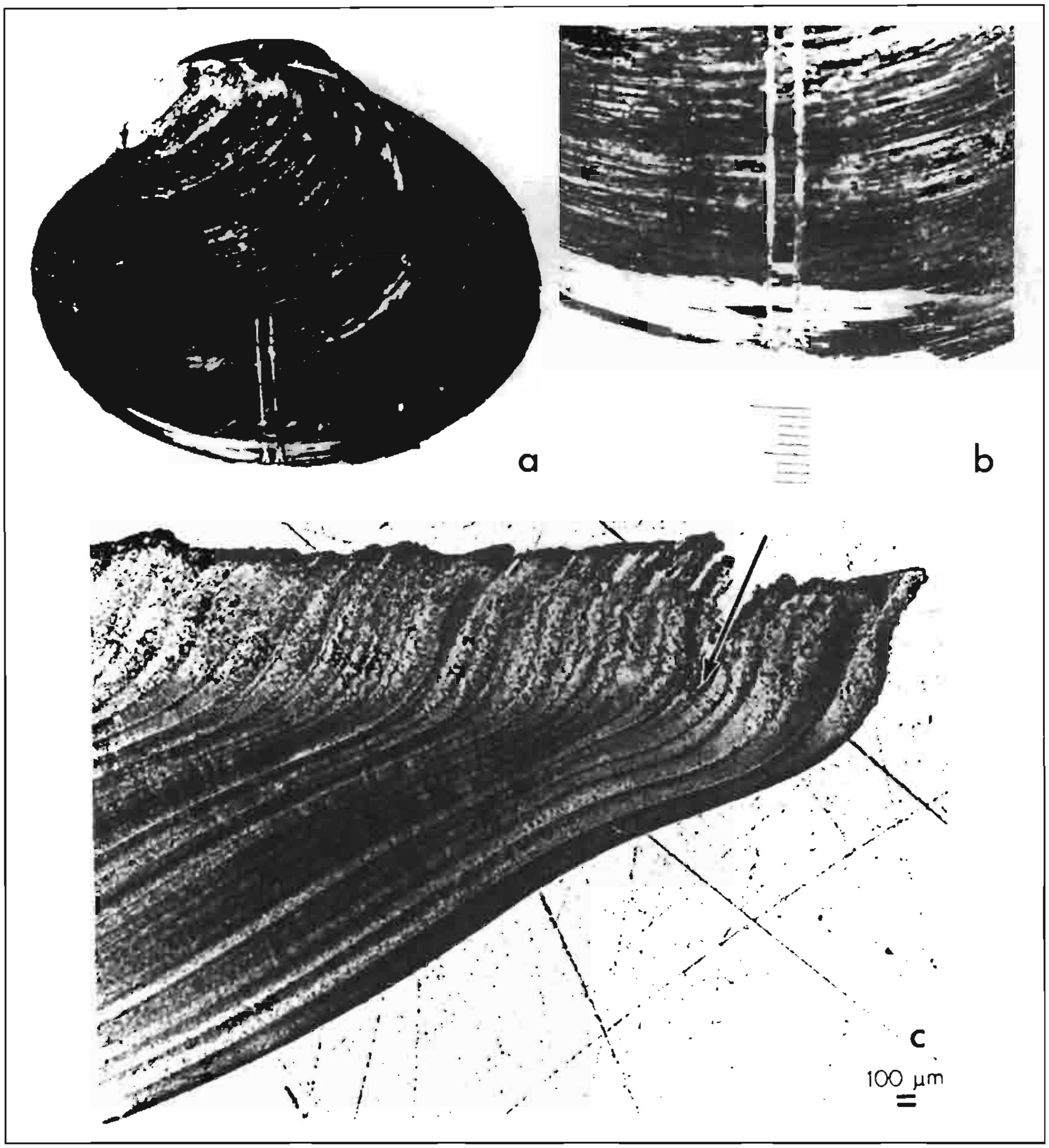

Figure 2

(a) A 110-mm (shell length), age-125 ocean quahog, *Arctica islandica*, released after marking in 1978 off Long Island, NY, and recovered 6 September 1983 before the annulus had formed for that year; **(b)** enlargement of the marked area; **(c)** photomicrograph of the valve margin showing the annulus formed soon after marking (arrow) and four additional annuli.

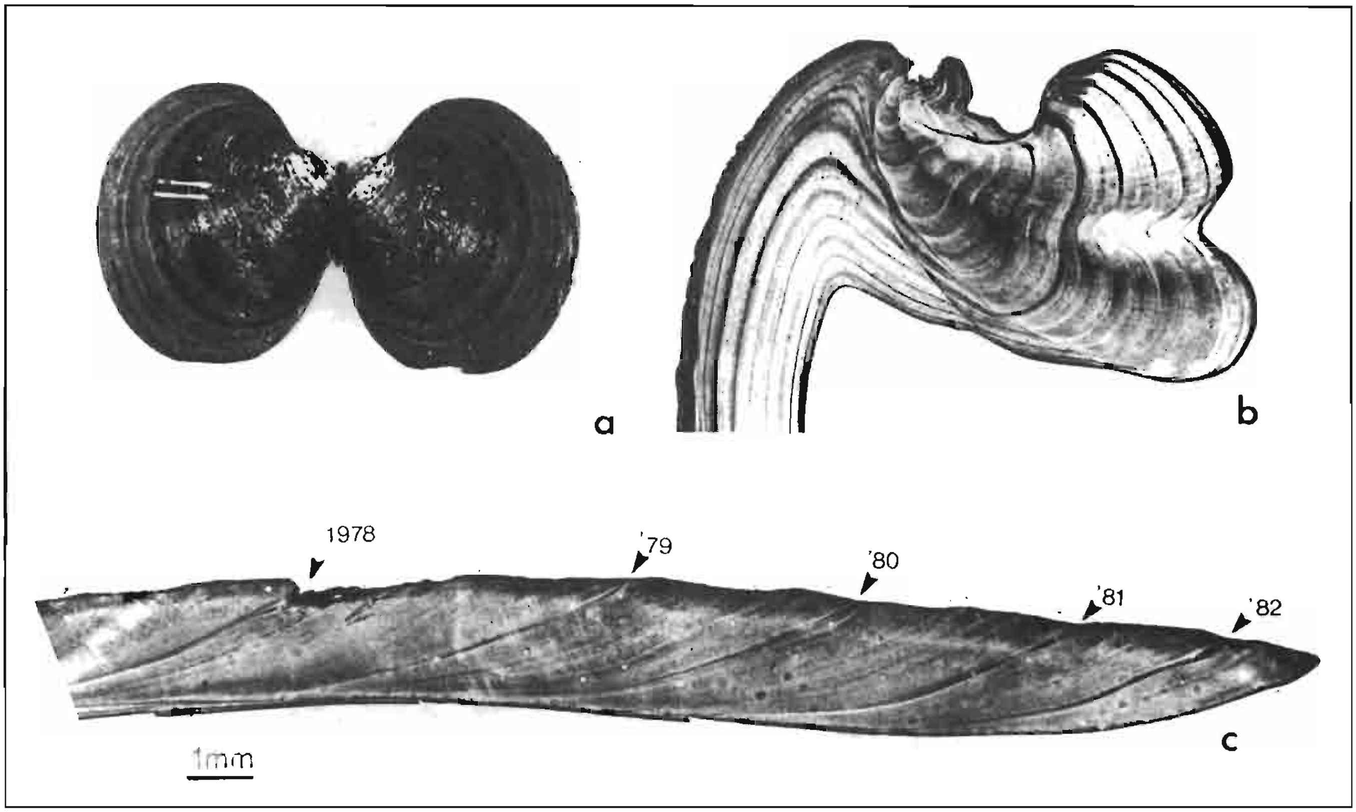

Figure 3

(a) A 62-mm (shell length), age-11 ocean quahog, *Arctica islandica*, released after marking in 1978 off Long Island, NY, and recovered 6 September 1983 before the annulus had formed for that year; (b) hinge tooth showing 11 annuli; (c) photomicrograph of the valve margin with each annulus identified by the year of its formation.

APPENDIX A
Age-structure preparation: Equipment and supplies

Fish processing

Coin envelopes for age samples

#1 ungummed, flap extended, printed.
Available from
Crest Envelope
2125 Highlands S.E.
Kentwood, MI 48508
or
Massachusetts Envelope Co.
30 Cobblehill Rd.
Sommerville, MA 02143

Thin-sectioning

Isomet low-speed saw with swivel-arm assembly and flanges.
Available from
Buehler Ltd.
41 Waukegan Road
P.O. Box 1
Lake Bluffs, IL 60044
Specifications or model numbers:
Isomet low-speed saw #11-1180
Swivel-arm assembly #11-1181
Chuck #11-1187
Flanges 2½″ diameter, recessed, set of two, #1180-S48

Diamond blades.
Available from
Norton Company
Worcester, MA 06106
(also available through local distributors)
Size and specifications of those currently used:
FO565977 Norton Diamond blade
3 × 0.006 × ½ Type 1A1
Blueprint ME 120929
M4D 10/20M1-N50M9-1/8

Paraseal Wax for canning and candles. Available in local markets.

Dennison Marking Tags - white, No. 12-105-1.
Available from office supply stores.

Carbon Decolorizing, Nuchar S-N (from coal) C-177.
Available from Fisher Scientific Co. and other scientific suppliers.

Calcium Oxide Powder, Technical C-114.
Available from Fisher Scientific Co. and other scientific suppliers

Scotch double-sided tape. Available from office supply stores.

Glycerin. Available from chemical supply companies.

Clear, liquid dishwashing detergent. Available from local markets.

Adjustable-temperature hot plate, Thermolyne, Type 1900, #HP-A1915B, 115V AC 700W.
Available from scientific suppliers.

Double boiler or egg poacher. Available from houseware stores.

Impressing scales

Jeweler's press, flat rolling hand mill.
Available from
Wm. Dixon Company
Carlstadt, NJ 07072

Cellulose Acetate plastic, 0.040 inch thick, 22×52″ sheets.
Available from
Commercial Plastics Co.
21 Main Street
P.O. Box 24
Cherry Valley, MA 01611

Laminated plastic
Curform Grade 7781, 8.0 mil polyester/0.1 mil saran/1.25 mil polyethylene.
Curform Grade 7770, 8.0 mil polyester/0.1 mil saran/0.75 mil surlyn.
Both available from
Curwood Inc.
718 High Street
New London, WI 54961
Curlam Grade 7780, 7.5 mil polyvinyl chloride/2.0 mil surlyn.
Curlam Grade 7710, 7.5 mil polyvinyl chloride/2.0 mil polyethylene.
Both available from
Tenneco Chemicals
Nixon Lane
Nixon, NJ 08817

Embedding otoliths

Permount, SP15-500. Available from Fisher Scientific and others.

Xylene X-5. Available from Fisher Scientific and others.

Molded plastic otolith trays, 10×5, circular depressions, jet black color.
Available from
Can-Am Containers
P.O. Box 340
Spring Hill, Nova Scotia B0M 1X0
or
Sheepscots Machine Works
P.O. Box 406
Boothbay, ME 04537

Baking otoliths

Toaster oven (any type that will heat to 550°F.)
or
Scientific oven, Dryco 20W 115V, or other types may also be used.
Available from
Dryco Products Inc.
65-37 Fresh Meadow Lane
Fresh Meadow, NY 11365

Bivalve processing

Raytech 10″ slab & trim saw, model #AL-P105, catalogue #20-023.
Available from
Raytech Industries, Inc.
P.O. Box 6
Stafford Springs, CT 06075

Diamond blades
For rough cuts: Red Blazer 10×028×038×⅝×½, cat.#33-090.
For fine cuts: Red Blazer 4×012×020×½ 4″ diam. cat.#33-061
or Red Blazer 5×012×020×½ 5″ diam. cat.#33-069.
Available from
Raytech Industries, Inc.
P.O. Box 6
Stafford Springs, CT 06075

Di-acetate sheets 19×24×0.005″ thick for acetate peels.
Available from
Commercial Plastics and Supply Corp.
352 McGrath Highway
Sommerville, MA 02143

Epon 815 and DTA hardener.
Available from
Miller-Stephenson Chemical Co., Inc.
P.O. Box 950
Danbury, CT 06810

APPENDIX B
Age-structure examination: Equipment and supplies

Kodak Photo-Flo 200 Solution.
Available from local photo supply stores.

Microprojectors

Contour bench projector with 12″ diameter screen.
  Cat.#900-377 - Leitz TP 300 Contour Projector
  Cat.#851-515 - 20:1 objective magnification
  Cat.#851-516 - 50:1 objective magnification
    Available from: local Leitz distributors

Microfiche reader #102568, ABR 914; #123599 dual lens accessory;
  #123645 lens; #123462 lens.
Available from
  Bell & Howell Company
  45 4th Avenue
  Needham Heights, MA 02194

Binocular stereomicroscopes

Leitz/Wild M3, M5, M8 stereomicroscopes.
Available from local Leitz/Wild distributors.

TV Camera & Monitor

S-NC65SC high resolution Newvicon camera
S-14VM993 Audiotronics 19″ monitor
S-BNC101 10′ cable
S-3665 Parfoclizer
Available from
  Spectra Instruments
  235 Great Road
  Littleton, MA 01460