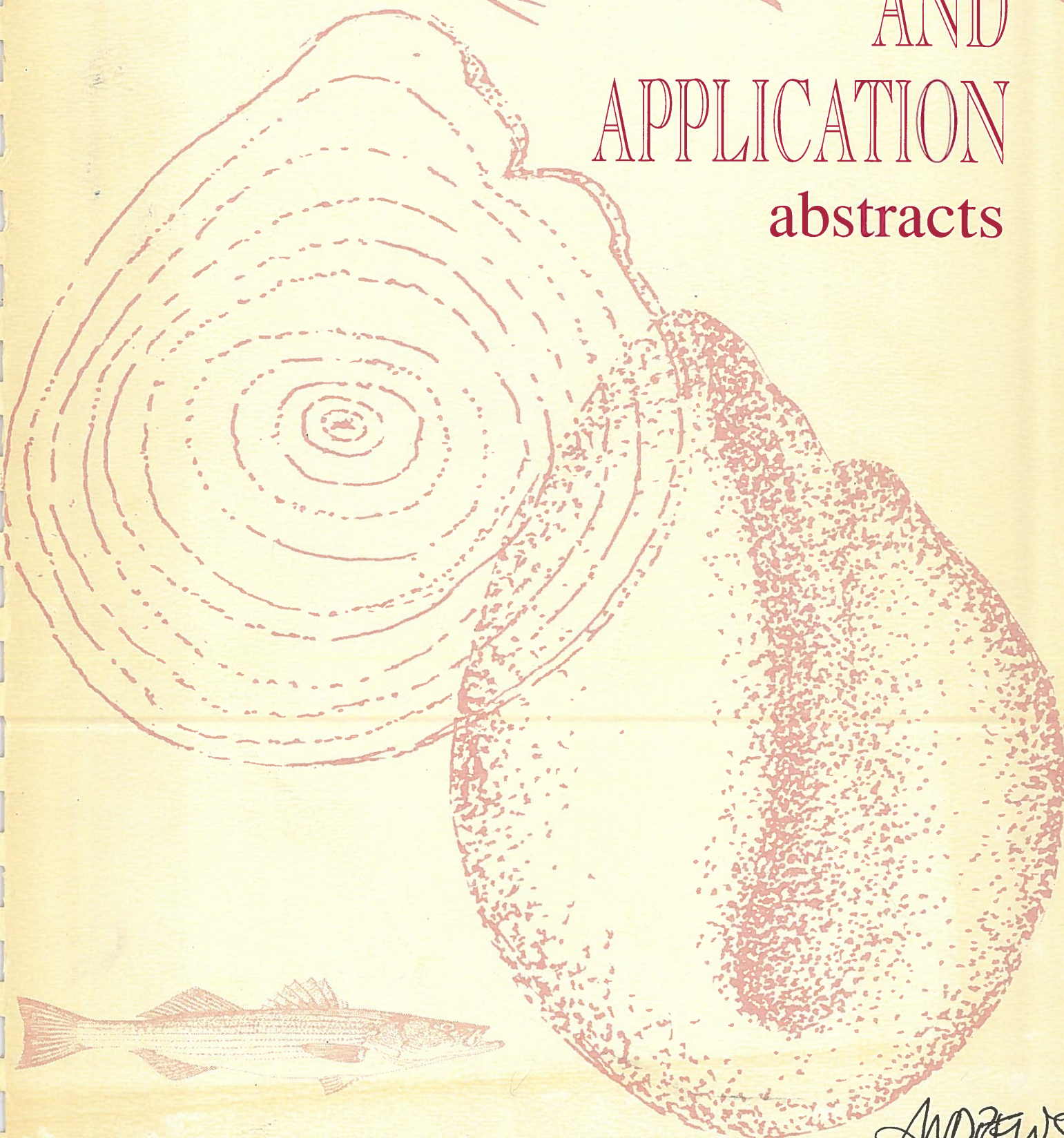


An International Symposium

# FISH OTOLITH RESEARCH AND APPLICATION abstracts



ANDREWS

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CHANGES IN PROGRAM - **Changes are made in bold type.**

SUNDAY, JAN 24

Otolith Growth and Morphology - Contributed Papers

Concurrent Session A - **Charles A. Wilson, Chairperson**  
**Louisiana State University**

Concurrent Session B - **John N. Casselman, Chairperson**  
**Ontario Ministry of Natural Resources**

2:00-2:15 T.E. Laren, P.E. Grotnes & J.E. Eliassen: Otolith pattern and shape as a **discrimination** of origin in various cod stocks (*gadus morhua*) along the north Norwegian coast. (41)

MONDAY, JAN 25

Estimation of fish growth - Contributed Papers

Concurrent Session A - **Edward B. Brothers, Chairperson**  
**Ithica, New York**

1:45-2:00 A.B. Thompson & A. Bulirani (80) - **Withdrawn**

2:00-2:15 G.R. Fitzhugh & J.A. Rice: Risk-calculation of size frequency distributions in southern flounder: A test of size-dependent processes in early life history. (81)

2:15-2:30 M.E. Waldron: **Age validation of the South African anchovy, *Engraulis capensis*, using daily growth increments.** (82)

3:00-3:15 W.J. Fletcher (85) - **Withdrawn**

3:30-3:45 H. Trodec (87) - **Withdrawn**

Concurrent Session B - **Audrey J. Geffen, Chairperson**  
**University of Liverpool**

3:00-3:15 S.A. Holt & B. Henley: Growth rates of two sciaenid fishes distributed across an estuarine tidal front. (96)



TUESDAY, JAN 26

Otoliths in Studies of Population Biology - Invited Lectures

9:15-10:00 K. Tsukamoto: **Otolith-tagging for stock enhancement program of Masu Salmon, *Oncorhynchus masou*, in the Kaji River.**

Otoliths in Studies of Population Biology - Contributed papers

Concurrent Session A - **John D. Neilsen, Chairperson**  
**CARICOM Fisheries Research Program, West Indies**

1:15-1:30 K.M. Bailey, S. Picquelle & S.M. Spring: **Mortality estimation of laval walleye pollock in Shelikof Strait, Western Gulf of Alaska. (115) Poster Only**

2:15-2:30 J.S. Bulak, J.S. Crane, J.M. Dean & D. Secor. (119) **Time change - moved to 3:45-4:00.**

3:15-3:30 E.C. Volk, S.L. Schroder, J.J. Grimm & H.S. Ackley: **A simple method for the assignment of thermally induced otolith banding patterns for mass marking using the interleaved two of five bar code symbology. (123)**

3:45-4:00 G.M. Wellington, L. Gutierrez & B.C. Victor. (125) **Time change - moved to 2:15-2:30.**

Concurrent Session B - **Erland Moksness, Chairperson**  
**Flodevigen Marine Research Station, Norway**

1:15-1:30 Y. Teklegiorgis & J.M. Casselman: **Increasing the precision of otolith age determination of tropical fish by differentiating biannual recruitment - a system developed and validated on tilapia, *Oreochromis niloticus*, from Lake Awassa, Ethiopia. (127)**

2:00-2:15 J.L. Butler (130) - **Withdrawn**

WEDNESDAY, JAN 27

Chemical Tags and Otolith Composition - Invited Lectures

10:30-11:15 J.N. Smith: **The use of radionuclide tracers in the ageing of marine fish.**



## Chemical Tags and Otolith Composition - Contributed Papers

Concurrent Session A - **Cynthia Jones, Chairperson**  
**Old Dominion University, Norfolk, Virginia**

1:00-1:15 D.A. Libby & K.D. Friedland (141) - **Withdrawn**

2:15-2:30 D.M. Monteleone, E.D. Houde, D.H. Secor & L.G. Morin: Comparison of alizarin complexone and tetracycline hydrochloride for immersion marking of otolith of fish embryos and larvae. (146) **Poster Only**

2:45-3:00 K.P. Severin, B.L. Norcross & J. Carroll: Chemistry of walleye pollock otoliths - a potential aid in stock assessment. (148)

Concurrent Session B - **Ron Thresher, Chairperson**  
**CSIRO Division of Fisheries, Australia**

1:30-1:45 P. Brown & J.H. Harris (153) - **Withdrawn**

## POSTER SESSIONS

### Otolith growth and morphology - Posters

14. B. Morales-Nin, S. Giro & A. Lombarte: Image analysis applications for otolith age determination.

16. D.G. Mortensen & M. Carls: Effects of whole oil ingestion on the growth and microstructure of juvenile pink salmon (*Oncorhynchus gorbuscha*) otoliths.

22. A.B. Thompson & A. Bulirani - **Withdrawn**

### Estimation of fish growth - Posters

48. C.P. Alvarez - **Withdrawn**

50. B.D. Bruce - **Withdrawn**

58. P.N. Kochkin - **Withdrawn**

71. C.G. Pineiro, N. Perez & J. Landa: Growth pattern of megrim (*Lepidorhombus whiffiagonis* and *L. boscii*) in the North-East Atlantic (ICES Div. VII<sub>jk</sub> VIII<sub>ab</sub> VIII<sub>c</sub> and IX<sub>a</sub>).

Additions to this poster session:

- 160. D.W. Ahrenholz, D.S. Vaughn & J.V. Merriner: Prerecruitment growth and relative survival among three seasonal cohorts of juvenile Atlantic menhaden from North Carolina estuaries.
- 161. G. Winkler: Age and growth of larval and juvenile *Barbus barbus* determined from otolith microstructure.
- 162. P.A. Garratt, A. Govender & A.E. Punt: Growth acceleration at sex change.

Otoliths in studies of population biology - Posters

- 103. G.W. Carder & J.A. Babaluk - Withdrawn
- 108. M.D. Malette, J.M. Gunn & J.M. Casselman: Withdrawn

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Jason Selong

Brent Sharrer

Todd Thuma

## ABSTRACTS (INVITED LECTURERS)

### Otolith Growth and Morphology

A. P. Wheeler

Y. Mugiya

D. Nolf (No abstract submitted)

### Estimation of Fish Growth

H. Mosegaard

R. J. Beamish

S. Ralston

C. Francis

### Otoliths in Studies of Population Biology

E. D. Houde

K. Tsukamoto

M. Yoklavich

D. Secor

### Chemical Tags and Otolith Composition

J. Kalish

R. Radtke (No abstract submitted)

J. N. Smith

J. Edmonds



# Regulation of Carbonate Biomineralization

Dr. "HAP"

A.P. WHEELER. Department of Biological Sciences, Clemson University, Clemson, SC-29634, USA.

"WHEELER@CLEMSON.EDU"

-1415 (OFF)

-3597 (LAB)

Carbonate calcification is a highly regulated process in that the microstructures for any type of deposit may be diverse. At the same time microstructure generally is conserved for the same deposit within a species or a group of closely related species. Recently, much of the regulation of crystal growth has been attributed to the polymers that make up the organic matrix of biomineral. One of the functions of matrix may be to provide an immobilized surface or a three-dimensional network or gel which favors nucleation from metastable biological fluids. In addition, the arrangements and types of immobilized reactive groups can affect microstructure of crystals which form on surfaces. Growth of crystals can be affected by soluble matrix components as well. It has been shown that the numbers and types of adsorption sites on crystals is dependent on polymer primary and secondary structure. Specific adsorption can lead to the development of certain crystal faces and axes at the expense of others. Less specific adsorption can lead to inhibition of crystal growth. Because carbonate formation often is cyclical, a repeating sequence in which matrix initiators followed by regulators and inhibitors arrive at the mineralization front may effect crystal layer formation.

C.C. Biomineral

1: Variety of Microstructures & Mineralogies

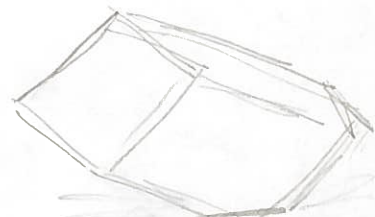
2: Microstructures Conserved



Coccoliths

each spoke one crystal

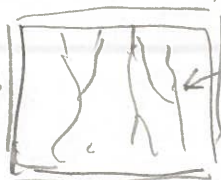
Rhombicidal ionic formation only



particular orientation

Insoluble Organic - Framework / Inter-crystalline

Soluble organic - intra-crystalline



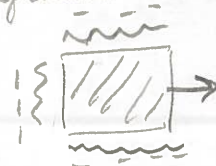
Nucleation Barriers - methods to stop development of crystal

C.C. formation = acidifying rxn.

Size of crystal - limited by compartment & / or protein Polyanion deposition

Trace elements: not known if incorporated or in protein matrix or both

Polyanions inhibit growth of crystal & create directional growth.



Considerations for Crystal Matrix Interactions

- 1) Structural
  - 1° spacing of charged groups
  - 2° beta sheet
  - 3° - microcomp or gels

2) Bonding

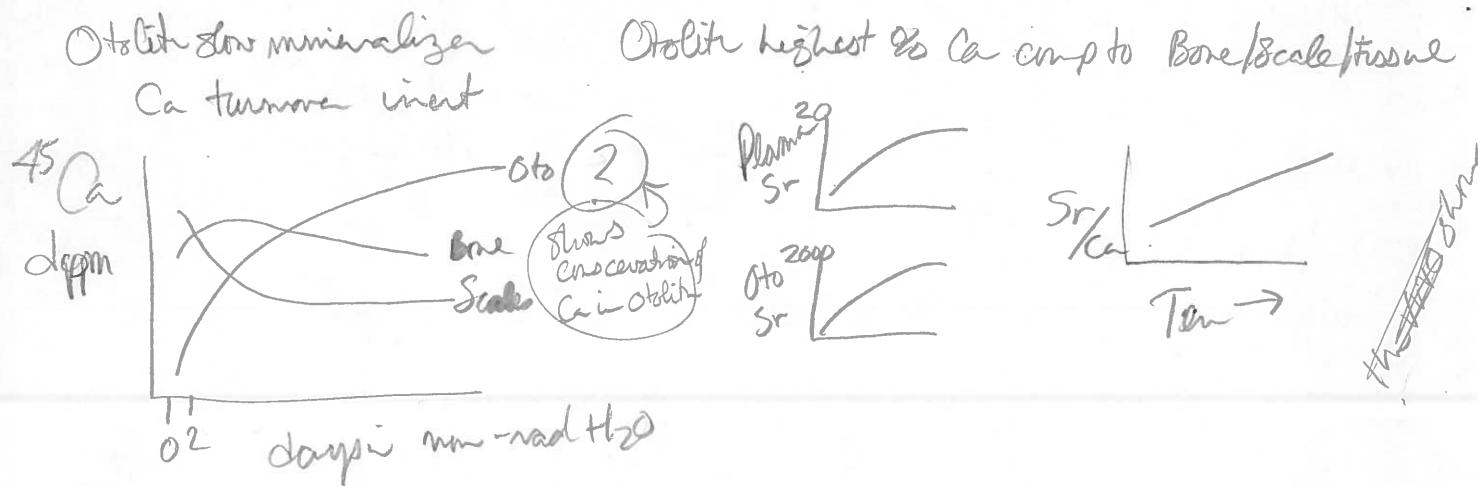
3)

# Calcium and Strontium Metabolism in Fish Otoliths

Y. Mugiya. Laboratory of Physiology and Ecology, Faculty of Fisheries, Hokkaido University, Hakodate 041, Japan.

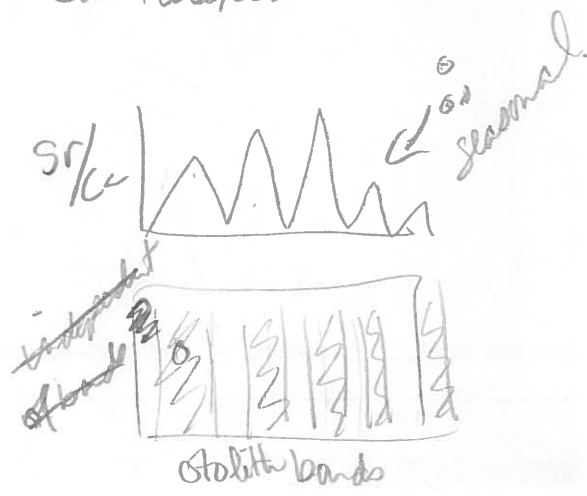
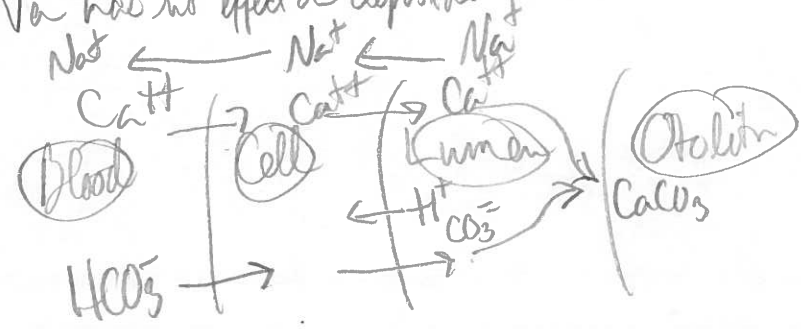
Teleost otoliths are highly calcified concretions which occur in the membranous labyrinth. Their growth will be controlled by two different processes of non-collagenous organic matrix formation and the succeeding calcification. Otoliths are a slow mineralizer and their calcium is metabolically inert, which characterize the calcium metabolism of otoliths compared with that of other calcified tissues such as scales and bone. Hypophysectomy reduces the rate of otolith growth only by half, resulting in an uncoupling between the somatic and otolith growth. Growth hormone therapy corrected the reduction. Calcium deposition occurs on the sulcus side of the otolith (sagitta) where it faces to the transitional and macular cells of the sacculus. Our recent observations confirm the presence of  $Ca^{2+}$ -ATPase in the membrane of transitional cells. A  $Ca^{2+}$ -ATPase inhibitor (W7) reduces the rate of calcium deposition by about 70%. The overall results suggest the presence of an active calcium transport system in the cells. Results of strontium incorporation into otoliths also will be presented in relation to the ambient temperature and strontium concentrations.

In-vitro exp produced same structures as in-vivo?



Calmodulin = Ca receptor protein activates  $Ca^{++}$  receptor

Na has no effect on deposition



H. Mosegaard: Statistical & Biological sources of error when  
back calculating fish ~~size~~ growth from otoliths



**A Discussion of the Possible Relationship Between Ageing Errors and the Decline in the Walleye Pollock Fishery: The Worlds Largest Fishery.**

BEAMISH, R. J. AND McFARLANE, G. A.\*  
Pacific Biological Station  
Nanaimo, B.C. V9R 5K6

The walleye pollock (*Theragra chalcogramma*) fishery in the 1980s was the largest fishery in the world. Annual catches were recorded to range from 6.0 to 7.0 million t, but may have approached 8.0 or 9.0 million t or approximately 10 % of the world catch of all marine fishes. In the Gulf of Alaska the fishery increased about 1982 and declined after 1985. In the international waters of the Bering Sea the fishery increased from approximately 300,000t in 1985 to 1.3 million t in 1987 and catches in 1992 were about 10,000t. We discuss how errors associated with the otolith method of age determination could be related to the decline in abundance of walleye pollock.

80s 7 million mt. annually  
10% of total world prod. of fish

$R^2$  values for adjusted (corrected) strong year classes were not signif.

is it valid to use a general equation to correct for reader bias?

**The Influence of Oceanographic Variables on the Growth of Otoliths in Pelagic Young-of-the-Year Sebastes.**

S. RALSTON\*, National Marine Fisheries Service, Southwest Fisheries Science Center, 3150 Paradise Drive, Tiburon, CA 94920, USA.

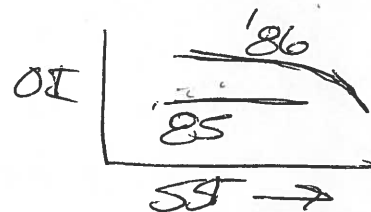
Larval growth rate is sensitive to environmental conditions and has a large influence on cumulative cohort mortality. Association of within season periods of enhanced and/or diminished growth with fluctuating oceanographic conditions may clarify the influence of physical factors on growth and reproductive success. To evaluate time-specific growth and its dependence on oceanographic conditions, back-calculated daily increment widths were used as a conservative measure of larval growth rate. Data from five species of rockfish (Sebastes spp.) collected during annual (1985-89) surveys of pelagic juvenile abundance off central California were available for analysis. The combined effects of larval age and date on growth rate were evaluated with ANOVA. The simple two factor additive model accounted for 85-95% of growth rate variability and there was little evidence of factor interaction. Time series of the date effect were calculated using least-squares means for each of the 25 species-year combinations. Results showed that, within any particular year, the growth time series of the five species were positively correlated, but distinct interannual differences in pattern were observed. Oceanographic variables (i.e., temperature, salinity, upwelling, sea level, and turbulence) were analyzed to account for these differences. Although temperature clearly is correlated with growth rate, the relationship is inconsistent. I conclude that no single environmental factor can be used to model within season trends in the growth rates of larval Sebastes spp.

40 Kt - annually (Sebastes sp.)

Sampled to 20mm

Oto index higher in el niño year ('86)

- index equivalent to cool year ('85) at higher temp



**The Analysis of Otolith Data - a Mathematician's Perspective:  
What, precisely, is your model?**

R.I.C.C. FRANCIS. Ministry of Agriculture and Fisheries, Box 297, Wellington, New Zealand.

The formulation and testing of models (whether mathematical, statistical, or conceptual) is a central part of all science. It is most important that these models be clearly and accurately described so that their assumptions (hypotheses) can be tested and results can be communicated. In many published analyses of otolith data the model on which an analysis is based is poorly or inconsistently described. As a consequence, the value of these analyses is limited. Examples are given from three areas of otolith data analysis: the estimation of growth, back-calculation of length, and age validation.

Von Bert - Population  $L_t = \text{mean length at age } t$

$L_{\infty} = \text{asymptotic length}$

Ch. individual  $L_t = \text{length at } t$

$L_{\infty} = \text{max length}$

$L_t, t$  must be defined

Francis '1988 Can. J. Fish. Aqu. Sci. 45: 936-942

- Tagging data; change in means of variables

Back Calc - Live =

$L_c$  - fish len at capture

$R_c$  - oto radius of capture

$R_1, R_2, \dots$  - radii marks in oto

Find =

$L_1, L_2, \dots$  - fish len at time of radius formation

Copmickal -  
Share origin



## How Have Otoliths Contributed to Understanding Fish Population Biology?

*E.D. Houde*

E.D. HOUDE\* and D.H. SECOR. Center for Environmental and Estuarine Studies, Chesapeake Biological Laboratory, University of Maryland System, P.O. Box 38, Solomons, MD 20688, USA.

Otoliths in fishes provide reliable records of age and past environmental histories. They are a primary method to determine population age structure and are an important source of life history information. This paper evaluates otolith-aging and other technologies, and their applications in population studies. Age-structured population models, because they delineate cohorts, conceptually are best to describe dynamics, explore life-history relationships, and apply to management. Otoliths record age as well as annual, seasonal and daily events in the lives of fishes. Application of daily increment analysis to early life studies arguably has been the single biggest advance in fish population studies in recent years. Population dynamics in early life, including survivorship, growth, and hatch-date analyses now can be undertaken with confidence. Fates of individual cohorts can be determined. No extant recruitment hypotheses have fallen as a consequence of emerging otolith technology, but critical insights into starvation, predation, size-selective mortality, and transport hypotheses have been gained. Sampling biases, misinterpretations of otolith microstructure, and poor understanding of relationships between otolith growth and body growth continue to constrain applications of otolith technology. Emerging applications combine otolith microstructure and microchemistry analyses to track environmental histories of individuals. Such approaches, if incorporated into time-series analyses and complemented by individual-based modelling, will significantly enhance fish population studies in the next decade.

*Ageing Errors - tend toward overest. mortality*

## Otolith-Tagging for Stock Enhancement Program of Masu Salmon, *Oncorhynchus masou*, in the Kaji River

K. TSUKAMOTO\* Ocean research Institute, University of Tokyo, Nakano-ku, Tokyo, 164 Japan

In order to determine the most effective method for stock enhancement of the masu salmon, a mark-recapture study was conducted for about ninety thousands fish of 10 different strains with various types of otolith-tagging in the Kaji River, Japan from March 1989 till March 1992. A major group in the experiment was 53985 alevin of native migrant strain with a single otolith-tag at embryonic stage released after emergency in March 1989. Other fish groups were released in autumn or next spring, or fish of resident strain or strains of different rivers. Recruitment of wild stock of the same year class was estimated about 274000 using Petersen method. There was no difference in growth rate between wild and released fish. Ninety one wild smolts of 1+ juveniles (0.033%) were collected during downstream migration in March-April 1990, whereas the recapture of the major group was only 7 (0.013%). About 1200 return fish were collected during upstream migration in January-June 1991. 888 fish were examined, of which 93(10.5 %) were released fishes and the rest of 795 fish, wild stock(return rate, 0.290%). Most of the released fish occurred from the major group (84 fish; 0.156%). Not a single fish of any released groups was recaptured in adjacent rivers. These results showed that the mortality during the stream life of the first year after emergency was more significant in released fish than in the wild. Therefore, it seemed most effective for stock enhancement in this river to protect the wild 2+ upstream migrants and to conserve the stream habitat for their maturation.

Intro Slide - Tree x-sect!

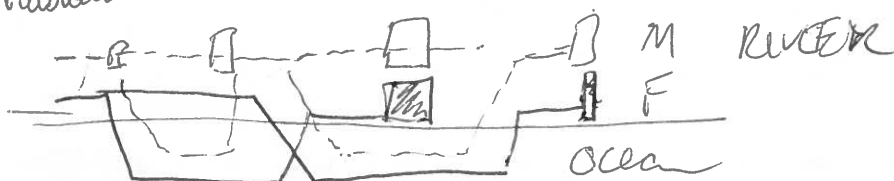
O. Keta sp → ocean net. 1 to 4 yrs (2, 3 ↑)

O. gorbuscha sp → ocean net 2 yrs (?)

Tetracycline

" Lysine complexes

O. masou



~~Like Salmon~~

Close link of Oncorhynchus to Salmon

## Long-term Variability In Growth of Rockfishes and Their Environment

M.M. YOKLAVICH. NOAA, National Marine Fisheries Service, Southwest Fisheries Science Center, Pacific Fisheries Environmental Group, P.O. Box 831, Monterey, CA, 93942 USA.

While long time series are needed to describe or predict response of marine fish populations to environmental variation, relatively few multi-decadal biological data exist and usually comprise changes in abundance from historical catch records. Likewise, temporal variation in fish growth is typically limited to short series of data. Recently, techniques have been described to extract historical growth rates from otoliths of long-lived (>60 years) rockfishes. These otoliths from recently-collected fishes act as chronometers, and measurement of early growth increments results in time series of annual growth.

Initial series of growth data from *Sebastes pinniger* (canary rockfishes) of ages 1-6 years spanned 1935-1979, and the dominant low frequency signal was increased growth after 1970. We originally interpreted this to be related to density-dependent factors associated with stock reduction from overfishing. However, development of even longer time series, currently extending to 1987, was important to resolve complex interactions among growth and biological and environmental factors (e.g., temperature, upwelling intensity, and acute effects like *El Nino*), methodological variability inherent in the initial study, and possible complications of increased growth and size-selective mortality at the end of the series. Description of the temporal variability in growth and possible links to the environment will be reviewed.

## Larval Mark-Release Experiments: Potential for Research on Dynamics and Recruitment

D.H. SECOR and E.D. HOUDE. Chesapeake Biological Laboratory, University of Maryland System, P.O. Box 38, Solomons, MD 20688, USA.

Millions of fish larvae can be marked by chemical-immersion or thermal-induction marking of otolith microstructure. Experimental releases and subsequent recaptures of such larvae can be used to estimate the effects of environment and biological attributes (e.g. age, size, genetic origin) on larval vital rates, movements, and recruitment potential. Larval mark-release experiments with striped bass (*Morone saxatilis*) were conducted in two Chesapeake Bay tributaries in 1991 and 1992. Approximately 7 and 13 million larvae (4-7 mm, SL) were marked by immersion in alizarin complexone solution and released into the Patuxent and Nanticoke Rivers, respectively. Recaptures of released larvae yielded estimates of abundance, vital rates, dispersal patterns and recruitment potential in relation to environmental factors. To predict sample sizes to accurately estimate relative abundance, we developed a nomogram for combinations of R (proportion of recaptured marked larvae) and P (confidence level for R). Larval mark-release experiments are feasible for many freshwater and anadromous species, where nurseries approximate closed systems. In marine (open) systems, we developed and evaluated hypothetical mark-release experiments for plaice (*Pleuronectes platessa*) and cod (*Gadus morhua*) larvae. Low recapture rates due to dispersal from release sites indicated that experiments on plaice or cod must be short ( $\leq 7$  days), but could be used in short-term experiments on larval dynamics and movements.

### Attrib. of Released Larval

- 1 #
- 2 Size Age Stage
- 3 Cond.
- 4 Parentage
- 5 Genotype

### Release Sites

- 1 Habitat Strat
- 2 Anthropogenic inputs
- 3 Patch simulation - Dispersal

### Release Times

- 1 Single / Multiple
- 2 Weather / Seasonal effects
- 3 Stage - Spec. Survival

Morone sax - Short temporal & spatial spawning

- well dev. hatchery  
(Chesapeake Bay)

Concentric Chem-tagging to diff. release  
circumferences

7 mill rel.

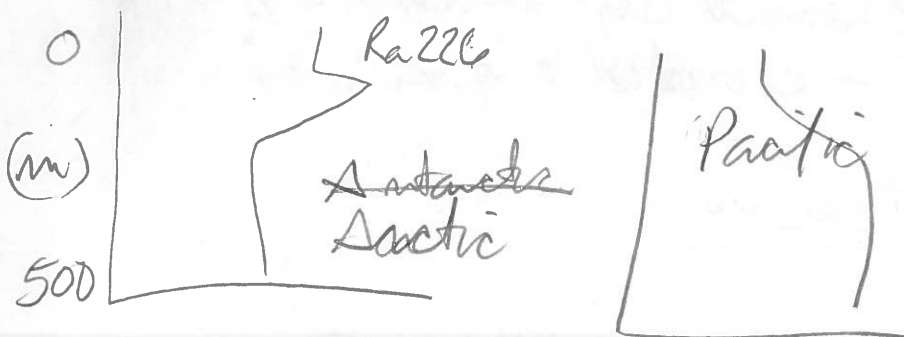
## The Use of Radionuclide Tracers in the Ageing of Marine Fish

J.N. Smith\* and S. Campana, Bedford Institute of Oceanography, P.O. Box 1006, Dartmouth, Nova Scotia, Canada B2Y 4A2.

Radionuclide tracer experiments offer a new and comparatively precise methodology for determining the growth rates of marine organisms. These radionuclide ageing techniques rely on two basic geochemical features of the naturally-occurring, uranium and thorium decay series, (1) the high solubility of radium in seawater compared to its parent and daughter radio-isotopes, and (2) the relatively efficient uptake of radium from seawater by marine tissues as a proxy for calcium. These conditions combine to insure that a state of radioactive disequilibrium is established initially by the incorporation of radium isotopes into the calcified tissues of organisms, relatively unaccompanied by parent or daughter radio-isotopes. Successful applications of the method then rely on a sufficiently high signal to noise ratio in the daughter/parent activities, as they decay to a state of radioactive equilibrium, that the determination of useful ageing information becomes feasible.

Measurements of Pb-210/Ra-226 dis-equilibria in the otoliths of redfish (Sebastes diploproa) have revealed that this species of fish can live to ages in excess of 75 years in coastal waters off Nova Scotia, Canada. Measurements of the Th-228/Ra-228 disequilibria in the otoliths of the much shorter-lived silver hake (Merluccius bilinearis) and flying fish (Hirundichthys affinis) may provide estimates of longevity on time scales of 0-10 years, which could then be used to evaluate the accuracy of currently-used, ageing models based on otolith annulus counts. The radioisotopic ageing technique relies on the extraction of a rectangular block centred around the core of the otolith whose analysis for radionuclide ratios permits an unambiguous estimate of the age of the fish. Age determinations of fish based on natural radioisotopes can result in significant improvements in the assessment and management of fisheries resources.

Slide desc. intake.

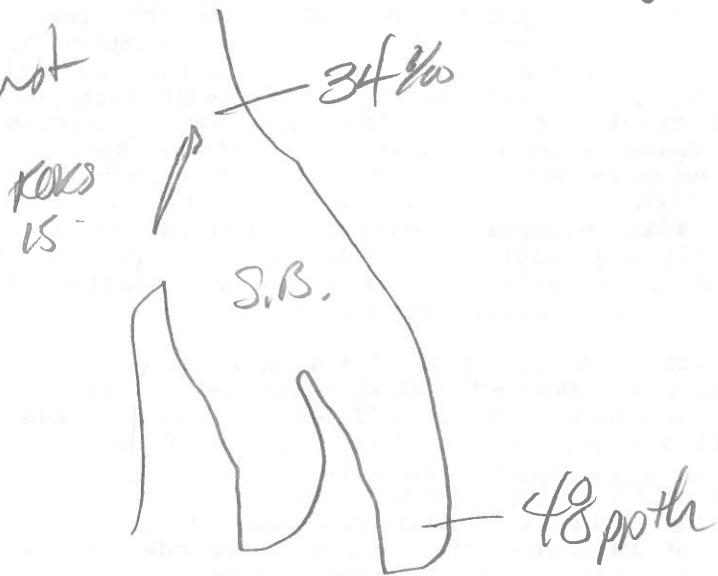


Measure  $\alpha$  from Ra directly? Short

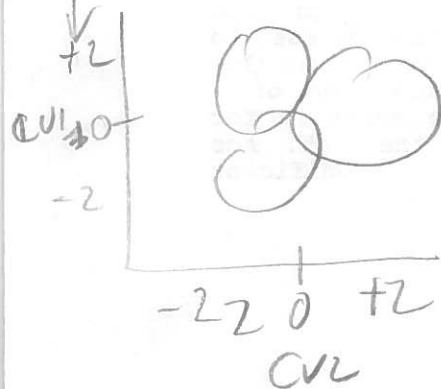
Sharks Bay Snapper (looks like Porgie!)

Edmonds: Minor & trace elem. in teleost  
 otos & use in pop discrimination. (Hg?)

Sharks Bay, Aust



Canonical Variate



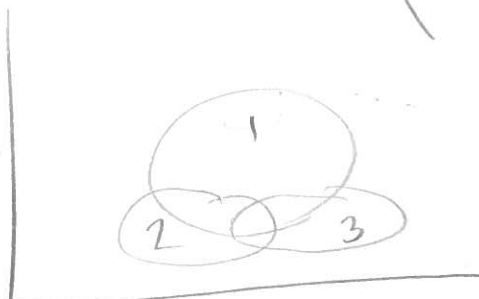
Elements dep. on fish size  
 - corrected & analysed.

Blue Emperor (Lethrinus)



Differentiation  
 among  
 locations

Bar



Pilchard  
 More sedentary than expected

**Comparative Analysis of Microstructure of Squid Statoliths of Families Ommastrephidae and Loliginidae and Fish Otoliths.**

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Microstructure of about 5 thousand statoliths of nektonic squid belonged to 18 species of family Ommastrephidae and 6 species of family Loliginidae were studied. Investigated squid represent different ecological life styles and inhabit shelf, slope and open waters of subantarctic, temperate, subtropical and tropical regions. Peculiarities of squid statolith microstructure were compared with those of fish otoliths obtained from literary data. Structure and resolution of daily growth rings in different phylogenetic and ecological groups of squid were examined. Patterns of statolith and otolith formation and development in early ontogenesis were investigated. Main growth zones were revealed in ground statoliths, possible reasons and period (ontogenetic stage) of their formation were described. Stress marks were examined. Main environmental and individual biological factors influencing their appearance in different growth zones were suggested. Similarity of statolith and otolith microstructure reflect principal resemblance of physiological processes in organisms of squid and fish developed during their co-evolution and formation of similar life styles.



**Morphology and Identification of Otoliths of Some Southeastern and Southern Brazilian Coast Species of Perciformes (Teleostei).**

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The present work aims the development of methodology which makes possible otolith (sagitta) identification to specific level, through strictly objective means. It is expected to get practical and accurate use of this structure peculiarities in assisting systematics, piscivore animal food habits and archaeological studies. Otoliths of 3955 teleosts, belonging to 30 species of 6 Perciformes families were utilized. The fishes were collected between 23 00'S and 29 21'S along the Brazilian coast. Otoliths of 12 individuals, from regularly spaced length classes were chosen in each species to be used in a morphological analysis. 70 Morphological characters and 14 morphometric relationships were examined on the inner face of each piece. These 14 relationships were used to separate the 30 species through multivariate discriminant analysis. A representative otolith of each species was photographed using a new technique that enhances otolith's morphological surface details. Tests performed with previously known otoliths resulted in 95.97% of correct identifications. Even in families whose species have very similar otoliths, like Haemulidae, 90.42% of correct identifications were gotten. The main motive of this method is to make otolith identification possible to researchers in other areas, not necessarily otolith experts, but who depend on the identification of their own pieces for the success of their work.

**Age and 45-Year Otolith Growth Chronology of Freshwater Drum,  
*Aplodinotus grunniens*, of Eastern Lake Ontario**

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During the past 34 years, freshwater drum (*Aplodinotus grunniens*) were collected during community index netting of eastern Lake Ontario and adjacent waters of the Bay of Quinte. A subsample of acetate replicas of transverse sections of otoliths of 550 fish (up to 868 mm TL, 11.9 kg TW) was examined. Annuli (narrow translucent lamellae), which were demarcated in June, were distinct and easily interpreted and measured by using a Calcified Structure Age and Growth data Extraction System (CSAGES). Since 1946, there have been three strong year classes--1955, 1983, and 1977, composing approximately 52% of the subsample. The 1955 year class made up 67% of the sample prior to 1977 and is now over 35 years old. Relative abundance of these year classes was correlated with the highest mean summer water temperatures, could be easily tracked in subsequent sampling, and was used to validate age-interpretation techniques. Otolith growth was strongly positively correlated with water temperature, slowest in 1958 and fastest in 1959. Two stocks were differentiated by otolith growth characteristics; the "bay" stock, which had a significantly lower growth potential, grew significantly more in 1979 after a catastrophic winterkill, showed greater overall annual variability, and zonation was optically different from that of the "lake" stock.

### **Morphological Characteristics of NAFO Division 5Ze Cod Otoliths**

**Maria-Ines BUZETA.** Marine Fish Division, St. Andrews Biological Station, N.B., EOG 2X0

The purpose of this study was to initiate investigation on topics discussed at the 1991 Georges Bank Cod and Haddock Ageing Workshop, and on other issues directly related to the ageing of 5Ze cod. Discrepancies between the Canadian and USA catch-at-age data have been of concern in the past, and improper ageing has been suggested by USA age readers as a possible cause. The possibility of misinterpretation of otolith features and incorrect age assignments was investigated. Whole larval otoliths and sectioned 1+ otoliths from commercial samples and groundfish surveys were used to measure settling checks, first annulus and second annulus cross-sectional (XS) widths.

The Image Analysis System facilitated the study by objectively quantifying their sizes. Settling check XS width averaged <1 mm, first annulus XS width ranged between 2 and 2.5 mm, and second annulus XS width averaged about 4.5 mm. The methods proved to be of practical application for "editing" age interpretations. Similar measurements should be done for each species and stock.

**Comparison Between Fish Length and Otolith Growth; An Investigation in the Potential Use of Individual Fish in the Study of Growth Pattern in Kapenta, *Limnothrissa miodon*.**

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Estimates kapenta growth had been obtained through length frequency analysis the accuracy of some of the results have been questioned. In order to validate the results an independent method of accessing growth was deemed to be necessary. Preliminary assessment indicated that daily rings on the otolith of kapenta could be used to determine growth and used for age back calculations. The method required that a large number of otolith be read. Investigations on whether individual otolith contained an accurate history of growth of the fish was carried out so that more growth information could be available from a single fish. The radius of every tenth ring, in the same axis for each otolith was measured at a magnification of X 2 000. The relationship between the otolith growth and the growth of the fish was used to transform the measurements on the otolith to fish length. The relationship in the radii of otolith in the axis used and that of fish length was linear. An average growth curve was plotted using the average length at each growth band. The curve obtained was similar to that obtained when only the total number of rings on each otolith were used.

## Ontogeny and Otolith Microstructure of Larval Bluefish (Pomatomus saltatrix)

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We are presently examining the use of otoliths as records of individual growth as part of a larger project studying survival of early life history stages of bluefish (*Pomatomus saltatrix*). Our purpose in this study was to examine the use of otoliths as records of life history transitions in bluefish. Our first step was to determine the time of first increment formation, and through examination of otoliths from egg, yolk-sac larvae and post yolk-sac larvae, we were able to demonstrate that first increment formation occurs at about hatching. To examine ontogeny of larval and juvenile bluefish, 15 morphological and meristic characters were measured for 70 individuals. In addition, otoliths from each individual were removed and both sagitta and lapilli increment number and widths were measured on at least three separate occasions. Multivariate analysis of the morphological and meristic data demonstrated two major transitions in young bluefish, not including hatching. We found that an increase in otolith increment width was correlated with the first transition. We also found an increase in increment width associated with the second transition, but more importantly secondary nuclei formation occurs at this transition. We believe that these otolith characters can be used to determine the timing of these two life history transitions and plan to examine whether timing of these transitions are important to the survival of young bluefish.

**Factors Influencing the Production of Daily Growth Increments in Otoliths of Young-of-the-Year Striped Bass, *Morone saxatilis***

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Young-of-the-year striped bass were raised under conditions of constant photoperiod and temperature to determine if an endogenous circadian rhythm influenced daily increment deposition. Growth increments were deposited daily for 98 d, 63 d of which were under constant environmental conditions. After 98 d, deposition rate decreased to an average of 0.9 increments  $d^{-1}$  for 43 d in both the constant light and dark experiments. Results supported the theory that daily increment deposition is regulated by an endogenous circadian rhythm. Furthermore, slower growing striped bass may eventually produce larger otoliths than faster growing fish of similar size. A stress mark, in the form of a distinct opaque zone, was observed on all otoliths at approximately 30 days of age, corresponding to shipment of fish. The results suggested that young-of-the-year striped bass can be accurately aged to approximately 100 days using daily growth increments and that average increment widths can be used as a relative indicator of somatic growth.

**Measurement of the Depth to the Nucleus under the Medial Surface of Otoliths in Larval Fish by Using Laser Scanning Microscope (LSM)**

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Larval otoliths (sagittae) were viewed with Laser Scanning Microscope (LSM), under epireflection and transmitted light, in order to obtain sequential measurements of calcium deposition from the medial surface, used as reference surface ( $Z = 0$ ), to the innermost increment. The morphological evolution of this base surface and of internal planes was checked using Scanning Electron Microscope (SEM). Otoliths from larvae of pelagic, demersal and benthic fishes, either of known age or field caught, were examined.

LSM sagittal and transverse sections showed plano-convex otoliths in early larvae. The medial (macula-adjointing) side of the otolith became slightly depressed in its center as larvae evolved. In such otoliths, penetration and scanning with a laser beam enhanced the resolving power in the nuclear and perinuclear areas, until LSM possibilities were limited by the increase in otolith size, i. e. for metamorphosing larvae. SEM investigations gave evidence for a morphological evolution governed by ontogenetic events, leading to the sulcus formation when metamorphosis is completed. LSM measurements demonstrated a marginal thickening, which prefigured the sulcus cristae, while there was lesser accretion in the area to be the sulcus groove, facing the macula. This morphology was stressed by the growth rate, making more concave otoliths in slow-growing larvae. Among the observed species, the nucleus (hatching check), identified from known-age specimens, appeared decentered toward the medial side of the larval otolith, in the first third of its thickness. The adjusted depth of focus to the nucleus ranged from  $\approx 5$  to  $25 \mu\text{m}$ , depending on the otolith size and thickness (from  $\approx 30$ - $200$  and  $20$ - $100 \mu\text{m}$  respectively), and with species-specific and growth rate variations. Changes in otolith shape, from late larvae to early juveniles, accentuated this pattern, leading to discuss technical procedures for examination of internal planes.

### **Otolith Characters as an Aid in Identifying Larval Rockfish (sebastes spp.)**

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Otolith characteristics (nuclear radius, first increment width, and nuclear shading patterns) were investigated for their utility in separating species of the genus Sebastes during the larval stage. Eight rockfish occurring off the coast of central California species were analyzed: S. auriculatus, S. enomelas, S. flavidus, S. goodei, S. jordani, S. mystinus, S. paucispinis, and S. saxicola. Three species were found to possess unique character combinations which allowed species identification by otoliths alone. When otolith data were used in conjunction with pigmentation, correct identification of species was greatly increased.



**The Ultrastructure of the Otolith of the Eel *A. anguilla*.**

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New aspects on the ultrastructure of the eel sagitta are presented. Examinations were made by means of TEM and SEM at 2 levels: crystals of aragonite and organic matter. TEM and SEM observations revealed that individual calcified layers (in the continental zone) were composed of crystals subunits. Between each calcified layer is an organic matrix layer: the interlamellar matrix composed of tightly packed fibers. In the calcified layer intercrystalline and intracrystalline matrix are observed. The centre (nucleus or marine zone) is more proteinic than the edge and separated from the continental zone by the transition ring.

**A Natural Tag on the Otoliths of Pacific Hake (Merluccius productus) with Implications for Age Validation and Migration.**

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A natural tag in the form of a reduced growth zone, was observed on the otoliths of offshore Pacific hake stocks off British Columbia. The reduced growth is attributed to the 1982/83 El Nino and its effect on productivity and ultimately growth of Pacific hake. This phenomena is most evident in the large year class of 1980, impacting the 4th summer zone on otoliths. The validity of the otolith burnt section ageing method was corroborated through recognition of the 1983 summer zone as a natural tag. Using the degree of predominance of this phenomena from south to north, we suggest a hypotheses to explain migration patterns.

### **Digital Image Processing to Age Long-finned Squid using Statoliths**

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Digital imaging techniques are currently being used to describe the inshore population age structure of long-finned squid, *Loligo pealei*. Video images of the prepared statoliths are digitized by an IBM® compatible PC equipped with a PC Vision® image capture board. Bioscan Optimas® software is then used to enhance the raw image, make morphometric measurements, and count growth increments. Hard-copy prints are made with a video printer (Mitsubishi®) of each field of the statolith as it is analyzed to map the increments which have been counted and for image archiving.

Thus far the ages of 77 squid, 2 to 41 cm mantle length (ML) collected inshore in late April and early May, have been determined. None were more than 285 days old at capture. Back-calculated hatching dates suggest the existence of 2 major spawning peaks, late July through early August, and the major peak in October and November. Two squid less than 3 cm long hatched in February. This finding suggests that spawning may occur throughout most of the year in some part of their wide latitudinal range.

**Effects of Light Regimes and Somatic Growth Rate on the Daily Increments Formation in Otoliths of *Salveinus alpinus***

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We aimed to test the effects of light and food regimes on the daily increment formation in otoliths of young landlocked arctic char (*Salveinus alpinus*). Newly fertilized egg were separated into three groups. One were put into hatching-jars and placed back onto the natural spawning ground (field-group). The other groups (laboratory group) were kept at a hatchery at dimmed and natural light period condition throughout the egg- and early plumage stage. They were then exposed to constant 24 hour LL-regime and a 16:8 hour LD-regime respectively. Measurements of growth zone in the otolith were made using a light-microscope and a separat PC-aided recording system. After fixation in 96% ethanol in a period of 2-3 months, we got a mean shrinkage of 8% ( $T_1=19-32\text{mm}$ ). The number of increments in the otoliths of the laboratory-groups were equal and showed one increment per day between 12 March and 9 May. ( $Y=24.145+0.9752 X$ ,  $r=0.981$ ,  $df=249$ ). This indicate the existens of a circadian rythm in the protein/calsium deposition with the pacemaker entrained at a developmental stage prior to the early yolk sack stage. The field-group showed a mean of 41 increment in a 32 days period, and with a negation specific growth rate and decreasing increment-breath from 1.25-1.15  $\mu\text{m}$ . This exhausting situation may have caused a more frequent resting period, which may have overruled the circadian 24 hour phase in the protein/calsium formation.

### **Image Analysis Applications for Otolith Age Determination**

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S. GIRO. MICROM Espana. S.A. Bertran 35, Barcelona, Catalonia, Spain.

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Fish age determination by means of daily increment enumeration is time consuming and tedious. The recent development on image processing hardware has made possible to develop methods to count and measure zonations in video-displayed skeletal structures of fish (McGowan et al. 1987; Methot & Kramner 1979; Moskeens 1989) some semi-automatic methods are capable of identifying growth structures in basis of differential optical densities, the output can be improved by the reader (Ralston & Miyamoto 1983, Marales-Nin & Ralston 1990).

Areas with unclear daily increments are frequent in juvenile and adult fish otoliths. These areas can be due to structural properties of the otolith or to the mounting medium. With the aim to solve this problem, a semi-automatic method has been adapted to an image analysis techniques.

The method of quantification through image analysis is carried out by mean of the MIP image processor by MICROM ESPANA S.A. and consists of measuring to the total radius of the otolith and subsequently counting rings from the sequential acquisition of consecutive images following the starting direction of the radius.

Once the image has been acquired, you can improve it by means of contrast filters in order to emphasize changes between bright and dark zones. With the filtered image, rings on which counting will be made are selected by segmentation. On the segmented image, a line is drawn in the direction of the original radius, so as to count automatically ring intersections on the line. It is sometimes advisable to mark interactively the intersections which have not been able to be segmented automatically. Such a process is followed for each of the images of the whole specimen.

Morphological measurements of otolith, (areas, perimeters, axis, radius, center of gravity) and otolith shape description and analyses (Lombarte and Castellon 1991) was added to the programme.

## DAILY GROWTH RINGS IN THE OTOLITHS OF Sparus aurata REARED IN DIFFERENT CONDITIONS

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Sparus aurata (Pisces: Sparidae) were reared in captivity under different conditions of light, food and density. In some experiments the spawning was induced with hormones and in others the spawning was natural.

Periodically a sample of larvae was collected in each experimental condition and fixed for posterior otolith studies.

Sagitta and lapillus otoliths were studied by means of light microscopy in order to prove the daily periodicity of the increments laid down in the otoliths.

The obtained results showed the daily formation of the increments in sagitta and lapillus otoliths. The increments were first laid down in the sagittas in the hatching day and within 3 days in lapillus.

The increment width was measured with an image analysis system. The data were grouped and time series analysis of growth rates for each experiment were carried out.

**Effects of Crude Oil Ingestion on the Growth and Microstructure of Juvenile Pink Salmon (Oncorhynchus gorbuscha) Otoliths**

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Juvenile pink salmon (Oncorhynchus gorbuscha) were fed a commercially prepared food contaminated with crude oil ranging from 0.335 mg/g (low) to 25.6 mg/g (high treatment). Over a period of eight weeks, growth and increment counts from sagittal otoliths removed from fish in these groups were compared to measurements on otoliths of fish in control and treated control (0.111 mg/g) (dichloromethane carrier) groups. Significant differences ( $p \leq 0.05$ ) in otolith growth between all oil levels and the controls were evident in the first week of the experiment while significant differences in increment counts between medium (2.49 mg/g) and high groups and the controls were produced by the second week of the experiment. By week six otolith growth in low, medium and high groups was significantly less than controls while increments counts remained significantly less in medium and high oil level than controls. All fish were fed clean food after week six, but distance and increment remained depressed in treated groups through week eight. Otolith growth and increment number were reduced with increasing oil dosage in a pattern similar to that of somatic growth. However, within group correlations of otolith growth and increment numbers was poor, even in control groups. While oil did have an affect on otolith growth and development, the usefulness of our data for determining the effects of oil ingestion on wild fish is limited. In control fish, major and minor otolith axis growth correlated significantly with somatic growth ( $r=0.71$  and  $r=0.74$  respectively). These relationships remain to be tested in the other treatment groups but may provide significant correlations with somatic growth.



**Otolith Growth in Field Caught Haddock *Melanogrammus aeglefinus* Larvae and Juvenile**

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Growth patterns attributed to daily increments were observed in otoliths of larval and juvenile Atlantic haddock *Melanogrammus aeglefinus* collected on Western and Emerald Banks during March to July of 1991. The study provided information on growth and development during the first 5 months of life for this species. The sagittae and lapilli were viewed under compound microscope with transmitted light equipped with a video system; resolving power was in the range of 0.3-0.5  $\mu\text{m}$ . Depending on the size of the otolith, magnification used ranged from 400x to 1000x. Lapilli from fish >30 mm SL were viewed with a scanning electron microscope (SEM). The nucleus had a mean diameter of 15.6  $\mu\text{m}$ , proceeding outward from the nucleus check were 4-8 increments bounded by a mark that correspond to the yolk-sac check described in other studies. The mean diameter of the yolk-sac was 24.31  $\mu\text{m}$ . No check was found corresponding to the time when the fish is expected to turn from pelagic to demersal life.

### **Validation of Daily Otolith Increments in Juvenile Striped Bass (*Morone saxatilis*)**

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The ability to accurately age juvenile striped bass by daily increments is vital to evaluating the role of density-dependent and density-independent factors on the recruitment process. Therefore, we completed a validation study to verify an otolith-aging methodology on known age hatchery-reared striped bass. Hatchery raised larvae were stocked in hatchery ponds at 5 days of age. Sampling was initiated when fish were 17 days of age. Thirty striped bass were collected weekly until fish were 136 days of age. At 45 days, the juveniles were removed to hatchery tank enclosures. We observed that a stress mark was deposited on all otoliths, corresponding to the transfer of fish into hatchery tanks. Otolith increment derived ages of all fish were determined by the mean of readings made by three separate readers. A total of 386 otoliths were read by all readers, however, 81 otoliths were discarded due to coefficients of variation greater than 10 percent. Mean age underestimated known age, with a general increase in underestimation with fish age. Analysis of mean increment count on known age provided a significant relationship which was not significantly different from zero; however, the slope was significantly different than one, indicating an underestimation of ages using daily otolith increments.

### **Age Determination Methods and Biological Substantiation of Statistical Year for Fishes from the Southern Hemisphere.**

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Our concept is based on the assumption that the beginning of the statistical year should be a date from which we read the specimen's age. We considered the following factors:

- (i) seasonal climate patterns in the Southern hemisphere
- (ii) fish reproductive cycles
- (iii) peculiarities of the seasonal growth

In the Southern hemisphere July 1 is considered to be the end of one growth year and the beginning of the following one and the beginning of the reproductive cycle when a new year-class appears. A definite picture can be read in the otolith illustrating growth inhibition in the winter period and laying down of the annulus.

As annual rings are laid down unsimultaneously all these individuals in May-June (up to June 31) will be regarded as 0-year-olds. In July-August (since July 1) they will be attributed to the age-group 1, etc. During the remainder of the year we determine age basing complete annual growth zones taking into account increments on scales or otoliths. To determine individuals age we should subtract the time difference (month or days) from the estimated age value. Thus, if only July we regard individuals as 1, 5 and 10-year-olds then individual age for these specimens will be 0.5, 4.5 and 9.5-year-olds. Age composition of catches from 1977 to 1991 inclusive was calculated using length composition data. Length-frequency measurements for 1, 837, 354 specimens and age determinations for 34, 950 specimens were used. Age data were broken down by a statistical period July-June i.e. in accordance with the seasonal pattern observed in the Southern hemisphere and hatching time for the new horse-mackerel year-classes and by January-December as it was earlier approved by ICSEAF. Age composition of catches for appropriate years varied greatly and depended on the date regarded as the beginning of the statistical year. It was especially true of the age groups ranging from 1- to 4-year-olds. The greatest discrepancies in age composition were found for 1990-91. Thus, abundance estimates of the 1-2-year-olds for the statistical year "January-December" was twice underestimated (123% versus 45%). Earlier assessments of strong classes for 1988-1989 and 1989-1990 turned out to have average abundances and VPA Analysis corroborated this. Total biomass varied by 25-40% depending on the date chosen as the beginning of the statistical year.

### **The Otolith Structure in the Ecologically Different Species of Baikal Cottoidei**

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The paper describes the otolith morphology and structure for 26 species of the Baikal Cottoidei. Close correlation is found between the product of the otolith length by its width and length of specimens of 8 species of benthic Cottoidei. Data obtained was used to compile a key to Baikal Cottoidei by the otoliths. Otoliths of Baikal Cottoidei can be used as a complimentary or even the only taxonomic criterion in the nutrition study of the Baikal seal and the commercial fish species. The connection are studied between the otolith structure in species of Baikal Cottoidei and the ecology and phylogenetical relations of species within the families. It is shown that the ecological factors influence the otolith sizes, its length and mass. For phylogenetical constructions use can be made of the form and sculpture patterns of the otolith surface..

**Comparison of Growth Rate and Formation of Otolith Increments in Juvenile Red Snapper Lutjanus Campechanus.**

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Juvenile red snapper, Lutjanus campechanus, were collected by trawl (9 m) in the northeastern Gulf of Mexico in July 1991. Fish were anesthetized with 50 mg MS222 L<sup>-1</sup> seawater and small (5 mm) tags attached with elastic thread. Fish were marked in 400 mg oxytetracycline dihydrate L<sup>-1</sup> seawater for 24 h. Fish were held under 14L:10D h photoperiod, 25 ppt salinity, 25 to 29 °C, in a 300 L tank that was partitioned into three 100 L sections (100 fish per section). Fish in each section were fed at different rates: low-ration fed every 6 d; intermediate-ration fed every 3 d; and high-ration fed twice daily. Fish were sampled at 14, 30, and 42 d after marking. No significant differences were detected in growth rates among feeding treatments (ANCOVA 0.05 level; low-0.14 mm/d; intermediate-0.18 mm/d; and high-0.17 mm/d). Growth rate of individual tagged fish is compared to increment deposition rates.

SYMPOSIUM ON FISH OTOLITH RESEARCH AND APPLICATION  
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DIRECT VALIDATION OF THE DAILY DEPOSITION OF GROWTH INCREMENTS ON  
SAGITTAE OTOLITH OF USIPA *Engraulicypris sardella* OF LAKE MALAWI/NIASSA

by

AB THOMPSON and A. BULIRANI  
UK/SADCC Lake Fisheries Project, Salima, Malawi

Usipa, *Engraulicypris sardella* is an exploited pelagic cyprinid of Lake Malawi/Niassa. In a study on growth and age of usipa, growth rings on sagittae otolith were used to estimate age of the fish. Growth ring counts were made at x400 magnification using a compound light microscope linked to a Bioscan Optimas image analysis computer program. Difficulties in ring identification were encountered with "coarse" and "fine" rings being observed at high magnification. Growth rates based on the fine ring counts were approximately 0.25 mm length per day. The sagitta, lapillus and asteriscus were examined and on average 12 and 28 more rings were observed on lapillus and asteriscus respectively compared to the sagitta showing that these three otoliths form at different times during the development of the fish. To verify the daily nature of the growth rings, experiments were undertaken on captive Usipa kept for a known number of days in a 1500 liter concrete tub and tetracycline was used to induce a uv fluorescent mark on the otoliths.

### **Annuli and Otolith Structure Formation in the Far Eastern Capelin *Mallotus Villosus Socialis*.**

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There is no common interpretation of all the well discernible hyaline zones on capelin otoliths so far. To make age estimation for capelin found off Sakhalin more accurate, the method of polymodal curve (Mina, 1973) was used, with individual curves being constructed for Tatar strait (Japan sea), Aniva bay and south-eastern Sakhalin (Okhotsk sea). Five peaks (1-3, 5, 6) were clearly seen at every curve. The fourth peak, was visible well only at the curve for Tatar strait. The investigations of variation of length growth rates in ontogeny showed that the rates monotonously decreased only in one case from 11 studied, where second and fourth zones were not taken as annuli. Back calculated average length at the first and third zones varied significantly over areas (1, 5-1, 7 cm), mean length at the second hyaline zone being of no great difference (0, 2-0, 6 cm). The formation of hyaline zones on the otoliths of adult capelin in the Tatar strait takes place mainly in May, and of juveniles evidently in winter. The occurrence of juveniles with one or two hyaline zones on the otoliths in different months allows to suggest that the second hyaline zone can be deposited during the summer. In the majority of juveniles such a zone is laid down in June, and in the slow growth rate fish the process can be retarded until the autumn. The above mentioned results support a hypotheses, according to which a second hyaline zone forms mainly as a result of profound metamorphosis inherent to the species at the second year of life.



Non-daily Otolith Increments and Seasonal Changes in Growth of a Pink Salmon (*Oncorhynchus gorbuscha*) Population in Auke Bay, Alaska.

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ABSTRACT

Wild juvenile pink salmon (*Oncorhynchus gorbuscha*) outmigrating from Auke Creek, Alaska were injected with coded-wire micro-tags and released into Auke Bay to identify their date of marine entry and validate the use of post-release otolith increments as a means to independently determine nearshore residence time. Examination of pre and post-release fish showed a distinctive transition check corresponding to the time of marine entry, with well defined otolith increments recorded after this transition. Otoliths from fish captured in the bay throughout the outmigration season were analyzed to examine seasonal changes in growth during the outmigration period and to test the hypothesis that otolith increments were recorded with a daily periodicity.

Fish released during the early part of the outmigration season grew more slowly than later outmigrants and this difference was clearly reflected in the growth of the otolith marine zone. Regressions of otolith increments on days were different between early outmigrating, slower growing fish and faster growing, later outmigrants. Otolith increments were recorded with a near daily periodicity in the slower growing fish, however, faster growing fish clearly recorded significantly more otolith increments than days during their period of marine residency. For all three years combined, increment periodicity was highly correlated with the growth rate of the otolith during the period of early marine residency. Where increment periodicity was less than one per day, SEM analyses showed that this was not necessarily due to counting errors associated with resolution problems. Where more than one increment was produced per day, there was no indication that sub-daily increments could be consistently distinguished from daily increments. This study suggests that otolith increment periodicity is variable over the outmigration season for Auke Bay pink salmon, that it is not always easy to determine the difference between "daily" and "sub-daily" increments, that in this situation, increment periodicity was most closely related with growth rate of the otolith and therefore otolith increment number did not represent a reliable indicator of time for determining early marine residency of pink salmon in Auke Bay, Alaska.

**FISH OTOLITH RESEARCH AND APPLICATION SYMPOSIUM, 1993.**

**POSTER PAPER PROPOSAL:**

**Studies on incremental structures in Antarctic fish otoliths**

**White M G and J R Ashford**

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About 270 teleost fish occur in the Southern Ocean and the majority of these are endemic demersal Notothenioid species. The marine environment is characterised by low but stable temperatures at or near the freezing point of seawater and a production cycle that is markedly seasonal and restricted to a short period during the summer. As a consequence most Antarctic fish have prolonged development cycles and slow but seasonal growth rates. Commercial-scale fisheries exploit both shelf and mesopelagic species but to date few studies have been undertaken to validate the age determination techniques applied to these or other ecologically important species. We report on an improved technique for examination of seasonal increments in large numbers of otoliths, the validation of annulus formation and the growth phase in Antarctic fish by use of time-series samples and the unexpected failure of an experiment to validate daily increments by use of chemical 'tags'.

### The Regulation of Otolith Increment Formation in Atlantic Salmon

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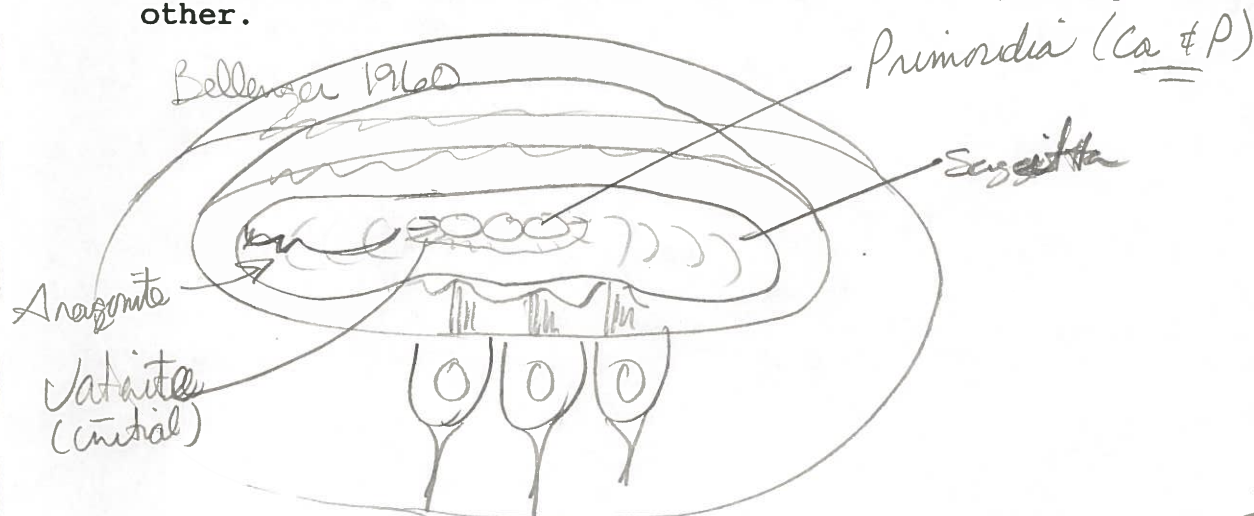
The regulation of increment formation in Atlantic salmon, *Salmo salar*, was investigated from environmental manipulation experiments, in which photoperiod, temperature and feeding regime were varied systematically. These experiments demonstrated that an endogenous rhythm, initiated around the time of hatching, regulated increment formation. An in vivo <sup>45</sup>Ca study of otolith calcification indicated that the endogenous rhythm was entrained to the light-dark cycle, net otolith calcification declining at night and resuming at dawn. The diel decline in otolith calcification coincided with a decline in plasma calcium concentration, suggesting that these two processes may be related. Support for a relationship between plasma calcium and otolith calcification was provided in an experiment in which hypocalcemia was induced. This experiment showed that a reduction in plasma calcium resulted in a net efflux of calcium from the otolith. The nature of the regulatory mechanism was also examined from investigations of somatic growth in individuals. These investigations indicated that the mechanism controlling increment formation was unrelated to somatic growth, since otolith and somatic growth was uncoupled in slow growing juveniles. Further, results from oxygen consumption and increment width measurements suggested that a process related to metabolic rate governed the rate of calcium accretion.

# Ultrastructure of Otolith Formation

(Fundulus)

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The saccular otolith begins development with the formation of primordia in close association with free cells in the endolymph. Primordia consist of a granular- fibrillar material, are high in sulfur, and contain dense nuclei 0.5 to 1.0  $\mu\text{m}$  in diameter. Nuclei do not increase in size but sometimes fuse with adjacent nuclei to form larger dense oblong granular structures. Primordia ultimately fuse and form the core of the otolith. During later growth new growth centers are initiated resulting in complex shapes in some otoliths. Electron diffraction analysis of seven higher teleost species indicates initial crystals of the primordia of the saccular otolith are vaterite with aragonite being the predominant mineral outside the core area. The lagenar otolith is predominantly composed of vaterite. The subcupular layer of the otolith membrane provides the organic matrix for calcification. Matrix deposition precedes crystal deposition. Aragonite crystals nucleate and grow in the matrix. Interlamellar matrix results from organic material compressed by two adjacent crystal layers growing towards each other.



larger / slower growing fish have large otoliths (papers?)

Acrescin = protein layer forms  $\rightarrow$  nucleation site protein  $\rightarrow$  crystal formation

primordia focus (which is best?)

primordia formed at sensory hair tips

Blue slides Bad

Low contrast.

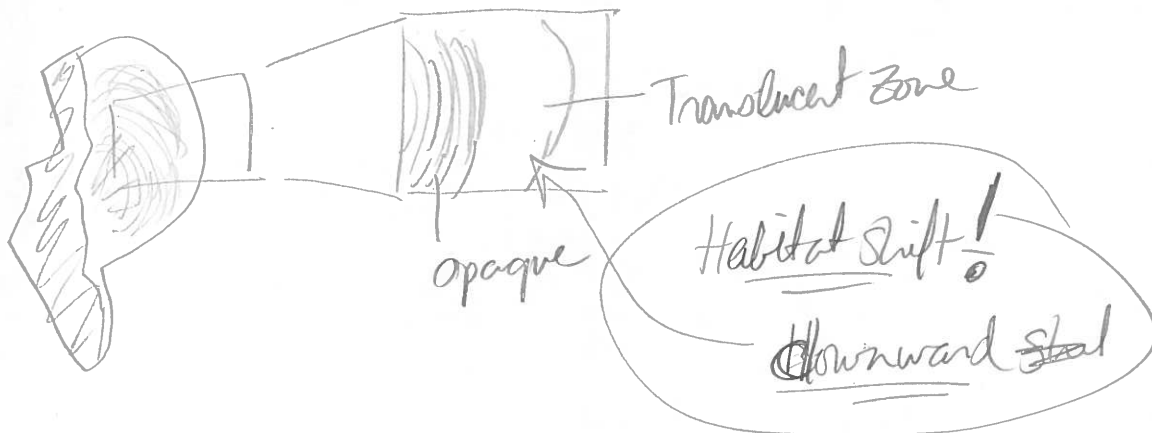
Bt (too much text on slides)

### Application of Otolith Microstructural Analysis Techniques to Squid Statolith Studies

G.D. JACKSON. University of Otago, Portobello Marine Laboratory, P.O. Box 8, Portobello, New Zealand.

The squid statolith is physiologically analogous to the fish otolith. Similarly squids have been shown to have periodic growth increments which can be discerned with the light microscope. Many of the techniques used in delineating daily periodic increments in fish otoliths have been applied to squid statoliths. Squid statolith analysis has included both the enumeration and validation of growth increments. Statolith increments have been found to be laid down daily in a number of tropical and temperate species. This has revealed that the majority of squid species are less than one year old, with the tropical species reaching maturity in less than 6 months. Because squids grow so rapidly, daily statolith increments can be observed across the whole of the life-span, a feature uncommon in fish otolith studies. Statolith derived growth curves for squids have revealed a non-asymptotic form of growth which is very different to fin-fishes. This is despite the widespread application of fin-fish growth models to squids in the past.

increments visible  
Oxy test validation of daily increments.



Starry Flounder, 1984

Tammya

Separation of Spotted Seatrout Collected from Two Areas Within Galveston Bay Using Scale and Otolith Morphology

ROBERT L. COLURA and TIMOTHY L. KING. Texas Parks and Wildlife Department, Perry R. Bass Marine Fisheries Research Station, HC 2 Box 385, Palacios, Texas 77465, USA.

The spotted seatrout (Cynoscion nebulosus) population of the Galveston Bay complex was examined to determine if separate sub-groups exist in the eastern and western areas of the bay system. Circuli spacing of scales and Fourier analyses of the shape of scales and otoliths were examined with the assistance of a computer software program. Discriminate analysis using the circuli spacing and Fourier harmonics correctly assigned 71.8 and 79.0% of the fish to the eastern and western areas of the bay, respectively and suggests at least some separation of the spotted seatrout population within the Galveston Bay system.

### **Relationship of Otolith Size to Growth in Spotted Seatrout and Red Drum Larvae**

G. Joan HOLT and Scott A. HOLT. The University of Texas Marine Science Institute. P.O. Box 1267, Port Aransas, Texas, 783731.

An investigation of otolith size and growth rate in spotted seatrout *Cynoscion nebulosus* and red drum *Sciaenops ocellatus* was carried out to determine the relationship of somatic growth to otolith growth, driven by the need to estimate live lengths from otolith size in wild caught larvae. Larvae grown under two different feeding regimes, well fed and pulse fed (fed every other day), were sampled on day 1 post hatch and then every other day throughout larval development. Anesthetized fish were measured for growth in length, the sagittal otolith was removed and video recorded, and tissues were processed for flourometric measurement of RNA and DNA concentrations. An Olympus CUE-4 image analysis system was used to measure several surface morphometric features of the otolith. Average diameter of the otolith was the most useful of the morphometric measurements. Pulse fed larvae had significantly reduced growth rate that was reflected in the RNA-DNA ratios but not in the otolith diameter to standard length relationship. Otolith diameter was significantly correlated with length through day 18 regardless of the larval growth rate.

**Fish Otoliths as Important Tools in Predator-prey Studies of Bottlenose Dolphins.**

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Bottlenose dolphins (Tursiops truncatus) are apex predators in marine systems, feeding chiefly on fish. The feeding ecology of a resident population of dolphins inhabiting the Indian River Lagoon System (IR) in the eastern coast of Florida is currently under study. The ultimate goal of the study is to quantify fish consumption at the population level, estimate the impact of the dolphins on the IR prey resources, and compare the figures with the takes of commercial and recreational fisheries in the area. Examination of stomach contents of 35 dolphins stranded in the IR indicates that they prey on a minimum of 25 species, mostly fish of the families Sciaenidae, Haemulidae, Sparidae, Mugilidae, and Clupeidae. Scanning electron micrographs of fish otoliths from the main prey species of bottlenose dolphins were taken as an effort to construct a pictorial field guide to otoliths. Regressions relating otolith length and fish length, as well as fish length and fish weight, were highly correlated. These parameters are crucial in estimating original size and weight of ingested prey, and therefore extremely useful in predator-prey studies.



# Variation in Timing of Annulus Formation in the Sagitta of Long Lived Estuarine Species of the Northern Gulf of Mexico

Charles A. Wilson<sup>1</sup> Daniel W. Beckman<sup>2</sup> and A. Louise Stanley<sup>1</sup>

<sup>1</sup> Coastal Fisheries Institute and Department of Oceanography and Coastal Sciences  
CCEER Louisiana State University, Baton Rouge Louisiana 70803 USA (504 388  
6283) <sup>2</sup> Department of Biology, Southwest Missouri State University, Springfield. MO  
65908 USA

The timing and duration of annulus (opaque zone) formation in the sagitta of several long lived estuarine dependent fishes was compared within and between species. We also examined the effect of temperature, salinity, state of maturation, and age on annulus formation. We hypothesize that annulus formation occurs in estuarine species during winter (colder) months and therefore should be in response to temperature minimums.

There was no difference in the timing of annulus formation between sexes or age groups within species. However the timing of onset and completion of annulus deposition varied between species and within species between years. Annulus formation in red drum (Sciaenops ocellatus) occurred between November and June (1985-1992). Annulus formation in black drum (Pogonias cromis) occurred between November and May (1984-1992). Annulus formation in sheepshead (Archosargus probatocephalus) occurred between March and June (1987-1989).

We conclude that annulus formation in sagitti of estuarine dependent fish examined is in part a result of cooler temperature. However, the formation of this opaque zone is also associated with temperature increase during early spring in the fish examined. Physiological and chemical explanations for this phenomenon will be offered

30-40 yrs

Blk span winter other opposite

Sheeps head variable in zone formation over one year

Blk Drum before Red

negative correlation of temp data

### Seasonal Timing of Annulus Formation in Fish Otoliths

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The periodic formation of annuli in otoliths of teleost fishes has been extensively applied in age determination, however causative factors resulting in the deposition of annual bands have not been well documented. Annulus formation has been purported to form as a response to seasonal environmental cycles or as a reflection of the fishes reproductive cycle. Marginal increment analysis, whereby the progression of annulus formation at the growing edge of otoliths is followed throughout the year, may be used to document the timing of annulus formation. A literature review regarding seasonal timing of otolith annulus formation in coastal fish species worldwide revealed inconsistencies in terminology which often made comparisons difficult. However available data generally supported concurrent seasonal formation of annuli in otoliths of species occupying geographically similar habitats. This suggests that environmental seasonality is the dominate factor affecting timing of annulus formation in fish otoliths.

*ambig oto terminology*

*Slow-growth vs fast-growth zones - rate only*

*White zone vs summer - time period*

*Ring, Mark, Band, Annulus -*

*Forming, formed, completed - when used (context)*

## Use of otoliths in ethnohistorical, zooarchaeological, and palaeo-ecological studies.

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Fishes constitute a diverse and dominant component of zooarchaeological materials recovered from prehistoric and historic sites. Use of these materials, especially otoliths, to gather a wealth of information about different human cultures, fish populations, and their changing environments is growing. However, zooarchaeological information is gathered with methods that are unfamiliar to most fishery biologists. The purpose of this paper is to examine otolith information gathered from zooarchaeological materials and its uses. We not only review the literature but also critically examine previous work and our own research with Atlantic croaker (Micropogonias undulatus), sea catfishes (Arius felis and Bagre marinus), and other fishes in the southeastern United States to evaluate information extracted from "old" otoliths. Preliminary evidence suggests that the biology of some estuarine fishes may have changed over relatively short time periods. Potential causes of these changes and some implications of these changes for species management are discussed. Assumptions and limitations of use of otolith information (fish size, age and growth) to understand fishing techniques and schedules of other human cultures are presented.

**Age Validation, Timing of Growth and Annulus Formation for English Sole (Parophrys vetulus) in Hecate Strait, British Columbia, Based on Otolith Interpretation.**

S.E. MacLellan\* and J. Fargo. Department of Fisheries and Oceans. Pacific Biological Station. Nanaimo, B.C., Canada V9R 5K6

We estimated time at liberty, using burnt otolith cross-sections, for 131 tagged and OTC-injected fish recovered at intervals ranging from 1 month to 5 years after the time of release. Estimates were accurate for 90.1% of the otoliths examined from fish ranging in age from 4-14 years. We estimated the timing of growth for the OTC sample and for a series of sequential samples from the commercial fishery by measuring the distance from the last annulus observed to the otolith margin. For mature fish, the period of greatest growth occurred during the March to June period. We suggest a hypothesis, related to the reproductive cycle, which could account for the timing of growth. We also recommend a preferred collection time for commercial samples to avoid ageing error associated with interpreting growth on the otolith margin.

**Use of Daily Growth Increments on Kokanee Otoliths to Assess Stockings of Hatchery-Reared Kokanees.**

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B. HOELSCHER. Idaho Department of Health and Welfare, 2110 Ironwood Parkway, Coeur d'Alene, ID. 83814.

Hatchery-reared kokanees Oncorhynchus nerka form discernible "stress" checks on their otoliths when they are stocked into lakes and rivers. We established the reliability of this mark for distinguishing age-0 hatchery fish from wild fish in Lake Pend Oreille, Idaho. Hatchery fish marked with dietary oxytetracycline (OTC) were recaptured 1-3 months after they had been stocked in the lake. All specimens with OTC marks on their otoliths also has stress checks; fish without OTC marks lacked stress marks as well. We also validated daily otolith increments for age-0 kokanees by comparing increment counts external to the stress check with known days between release and recapture of hatchery fish. Then, counts of daily increments allowed us to correctly identify fish from several co-occurring release groups that had been stocked at different times in the same season. Such discrimination will enhance evaluations of alternative stocking strategies. Because some ring counts varied slightly from days at large, and because fish released in cold, food poor, or otherwise unfavorable sites apparently did not resume otolith growth immediately, we recommend that groups of kokanee be released at least 7 d apart if they are to be subsequently identified by otolith analysis.

**Analysis of Winter Flounder (*Pleuronectes americanus*) Larval Otoliths.**

MARIA BEMIS\*, EINAR HJORLEIFFSON, ANN DURBIN and TED DURBIN. The University of Rhode Island Graduate School of Oceanography, Narragansett Bay Campus, South Ferry Road, Narragansett, RI 02882.

Larval winter flounder otoliths contain poorly defined structures, and increments are difficult to discern. We have developed procedures to make possible the visualization and enumeration of these increments. Methods will be presented. An experiment was carried out where larvae of known age were reared in large outdoor mesocosms in semi-natural conditions. Relationships between growth and otolith size and increment number and width will be presented. Otoliths were found to have a variable number of initial narrow increments (0.2-0.6 $\mu$ m) after yolk sac absorption and during the first feeding period. As larvae emerged from this critical period they began to grow rapidly and otolith increment widths increased to 1-3 $\mu$ m.

# **Identification of Hatchery Reared Red Drum Using Discriminant Analysis of Otolith Banding Patterns**

J.J. ISELY\*, C.B. GRIMES, and A.W. DAVID. National Marine Fisheries Service, 3500 Delwood Beach Road, Panama City, FL 32408, USA.

To evaluate the efficacy of stocking programs we developed a procedure to discriminate hatchery reared and wild juvenile red drum, *Sciaenops ocellatus*, using otolith microstructure. Because hatchery conditions do not approximate natural conditions, growth histories of hatchery and wild fish are significantly different. Otolith banding patterns record the growth history of fish and therefore, provide a method of separating hatchery from wild fish. We measured asteriscus radius from the primordia to each successive daily increment for the first 20 increments in 152 hatchery and 185 wild fish. Change in radius or increment width was used to classify juvenile red drum into hatchery and wild groups. The discriminant function correctly classified 100% of hatchery fish and 100% of wild fish. Hatchery fish were further correctly classified into production period. Up to 94% of wild fish were correctly classified to collection date. We conclude that discriminant analysis of otolith banding patterns provides an accurate means of evaluating stocking programs.

### **Computer Image Analysis of Otoliths. Image Enhancement and Presentation.**

K.W. ESTEP, K. NEDREAAS, and F. MacINTYRE. Institute of Marine Research. Postboks 1870, Nordnes, 5024, Bergen, Norway.

Otoliths are important for the assessment of age and stock in fisheries of biology, and therefore great labor is expended world-wide in their analysis. The analysis of otoliths is time consuming and must be performed by trained personnel, due to the unclear nature of the annuli that are identified in the analysis of these structures. Though video has been used to present, and sometimes to assist in the analysis of otoliths, the full effectiveness of image processing has not been used to improve the quality of otolith video images, or to further automate and objectivize the process of their examination. Using an image-analysis system developed in Bergen, results are presented of image capture and enhancement. Using the techniques, otolith microstructure is more clearly defined. The techniques presented here are useful both for manual analysis on a video screen and for the further analysis of these structures with computers.

C. nepestris



# Use of Statolith Banding to Determine Age of Lamprey

*Don't use larger so much*

F.W.H. Beamish and J.A. Lane\*. Department of Zoology, University of Guelph, Guelph, Ontario, Canada, N1G 2W1.

In the phylogenetically primitive lampreys structures analogous to the teleost otolith, called statoliths, display a banding pattern which can be used to assign age. In northern species, a broad range of environmental conditions causes variation in growth resulting in a narrow band for slow growth and a wider, opaque band for increased growth. Growth in southern species is relatively constant throughout the year and in these populations the banding pattern has not been observed. In their final year of larval development lamprey typically enter a phase of slowed growth during which lipid reserves are increased at the expense of tissue synthesis, in preparation for metamorphosis. During this non-trophic phase an annulus is not formed, thus an additional year must be added when aging post-metamorphic lamprey.

The process of aging lamprey using statolith banding is being validated using intraperitoneal injections of oxytetracycline which cause statoliths to fluoresce along their outer margins. Fluorescence persists for at least 12 months and a complete annulus is formed immediately beyond the marked area.

Currently, the influence of acute and chronic stress on 'annulus' formation is under study, using temperature change and implantation of silastic pellets impregnated with corticosterone.

*Lamprey - parasitic & non parasitic*

*Some varieties consume whole fish.*

*Statolith - analogous to Otolith*

*- Ca PO<sub>4</sub>*

*Size classes overlap*



*OTC inserted & dipped*

*Validation -*

*annulus formed*

*- (annual ?)*

*temperature found to be*

*~~dependent~~ cause of*

*increments*

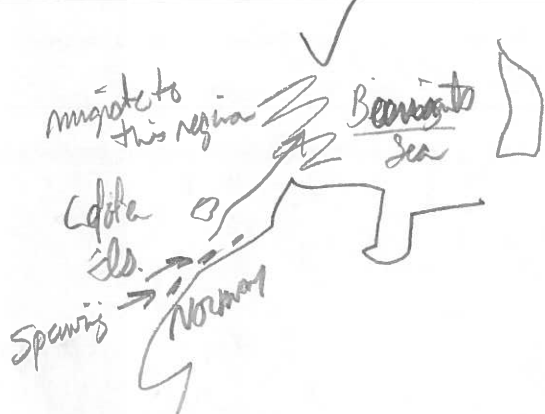
*1 year added for no growth period.*

*Summary*

*- Bands good*

*- Validated*

*- Second growth resp for incr.*



-41-

Now as old as Tyne.  
Older before war (II)

# Otolith Pattern and Shape as a Discrimination of Origin in Various Cod Stocks (*gadus morhua*) Along the North Norwegian Coast

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J.E. ELIASSEN. Norw. Inst. of Fishery and Aquacult.

A programme of devising quantitative methods for cod stock discrimination based on otolith morphometry has been initiated at the Fishery Research Unit and the Norwegian College of Fishery Science in Tromso.

Samples of otoliths of several fjord stocks and of migrating Arcto-Norwegian cod have been studied to devise a quantitative and objective method for determining stock characteristics. To facilitate and improve precision a simple and low-cost computer assisted visual otolith reader (CAVOR) has been devised.

The results confirm the work done by Rollefson in the early thirties, but also points to certain differences between stocks in the general outline of the otolith, that may be more promising. As the outline changes with the size of the fish, a normalizing function has been devised. This function may also be used for enhancing the otolith zone pattern, and for automatic measurement of zone width.

3 types of stocks - Fjords & off shore & ?

Rollefson - studied cod oto diffs. (1925)

Migrate from sea to fjord

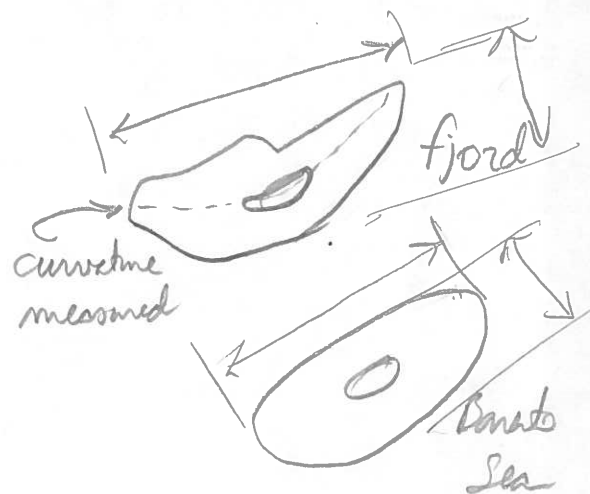


thickness

Curvature through otolith tips



curvature



Lofoten only for Barents  
Fjords cods separate

**Effects of Variable Temperature and Feeding Rate on Daily Increment Deposition and Growth of Otoliths of Colorado Squawfish, Ptychocheilus lucius**

K. R. BESTGEN\*. Larval Fish Laboratory, Department of Fishery and Wildlife Biology, Colorado State University, Fort Collins, Colorado 80523 USA.

Eggs and larvae of federally endangered Colorado squawfish, Ptychocheilus lucius, were reared in the lab to validate use of otoliths to determine age and growth histories. Eggs were hatched and larvae were reared in constant (22° C) and fluctuating (18, 22, and 26° C,  $\pm 2.5^\circ$  C) water temperature regimes to determine the effects of diel temperature change on increment deposition. Effects of variable feeding rate and food deprivation on increment deposition were also investigated. Otoliths (sagitta and lapillus) of Colorado squawfish form prior to hatching but do not deposit daily increments until after hatching. Daily increments were deposited in otoliths of fish in all temperature treatments and the number of increments generally corresponded with post-hatch age. However, increments in otoliths from fish reared in fluctuating temperature regimes were much clearer and age determinations more accurate than in otoliths from fish reared in constant temperature. Fish length and otolith growth generally correspond. Otolith microstructure analysis appears to be a useful tool for determination of age and growth parameters in early life stages of Colorado squawfish and is currently being applied in recruitment studies of wild populations.

**Bar Codes in a Billion Salmon: Alaska's Venture into Thermally Manipulating Otoliths.**

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Alaska salmon hatcheries have embraced otolith thermal marking as a cost effective method to evaluate release strategies. Alaska fisheries managers have identified it as the best method available to separate wild and hatchery salmon on the fishing grounds for in-season management. Negotiators for the U.S.\Canada treaty have stipulated that it be used to allocate salmon that have been reared as part of treaty agreements. As a result, literally millions of marked fish are out at sea ready to return within the next two years and there are plans to release hundreds of millions more. This paper summarizes the efforts in otolith research, otolith processing, pattern recognition, and sample size determination, to prepare for these returns. The need for cost effective mass marking, the analogy of bar codes, and the tendency of researchers to show off their best otoliths, has directly contributed the situation in Alaska where currently a very large tail is wagging the dog. Despite some pitfalls and unanticipated problems, the bar code analogy is appropriate and the prospects are encouraging for the development of mass marking programs provided that research continues and standards in marking protocols are followed.

### Three Dimensional Imaging of Internal Structure of a Juvenile Walleye Pollock Otolith

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A 3-D model of a juvenile walleye pollock otolith was constructed from 32 serial, frontal plane sections of a single sagitta. Acetate peels were made of polished and etched preparations every 25 or 50  $\mu\text{m}$  during sequential grinding of the otolith. Prominent increment checks, accessory primordia and other structures of interest were digitized from the serial sections. The otolith was reconstructed using an image processing package (SKANDHA, Dr. J. Prothero, Department of Biological Structure, University of Washington). The resulting 3-D model can be viewed, rotated, and sectioned in any plane, highlighting various internal structures. The model has been very helpful in visualizing otolith structure and growth patterns. Its utility in determining backcalculating procedures and detecting bias in backcalculations will be discussed.

**Geographic Variations in Otolith Morphology, Microstructure and Growth Patterns of Cosmopolitan Lanternfish, Nominally *Ceratoscopelus warmingii* (Myctophidae).**

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Morphology, microstructure and growth patterns of *C. warmingii* otoliths collected over its entire Atlantic distributional range were studied. At least five distinct geographic forms were recognized, i.e. northern-subtropical, tropical-equatorial, northern-gyre, southern-gyre and southern-subtropical. The forms differed in otolith morphology, microstructure and growth patterns. This was in agreement with the results of earlier studies describing the existence of such geographic forms of *C. warmingii* on the basis of other taxonomic characters. Otoliths grow relatively slower than body during rapid growth and relatively faster during slow growth. According to that rule a slow-growing fish should deposit larger otoliths than a fast-growing. On the contrary to that, the fast-growing tropical-equatorial Atlantic form of *C. warmingii* was characterized by the largest otoliths, while the slow-growing forms, i.e. northern-gyre and southern-subtropical, were characterized by the smallest otoliths. In addition to the Atlantic materials some samples from Pacific and Indian Oceans were also examined and similar differences were observed. These findings suggest that this previously recognized cosmopolitan species may comprise a multispecies-complex.

Comparative Otolith Growth in Carangoid Fishes: Evolutionary or Ecological Relationships

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Historical literature on otolith structure has placed strong emphasis on elucidating evolutionary relationships. Other studies suggested that "form follows function" and that ecological similarities, not kinship could be the main force in producing similarly structured otoliths.

Kijima et al. (1986), Laroche et al. (1984), and Smith-Vaniz (1984) have provided a phylogenetic framework for Carangidae and several related families such as Rachycentridae, Coryphaenidae, and Echeneidae. The wide range of life history and ecology of these species provides a test for this evolution vs. ecology hypothesis.

As examples: there are several distinctive otolith shapes: a) round/oval - Trachinotus, b) elongate/round - Elagatis, c) elongate/robust - Caranx, and d) elongate/slender - Seriola, Alectis. Rachycentron is similar to Seriola, while Nematistius resembles Caranx.

The otolith growth pattern, however, is quite uniform and seems to cross phylogenetic lines, with growth occurring primarily in one plane. This produces an otolith with the core on one side and growth increments forming on the opposite surface as the otolith becomes thicker with age.

**Otolith Daily Increments in the Myctophid Fish Lampanyctus parvicauda Parr, 1931 in the Northern Chilean Coast.**

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Myctophids are very important fishes in oceanic trophodynamics, being preys for several fishes and marine mammals, and have been also considered as an important potential fishery resource worldwide. It is therefore important to study their biology, including age and growth, before eventual fishing activities may start. One of the methods for this purpose is to study the growth rings present in fish otoliths, technique which has already being used in several myctophid species.

Lampanyctus parvicauda is one of the most abundant species found in northern Chilean waters. The specimens used in this study were captured at night with an anchovy purse seine and a midwater trawl off Arica (18°30'S) and Pisagua (19°29'S) during February 1988 and 1989, and preserved in alcohol. The sagittal otoliths were treated with a H<sub>2</sub>SO<sub>4</sub> solution for five minutes and grinded with a # 0 and # 2 sandpaper until a thickness where the increments were clearly seen. Otoliths were then mounted and examined under a compound microscope.

Daily, fortnightly, monthly and annual bands, having the last ones a mean number of 380 increments, were recognized in the otolith, suggesting that the method can be a valuable tool to study age and growth of this species. According to our findings L. parvicauda is a short-lived fish, being the largest analyzed about three years old and the probable age at first maturity two years.



**Study on juvenile growth pattern of European hake (Merluccius Merluccius L.) using whole otoliths and length frequency distributions.**

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Growth pattern of juvenile European hake is studies on the basis of comparative analysis of fish length frequency distribution obtained from backcalculating rings measured on the whole otoliths and fish length frequency distributions of groundfish surveys carried out between September 89 and September 90. The results of this study permit identify the first annual ring. From observation of the edge of otoliths throughout the study period the location and time of formation of this annual is determined.

### **Ageing of squid *Loligo forbesi* from growth rings in statoliths**

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Ageing of loliginid squid using growth rings in statoliths has recently been validated. Rings are believed to be daily, perhaps related to feeding events.

In previous studies on the North East Atlantic loliginid *Loligo forbesi* in UK waters, lifespan has been inferred from analysis of length-frequency and maturity stage data, which suggests an annual life-cycle, semelparity and a maximum lifespan of 12-15 months. Recent results however point to the existence of 'short' and 'long' life history strategies and confirmation using an ageing technique is urgently needed.

The present paper describes methods for the preparation of statoliths from *Loligo forbesi* and presents preliminary results of counts of putative daily growth rings. Squid from Scottish and Azores populations were sampled. The main findings are that age and size are very poorly correlated, indicating wide variation in growth rates, and that some animals may live for 18 months or more.

### **Age, Growth and Early Life History of King George Whiting larvae.**

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The age and growth of King George whiting was examined in both field collected and laboratory reared material in order to validate the daily nature of increment formation and compare interannual growth rates of larvae and newly settled juveniles. Tetracycline marking of newly recruited juveniles was undertaken in cages in nursery areas.

Back calculated spawning dates for whiting range from late February through to mid July with peaks during April and May. Peaks in spawning dates indicate some lunar periodicity. King George whiting eggs hatch after 48-60 h at 16.5-18.7 deg. C. The larval phase is protracted, with larvae remaining planktonic for 50-80 days prior to recruitment to nursery habitats at 15-20mm. Growth of whiting larvae in the laboratory was similar to estimated growth rates of field collected fish. Otolith microstructure was less well defined in reared larvae although daily increment formation was observed.

### **On the Use of Otolith for Modelling the Sea Temperature - Growth Relationship and its Possible Application on Fishery Management**

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An autoregressive model with exogenous input was used to explore the relationship between sea surface temperature and blue whiting growth parameters. Sea temperatures in April, May and June were used as input because water stratification in the Western Mediterranean and blue whiting recruitment to the fishing ground take place during those months. The otolith nucleus diameter and annulus width from age groups 1 - 3 were selected as output because the fishery is supported by those age groups. A sample of 4767 individuals were selected for aging from 145,685 fishes collected between 1950 and 1987 in the Catalanian coast. 867 otoliths were embedded in plastic resin and sectioned for measuring the nucleus diameter and annulus width. The best model is expressed by the equation:  $A W_{1(t)} - 0.6379 A W_{1(t-1)} = 0.1761 T^{\circ}C \text{ May}$ , and it was obtained using sea temperature in May as input and annulus width of age group 1 as output. The model states that if temperature rises, the individual will grow faster and this is supported by the cross-correlation function for CPUE and sea surface temperature in May. It is suggested that this kind of models could be useful for predicting harvests in spite of their ecological constrains and emphasizes otolith use in population dynamic studies.

**Early Growth of Yellowtail Flounder, *Limanda ferruginea*, on the Scotian Shelf**

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More than 1000 yellowtail flounder larvae (4 to 18 mm standard length) were captured from July to September 1991 on Sable Island Bank, Scotian Shelf. A light microscope combined with an image analyzer were used to examine the otoliths of a subset of the larvae ranging in development from hatch to metamorphosis. On average, the larvae increased in size by 1.4 mm SL between July and August and 1.3 mm SL between August and September. Estimates of growth based on length frequency distribution at date and length at age data are compared.

## Age and Growth of Juvenile Tautog (*Tautoga onitis*, Family Labridae) in Narragansett Bay, Rhode Island

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Tautog (*Tautoga onitis*) is an important recreational and commercial species in New England coastal waters. Exploitation in Narragansett Bay, Rhode Island has increased with closure of the winter flounder (*Pseudopleuronectes americanus*) fishery. No information is currently available on the age structure and growth of post-settlement young-of-the-year juveniles in Narragansett Bay. Daily growth increments from lapillae of young-of-the-year juvenile tautog (TL 26-98 mm, weight 0.2-18.3 g) collected from Narragansett Bay were examined using image analysis to estimate age and growth of early life history stages. Increase in otolith length and width was linearly related to increase in fish total length (TL) and fish wet weight. Increment width varied with age. A mark corresponding to estimated time of settlement was consistently found at approximately 3 weeks of age. Juveniles were estimated to be between three and four months of age corresponding to hatch dates during late spring and early summer. Age-specific growth was estimated from length-at-age data and from back calculation. Positive relationships were present between absolute growth rate (TL/number of increments, weight/number of increments) and fish TL and weight.

**Daily Growth Increments: A Tool for Validating Annuli in Sagittal Otoliths of the Chilean Sardine (*Sardinops sagax*).**

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The objective of this research is to show that daily growth increments can be used to validate annuli in sagittal otoliths of the Chilean sardine (*Sardinops sagax* Jenyns) in northern Chile. From 1,161 Chilean sardine aged, 200 were subsampled for validation. Annuli were validated through daily growth lines in two ways: from the time between estimated date of hatching and the end of each winter, and counting the daily lines between the different hyaline rings. Consistent estimates were found with both methods but not with the traditional methods used by the government management institution. The modal value of date of winter annulus formation varied between July to September; the summer ring between December to February. The radius at the first hyaline mark showed great variability (mean = 323  $\mu\text{m}$ , C.V. = 118.23) directly related to the individual's season of hatching. Maximum age of *S.sagax* is 6 years. From the 3rd to the 6th year, two hyaline rings are deposited each year: the first in winter at the time of the main spawning period, and the second during secondary spawning in summer. The management implications of the different methods of age determination are discussed.

**Growth of juvenile black sea bass, Centropristis striata, during an upwelling event on the New Jersey continental shelf.**

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Periodic stratification and upwelling events appear to be regular oceanographic features along the New Jersey coast in summer, but their effects upon coastal fishes are largely unknown. We examined growth records on sagittal otoliths of black sea bass caught in 1991 and conducted several laboratory experiments to 1) validate the daily formation of otolith increments across a range of environmental conditions; 2) assess the utility of otolith increments as a record of stratification and upwelling events; and 3) test several hypotheses about effects of these events on growth of black sea bass. Validation of daily otolith increments in juveniles (up to 98 mm TL) was established by rearing experiments and recapture of tetracycline-marked individuals. Experimental manipulations in the laboratory are being performed to determine if temperature changes during upwelling produce distinct marks on otoliths. Mark-formation on otoliths of free ranging black sea bass caught before, during, and after the upwelling event are being compared. Results of these experiments should provide a better understanding of the complex effects of stratification and upwelling on growth of juvenile C. striata and other fishes that use the inner continental shelf as a nursery area.



**Age Validation of Southern blue whiting (*Micromesistius australis*) in New Zealand.**

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Age and growth data are fundamental to the understanding and modelling of fish populations. The data are important when assessing the optimum exploitation rate of a stock and can also be used to estimate stock size using cohort analysis or virtual population analysis (VPA). Recent papers have emphasised the importance of using reliable ages when carrying out these analyses.

In the present paper ages of juvenile fish are validated by following the monthly progression of modes in length frequency data and by counting presumed daily increments in the smallest fish. Ages of older fish are validated by the ability to follow strong year classes through the fishery using both otolith ages and length frequency data. A dominant mode in the length frequency distribution of the fish could be followed from 1981 to 1989. Ages derived from otoliths suggested this mode comprised fish of two year classes, born in 1979 and 1980, which entered the fishery as two year olds and were 9 and 10 years old in 1989. It is concluded that otolith ages are validated up to at least age 10.

### **Optimalization of the Back-Calculation Method.**

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The present work describes three component parts of the optimalization of the back-calculation, i.e. sampling, validity of age determination, ways and methods of counting growth rate. The first part concerns catching and sampling time, sample size, measurements of fish length, sampling of calcified structures and their measurements. The second part regards the validity of age determination. This can be made by two methods; direct methods like tagging (e.g. plastic marks and tetracycline) or indirect methods like comparison of age-determination techniques between other calcified structures by individual readers. With ten to twenty counting growth rate methods (third component part) only three formulas have been selected: Fraser-Lee with constant "C", Fry (1943) with constant "C" and Dahl-Lea. In the first two examples the constant "C" is the real fish length, when the examined calcified structures appear. When the species grows in the first year to about 40 % of the maximum body length Dahl-Lea formula can be used. Usually we have some differences if we count growth rate to compare back-calculation and direct measurements methods. These results give various estimated parameters of von Bertalanffy equation. These differences can not be ignored. The suggested validation of the back-calculation method should be recommended as correct to count growth rate.

### **Age Determination and Growth Rate Assessment for the Peruvian Horse Mackerel**

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The study of the inner structure on thin slices of otoliths of horse mackerel - *Trachurus symmetricus murphyi* (Nicols) taking into consideration the sizes of young fish caught in 1980-1990 made it possible to define more precisely the location of annual growth marks and determine the time of their formation.

A new approach is suggested to the growth backcalculation of fish growth taking into consideration the curved dependence between the fish length and the otolith size.

The age and linear growth of the horse mackerel more corrected. It was shown that the reduction of the horse mackerel growth rate is observed in 3 year olds when fish reach average length of 25,6 cm.

The Use of Otolith Microstructure to Resolve Issues  
of Spawning Seasonality and First-year Growth of  
White Hake, *Urophycis tenuis*, in the Gulf of Maine-  
Georges Bank Region

by

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The white hake is a commercially-important gadid of the Gulf of Maine-Georges Bank region, but a question exists as to whether or not the species spawns successfully in this region. In addition, no age validation study has been conducted for the species to date. In this study, larval and juvenile white hake otolith microstructure is examined to evaluate time of spawning and the formation of: 1) a 'settling check' (a mark associated with a change in habitat as pelagic larvae assume a demersal juvenile existence); 2) a check occurring in individuals of certain estuarine systems presumed to be associated with high late summer temperatures; and 3) the annulus marking the first year of growth.

Specimens examined were collected from numerous locations including coastal waters of Maine and Massachusetts as well as Georges Bank. Our results tend to support the recent hypothesis of Fahay and Able (1989) that white hake in this region are recruited from a spring-spawning population inhabiting the continental slope south of Georges Bank and southern New England. Additionally, we validate an otolith-based ageing methodology to describe first-year growth for the species.

**Biometric Features and Age of Glass Eels *Anguilla anguilla* (L.) ascending the Vilaine estuary (France)**

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The biometric features and the age (daily age) of ascending glass eels were studied through measurements of total length, weight, condition factor, development of pigmentation and SEM examination of sagittal otoliths. Glass eels were sampled monthly from the Autumn 1991 to the Summer 1992. Additional catches were made on the continental shelf, down to 80 m depth. The arrival of newly glass eels from the ocean was continuous as indicated by the relative percentage of transport individuals. The mean age of these individuals ranged from 7 to 9 months, less than one year in contradiction with Schmidt's hypothesis. Results are discussed in term of local/wide scale variability in age and in migration processes.

Validation of Age and Growth Rates in Juvenile American Shad (*Alosa sapidissima*). Karin E. Limburg, Cornell University, and Edward B. Brothers, EFS Consultants, Ithaca, NY.

Abstract: The validity of determining fish age and growth rates by examination of otolith microstructure was tested on juvenile American shad whose age and rearing history was known, thus extending the work of Crecco and Savoy (1985) on larval shad. Sagittal otolith increment counts underestimated true age by 1.5% to 3.5%. This was primarily due to the occurrence of a blurred zone on the otolith that corresponded to the time of transfer of the fish from an outdoor pond to an indoor growth chamber; this transition zone contained indistinct increments that were difficult to resolve with a light microscope. However, increment widths measured with a digitizer did correspond well with the growth history of the fish, and additional, temperature-induced marks on the otoliths of certain fish provided an accurate time mark that validated the daily increments in the lab. Increments laid down in the laboratory were less distinct than those laid down in the outdoor pond; in addition, subdaily increments were formed during the period of most rapid growth (outdoor pond).

Age of giant squid Architeuthis, based on statolith readings:  
a case in the point

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The giant squid Architeuthis is a largest living invertebrate. Its growth and longevity was unknown so far. It is not surprising as this squid is very rare and statoliths were not frequently investigated. The case of the giant squid, albeit isolated, may be important in resolving the present discussion concerning the growth of cephalopods: is it fast or slow (especially in deepwater oceanic cephalopods)?

In the study three pairs of statoliths of mature giant squid females were investigated. Their external and internal morphology is described. The statolith structure is rather complex. Number of growth rings in both cases do not exceed 1000, suggesting the very quick growth rate (mantle lengths 1400, 1680 and 1850 mm). These counts were not validated, however. The problem how to count and believe when the direct validation is impossible is discussed.

**Analysis of Sectioned Otoliths to Determine Age and Growth of Wreckfish, Polyprion americanus, off the Southeastern United States**

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Sectioned sagittae were used to estimate the age of wreckfish Polyprion americanus, a large, deep-water temperate bass which is the target of a rapidly developing commercial fishery on the Blake Plateau off the southeastern coast of the United States. The oldest fish examined was estimated to be age 31 (years); it measured 1,460 millimeters total length and weighed 51 kilograms. Back-calculated mean lengths of 543 fish ranged from 319 mm to 1,437 mm for fish aged 1 and 31 years, respectively. The von Bertalanffy growth equation was fitted to back-calculated lengths:  $L_t = 1,412 (1 - e^{-0.09(t+2.57)})$ , where  $L$  = total length (mm), and  $t$  = years. Wreckfish were recruited into the fishery at age 4. Most fish harvested were 8-12 years old. Catch curves were constructed from species age-length keys in order to estimate total instantaneous mortality rates ( $Z = 0.22 - 0.50$ ) by year, 1988-1991. Mortality increases drastically after age 15. Natural mortality rates ( $M$ ) were calculated based on the relationship between the theoretical growth parameters and mean water temperature ( $M = 0.15 - 0.30$ ). The length-weight relationship is  $W = 2.2 \times 10^{-8} L^{2.956}$  where  $W$  = weight in kilograms, and the whole weight (TW)-gutted weight (GW) relationship is  $TW = -0.3667 + (1.141) GW$ .



**Age and Growth of *Acanthurus lineatus* and *ctenochaetus striatus* Determined Using Otoliths from Kavieng and Port Moresby Sites in Papua New Guinea.**

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This study attempts to determine the ages of two fish species (*Acanthurus lineatus* & *Ctenochaetus striatus*) using otoliths from the Kavieng and Port Moresby sites of Papua New Guinea. It also attempts to compare growth between the two areas. The method used in this study to age the different species was slightly modified from that developed by Ralston and Mivamoto (1981, 1983) and Ralston (1985). Ages of *A. lineatus* from Kavieng ranged from 460 days for a 15.6 cm (FL) individual to 535 days for a 17.8 cm (FL) individual. At Port Moresby, ages ranged from 540-1190 days for 13.9 cm (FL) to 23.1 (FL) individuals respectively. Age for *C. striatus* from Kavieng ranged from 470 days for a 13.4 cm individual to 795 days for a 17.3 cm (FL) individual and the age for the species from Port Moresby was found to range from 440-740 days for individuals of 14.2 to 19.1 cm (FL) respectively. Estimated age in days was significantly correlated to fork length for the species, *A. lineatus* and *C. Striatus* (sexes combined) from both sites. The two species, *A. lineatus* and *C. striatus* had the same growth rates at the two different sites sampled.

## **Otolith Pattern as an Index of Eel Habitat in the River Loire (Brittany, France).**

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Ability of otolith pattern to discriminate between estuarine and fluvial stock units in eel population of the river Loire were tested (100 eels sampled in each fishing site). Rest lines were revealed on ground "Sagitta" with toluidin blue and their microstructure described using scanning electronic microscopy. Biometric criteria cannot be used to distinguish both samples, whilst number and microstructure of rest lines have been proved efficient to characterize each biotope. Rest lines were less numerous on "estuarine" otoliths ; through S.E.M. observation, most of them appeared as "large and twinned" grooves supposed to be annual marks. "Fluvial" otoliths displayed as twice as many rest lines, including annual twinned lines and supernumerary narrow checks as well. Results are discussed with reference to validated data on a dammed river (river Vilaine). The complex pattern of fluvial otoliths is assumed to describe upset growth related to more fluctuant environmental conditions. As a fisheries application, two age-length keys should be built and used to estimate age structures of both samples. We conclude that otolith pattern analysis could be of great interest in stock identification for amphihaline species.

**Age and Growth of Larval Kapenta *Limnothrissa miodon* as Determined by Otolith Daily Growth Increments.**

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Growth increments in otoliths were used to estimate the age of larval *Limnothrissa miodon*, a tropical freshwater sardine from Lake Kariba on the Zambezi River between Zambia and Zimbabwe in January to May 1992. Increment initiation was assumed to commence at yolk-sac reabsorption to coincide with the onset of exogenous feeding. Age therefore was represented by the number of increments counted ignoring the period between hatching and yolk-sac reabsorption which for tropical fish is considered very short. Age was found to vary from 12 to 80 days (mean 32; sd=12.75) for fish of 5.5 to 15.5 mm (mean 6.8; sd=1.48) in length. There was a close correlation ( $r^2=0.91$ ) between standard length and otolith growth at a daily level which was used to backcalculate instantaneous growth rates. Increment widths were narrowest for the first 10 days (less than 1.5  $\mu\text{m}$ ), increasing rapidly to increment widths of greater than 2.5  $\mu\text{m}$  from 10 to 45 days followed by a decline to between 1.5 to 2.5  $\mu\text{m}$  from 50 days onwards. Increased ability by the larvae to capture prey could explain for the rapid increase in growth after the first 10 days.

**Differential growth of larval cod - evaluation of interpretations drawn from otolith microstructure analysis.**

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The early life of larval and juvenile cod in the northeastern North Sea was studied during field investigations May 1992. At a series of stations the condition and growthrate of cod were investigated using different methods. Morphology, lipid composition, RNA/DNA ratio and otolith microstructure were analysed and findings compared. Growth rate of cod differed significantly between the stations in the investigated area, and in general the interpretations drawn from otolith microstructure were consistent with the findings using the other methods. The implications of the results are discussed.

## Potential for Age and Growth Investigations of Exploited Fish Stocks in the Eastern Caribbean Region

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Relative to temperate species, comparatively few data exist on age and growth of exploited stocks in the Eastern Caribbean. Given current interest in length-based approaches to stock assessment and the frequently acknowledged need to validate length-frequency data with age determinations, such a shortcoming is a serious impediment to development of an adequate understanding of the population dynamics of exploited fish stocks in the Caribbean region. In an exploratory survey of several families of commercially-significant fish (including Coryphaenidae, Scombridae, Carangidae, Lutjanidae, Holocentridae, Scaridae and others), the utility of otoliths, fin ray sections, vertebrae, and scales for studies of age and growth is assessed at the annual level of precision. When interpretations of age and growth are available through length-based modal progression techniques or from mark-recapture studies, such inferences are compared with age-based data derived from examination of hard parts. In the instance of otoliths, the presence of presumptive daily growth increments is also noted.

**Age and Growth of King Mackerel, Scomberomorus cavalla, in North Carolina**

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Declines in king mackerel landings in the 1980's led to recreational and commercial conflicts and management efforts to resolve them. Data on the age and growth of king mackerel off North Carolina are necessary for reliable stock assessments and to determine whether implemented management measures accomplish desired results. Sagittae from king mackerel captured in North Carolina's hook-and-line fisheries were examined using reflected light. Measurements were made from an image on a high resolution monitor with a sonic digitizer from the focus to the distal edge of each opaque ring and to the otolith margin. Otoliths were classified into age groups based on the number of opaque nonmarginal marks. Ages were determined from 521 whole and 896 sectioned otoliths. Females ranged in size and age from 460 mm to 1,520 mm FL and 1 to 26 years, and males ranged from 420 mm to 1,245 mm FL and 1 to 20 years.<sup>2</sup> Correlations of fish length with otolith radius were significant ( $0.38 < r^2 < 0.80$ ). Frequency distributions of distance from focus to each annulus for successive age groups were unimodal. Mean back-calculated lengths at age were greater for females than males in each age group. Growth increments were largest for the first three years, after which they gradually decreased. Asymptotic length from von Bertalanffy equations was greatest for females from sectioned otoliths ( $L_{\infty} = 1,370$  mm FL). Attempts to validate annual ring deposition by using marginal increment analysis and mark-recapture studies were inconclusive. Data from this study were utilized in annual stock assessments by federal management agencies.

**The Essential Use of Otoliths in the Determination of Growth of 3 Central American Cichlids in a Tropical Floodplain River from South Mexico**

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The objective was to describe the characteristics of fish growth in a tropical floodplain river. The study turned on 3 Central American cichlids from San Pedro river, South Mexico: *Petenia splendida*, *Cichlasoma synspilum* and *Cichlasoma managuense*. Fish captures were made during 1988-1990. The extended spawning seasons, the great variation in maturation sizes and final sizes of males and females, and the deposition of multiple marks on fish scales forced us to use otoliths instead of length-based methods or fish scales examination. Cross acid-etched ground sections of sagittal otoliths were examined under scanning electronic microscope (SEM). The daily nature of the increments was first verified. Growth rates and estimated hatching dates of young of the year were studied. The amount of growth in the first year was measured. Change in otolith increment width was noted. Correlation with environmental factors was examined. Fish otolith studies appear essential to understand the dynamic of fish growth in a tropical floodplain river environment.

**Growth pattern of megrim (*Lepidorhombus whiffiagonis* and *L. boscii*) in Nord-East Atlantic (ICES Div. VII<sub>j-k</sub> VIII<sub>ab</sub> VIII<sub>c</sub> and IX<sub>a</sub>).**

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Growth pattern of both species of megrim, (*Lepidorhombus whiffiagonis* and *L. boscii*) in Nord-East Atlantic area is analyzed as a function of latitude and deep to estimate the different growth parameters and the influence of these values in the stock assessments. A sample of 4719 megrim otoliths is examined in waters of ICES Divisions VII<sub>j-k</sub> VIII<sub>ab</sub> VIII<sub>c</sub> and IX<sub>a</sub> and the growth parameters is determined to test the significant differences.



**Age Validation of Two Tag-Recaptured Atlantic Albacore, Thunnus alalunga, Based on Dorsal and Anal Spines, Pectoral Rays, Vertebrae, and Otoliths**

E. D. Prince<sup>1\*</sup>, D. W. Lee<sup>1</sup>, J. L. Cort<sup>2</sup>, G. A. McFarlane<sup>3</sup>, and A. Wild<sup>4</sup>  
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Transatlantic movements of two Atlantic albacore (Thunnus alalunga) were established based on tag recoveries from fish released in the Bay of Biscay in the eastern Atlantic and recaptured 6.4 and 4.5 yrs later off the coast of New York in the western Atlantic. When recaptured, one albacore weighed 23 kg, measured 109 cm fork length, was a male, and carried a Spanish tag; the other recapture weighed 19 kg, measured 106 cm fork length, and carried a French tag. Both fish were originally tagged, measured for fork length, and released by Spanish and French scientists, respectively. We obtained dorsal and anal spines, otoliths, vertebrae, and pectoral rays from the Spanish recapture and vertebrae from the French recapture for evaluation of accuracy and precision of age determination techniques.

Age of both albacore were determined from tagging records (relative known age) based on the estimated age for the measured size-at-release and the time-at-large. Tagging record age for the Spanish recapture was 8-9 years and for the French recapture was 6-7 years. These data were compared to the estimated ages from eight hard structures from the Spanish recapture and one structure from the French recapture for validation of ageing techniques. The precision of age estimates from five different readers were also evaluated using the average percent error method.

**The Use of Otoliths for Age Determination of Eastern Bering Sea Walleye Pollock  
(*Theragra chalcogramma*)**

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NORWAY

Otoliths were used for age determination of walleye pollock (*Theragra chalcogramma*). Ages determined from burnt otolith sections were generally greater than those obtained from otolith surface. This difference increased substantially with age. Interrelationships among age, fish standard length, otolith length and otolith weight were examined. The author also discusses the ageing methodology.

## **Estimating Otolith Ages from Two Tag-recaptured Lake Trout (*Salvelinus namaycush*)**

**S.T. SCHRAM \***

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Aging lake trout with scales has been unreliable due to crowding of annuli on the outer margins. This study estimated otolith ages from two wild Lake Superior lake trout that were marked as adults and recovered after being at liberty for 14 and 17 years. Sagittae otoliths were polished with 600-grit abrasive paper to the sagittal plane and examined under a Bausch & Lomb compound microscope at a magnification of 4-40X. Both fish had estimated ages of 26 years, however the first and second annuli were difficult to distinguish. Tag returns, combined with age validation, indicated lake trout were older than previously thought.

Aging methodology must be continually validated as the population increases, growth slows, and fish become older. Reliable aging of a slow-growing, long-lived fish like lake trout is an essential management prerequisite.

### **Age Determination Methods for Nova Scotian Hake.**

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The work finishes the 26-year-long scientific discussion between Canadian and Russian scientists on the problem of age-length relation for the age group 1 of Scotian hake (Noskov, 1966; Sauskan, 1966; Senina, 1966; Hitchi, 1965.) The Canadian point of view was adopted as an official one (Hunt, 1976, 1979) according to which fish up to 25 cm in-length with moda of 20-23 cm is considered as 1-year-old. We have shown at diurnal level that fish of 20-25 cm, caught in April, was aged as 500-550 days old, and belonged to the last but one spawning, not the last one, as assumed before. Thus, the first age group, presented by Hunt (1978, 1979), is acutally divided into 2 year-classes of 1-year-olds and 2-year-olds. Estimates of all annual growth zone radii of otolith, fish mean length-at-age, length-age relation in days, visual signs of annual and additional rings are presented in the article. Results in express-analysis of fish age.

## Age Structure of Adult Red Drum Collected in Georgia

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The red drum, Sciaenops ocellatus, is a popular marine sportfish throughout the southeastern United States. Fishing mortality of immature fish is high, particularly during the first summer of life. Regional investigations within the South Atlantic Bight indicate that recruitment to the spawning stock is compromised by this intense harvest. Red drum leave inshore waters at maturation and join aggregations of spawning adults in the open ocean. This emigration has confounded efforts to quantify survival to adulthood. Sagittal otoliths taken from large red drum collected during 1988-1991 were used to describe the age structure of the adult stock utilizing continental shelf waters off Georgia. While some young adults (Age 4-12) were collected, older individuals (Age 12-28) were most abundant confirming that survival to adulthood has been poor in recent years. Relative abundance within these samples suggests that survival to adulthood may also be influenced by climatic conditions during the first winter of life.

**Seasonal Variation in Age at Recruitment and in Early Life History Growth Rates of *Sicydium plumieri* (Bloch) in Dominica, W.I., as Inferred from Otolith Analysis.**

K.N.I. BELL\* and J.A. BROWN. Biology Department and Ocean Sciences Centre, Memorial University of Newfoundland, St. John's, Newfoundland, Canada A1B3X9.

*Sicydium plumieri* is an anadromous gobiid whose eggs hatch in rivers, and whose larvae immediately move to the sea where they remain through larval and postlarval phases until returning to fresh waters and settling as juveniles. In this first early-life-history study of this species, egg and larval production was found to be continuous throughout the year, and recruitment to be episodic and predominately lunar-synchronised. Ages at recruitment were inferred from sagittal otolith increment numbers, and increment widths were used to infer daily growth rates. Otoliths are very readable, but utility of increment width as an index of daily growth may be confounded by the highly structured relationship between the increment's number and its width; use of deviations from the pattern is considered as a possible index of deviations in a daily growth at age. Larval production was estimated by river plankton sampling, while local fishery yields and passive traps were used to estimate recruitment volumes; ratios of recruit-per-larva ratios indicate relative survival at different times. This system, because of its spatial confinement, pan-seasonal reproduction and short recruitment cycle, represents a unique opportunity for examination of the effects of seasonal environmental variables on recruitment in single species.

**Density, Temporal Spawning Patterns, and Growth of Young-of-Year Largemouth Bass in Vegetated and Non-vegetated Areas of Lake Guntersville, Alabama.**

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In Lake Guntersville Alabama (USA), a 28,000-hectare impoundment, we examined age-0 largemouth bass (Micropterus salmoides) density, temporal patterns of successful reproduction, and growth in relationship to the presence or absence of submersed aquatic plants. Densities of age-0 fish collected with cove rotenone and catch-depletion sampling averaged 387/hectare in vegetative stands of Eurasian milfoil (Myriophyllum spicatum) compared to 33/hectare in non-vegetated littoral regions in August 1991. Based on daily increments deposited on otoliths, 73% of the surviving fish displayed swim-up dates between 10 May and 8 June and successful spawning was continuous. A greater proportion of smaller, younger, and slower growing fish occurred in the vegetated areas. Age (days) was positively correlated ( $r = 0.77$  and  $0.83$ ,  $P < 0.01$ ) to length in both regions. For fish hatched in non-vegetated areas, daily growth remained constant among different age fish, but progressively decreased for younger fish inhabiting Eurasian milfoil. Back-calculated lengths indicated that differences in growth between regions occurred as early as 10 days old. Aquatic plants appeared to provide important nursery areas for young largemouth bass, but slower growth may increase size-dependent mortality. Thus, expected recruitment to the sport fishery may not be fully achieved.

## Ageing Coral Reef Fish of Australia - Preliminary Studies

A.J. FOWLER\*. Australian Institute of Marine Science, P.M.B. No. 3, Townsville M.C. 4810, Queensland, Australia.

The application of fish ageing in population studies is a new field of research for coral reef fish of the Great Barrier Reef of Australia. Here, I present results from some preliminary investigations into the assessment of the usefulness of otoliths for ageing such fish. Two criteria which otoliths must fulfill in order to be used for ageing are considered: otoliths must display an interpretable pattern of increments; and such increments must be formed on a regular and determinable time scale.

The criteria were considered for numerous species of coral reef fish from within the latitude range of 19 - 23°S. The structure of otoliths was determined from transverse sections, whilst validation was assessed by treatment of fish with tetracycline, and secondarily by marginal increment analysis.

Otoliths from 28 species representing 8 families have been examined. Most otoliths demonstrated an interpretable pattern of alternating opaque and translucent zones, whose clarity and interpretability varied on two scales:

- 1/. there was considerable variation amongst species, even amongst congeners;
- 2/. there was considerable variation amongst conspecifics collected from reefs in different latitudes.

Validation of the period of increment formation is now complete for a few species, indicating in each case that the increments represented a yearly pattern of growth. Furthermore, deposition of the opaque region of the otolith was initiated in Spring, whilst the translucent region was deposited from at least late Summer to late Winter.

These achievements are modest and preliminary. Wholesale progress in this field is still limited by the poor understanding of the relationship between the growth of otoliths of tropical fish, their structure and the factors that control growth be they exogenous or endogenous.

*Clear. vary w/ in species at dif places*



SYMPOSIUM ON FISH OTOLITH RESEARCH AND APPLICATION  
24-27 JANUARY 1993

COUNTING RINGS USING THE BIOSCAN<sup>®</sup> OPTIMAS<sup>™</sup> IMAGE ANALYSIS SYSTEM

by

AB THOMPSON and A BULIRANI  
UK/SADCC Lake Fisheries Project, Salima, Malawi

Rings seen on sections of otoliths can be counted using several different methods. This paper discusses the use of the BioScan<sup>®</sup> OPTIMAS<sup>™</sup> system to produce a magnified image of the otolith on a video monitor. The image can be enhanced by altering the optical arrangement of the microscope or by using various electronic filters operated through a computer. Rings can be counted directly off video monitor, or semi-automatically using the supplied software. The method of data acquisition, such as number of rings and ring increment widths, on to a computer for subsequent analysis will be presented. This work is demonstrated using sagittal otoliths from the pelagic cyprinid Engraulicypris sardella.

**Back-calculation of Size Frequency Distributions in Southern Flounder: A Test of Size-dependent Processes in Early Life History**

G. R. FITZHUGH\* and J. A. RICE. Department of Zoology, Box 7617, North Carolina State University, Raleigh, North Carolina 27695, USA.

Otolith-based back-calculation methods were evaluated for testing size-dependent dispersal/loss rate of juvenile southern flounder from estuarine creek habitats. Periodicity of otolith increment formation was validated by sampling calcein-tagged juvenile flounder from an outdoor tank over a 106-day interval. In a second 2-month rearing trial, juveniles were individually marked in order to estimate error in back-calculation. A size hierarchy developed with growth rates ranging from 0.08 to 1.4 mm/d, comparable to the variability observed from juveniles sampled across an estuarine depth gradient. Mean back-calculation errors for one- and two-month intervals were estimated to be 8% and 5% respectively. From ANOVA simulations, a shift of at least 10% between the mean of an observed length distribution and a back-calculated distribution (e.g. mimicking size-dependent removal of larger individuals) was detected with 0.9 statistical power, however power declined as back-calculation uncertainty was incorporated into simulations. The amount of shift in the mean can be related to the instantaneous loss rate as a function of size, to provide a quantitative measure of the magnitude of size-selection.

Age validation of the South African anchovy, *Engraulis capensis*,  
using daily growth increments.

M.E.WALDRON. Sea Fisheries Research Institute, Private Bag X2,  
Cape Town 8012, South Africa.

South African anchovy, *Engraulis capensis*, usually deposit several check rings per year which obscure annual zones. It was tested whether the macroscopically identified growth zones on the otoliths are laid down approximately one calendar year apart by counting the total number of daily increments on the otolith. More than 100 otoliths, exhibiting supposed annual growth zones, were mounted in dental resin, ground, polished with 0.05um alumina powder, etched in Hydrochloric acid and coated with gold palladium alloy. Otoliths were examined under a Cambridge S200 SEM, at variable magnification between 600 and 800X. The results justified the assumption that the growth zones were annular. Difficulties in interpreting some of the microscopic structures are discussed. It is concluded that growth and recruitment predictions, based on the assumption that the identified annual growth zones on the otoliths of the South African anchovy are annular, can be made with confidence.

AA  
Age and growth of the Ceratoscopelus cf. warmingii (Pisces: Myctophidae) population in the eastern Gulf of Mexico, based on daily growth increments in sagittal otoliths.

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Sagittal otoliths were examined from Ceratoscopelus cf. warmingii, collected in the eastern Gulf of Mexico during six sampling periods between September 1984 and May 1986 and ranging in size from 15 to 72 mm SL, which included the entire post-metamorphic size range of the eastern Gulf population. The size range of the species was subdivided into 5 mm SL groups and otoliths were selected randomly within groups with regards to size of individual, but allowing for equal representation of gender within each size group. The sagittal microincrements were arranged in three distinct zones, an innermost larval zone (LZ), surrounded by a broad, dark post-larval zone (PZ) accompanied by numerous accessory primordia, and an outermost post-metamorphic zone (PMZ). A size-related sexual dimorphism was present in the eastern Gulf Ceratoscopelus population, with males attaining a maximum length of 55 mm SL. Based on daily growth microincrements, which were validated using marginal increment analysis, the larval period (LZ) averaged 30 days. The smallest post-metamorphic individual (15 mm SL) possessed only LZ and PZ rings and was 45 days old. The largest male examined (55 mm SL) was 239 days of age, whereas the largest female (72 mm SL) was 457 days old.

dark border (zone) around nucleus reps a  
habitat  $\Delta$  surface to deep.  
Marginal inc anal to validate

Automatic ageing of North Sea plaice (*Pleuronectes platessa*) by means of image analysis.

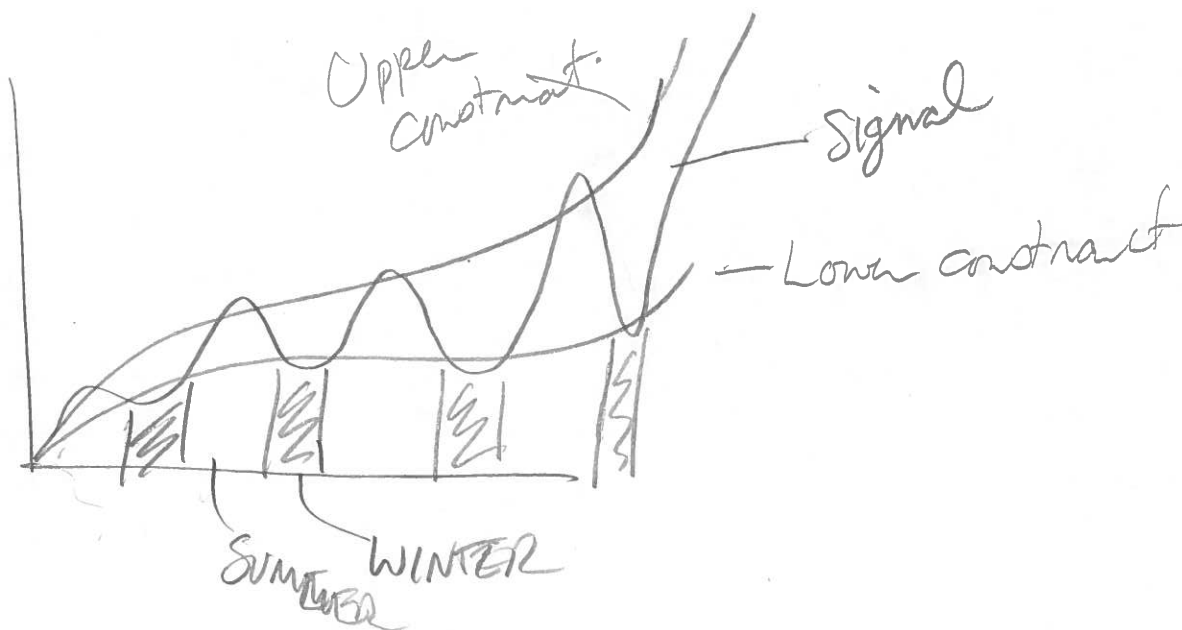
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**Abstract**

This paper describes a method to extract age information from growth patterns in otoliths by means of image analysis techniques. Emphasis is laid on a full automatic procedure without pre-processing of the otoliths. The automatic system has the advantage of eliminating subjective interpretations, such as a shift in age reading by human experts in time and differences between readers.

The method is partly based on standard image processing techniques and partly based on *a priori* knowledge of the growth pattern of plaice.

Special attention is paid to the validation of the results when applied to known-age plaice.



**Otolith Weight and Age: An Examination Using Weight Frequency Distributions for *Sardinops sagax neopilchardus***

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An analysis of the otolith weight frequency distributions for the pilchard, *Sardinops sagax neopilchardus* from the Albany region, south west Australia, was made over a two year period from September 1989 to September 1991. The modes in the September 1989 sample, which were known to correspond with separate year classes, persisted in subsequent samples with the position of the modes progressing in a manner consistent with them representing different year classes. In contrast, there was no consistent pattern showing modal progression when the length frequencies for these samples were examined. The average weight of an otolith for each of the year classes 2-8 were determined, from which the growth rates of males and females were calculated. Following sexual maturity, at age 2, males grow less than females, having an  $L_s$  of 162 compared with 172 for females. Analysis of the age structure of the catch for the fishery during 1989-91 indicated that there are variations in year class strength and these were consistent among years. Recruitment to the fishery begins at age 2 but is not complete until age 4. Finally, total mortality increased from 0.83 to 0.93 during the period of sampling, the possible reasons and implications for the future management of this fishery are discussed.

**Analysis of Otoliths and Vertebrae from Nine Tag-Recaptured Atlantic Bluefin Tuna (Thunnus thynnus)**

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Biological samples were collected from nine tag-recaptured Atlantic bluefin tuna (Thunnus thynnus) for which chronological ages (6 to 16 years old) could be closely approximated from tagging records. The 35th vertebrae and otoliths (sagittae) were collected from six of these fish, while otoliths or vertebrae were retained from the remaining three tuna. Analysis of the whole vertebral ageing method substantiates previous studies that the periodicity of growth zones on the cone surface, including the crowded bands near the outer margin, are deposited annually for tuna up to 13 years of age. Overall results from analyses of accuracy, precision, and structure preparation suggested that vertebrae are preferable to otoliths as the most suitable structure for use in routine ageing of Atlantic bluefin tuna. The average percent error values for both vertebrae and otoliths were less than 5%, indicating good precision compared to other methods of ageing. However, error rates from otolith analysis were higher than from vertebrae analysis. The microstructural record of otoliths was difficult to read and as a result there was also greater variability associated with estimated ages from both the long and short arms of sagittae cross sections compared to vertebrae.

~~The~~ 1<sup>st</sup> documentation of transatlantic travel of  
Bluefin tuna. (NW Atl. to Bay of Biscay)

Spines produce very good results. (Anal <sup>#4</sup> APE 0.0%  
6 readers)

Atl. Bluefin  
LEE

Acetate replicates prod. similar results  
— comment from audience.

## **Retrieving the Individual Growth Pattern from the Modulating Function**

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The radial growth of an otolith could be assimilated, from a signal theory point of view, to a periodic signal which frequency is modulated by a growth function. This proposition is compared to the recent biological knowledge about calcified structures morphogenesis mechanisms. The use of an *a priori* growth pattern might allowed, under favorable conditions (addequacy of the growth model, monophased growth), an age estimation and the deduction of a relative individual growth function, but the constraints linked to this approach lead us to search for methods that do not require any *a priori* growth pattern. The exact modulating function could be obtained by the computation of the instantaneous spectrum which represents the signal frequency versus time. The ability of this signal processing tool to estimate an individual growth pattern without any *a priori* knowledge is presented and illustrated on several larval and adult fishes otoliths.



### Growth Compensation in Fish - a General Phenomenon?

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It was hypothesized that growth compensation is common in fish and that otolith increments provide evidence for this phenomenon. Otolith measures originated from some 11,000 specimens comprising 3 migratory North Sea species: horse mackerel (*Trachurus trachurus*), anadromous smelt (*Osmerus eperlanus*), and katadromous flounder (*Platichthys flesus*). Annual increment widths were measured along postrostral axes at a 15x magnification. Otolith size - fish length relationships were quantified for all species. Consecutive radii were regressed on each other and on the preceding accumulated radii, using original values and relative measures compensated for differences in otolith size. In otoliths of the same size growth compensation was evident in all species and at all ages. Small otoliths exhibited particularly large annual increments (and vice versa) the following year. Regressions using relative measures of consecutive radii proved that compensation is pronounced between ages 2 and 1 but little or no correlation existed between increments at ages 2 and 3. R4 (radius 4) measures, however, increased with increasing R3 values, indicating that after age 3 growth differences tend to be carried along. It is concluded that growth compensation is a general phenomenon in fish and that it needs to be considered in population studies employing back-calculation methods and radius measures for stock separation.

Comparisons of growth and smolt migration between wild and domestic masu salmon

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In order to determine differences in growth and smolting between wild and domestic masu salmon (*Oncorhynchus masou*), eyed eggs from domestic salmon which have been reared for generations in the Hokkaido Fish Hatchery's freshwater pond since 1965, were marked with fluorescent substance, Alizarin complexsone (ALC) and buried in artificial redds in a Hokkaido stream. Wild and domestic juveniles after emergence were collected at three stations by electrofishing and cast nets during the growing season. In the following spring, smolts were collected at downstream site by cast nets. Otoliths were examined under ultraviolet light microscope to detect the domestic fish with fluorescent marks resulting from incorporation of ALC. There were no significant differences in body and otolith growth between wild and domestic masu salmon from April to August during the growing season. After this period, however, wild fish grew more rapidly than did domestic fish. Wild smolts was larger than domestic ones. Time of downstream migration in wild smolts was one month earlier than that in domestic ones. These results are interpreted as showing that even though domestic masu salmon swim up naturally from the stream bed as well as wild salmon, a growth performance of domestic masu salmon is lower than that of wild masu salmon in a natural stream.

**Spatial and Temporal Variation in Growth of Larval and Juvenile Gizzard Shad: An Assessment Using Otolith Microstructure**

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Gizzard shad are a prolific limnetic clupeid and an important prey species for piscivores. Large variation among years in gizzard shad size and abundance affects their availability to predators. To evaluate mechanisms influencing gizzard shad growth, five evenly spaced sites were sampled in an Illinois reservoir (4500 ha) over a four year period. Sites were distributed along a trophic gradient from eutrophic uplake regions to more mesotrophic areas downlake. Sampling gear bias prohibited the use of length frequencies to characterize larval and juvenile population growth rates; however, by aging otoliths, we were able to effectively determine growth and assess recruitment mechanisms. Growth rates were determined for a range of gizzard shad sizes by calculating slopes of fish length against age. Differences in rates of growth were observed among sites and years (ANOVA  $p < .05$ ). Fastest increases in size were observed at mesotrophic downlake sites and slowest increases in size were observed at eutrophic uplake sites. Biotic factors such as fish density and zooplankton availability were highly correlated to growth and may affect recruitment. Within a system, gizzard shad experience spatial and temporal variability in growth and future population assessments should account for this variation.

Age, Growth and Natural Mortality of Large Tropical Reef Fish in the Gulf of Carpentaria, northern Australia

D.A. MILTON\*, S.J.M. BLABER AND M.F. O'NEILL. CSIRO Division of Fisheries, Marine Laboratories, P.O. Box 120, Cleveland, Queensland 4163, Australia.

The difficulties in ageing large tropical marine fish are well known. Many species do not have reduced growth in the cooler months and no rings (annuli) form in their calcareous structures. We present the results of a study on the age and growth of several species of large reef fish (Families Lutjanidae, Lethrinidae and Haemulidae) from tropical northern Australia. We examined the ring formation in whole, sectioned otoliths, vertebrae and scales in an attempt to identify the age structure of each species. There was wide discrepancy between these structures in the number of rings, and the results were different from those of other studies of similar species. An independent estimate of the ages of three species was determined using  $^{210}\text{Pb}$ : $^{226}\text{Ra}$  disequilibria in whole otoliths. The results highlight the need to validate the rate of ring formation in calcareous structures of tropical fishes. All species grew slower than reported previously. This slow growth may be due to relatively low densities of the lower order consumers that form the natural prey of these species in the Gulf of Carpentaria. Natural mortality estimates, based on catch curve analyses, were also lower than previously reported and hence may influence fishery yields from these species.

An Evaluation of Computer-aided Age Determination of Otoliths from the Bank Rockfish, *Sebastes rufus* off California

G.M. CAILLIET<sup>1\*</sup>, L.W. BOTSFORD<sup>2</sup>, J. BRITTNACHER<sup>2</sup>, R.G. KOPE<sup>2</sup>, G. FORD<sup>3</sup>, D. WATTERS<sup>1,4</sup>, A. KING<sup>1,5</sup>, and M. MATSUBAYASHI<sup>3</sup>. <sup>1</sup>Moss Landing Marine Laboratories, P.O. Box 450, Moss Landing, CA 95039; <sup>2</sup>Gruppo Popolazioni, Dept. of Wildlife and Fisheries Biology, University of California, Davis, CA 95616; <sup>3</sup>Image Processing Laboratory, Dept. of Electrical and Computer Engineering, University of California, Davis, CA 95616; <sup>4</sup>Present Address: California Department of Fish and Game, 2201 Garden Road, Monterey, CA 93940; <sup>5</sup>Present Address: Office of Fisheries Conservation and Management, NMFS, 13356 East-West Highway, Silver Spring, MD 20910.

We have developed a computer-aided system to analyze periodic zones in fish otoliths (or other structures) for age estimation. The image analysis program first scans the image of an otolith thin section, perpendicular to the zones (user specifies orientation). Adjacent scans are averaged and filtered (using Fourier transformation or spatial domain convolution). Valleys are detected, based upon heights of adjacent peaks, and are marked and summed on the screen. We are evaluating this new technique using thin-sectioned otoliths from the bank rockfish (*Sebastes rufus*). The time and effort for cleaning, preparation, sectioning, and mounting are the same for both traditional and computer-aided techniques. The software was designed to reduce the time and tedium of counting zones, and it provides counts very quickly. This is an interactive system, allowing the user to make subjective decisions about ageing criteria. Both approaches are dependent upon subjective criteria and produce similar readings, but computer-aided estimates are more precise than traditional readings, and require less analysis time. We are continuing to evaluate this technique to determine if sample size and precision can be increased without jeopardizing accuracy.

**Differences in the Sagitta, Lapillus, and Asteriscus in Estimating Age and Growth of Juvenile Red Drum**

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Age and comparative growth rates of juvenile red drum, *Sciaenops ocellatus*, were determined from sagittae, lapilli, and asterisci, and compared in order to determine accuracy and precision of back-calculated size and age information. Otolith diameter was regressed on standard length for each otolith from 50 fish through a range of sizes between 10-50 mm SL. Sagittae provided the highest correlation ( $r^2 = 0.956$ ). Lapilli and asterisci were similar in their ability to predict standard length, but accounted for a lower proportion of the variability ( $r^2 = 0.880$  and  $0.899$  respectively).

Age estimates in days were determined from otolith increment counts from known age fish. The asteriscus provided the most accurate estimate of age. Both sagittae and lapilli underestimated the true age by 18 and 20 days, respectively. Errors in estimating age varied by  $\pm 3$  days on average. Underestimations for sagittae and lapilli were due primarily to low differentiation of rings in the nuclear region. We conclude that the asteriscus should be used for age estimates in juvenile red drum.

*Sagittae are dense & unclear / inner ring.*

*Lapillus cleaner*

*Good Slides DK blue fade to blk. (Yel & wt text & figs)*

COMPARISON OF SOMATIC AND OTOLITH GROWTH IN NORTH SEA HERRING (*CLUPEA HARENGUS* L.) LARVAE; EVALUATION OF GROWTH DYNAMICS IN MESOCOSMS.

by

Erlend Moksness (1), Kjetil Rukan (2), Lars Ystanes (2), Arild Folkvord (2) and Arne Johannessen (2)

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Abstract

Autumn spawned North Sea herring eggs were fertilised artificially, incubated in the laboratory and released into two separate mesocosms (A and B) in September 1991 as 1-day-old larvae (hatched within 24 hr). The duration of the experiments was 60 days. The released larvae experienced high temperature initially ( $\sim 18^{\circ}\text{C}$ ) in both mesocosms, decreasing to approximately  $10^{\circ}\text{C}$  at the end of the experiments. The temperature in mesocosm A was on average  $0.7^{\circ}\text{C}$  higher compared to mesocosm B. The prey density was higher initially in mesocosm B ( $> 1 \text{ l}^{-1}$ ) compared to mesocosm A ( $< 0.1 \text{ l}^{-1}$ ) while the situation was reversed around day 23 resulting in higher prey densities in mesocosm A ( $> 3 \text{ l}^{-1}$ ) compared to mesocosm B ( $< 0.1 \text{ l}^{-1}$ ). Differences in somatic and otolith growth rate of the herring larvae were observed, reflecting the pattern of prey densities in the two mesocosms. However, otolith growth rate was delayed with approximately a week compared to the somatic growth rate, indicating a delayed response to changes in body growth rate. The results show that starving and slow growing herring larvae have a relatively higher otolith growth rate than somatic growth rate once feeding has commenced. The effect of observed changes in otolith and body growth on the otolith size:body size ratio is presented and discussed.

**Back-calculated Growth in Larval King Mackerel and Gulf Menhaden,  
an Examination of the Influence of Size-Selective Mortality**

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We back calculated lengths at ages for wild king mackerel, *Scomberomorus cavalla*, and wild and lab-reared gulf menhaden, *Brevortia patronus*, using standard procedures, then constructed increment number (columns) by observed age (rows) half-rank matrices of mean back-calculated lengths at age. Columns in the matrices of both wild and lab-reared fish show trends of increasing back-calculated length at age for older larvae (but more so for wild), suggesting size-selective mortality on the smallest larvae of a given age. Predation is indicated as the main cause of size-selective mortality in the wild and starvation (and perhaps cannibalism) in the lab because wild larvae were exposed to predation and lab-reared larvae were not. Column slopes of the matrices indicate the time trend and intensity of size-selective mortality; in wild larvae it began at day 2, increased to day 15 then declined suggesting that the influence of predation was mainly expressed during this period. Starvation in lab-reared fish began at day 15. Size-selective mortality causes average growth (mean back-calculated or observed length at age) to appear higher because the smallest larvae of a given age are removed. We adjusted back-calculated growth by removing the effect of size-selective mortality with ANCOVA and estimated that the observed growth rate was 27% higher than the adjusted rate for king mackerel larvae.



### **Growth Rates of Two Sciaenid Fishes Distributed Across an Estuarine Tidal Front**

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Estuarine fronts may be areas of high productivity though these areas have not been as well studied as oceanic fronts. We designed a study to test whether larval sciaenids congregated (or were concentrated) near a tidal front within the estuary and whether their position on either side of the front affected their growth rate. Atlantic croaker larvae (3-6 mm) were consistently more abundant on the oceanic side of the front than on the estuarine side while densities of spotted seatrout larvae (2-5 mm) showed no particular pattern relative to the tidal front. Densities of both species were occasionally, but not consistently, elevated in the immediate area of the tidal front. There was no difference in the age of spotted seatrout larvae between the oceanic and estuarine side of the front while Atlantic croaker were generally older (and larger) on the estuarine side. There was no difference in growth rate of Atlantic croaker between environments but spotted seatrout larvae appeared to have higher growth rates in estuarine water.

### **Individual Growth History as an Indicator of Environmental Heterogeneity for Young Yellow Perch.**

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The size of young fish relative to both the size of their prey and of potential predators are important aspects of their ecology because they influence feeding success and risk of mortality due to predation. Since growth is closely related to important ecological relationships, growth history reflects past environmental and ecological conditions. Variation in individual growth history is an indication of the degree that individuals experience similar or dissimilar ecological conditions. Larval and juvenile yellow perch were collected from Red Lake, Minnesota. Daily otolith growth increments were measured for fish from several locations and individual growth histories determined. The effects of age and date on growth were separated statistically in order to establish temporal growth signatures for fish of different ages and different sizes. Temporal growth signatures were used to evaluate the degree that environmental conditions vary between locations for fish of different ages and sizes. Age and size distributions of fish vary between locations. Fish are segregated by size and larger fish tend to be found where abundance is locally low. This suggests that the population of young perch is not homogeneous, that they do not all experience the same environmental conditions, and that the effects are density dependent.

## **Temporal and Parental Spawning Origin of Newly Forming Atlantic Menhaden Year Classes in North Carolina Estuaries**

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The geographic and temporal spawning patterns of Atlantic menhaden (Brevoortia tyrannus) are complex: spawning occurs during most months of the year, the adult population annually migrates north and south along the U.S. Atlantic coast, and the population is differentially distributed by age and size latitudinally from late spring through early fall. Thus, progeny from different spatio-temporal spawnings are likely to encounter different environmental and spawning stock density conditions during critical phases of their early larval and juvenile life history. Lack of daily age and birthdate information for juvenile Atlantic menhaden greatly impeded earlier research attempting to estimate prerecruitment year-class size from estimates of juvenile abundance. Herein, we examine the reproductive condition of adult Atlantic menhaden caught by commercial purse seines; determine otolith-based birthdate distributions of larvae migrating into estuaries; and compare length-frequency and otolith birthdate distributions of estuarine resident juveniles. These data demonstrate the presence of two spatio-temporally separated age groups of spawners and as many as three seasonal cohorts of young of the year in North Carolina waters.

**Calculating Size-dependent Relative Risk of Mortality from Samples Taken Before and After Selection.**

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It is generally assumed that small fish are subject to size-selective mortality and that the magnitude and form of risk change over time driving variation in recruitment. I show the form and relative magnitude of the size-dependent risk function can be described from otolith or scale samples taken before and after selection using a semi-parametric spline. A series of samples may thereby identify episodes of selective mortality. This use of the spline is an extension of the model developed by Schluter (1988 Evolution) for longitudinal studies of natural selection, and could replace some use of quantile plots and electivity indices in fish ecology. I show the derivation of an appropriate transformation for cross-sectional studies and develop a bootstrap confidence band. In principle, the spline can be used to describe risk as a multivariate response surface, dependent on otolith size and growth rate for example.

**Anchovy Larvae Daily Mortality (*Engraulis ringens*, Jenyns) in the north of Chile during 1990.**

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The instantaneous rate of daily mortality in anchovy larvae was estimated from captures obtained during three cruises corresponding to INPESCON - Program 1990 (March, May and September) carried out aboard B/I Carlos Porter in the north of Chile.

The converted length frequency method was used to mortality estimation. It consists of transforming, by means of an inverse relation of a growth model, the class mark of each length interval into age in days. Valuing the age by means of the otoliths daily growth increments readings.

The mortality rates for each cruise were of 0.187, 0.162 and 0.143  $d^{-1}$  respectively, these values were found among the estimation rank carried out in marine fish larvae. The values estimated and the assumptions of the method are discussed.

IDENTIFICATION OF TWO STOCKS OF ANCHOVIES (*ENGRAULIS ENCRASICOLUS* L.1758) IN THE ADRIATIC SEA BY MEANS OF OTOLITH READING

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ABSTRACT

The problem of the existence of one or more stock units of anchovies in the Adriatic Sea has been debated among scientists in the region for a long time.

Catches of anchovies in the Adriatic average about 30,000 metric tons per year therefore the identification of two stock units can be very important on a fishery management perspective.

The use of otolith reading as a possible mean to sort out this problem is here presented.

Otoliths of anchovies were read and the outcoming age-length keys were statistically processed in order to discriminate different sets of data.

The evidence collected strongly suggests that more than one unit of population have been identified.

The core of the final discussion is on the possible correlation between the above evidence and the oceanographic structure of water masses in the same area.

### **Abnormal Lake Trout Otoliths: Applicability to Stock Identification**

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Lake trout (*Salvelinus namaycush*) from the Great Lakes exhibit a high percentage of abnormal sagittae. Although the presence of these structures is regarded as a response to stocking and has been used to separate hatchery-reared trout from wild lake trout, little has been done to identify the cause of these abnormalities or to establish to what extent they occur in wild fish. To characterize these abnormalities, we compared external morphology of sagittae from 876 hatchery-reared lake trout ranging in age from 5 months to 12 years with that of 303 wild lake trout from Lake Superior and lakes in the Northwest Territories. Half of the hatchery fry showed signs of abnormal sagittal growth after their first handling. After two or more years, sagittae were abnormal in 67% of fish from Lake Superior, 76% from Lake Huron, and 86% from Lake Ontario. For wild lake trout from Lake Superior, 38% were abnormal and for fish from the N.W.T. 51% were abnormal. Abnormal sagittae were composed of  $\text{CaCO}_3$  in the vaterite morph. The high percentage of abnormal sagittae in wild and hatchery lake trout made it difficult to determine the origin of individual trout and their occurrence may have less diagnostic usefulness than previously believed.

**Preliminary Analysis of Otolith Shape for Arctic Charr, Salveinus alpinus (L.), Stock Differentiation**

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To determine the relative contribution of each stock to a mixed-stock fishery, managers need to be able to identify fish in the catch according to their stock of origin. Differences in otolith shape may serve as stock identifiers. In this preliminary study, we compared shapes of Arctic charr, Salveinus alpinus (L.), sagittal otoliths between two geographically distinct stocks (one each from near the eastern and western extremities of the species range in Canada). Otolith shapes were described using the Fourier series. Age and year-class effects on otolith shapes were assessed. Preliminary results suggest that differences in otolith shape may be useful for differentiating geographically widely separated Arctic charr stocks.



**Use of otolith microstructure to discriminate among hatchery and wild incubated sockeye salmon (*Oncorhynchus nerka*) juveniles in Tustumena Lake, Alaska.**

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Analysis of otolith microstructure was used to test the feasibility of developing a model to discriminate among hatchery and wild-incubated sockeye salmon (*Oncorhynchus nerka*) fry in Tustumena Lake, Alaska. Samples of fry migrating from natal streams into the lake were collected over a 3-year period (1990 - 1992). Hatchery-incubated fry were sampled just prior to their release into the lake over the same time period. The sagittae were polished and an image analysis system was used to quantify banding patterns (band counts, widths, and distances between bands). Methods to standardize measurements (reference and transect lines) were developed and tested. Preliminary analysis indicated that the number of bands and the distance to the hatching check are potential variables for discrimination. Naturally occurring banding pattern differences may result in refined measures of early-life-history growth rates, survival, and distribution.

**Otolith Microstructure and Tidally Induced Resource Limitation in Juvenile Flatfish**

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We investigated the possibility that temporally limited food resources affect juvenile flatfish in shallow-water nursery grounds. The otoliths of newly settling plaice were examined to test the hypothesis that tidal and light cycles can cause disruptions in growth through disruptions in the daily feeding cycles. Juvenile plaice were sampled from a variety of nursery areas with different tidal cycles and prey distributions. Plaice diets and times of feeding were determined and related to the pattern of otolith checks. Feeding incidence in juvenile plaice *Pleuronectes platessa* was influenced by the tidal cycle, mean stomach fullness at low water was higher for spring tides than for neap tides. Restricted movement during neap tides may deprive plaice of access to feeding areas, and result in a regular pattern of otolith checks. Although the structure of juvenile flatfish otoliths is similar for many species, the mechanism of check formation may differ, especially between intertidal and subtidal species.

**Herring (Clupea harengus) Otoliths in Back Calculation of Size at Age AND Differences in Otolith Morphology in Different Stocks.**

H.E.B. HALLBECK, A. KONRADSSON, and P.E. GROTNES. Norw. Coll. of Fish. Science, Univ of Tromso, Norway.

Samples of otoliths of a reproductively isolated fjord stock of herring and of Atlanto-Scandic herring have been studied to devise a quantitative and objective method for determining stock characteristics. To facilitate and improve precision a simple and low-cost computer assisted visual otolith reader (CAVOR) has been used. The study also sought to find the best suited axis for back-calculation.

The results partly confirm earlier studies of otolith morphology, and determines which measurements discriminate best between stocks. The morphology changes with the size of the fish. Hence a normalising function has been devised. The function is also used for enhancing the contrasts of the otolith zone pattern, and for automatic measurement of zone width.

The axis from the centre towards the post-rostrum gives a direct proportionality between fish and otolith dimensions. Other axes may be used, but only by applying a more complex function. The analysis of different axes is also presented.

**The Importance of Estuarine Habitats of the Skagit River, Washington, to Juvenile Chinook Salmon, Oncorhynchus tshawytscha.**

K.A. LARSEN\* and R.R. REISENBICHLER. National Fisheries Research Center-Seattle, Bldg. 204, Naval Station, Seattle, WA 98115, USA.

Juvenile chinook salmon rear in estuaries to a greater extent than do most other species of Pacific salmon; however, the importance of estuary rearing for chinook salmon is unknown in most river systems. This study is to investigate the importance of the Skagit River estuary for production of chinook salmon and to examine the use by juvenile chinook of various types of habitat within the estuary. The objectives are (1) to estimate, using daily growth increments on otoliths, the number of days that adult chinook salmon returning to the Skagit River spent in the salt marsh and in the lower estuary as juveniles; and (2) to determine whether these estimates vary among the three seasonal runs. Work to date has consisted of collecting juvenile salmon throughout the spring and summer of 1991 from various habitats (Skagit River above the salt marsh, the salt marsh, intertidal cobblestone beaches of Skagit Bay and offshore waters of Skagit Bay and Saratoga Passage), and examining cross sections of the sagittal otoliths of the juveniles collected from these habitats with an image analysis system. At this symposium we will report on whether juvenile salmon from these various habitats display distinct growth patterns, and on the basis of these results, will discuss the feasibility and the degree of specificity for the remainder of the study.

**Migration and Growth History of Anadromous Brook Trout (Salvelinus fontinalis) Populations of Hudson Bay Determined by Otolith Analysis.**

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Sutton River brook trout (Salvelinus fontinalis) off the coast of Hudson Bay appear to be an anadromous population. Hydroelectric development is planned for many rivers in Northern Canada where interactions with anadromous populations of fish is inevitable. A total of 508 fish were collected in 1990 and 1991 from the Sutton River and its headwaters. A total of 177 fish were collected at the mouth of 5 rivers on the Quebec side of James Bay. Otoliths were used to determine age and growth of the brook trout and shape analysis was used for stock differentiation. The average age for Sutton River brook trout collected in 1990 and 1991 was  $3.86 \pm 1.50$  (n=186) and  $3.96 \pm 1.54$  (n=208) respectively. Whole otolith chemical analysis (Ca, Mg, Sr, Na, Zn, K, Mn) and S.E.M. scans of prepared sections of otoliths were conducted to allow the identification of anadromous individuals in these populations. These results will be used to assess the role of anadromy in the bioenergetics of these northern stocks of fish.

**Comparative Early Life History of Prionotus carolinus and P. evolans (Triglidae) in New Jersey, USA**

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Prionotus carolinus and P. evolans are sympatric in the Middle Atlantic Bight and abundant in trawl and plankton surveys. The early life history of these triglids is poorly understood because 1) reliable diagnostic characters have not been available for eggs and larvae, and 2) it is difficult to interpret juvenile length-frequency data because small individuals (< 50 mm TL) are present for long periods (April-December). We examined daily rings of sagittae to compare interspecific patterns of reproductive seasonality, spawning and settlement sites, age at settlement, and juvenile growth rates. Both species spawned from June to October. Spawning of P. carolinus was bimodal (i.e. peaks in June, Sept), based on backcalculated birthdates, but birthdates of P. evolans were distributed more continuously during this period. Both species settled from the plankton 2-3 weeks after hatching, but P. carolinus settled at a smaller size and grew slower between 10 and 70 days age. Juvenile Prionotus carolinus was more abundant in continental shelf habitat, while P. evolans was more abundant in the estuary. Thus, based on analyses of otoliths and supplemental distribution data, these two species differ in aspects of spawning periodicity, size at settlement, juvenile growth rate, and juvenile distribution.

Hatch Date Estimation of Juvenile Walleye Pollock Collected in the Eastern Bering Sea, in 1989 and 1990.

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It is known that there are separate spawning stocks of walleye pollock (Theragra chalcogramma) in the central and eastern Bering Sea. One is located in the Aleutian Basin (depth > 2,000m) and the others are located on the continental shelf (depth < 200m). These adult stocks have different biological characteristics, including spawning period, but linkages between basin and shelf components of pollock resources are not understood. Surveys in the Bering Sea, in the summer of 1989 and 1990, showed wide distribution of juvenile walleye pollock in the eastern continental shelf area, and limited distribution in the Aleutian Basin area. The collected juvenile fish were measured and weighed, and otoliths (sagittae) were dissected out. The otoliths were ground perpendicular to the otolith surface along the long axis of a plane from both sides, and daily growth increments were counted by light microscopy. Back calculated hatch dates ranged mainly between mid-April to early June, and the peak was observed in late April to mid-May. Taking account of the days required from spawning to hatching, spawning was estimated to have had a peak in mid-April. This spawning period was in accord with the spawning period in the southeastern continental shelf. Although it is known that spawning takes place from February to March in the Aleutian Basin, only a few fish were estimated to be spawned in this period. Geographical differences in survival and growth were suggested between juvenile fish spawned in the continental shelf and in the basin.

**A 100 Year History of Growth and Recruitment of Freshwater Drum (*Aplodinotus grunniens*)  
Constructed from Annular Otolith Increments**

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P.K. CUNNINGHAM. Wisconsin Department of Natural Resources.

D.J. CONNER. Red Lake Band of Chippewa Indians.

Freshwater drum in the Red Lakes, Minnesota are long lived and grow slowly. Recruitment appears periodic, with year classes detected in the commercial fishery approximately every seven years. We used contemporary samples from the commercial fishery and archived samples to develop a growth history extending from 1879 through 1989. We detrended allometric growth of sagittal otolith increments with an exponential model. We then aligned residuals from individual fish to time (years) to establish a growth history. We draw inference on stock dynamics through analysis of this growth history as well as through patterns in the parameters describing otolith growth for separate year classes. Finally, we use otolith growth history as a biochronology to infer past environmental conditions and establish correlation with climate.



**Length of Spawning Season and Cohort Production in Bluntnose Minnow, *Pimaphales notatus*, Central Ontario, using otolith microstruce**

P.M. POWLES<sup>1\*</sup> and R.A. CURRY<sup>2</sup>. <sup>1</sup> Department of Biology, Trent University, Peterborough, Ontario, Canada K9J 7B8; <sup>2</sup> Department of Zoology, University of Guelph, Guelph, Ontario, Canada N1G 2W1.

Ageing by means of otolith daily growth increments (DGIs) was employed to determine the number of cohorts and the length of the spawning season in bluntnose minnow (*Pimaphales notatus*), a fractional spawner. Ages (in days) were established for mature fish, and batch fecundity computed by the Conover model. The main hypothesis of the study was that fewer cohorts would be produced in northern lakes because of the shorter spawning season. The second hypothesis was that fecundity (by weight) would not differ between the northern and southern populations of bluntnose. Ages were validated by following two early length modes, back-calculating to the known spawning date, and by laboratory experiment. Subdaily rings were present in yoy fish, inside the presumed hatch date, and sometimes beyond. These resulted in overestimating ages of you fish by as much as 10%. Subdailies were not significantly numerous on otoliths of fish maintained in the laboratory. The spawning season in the northern study areas (Chandos Lake) was marginally shorter (early May – mid August) compared to the southern sites (Riche Lake, end April – mid August). Fish to the north were smaller, resulting in lower fecundity values. However, the batch fecundity values per female ranged from 96 – 412, with considerable overlap between northern and southern lakes. Between 8 – 14 cohorts were produced in the southern sites, and 6 – 12 to the north, supporting the main hypothesis. Our model may help to explain the limitation of northward distribution in fractional spawning cyprinids.

## **Growth Trajectories and Associated Sexual Maturation in Atlantic Cod as Inferred from Otolith Microstructure**

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We will assess (i) if onset of sexual maturation in Atlantic cod (*Gadus morhua*) is a function of growth rate during immaturity and (ii) whether there is a somatic cost associated with gametogenesis that is expressed as lower somatic growth in reproducing compared to non-reproducing members of a stock. To test these hypotheses we will estimate first, second and third year growth by backcalculation procedures using otolith cross sections of immature and mature cod of Georges Bank origin. The interannual growth of individuals will be documented from spacings between annual marks. Several sections will be made in the vicinity of an otolith's focus to examine for variability in measurements attributed to the difficulty in making cross sections in precisely the same place.

Spatio-temporal Patterns of Growth and Movement of Subyearling American Shad (*Alosa sapidissima*) in the Hudson River Estuary. Karin E. Limburg, Section of Ecology and Systematics, Cornell University, Ithaca, NY 14850.

Abstract: Young-of-year American shad were collected fortnightly from late May through early November 1990 at sites spanning most of the length of the tidal Hudson River. Length and weight measurements were taken on each fish and sagittal otoliths extracted. Otolith increments were counted and measured to provide fine-scale information about the growth histories of individual fish. This demographic information, in conjunction with environmental and ecological data, is used to test models of out-migration from the natal river.

## **Mortality Estimation of Larval Walleye Pollock in Shelikof Strait, Western Gulf of Alaska**

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Early developmental stages of walleye pollock spawned in Shelikof Strait, Alaska are a model population for transport and mortality studies. Pollock spawn in a predictable and geographically confined area within a restricted time period, about 2 weeks in early April. Eggs and larvae drift in the Alaska Coastal Current as a relatively coherent aggregation. Daily cohorts of larvae are identified from otolith increment analysis and are tracked during several sequential samplings of the larval aggregation. Our methods for estimating mortality have been undergoing refinement over the past 5 years. We will discuss our techniques to study mortality of daily cohorts of pollock larvae over coarse (100 km, 30 d) and fine mesoscales (25 km, 3 d).

**Recruitment Processes of the Strong 1991 Year-Class of Norwegian Spring Spawning Herring (*clupea harengus* L.) Derived from Otolith Monostructure Examination.**

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The spawning-stock of Norwegian spring-spawning herring have increased since 1988 when the strong 1983-year-class recruited to the spawning-stock. Since then large numbers of yolksac larvae, with peak values of more than 5000 m<sup>-2</sup>, have observed over the spawning beds. During March-April 1991 an intensive sampling program for microzooplankton and yolksac herring larvae was carried out in the hatching area. Additional investigations were carried out on the Norwegian shelf in May and June the same year for older herring larvae. The otolith microstructures was studied in these larvae. Their hatching date and daily growth rate were back-calculated. The hatching date distribution of the surviving larvae was compared with the observed hatching over the spawning beds. The present results indicate a good match between the abundance of first feeding herring larvae and their prey, resulting in the largest year-class since 1983. The observed and back-calculated hatching date distribution in 1991 peaked two weeks earlier than the previous years, 1985, 1989, and 1990. A significant lower daily growth rate was estimated for the 1991-year-class, compared to the previous years, indicating a density dependant effect on growth rate of the early larvae.

**Advances in Larval Fish Ecology: New Concepts Resulting from Application of Otolith Analyses**

A.J. GEFFEN. Port Erin Marine Laboratory, University of Liverpool, Port Erin, Isle of Man, UK

The number of publications on the use of primary increments for larval fish studies has increased rapidly in the past two decades. The number of institutions which regularly collect data on otolith increments has also increased. Some applications of otolith analysis have made substantial contributions to our understanding of larval fish ecology, answering questions that were difficult to approach without this technique. Several examples will be reviewed, highlighting the improved information available and how changes are integrated into traditional views of larval fish ecology. In other cases otolith analysis may not be the right tool for the job, since the level of resolution obtained is no better than other methods.

## Episodic Recruitment of a Long-Lived Fish as Revealed Through Otoliths

C.M. JONES\*. Applied Marine Research Laboratory, Old Dominion University, Old Dominion University, Norfolk, Virginia 23529-0456.

Black drum, *Pogonias cromis*, has a life span of close to 60 years. Indeed, we still see members of the 1933 class. The average age in the catch in Virginia is 27 years and many older fish are routinely landed. Ages were validated with Pb/Ra dating because more traditional methods could not be employed. The long life span as shown by otoliths and other population parameters reveal that this species historically depends on the occasional large year class to maintain stock size and is vulnerable to recruitment overfishing. However, the age distribution of the catch reveal that mortality has been low and the stocks are not presently overexploited. The information available from otoliths also reveal a pattern of expatriate spawning and extensive migration. The long life reveals information about the population biology of this species that would not be discernable in a species with a typically shorter life span.

Target of Biologists

Sagittae, Otoliths, & Rectal F.R. — for verification  
    \ clear    \ Difficult

100% argut

Other 2 underest. micr # in otoliths

Counts & Pb/Ra correlate

Juveniles leave Bay

Matures at ~5 yrs.

1931 & 1942 year classes still represented!

$L_{\infty} = 1186$

Very asymptotic growth  
past 1100 mm  
Resilient population—

**Use of Otoliths to Determine the Relationship between Egg Production and Striped Bass Recruitment in the Santee-Cooper System, South Carolina, 1986-1990.**

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The relationship between the timing and magnitude of egg production and resulting recruitment of striped bass is not well understood. This study's objective was to correlate spawning date of recruited juveniles with egg production events. Eggs were collected a minimum of two times per day in spawning tributaries. Juveniles were collected from nursery areas and their otolith were removed for hatch date determination. Hatch date was determined by a minimum of two independent counts of daily rings. The precision of daily counts varied among readers and suggested that known age specimens be used to certify the accuracy of obtained counts in future efforts. Obtained results indicated that recruitment is not directly correlated with egg production but results from relatively high survivor of eggs and larvae during windows of time. During the study years, these windows were generally associated with the latter half of the egg production cycle. Since the timing and transport of eggs is tied to hydroelectric production schedules, the data suggests an opportunity to manage water releases to maximize survival potential.



### Recruitment of Coral-reef Fish Larvae to Bermuda: Local Retention or Long-distance Transport?

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The planktonic larvae of coral reef fishes are capable of prolonged dispersal, but those that settle to the reef may mostly be those that remained in the open waters near the island at which they were spawned. The island of Bermuda is distant from any other putative source for recruits and would require an uncharacteristically long period of planktonic dispersal. We inferred the planktonic larval duration (PLD) from otoliths of reef wrasses (Labridae) captured in Bermuda to determine whether their larvae are retained locally or transported from elsewhere. One species (*Thalassoma bifasciatum*) displayed a high proportion of individuals that delayed metamorphosis, in addition to long PLDs (mean = 50 days) that are characteristic of the species. The other wrasses showed short PLDs (around 3 weeks) and no evidence of delayed metamorphosis. We conclude that most wrasse recruits to Bermuda were spawned locally.

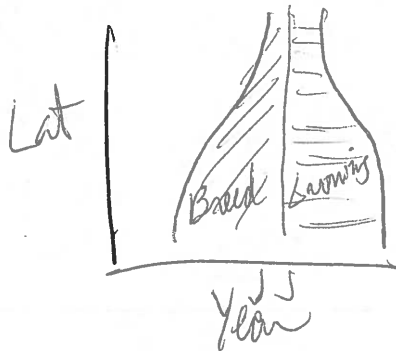
Could core ring transport. Trans time Cape Hatteras (~80-360d)  
(model? Ring translation)  
Entrainment Model - ~40d

2-3 yrs bet. events to Bermuda  
Larval duration det. by settlement marks in Otoliths  
Entrainment model allows transport by to Bermuda  
to allow recruitment after for wrasses

**Latitudinal Variation in Hatch Dates, Growth Rates, and Body Sizes of Male and Female Silversides (*Menidia menidia*): Evidence From Otoliths**

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Prior laboratory studies have demonstrated that local populations of the Atlantic silverside vary genetically with latitude in two major ways. First, temperature during larval development influences primary sex differentiation but the level of thermal control is greater in low than in high latitude populations. Second, the innate capacity for growth increases with latitude. We used otolith ageing techniques to explore the effect of these genetic differences on young-of-the-year fish in the field. In South Carolina fish where temperature has a strong influence on sex determination, hatch dates of females averaged about 10 days earlier than did those of males. The sexes grew at similar rates but females were larger by virtue of earlier birth. In Nova Scotia fish where temperature has no influence on sex differentiation, the mean hatch dates, growth rates, and body sizes of the sexes did not differ. New York fish display an intermediate pattern. Overall growth rates increased with latitude (0.5 mm/d in SC, 0.75 mm/d in NY; 1.0 mm/d in NS). These results verify that genetic variation plays a major role in determining growth rates and body sizes of fish from different latitudes.



highest lat = greatest Var in size!  
= equal sexes.  
lower lat = females larger.

**Size Selective Mortality of O-group Cod (*Gadus morhua*) During Transition from the Pelagic to Benthic Life as Demonstrated by the Change in Otolith Centre Size Distribution.**

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Samples of otoliths from one cohort in the cod stock in the cod stock of Balsfjord (North Norway) have been taken monthly during late summer and during the subsequent years, annual samples of otoliths from the same cohort have been studied. The opaque centre of the otolith, representing the pelagic summer growth until the juveniles seek the benthic environment, exhibit a marked Rosa Lee phenomenon.

To measure the otolith centre as precisely as this study required a simple and low-cost computer assisted visual otolith reader (CAVOR) has been developed.

The results indicate that the autumn is a period of exceedingly high size selective mortality for the O-group, perhaps being more decisive for the recruitment of cod stocks than formerly thought. The selective mortality also seen to act as a pruning on the O-group size distribution, transferring environmental effects on variation in summer growth to a change in number instead.

The cause of this large mortality is discussed, and cannibalism within the cohort is proposed as the culprit.

A Simple Method for the Assignment of Thermally Induced Otolith Banding Patterns for Mass Marking Using the Interleaved Two of Five Bar Code Symbolology.

Eric C. Volk, Steven L. Schroder, and Jeffrey J. Grimm

Washington State Department of Fisheries  
Olympia, Wa.

and

H. Sprague Ackley

Intermec Corporation  
Everett, Wa.

ABSTRACT

Over the past five years, the Washington Department of Fisheries has mass-marked some 35 million juvenile salmonids at several of its hatcheries by inducing specific and unique banding patterns to their otoliths with short-term incubation water temperature changes. We have successfully marked all five species of pacific salmon and several broods of adult returns have demonstrated the persistence and recoverability of these marks for at least five years. Otolith patterns may be created by this method in a number of different ways, however, as the number of treatment groups, brood years and facilities to be marked grows, some organized system of pattern assignment is desirable for convenience and accuracy. Using systematic rules developed for the assignment of patterns in the bar code industry, we have created a method for the assignment of up to 1000 different patterns in the otoliths of incubating salmonids. Patterns are based upon the interleaved two-of-five bar code symbology rule, requiring six thermal events to produce the six band pattern. Pattern recognition is based upon relative spacing of adjacent bars.

Repeat blind tests among readers of broadly different experience levels showed that pattern recognition was virtually error free and that only the time required to perform the decode was different among readers. We also wrote a decode algorithm to be used in conjunction with an automated image analysis system to decipher the patterns. This process was also very accurate in correctly decoding patterns however, it took nearly three times as long to perform as the visual method. Since the symbology is based upon relative spacing of bars, the pattern can be deciphered along any clear portion of the otolith and is not dependent upon a consistent measurement axis. As a result, relatively gross, rapid otolith preparation procedures are adequate for code detection.

A method to determine the onset of sexual maturity from back-calculated growth curves from otoliths of individual female North Sea plaice,  
*Pleuronectes platessa* L.

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1970AB IJmuiden  
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### Abstract

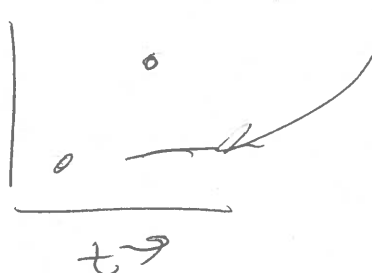
This paper explores two simple methods to determine the age at first maturity from the discontinuity in the pattern of otolith growth of female plaice. One method fits a discontinuous linear regression, whereas the other fits a surplus production allocation model through back-calculated growth data. Both methods are applied to estimate the age of first maturity of sixty-one females of year class 1963 and 1969 sampled at age VIII and X. The results are compared with direct observation. Accuracy and precision of the method, and the sensitivity for random variation in surplus production, are explored using a simulated growth data. It is concluded that a growth discontinuity can be located that corresponds to the onset of sexual maturity, but further work is necessary to study the possible bias in the method.

**Sources of variation in size at settlement in coral reef fishes inferred from otolith microstructure.**

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We investigated the effects of age and growth to explain sources of variation in size at settlement in three species of coral reef fishes with different mean planktonic larval durations; the Hawaiian dascyllus *Dascyllus albisella* (25 d), yellow-tailed surgeon *Prionurus laticlavus* (40 d), and bluehead wrasse, *Thalassoma bifasciatum* (46 d). Correlations between size at settlement and planktonic larval duration (age) were significant for the Hawaiian dascyllus and the bluehead wrasse but not for the yellow-tailed surgeon. In all cases, no more than 50% of the variance in size at settlement was explained by age alone. Analysis of settlement size versus otolith length (growth) showed a positive and significant correlation in all species yet only accounted for 25 - 50% of the variance. The period where growth varied during the larval stage was determined by correlating the residuals of the regression line for size at settlement and planktonic larval duration (to account for age differences) with mean daily otolith increment width at each day during the planktonic period. Results show significant positive correlations (i.e. larger settlers growing faster than smaller settlers) occurring during the early, middle and late larval stage in the Hawaiian dascyllus, yellow-tailed surgeon, and bluehead wrasse, respectively. We conclude that variation in size at settlement is largely due to individual differences in growth rates, and its magnitude dependent upon when these differences occur during the larval phase.

Wrasse (*Thalassoma*) longer settlement period than  
Prion & Dascy  
Grow faster in plankton?



Interannual variation in growth rates and back-calculated birthdate distributions of Pacific hake juveniles.

"Whiting"

David Woodbury<sup>1</sup>, Anne Babcock Hollowed<sup>2</sup> and Julie Pearce<sup>2</sup>

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The coastal stock of the Pacific hake (also known as Pacific whiting) (Merluccius productus) exhibits large variations in year-class strength. The coefficient of variation in recruitment of Pacific hake is 1.29, and adjacent year classes may vary tenfold to thirtyfold in abundance. The early life history stages of Pacific hake are spent in waters off the coast of California, where the oceanic environment is dynamic. The strong signals in both recruitment variability of Pacific hake and oceanic conditions make hake an ideal species to study stock response to interannual variations in ocean conditions.

Analysis of growth rates and back calculated birthdate distributions of young-of-the-year Pacific hake juveniles were estimated from samples collected during the surveys conducted in late May and early June in 1987 and 1991. These estimates differed significantly between years. Mean length of juveniles collected in 1987 was 83 mm compared to 32 mm in 1991. Growth rates of larvae captured in the field from cruises off the California coast in 1977, 1978 and 1979 are compared with those derived from the 1987 and 1991 samples. Specifically, growth rates observed during years that produced strong year classes (1977 and 1987) are contrasted with growth rates during years that produced poor year classes. Estimates of larval mortality rates are derived and applied to a life history table simulation to examine hypotheses concerning factors underlying production of strong or weak year classes.

Largest prod. on West Coast.

Spawn off shore Pt. Conception to off Baja

Migrate North to near shore sites (Monteary to Vancouver)  
juv. → Large Adults

**Increasing the Precision of Otolith Age Determination of Tropical Fish by Differentiating Biannual Recruitment--A System Developed and Validated on Tilapia, *Oreochromis niloticus*, from Lake Awassa, Ethiopia**

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Over a 12-month period 62,781 tilapia were sampled from Lake Awassa, Ethiopia. An analysis was conducted on the translucent macrozonation and microzonation of a subsample of 578 otoliths of immature fish. Age assessment based on the interpretation of seasonally formed translucent macrozones was verified by independent daily age interpretations. Microzonal estimates of daily age, validated by tetracycline marking techniques, were extrapolated from date of capture to determine time of hatch, confirming two primary breeding seasons peaking during March and May (80% total annual production), with secondary activity during September and October (20%). Two translucent macrozones associated with *biannuli* (a type of annulus formed twice a year) were formed each year during January and February and during July and August. We describe a model to assign otolith age more precisely by discriminating between the two recruitment cohorts by using number and location of biannuli, conditions on the edge, and time of capture. The method was validated with microzonal analysis and showed excellent agreement ( $r = 0.95$ ) when correlated with actual date of hatch. The procedure is especially applicable to tropical fish but can be applied to any species that show biannual reproduction and otolith growth cycles.

*Biannual recruitment*  
*Validated daily w/ OTC*



**Determining Stock Composition of King Mackerel in a Mixed Stock Fishery Using Discriminant Analysis of Otolith Growth Data**

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Based on results of a tagging study conducted during 1975-78, fishery managers have assumed that king mackerel, *Scomberomorus cavalla*, caught off most of the east coast of Florida during November-March belong to the Gulf of Mexico stock, while those caught during April-October are Atlantic Ocean stock. Electrophoresis of nuclear genes has been used successfully to separate east and west Gulf stocks, but was unable to distinguish Atlantic from east Gulf fish. However, age and growth data indicates significant differences in growth between females from the Atlantic and east Gulf, especially above age 3. The objective of this study was to investigate the feasibility of using discriminant analysis of otolith annulus measurements and growth data to classify fish to Atlantic (North Carolina to Jacksonville, Florida) and east Gulf (Louisiana to northwest Florida) stocks, enabling us to calculate mixing rates of the two stocks in the east Florida winter fishery. Annulus measurements were made on sectioned otoliths from 100 (50 from each region) females >90 cm FL collected during 1986-89 using an image analysis system. Measurements were made from the second to the fourth, fifth, and sixth annulus. Using the three measurements and a fourth variable which was simply FL/age, we produced a quadratic discriminant function which correctly classified 72% of Atlantic females and 68% of east Gulf females.

New England → Gulf → Rio de Janeiro

Separate Stocks -

W Gulf, E Gulf, Atlantic, Mixing Area (Winter)

All E Gulf fish not diff by electrophoresis

# **Statistical Analysis of Reconstructed Life Histories from Otoliths: Special Properties of Longitudinal Data**

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Otoliths have great utility in providing information about a fish's history. This very utility, however, carries with it statistical properties that could complicate analyses if the investigator plans on obtaining estimates beyond ages of fish or other characteristics derived from single observations. Analyses that require multiple measures, such as reconstructing and comparing growth histories, do not conform to assumptions of commonly used linear statistical models. In this paper we present the basis of the problem of repeated measures on individuals and, using a model otolith data set, demonstrate the consequences of violating the assumptions of independence of observations on otoliths and random sampling of fish. Lastly, we provide a decision tree of methods for appropriate analysis of otolith-based data.

THE USE OF OTOLITH DAILY INCREMENTS AND THE LEFTKOVITCH MATRIX TO  
EXAMINE THE NATURAL VARIABILITY OF ANCHOVY AND SARDINE  
POPULATIONS

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ABSTRACT

Growth of larval and juvenile northern anchovy (*Engraulis mordax*) and Pacific sardine (*Sardinops sagax*) were studied using daily increments in otoliths. Increment widths and back-calculated growth rates of northern anchovy differed during the El Nino of 1983 from previous years. Growth rates of Pacific sardine were greater than those of northern anchovy during El Nino. The effect of variable growth rates on the population dynamics of two species was examined using a Lefkovitch matrix. The Lefkovitch matrix uses the duration, mortality and fecundity of life stages to determine population growth rate. Natural variation in the duration and mortality rate of the early larval stage have the greatest impact on the northern anchovy population. Natural variation in the vital rates of the late larval stage have the greatest impact on the Pacific sardine population. Lefkovitch matrices in combination with daily otolith growth studies provide a powerful tool for the investigation of environmental change and recruitment variability.

## Young-of-Year Striped Bass Survivorship in Albemarle Sound, North Carolina, Based on Otolith Daily Rings

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The objective of this study was to examine the relationship between the estimated egg production of striped bass, *Morone saxatilis*, in the Roanoke River and recruitment to the year class forming in Albemarle Sound as functions of seasonal patterns in river flow and age structure of the spawning stock. Daily rings formed on sagittal otoliths of juvenile bass were used to calculate individual spawning dates, which were then compared to daily egg production downstream of the spawning grounds, river flow conditions, and the age distribution of female striped bass sampled near the spawning grounds in 1990 and 1991. Previous interpretations of annual egg production, the Juvenile Abundance Index (JAI), and the estimated spawning population assumed a uniform mortality rate; no correlations among the three data sets are evident. Estimated birth dates of surviving juveniles recruited to the year class indicate a dramatic non-uniform mortality rate of eggs and larvae during the spawning season; over 50% of all juveniles collected in the annual JAI survey were from less than 5% of all eggs produced, primarily at the end of the spawning season. Also, the spawning window is longer than previously thought: March into July, rather than late April to early June.

## OTOLITHS IN SALMONID RECRUITMENT STUDIES: AN EXAMPLE TESTING THE EFFECT OF RELATIVE EMERGENCE TIME ON SURVIVAL AMONG FRY GROUPS

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The effect of relative emergence time on recruitment success among two groups of migratory brown trout (*Salmo trutta*) fry was investigated. We tested the hypothesis that fry which emerge first in a given swim-up area will dominate numerically and attain larger sizes than fry emerging later within that area. An experiment was conducted where 160 early and 50 late emergers (progeny of different females) were planted in the gravel as alevins at each of five swim-up sites in a small stream. To distinguish between the groups following capture at a later date, sagittal otolith microstructure formation in the early emerger group was manipulated by varying water temperature to imprint coded sequences of identifiable check marks. The difference in mean emergence times between the two groups was ~4 d. Late emergers were significantly larger (in weight) at emergence than early emergers, presumably a parental effect. Mark legibility in otoliths was excellent, and contrary to our predictions, after 54 d early emerging fry were recruited in proportions less than or equal to an expected value assuming proportionate survival, and their size in weight was on average significantly lower than that of late emergers. Otolith microstructures were analyzed both within and among groups in relation to these results.

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#### **Location and Time of Spawning of White Bass (*Morone chrysops*) in Lake Erie**

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White bass have two principal spawning groups in Lake Erie: a riverine group and an open-lake reef group. The riverine group is fished heavily in the major tributaries each spawning season and appears to be vulnerable to exploitation at that time. Our objectives were to determine the relative contribution of the two principal spawning groups to the Lake population of age-0 white bass. We accomplished this by examining the age, and hence approximate spawning dates, of age-0 white bass collected from the Lake population. We also looked for indications of growth advantages that might be associated with observed age differences. Age-0 white bass were collected from the western basin of Lake Erie in 1984. Their length and weight, and daily age from otoliths were determined. The otoliths were found to have daily growth increments; however, the formation of accessory primordia limited aging to the first 90 days. Analysis of age-frequency distributions revealed two spawning cohorts--one associated with riverine spawning and the other the open lake. The lake-spawned cohort accounted for 70% of the age-0 white bass in 1984. Average daily growth rates between the cohorts were not significantly different.

**Variation in hatch date distribution of *Ammodytes marinus* in Shetland waters, and its influence on early growth and survivorship**

**P.J. Wright** SOAFD Marine Laboratory, Victoria Road, Aberdeen, AB9 8DB, Scotland, UK.

This study examined how growth opportunity was influenced by hatchdate in the lesser sandeel (sandlance) *Ammodytes marinus*. Daily increment deposition was validated for this species by rearing and sequential sacrifice experiments. Changes in sagitta microstructure were also found to provide a chronological record of the time of hatching and larval metamorphosis. Sagitta microstructure was used to compare the hatch date distribution and early growth of juvenile sandeels sampled from Shetland waters in years of relatively low (1990 & 1992) and high (1991) recruitment. This comparison revealed marked differences between the hatching period and growth rate of sandeels that survived beyond larval metamorphosis in the three years. Based on known and back-calculated hatch date distributions, the peak hatching period was late February in 1990 and 1992 and late March in 1991. Nevertheless, the 1991 year class grew significantly faster, metamorphosed earlier, and attained a slightly larger size by late June than either the 1990 or 1992 year-classes. The significance of this difference in hatching time and subsequent growth is discussed in relation to the timing of larval prey production. Further, the relationship between hatching time, as assessed from larval occurrence, and hydrographic conditions are considered using information derived from historical data sets for the Shetland region.

**Recruitment Determination in Atlantic Cod: Preliminary Results  
Based on the Hatchdate Frequency Distribution of Juveniles**

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Monthly ichthyoplankton surveys were conducted over Emerald and Sable Island Banks, Scotian Shelf, from March 1991 to April 1992. Atlantic cod (*Gadus morhua*) eggs, larvae and juveniles were sampled with rectangular midwater trawls, Tucker trawls, and bottom trawls. Early stages of cod were captured between October 1991 and April 1992. A nucleus of the population was retained in a well delineated 50 x 50 km area of Sable Island Bank up to the demersal juvenile stage. Based on date of capture and otolith age, hatchdates were assigned to juveniles in the range 20-25 mm standard length. The hatchdate frequency distribution of the juveniles will be compared to egg production at date to determine if survival windows leading to more intense recruitment existed during the spawning season of cod.



# Recruitment Forecast of the Pacific Saury based upon Otoliths Information

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<sup>1</sup>National Research Institute of Fisheries Science, 5-5-1, Kachidoki, Chuo-ku, Tokyo 104, JAPAN; <sup>2</sup>Tohoku National Fisheries Research Institute, 3-27-5, Shinhamma-cho, Shiogama-city, Miyagi 985, JAPAN.

Daily ring analyses of otoliths revealed that the Pacific saury, Cololabis saira, became adult size in one year. Among commercial size categories of saury caught in the fishing season (late August - early December), age of large fish (29 - 32 cm in knob length) is estimated to be around a full year, medium fish (24 - 29 cm KnL) is about nine months and a small fish (20 - 24 cm KnL) six months, which correspond to autumn, winter and spring spawned groups. The stock level and size composition of saury are determined by recruitment of three season groups. Recruitment of saury can be calculated from three parameters, larval production at hatching, growth rate and mortality rate during larval and early juvenile periods. These parameters are estimated from quantitative larval survey and otolith analyses. As 50 mm juveniles start to show schooling behavior, we use daily production of 50 mm juveniles as an index of recruitment, which is calculated from a mortality formula. We could predict trends of stock level and size composition of saury in the last two years using seasonal recruitment indices. We found that the recruitment estimates based upon otolith information gave us a good method in assessing the stock level and the size composition of short-lived species.

### **Elemental Composition of Pacific Halibut Otoliths**

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Trace elemental analysis of otoliths was evaluated as a method to identify Pacific halibut to their nursery area origins. Otoliths were collected from age zero and one halibut in three widely separated near-shore areas in Alaska. In addition, comparisons were made with otoliths collected from one of the areas 20 years earlier. Using a wavelength dispersive X-ray microprobe, exploratory work indicated only Sr, Na, K, S, and Ca, were in quantities above the detection level of the equipment (ppt). The distribution of Na, Sr, and K within select otoliths, showed correspondence to structural features that reflect life history changes. Nested analysis of variance of Na and Sr/Ca ratios indicated significant between area and between individual variation, though no significant within individual variation. The percent variation explained by area and individual however was small in relationship to within otolith variation. Halibut from the same area collected 20 years apart appeared to have significant differences in elemental concentrations. Additional investigation is warranted by the indication of significant area effects even with small sample sizes. The application for stock separation of halibut however, is restricted by the detection level of the equipment, and by the indication that for the elements observed the method may not be free from interannual variation.

**Radiometric Age Verification for Two Deep-Sea Rockfish (*Sebastolobus altivelis* and *Sebastolobus alascanus*)**

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Increment patterns visible in transverse otolith sections from two congeneric species of deep-sea rockfish (*Sebastolobus altivelis* and *S. alascanus*) are irregular and difficult to interpret. Different section reading techniques result in longevity estimates which differ by as much as a factor of two. Radiochemical assay was used to independently estimate longevity and verify an appropriate reading technique for these two species. Determination of the  $^{210}\text{Pb}/^{226}\text{Ra}$  disequilibria in otolith cores revealed ages of at least 30 years for *S. altivelis* but were inconclusive for *S. alascanus*. Section increment counts using reflected lighting at 50X magnification most closely agreed with radiochemically assayed ages for *S. altivelis*.

**Evaluation of Calcein and Oxytetracycline as Chemical Markers of Summer Flounder**

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Although several researchers have aged summer flounder (*Paralichthys dentatus*), the method of ageing for this economically important species has not been validated using chemical markers. Summer flounder were injected intramuscularly with calcein or oxytetracycline at dosages of 25 mg/kg or 50 mg/kg of body weight. Fish were held for 36 to 194 days following injection. After 18 months in storage, sagittal otoliths and dorsal fin rays were examined for fluorescent bands. No bands were seen on dorsal fin rays. Both fluorophors produced readable bands on summer flounder otoliths when injected intramuscularly at 50 mg/kg and at 25 mg/kg. No dosage effects were observed. However, calcein bands were much more intense than those of oxytetracycline. After injection with oxytetracycline, summer flounder stopped feeding and their activity decreased. No deleterious effects were observed in fish injected with calcein.

## **Can Otolith Strontium Chart Migration Histories in Anadromous Striped Bass Populations?**

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Electron micro-probe analysis of otoliths of several large Chesapeake Bay striped bass showed annual patterns in strontium level which was consistent with exposures to varying salinities due to upriver spawning migrations. Accurate prediction of ambient salinity based on otolith microchemistry could be used to retrace individual migration histories across estuarine and marine habitats. The effects of salinity and temperature on otolith microchemistry were investigated in rearing experiments on juvenile striped bass. Juveniles were exposed to six salinity treatments (0, 5, 10, 15, 20, 30 ppt) nested within two temperature treatments (15 and 25°C) for a period of three weeks. Analysis of covariance was used to determine whether temperature affected the relationship between ambient salinity and otolith strontium. A second experiment was designed to investigate the spatial resolution within the otolith's microstructure to detect exposure to differing levels of salinity. Juveniles were exposed to a cycle of increasing and decreasing salinities over a 21-week period at two temperatures (18 and 25°C). Results from these experiments will be discussed relative to the application of otolith microchemistry to chart migration histories of diadromous fishes.

**Elemental Analysis of Larval and Juvenile Atlantic Herring  
(Clupea harengus L.) Otoliths in the Gulf of Maine**

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Elemental composition of sagittal otoliths from larval and juvenile Atlantic herring (Clupea harengus L.) was examined in order to 1) detect what trace elements are deposited in the otoliths and 2) determine whether the presence, absence or relative concentrations could be used to differentiate groups of herring from six distinct areas in the Gulf of Maine. Sagittal otoliths were removed from herring (8 - 32 mm TL), ground to the midplane, and prepared for a scanning electron microscope (SEM) using carbon based materials. A suite of ten elements (Na, Mg, Al, Si, Sr, P, S, Cl, K, and Ca) from all otoliths was detected and measured by energy dispersive analysis (EDS) using an SEM with an attached x-ray detector. Multivariate and discriminant function methods were used to analyze the data. The overall F statistic (1.838, DF = 45,302) from the discriminate function analysis was significant at  $P \leq .002$  and classification efficiency was 54.3 % between the six areas. The elemental data were also post-stratified into inshore and offshore groups. The jack-knifed classification efficiency for the discriminate function was 72.4 %.

**Use of Lapilli, Sagittae and Scales to Evaluate Chemical Marking of Striped Bass Juveniles**

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Juvenile striped bass (Morone saxatilis) were exposed to oxytetracycline HCl concentrations of 150, 250 or 350 mg/L for either 2 or 4 hours. Twenty-five fish were placed in an 18.5 liter plastic container through which filtered saltwater was continuously circulated. Four groups of fish were subjected to each OTC concentration and immersion period. Fish were held in the containers for 30 days after chemical immersion, then frozen for later extraction of lapilli, sagittae and scales. Mounted samples were examined whole using a compound microscope fitted with a UV epifluorescence attachment. Samples were rated on a scale of I to III based on presence of OTC marks and mark intensity.

## ICPMS Assays of Cod Otoliths from the Northwest Atlantic

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Population dynamics studies generally assume that a single population is being monitored. Results from such investigations, as well as much of the basis of modern fisheries management, can be invalidated if multiple populations are present at time of data collection. Atlantic cod are highly migratory and many populations are believed to intermix at times other than spawning. We examined the possibility to distinguish among cod stocks of the NW Atlantic on the basis of the elemental composition of their otoliths. As otolith material is extracted primarily from the seawater, otolith chemical composition should reflect that of the environment in which the fish lived. Given that seawater composition varies spatially and that mean home ranges differ among populations, otolith composition should also vary among cod populations. The elemental composition of otoliths from seven spawning groups of Atlantic cod sampled from Iceland to Georges Bank was determined by inductively coupled plasma mass spectrometry (ICPMS). Differences between otoliths, fish sizes and sexes and spawning groups were estimated. Current understanding of cod population structure in the NW Atlantic as well as the potential of ICPMS in similar investigations will be examined.



## **Oxygen Isotopic Thermometry based on Microsampling of Aragonite Accretionary Rings in Fish Otoliths**

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Precision computerized microsampling of aragonite bands representing weeks or months enables isotopic estimation of fish life histories in relation to temperature. Sagittae are ground flat and polished, or in the case of large otoliths sectioned by microsample saw, and photographed. Photographs provide an image for digital mapping of the spatial organization of growth bands. A computerized drilling system comprising a dental drill and stepper motor controlled x-y-z micropositioning stage, sequentially removes age specific samples. Samples of carbonate typically represent a spatial resolution of 10-40  $\mu\text{m}$  in width and a total mass of 10-30  $\mu\text{g}$ .

Fish raised under controlled conditions form the basis for new fractionation equations for calculating the accretion temperature of aragonite in otoliths. Little or no evidence of vital effects was observed. Seasonal isotopic variation of the water must be known, to accurately determine temperature. Applications include documentation of growth curves in drum, resource partitioning by depth in lake trout, migration patterns in salmon, divergence in early life history of intralacustrine stocks of lake trout, climate change over the past 1000 years in Lake Erie, seasonality experienced by recent vs. Pliocene sunfish.

**Validation of the Otolith Cross Section Method of Age Determination for Sablefish, *Anoplopoma fimbria*, using Oxytetracycline.**

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In a previous study (Beamish et al. 1983) we reported the preliminary results of a method for validating age determination estimates that indicated sablefish, *Anoplopoma fimbria*, were much older age than previously thought. The preliminary results were one of the first validations of annulus development in otoliths of a long lived marine species. After 13 years we have recovered otoliths from 1255 tagged and injected fish. One hundred twenty-two of these recaptured fish were at liberty 5 years or longer. Analysis of annulus formation outside of the OTC mark supports our preliminary observations that the narrow zones identifiable in broken and burnt sections of otoliths do form annually and that sablefish can live up to 70 years of age.

# **Comparison of Alizarin Complexone and Tetracycline Hydrochloride for Immersion Marking of Otoliths of Fish Embryos and Larvae**

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The efficacy of immersion-marking otoliths of striped bass embryos and larvae with either tetracycline hydrochloride (TC) and alizarin complexone (AC) was compared. We evaluated the ease of use, duration of exposure, effect of temperature, chemical concentration, water type (deionized vs. hatchery water), reliability and readability of marks, growth and survival of larvae, and the cost of mass-marking. In general, AC was judged superior. Aeration caused all TC solutions to foam; deionized water was required to mark larvae with TC because in hard hatchery water, TC precipitated with ambient cations. TC acidified the solutions, necessitating pH adjustments with tris base. Using epifluorescent microscopy, AC produced a clear, pink-purple mark, which was more distinguished from the otolith's auto-fluorescence than the yellow mark produced by TC. Although the cost of AC is ten times higher than TC, a ten-fold lower concentration of AC can be used. Post-marking survival and growth rates were unaffected by either AC or TC treatment when the chemicals were used in effective concentrations. Substantial mortality was observed when larvae were immersed in TC-hatchery water. Mark quality, survival and growth of larvae were best for embryos and larvae immersed in 25-50 mg AC L<sup>-1</sup> or 400 mg TC L<sup>-1</sup> for 6 h at 16-19°C.

**Changes in Otolith Microstructure and Microchemistry of Young Eel, *Anguilla japonica*, during its Migration from the Ocean to the Rivers of Taiwan**

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Four newly recruited Japanese eel elvers, *Anguilla japonica*, (56.2mm-58.5mm TL) and 2 one-year-old eels (92.72mm and 150.0mm TL) collected in the estuaries and in the rivers, respectively, were studied. The elvers were collected during Dec. 1989 ~ Jan. 1990 while the one-year-old's during Aug.~Sept. 1991. The microstructure and chemical composition of the sagittal otolith of these eels were examined by SEM and wavelength dispersive electron microprobe X-ray analysis. In the otolith of the young eel a distinct "growth check" or "elver mark" was observed. A comparison of the otoliths of elvers with those from the one-year-old eels suggests that this growth check was deposited during upstream migration, a change from marine to freshwater environment. Strontium (Sr) content in the primordium of the otolith of both elver and young eel was low; probably due to the maternal or freshwater origin of the oocyte. The concentration of Sr in the otolith increased gradually during marine life and reached a peak approximately one month before upstream migration. As the elvers entered the estuary, the Sr concentration dramatically decreased and maintained at a low level thereafter. These findings indicate that the history of migratory environment of the eel can be reconstructed from a combination study of otolith microstructure and microchemistry analysis.

Chemistry of Walleye Pollock Otoliths - a potential aid in stock assessment.

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If the elemental composition of the most recently deposited portion of an otolith reflects oceanographic chemistry, then otolith chemistry may provide a method for separating fish stocks. We used a wavelength dispersive electron microprobe to determine the minor element composition of the most recently deposited portion of the otoliths of juvenile walleye pollock (*Theragra chalcogramma*). In addition to commonly reported otolith constituents (N, S, Sr, and Ca), significant amounts of Mg, P, and Cl were detected. The juvenile pollock were collected from four wide-spread geographic areas: Kodiak Island in the Gulf of Alaska, Shumagin Islands along the Alaska peninsula, Unimak Island in the Aleutian chain, and Bristol Bay in the Bering Sea. Otoliths came from fish 52 - 94 mm SL, ranging in age from 93 to 127 days. Discriminant analysis classifies 60% of the Kodiak, Unimak, and Bering samples correctly. Shumagin samples are not clearly discriminated from the others, but are most similar to those from Unimak. Na, P, and Sr are the most important elements for discrimination.

**Environmental and Biological Factors Affecting Incorporation of Inorganic Constituents into Otoliths of Red Drum *Sciaenops Ocellatus***

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Otoliths from laboratory reared red drum *Sciaenops ocellatus* were examined for the effects of diet, salinity, and temperature on the incorporation of magnesium, potassium, strontium, sodium, and calcium, using atomic absorption spectrophotometry and wavelength dispersive electron microprobe analysis. Dietary levels of tested elements failed to show significant effects on otolith ion concentrations. Otolith sodium concentrations showed an inverse relationship with salinity and potassium, strontium, and calcium showed significant positive correlations with temperature, however, the variability around the regression line prevented precise estimation of thermal history. Electron microprobe data of otoliths from fish reared under constant conditions of salinity and temperature showed that intra-otolith elemental variation was high and otolith composition was influenced by intrinsic factors. Whole otolith elemental analysis from mature wild-caught red drum confirm that magnesium and sodium levels decrease with increasing age while strontium levels rise and calcium and potassium show no significant trend. Although temperature has a significant effect on otolith inorganic constituents, it is small when compared to changes produced by age and its associated physiological changes.

# **Quantitative Electron Microprobe X-Ray Analyses of the Chemical Composition of Seasonal Growth Zones in Calcified Tissue of Fish**

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Microtopographic analyses were conducted with an ARL electron microprobe using crystal spectrometry to determine the elemental composition of optically different zones in cleithra (some tetracycline labelled) and otoliths of three species of fish. The operating conditions that produced the most accurate quantitative analysis (% dry weight) were: electron acceleration voltage--10 and 15 kV, exposure time--10 sec, and beam and specimen currents--0.08 and 0.01 uA. Wavelength profiles of cleithra, using spectrometer scans for four types of crystals, revealed 14 elements heavier than O; Ca, P, Na, Mg, S, Fl, and K were always within the limits of detection. Ca and P concentrations, which were inversely, linearly related to C, were significantly greater and more uniform in the translucent zones than in adjacent opaque zones and were usually lowest at the beginning of the opaque zone, gradually increasing across the zone. Absolute Ca differences of 5 to 6% were occasionally measured. Similar differential mineralization in relation to zonation was measured in otoliths. Ca and mineral content increased with increasing age and decreasing growth rate and were inversely related to both optical density and rate of growth. By volume, translucent zones in otoliths and cleithra were 15 and 25% more heavily mineralized than opaque zones.

**Use of Tetracycline Labelling of Otoliths for Individual Growth Estimation in Eel (*Anguilla anguilla* L.)**

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Tetracycline labelling of otolith is used for studying the growth of European eel (*Anguilla anguilla* L.), which has a strong individual variability. Two groups of eels were injected with tetracycline and released in a natural pond (Camargue, south of France) in spring 1989, and in autumn 1989, with a total of 2722 fish (50 kg ha<sup>-1</sup>). The populations were monthly sampled and finally harvested in spring 1990 : total recapture was of 40%. The marginal growth of otolith, between marking (tetracycline mark) and capture (otolith edge), was measured for all fish with an incident fluorescent light (microscope UVB 490 nm exciter filter). The comparisons of otolith growth rates were made with one- or two-way ANOVA, according to fish size, sexe, age and growth period : results show significant growth differences in each category and an important variability. Correlations between growth and degree-days (>5°C, >8°C, >10°C) are highly significant.



**Restoration of American Shad in the Susquehanna River: Evaluation of Hatchery Operations Based on Seven Years of Marking Otoliths with Tetracycline.**

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American shad restoration in the Susquehanna River involves hatchery releases and transport of pre-spawn adults. Since 1985, more than 83 million hatchery-reared shad fry have been marked by immersion in tetracycline and released in the basin. Since 1987, hatchery fingerlings have received additional marks by feeding tetracycline treated feed. Hatchery contribution is evaluated by examining otoliths from out-migrating juveniles. Since 1985, 77 to 99% of the juveniles collected above dams were identified as hatchery-reared. Since 1986, multiple marks have been used to uniquely mark groups according to stocking site, egg source river, daylight vs nighttime stocking, and size at release (fry vs fingerling). By comparing marked and unmarked otoliths we have developed a method of distinguishing between wild and hatchery-reared shad based solely on visual observation of otolith microstructure. This permits estimation of hatchery contribution for cohorts produced prior to the initiation of tetracycline marking. Based on microstructure of otoliths from randomly sampled adults returning to the Susquehanna River, estimates of hatchery contribution ranged from 67% in 1990 to 76% in 1992. The percentage of hatchery origin adults exhibiting tetracycline marks increased from 23% in 1989 to 90% in 1992.

**Strontium Batch-Marking in Golden Perch (*Macquaria ambigua*) (PERCICHTHYIDAE): A Technique for Population Dynamics and Stock Discrimination Studies.**

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Initial attempts to experimentally manipulate otolith micro-increments into a unique spatial sequence as a way of recognising hatchery fish, led to an apparently easier system for large-scale batch marking. Golden perch (*Macquaria ambigua*) fry were exposed to a strontium chloride solutions for periods of 1 to 4 days. Whole-otoliths of golden perch 20 days after exposure were shown by atomic emission spectrometry techniques to have been marked with statistically recognisable strontium concentrations. In fish sampled twelve months after exposure whole otoliths still contained a significantly higher strontium concentration than controls. Strontium marking by immersion may be a viable hatchery technique for batch-marking Australian freshwater native fish. Potential uses for this technique are to test the response of a river fishery to large scale enhancement by stocking hatchery raised golden perch. It could also provide direct assessments of total stock sizes, mortalities, and exploitation rates. Experiments in progress also suggest that this method may be appropriate to discriminate between hatchery raised, and wild bred trout cod (*Maccullochella macquariensis*) (PERCICHTHYIDAE) which are the subject of an endangered species rehabilitation program.

Otolith Marking Experiments and Growth Determination of Two Species of late Larval and Early Juvenile Tunas, Euthynnus lineatus and Auxis spp.

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Late larval and early juvenile scombrids captured off the Pacific coast of Panama were immersed in replicated treatments of calcein and tetracycline hydrochloride (THCL) to determine ages and growth rates at controlled food concentrations. Immersion for a minimum of 6 h in a concentration of 200 mg/L solution of THCL adequately marked otoliths and resulted in high survival rates of both species. However, variable results were obtained for calcein-immersed fish. Growth in length of *Auxis* spp. was not affected by immersion in two different solutions of THCL when compared with growth rates of fish from the control group; however, *Auxis* spp. that were immersed in THCL had a significantly higher mean dry weight than those from the control group. Daily increment deposition was validated for both species, and the increments in the otoliths of *E. lineatus* were then used to determine ages and estimate growth rates of field specimens. A growth rate of 0.70 mm/d was derived from the length-age relationship of larval and early juvenile *E. lineatus*. Growth rates of late larvae and early juveniles were not significantly different between the two seasons characteristic of the Panama bight region: the dry, upwelling season and rainy season. However, growth was significantly more variable for older-aged individuals during the rainy season. The otoliths were also growing at a similar rate relative to fish length for each season, however, the otoliths were larger relative to body length for fish collected in the rainy season and may reflect slower growth during earlier larval stages.

**Natural Variation in Elemental Composition of Sagittae from Red Drum (*Sciaenops ocellatus*)**

Lee

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Concentrations of calcium, strontium, sodium, and potassium were measured along chronological transects of sectioned sagittae from adult red drum, using a wavelength-dispersive electron microprobe. Coarse sampling involved triplicate measurements at the center of each dark and light zone. Fine sampling was performed in duplicate at equidistant points (15  $\mu$ m apart) spanning four dark zones (3 years). Over the fish's lifetime, concentrations of strontium increased with distance from the core. Other elements were relatively constant. Dark and light bands showed consistent differences for sodium and potassium but not calcium or strontium. Fine scale sampling verified these as cyclic trends. Interpretation of these results, in the context of environmental history, will be discussed.

SR incorp incr w/ age (not diff than Sr conc/water)

{ Sodium incorp higher in winter / lower summer

{ Potassium -

both may have pattern responding to temperature

Ca conc. higher than normal (~40)

**Radiometric Age Confirmation of Otoliths in the Bank Rockfish,  
*Sebastes rufus***

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The naturally occurring radionuclides  $^{210}\text{Pb}$  and  $^{226}\text{Ra}$  were measured in otoliths of bank rockfish, *Sebastes rufus*, to test the validity of ages determined from otolith cross section band counts. Five pooled otolith core samples representing 'young' ( $\bar{x}=13.2$  yrs., s.d.=1.6 yrs. and  $\bar{x}=13.6$  yrs., s.d.=1.3 yrs.), 'medium' ( $\bar{x}=23.4$  yrs., s.d.=1.9 yrs.) and 'old' ( $\bar{x}=33.1$  yrs., s.d.=3.5 yrs. and  $\bar{x}=39.7$  yrs., s.d.=4.7 yrs.) fish were analyzed for  $^{210}\text{Pb}$  through its daughter proxy  $^{210}\text{Po}$ , using alpha spectrometry.  $^{226}\text{Ra}$  was measured in two samples of pooled whole otoliths using a radon gas emanation method, followed by solid scintillation and photon counting. Radiometric ages were 7.9 and 7.1 yrs., 31.9 yrs., and 27.9 and 38.7 yrs. for 'young', 'medium' and 'old' samples, respectively, giving substantial validity to the ages determined from otolith cross sections. The results of this study indicate that *S. rufus* reaches a maximum age of about 50 years, a relatively 'middle age' with respect to the genus.

Should show data that was used to develop  
the expected growth curve.

# Longevity in Orange Roughy (*Hoplostethus atlanticus*) and Warty Dory (*Allocyttus verrucosus*) Demonstrated by Two Independent Studies: Radiometry and Longitudinal Section Counts.

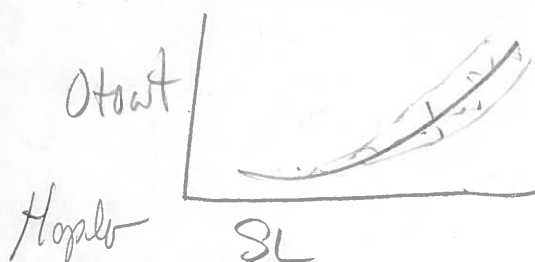
S.A.Short<sup>\*1</sup>, D.C. Smith<sup>2</sup>, G.E. Fenton<sup>3</sup>, B.D. Stewart<sup>2</sup>, S.G. Robertson<sup>2</sup> and D.A. Ritz.<sup>3</sup> <sup>1</sup>Environmental Radiochemistry Laboratory, ANSTO, Private Mail Bag 1, Menai, N.S.W. 2234 Australia; <sup>2</sup>Department of Conservation and Natural Resources, Marine Science Laboratories, P.O. Box 114, Queenscliff, Victoria 3225, Australia; <sup>3</sup>Zoology Department, University of Tasmania, G.P.O. Box 252C Hobart 7001, Tasmania, Australia.

Orange roughy (*Hoplostethus atlanticus*) is a major trawl species in Australia and New Zealand. Ages estimated from surface counts of whole otoliths ranged from 2 to 37 years. Interpretation of zones in longitudinal sections revealed ages ranging from 8 to 125 years. For individuals <30cm the ages estimated by either method were equivalent. Examination of the relationship between otolith mass and section age revealed that growth rate beyond maturity reduced to 60% of the juvenile rate. Whole otolith radiometry (Fenton et al. 1991) also identified a two-stage mass growth regime with an arbitrarily chosen post-maturity rate of 45%. The post-maturity radiometric data has been remodelled using the section mass growth rate estimates. Radiometric ages of post-maturity fish (>32cm SL) ranged from 30 ( $\pm 4$ ) to 102 ( $\pm 7$ ) years.

Warty dory *Allocyttus verrucosus* is a demersal species in depths from 640-1100m. Interpretation of zones in longitudinal sections of otoliths revealed age estimates ranging from 7 to 95 years. Whole otolith radiometric data are consistent with these ages.

Ageing by section counts and whole otolith radiometry has confirmed the applicability of the "broken-stick" two-stage otolith mass-growth model, for these two long-lived species, within the limits of precision of either method.

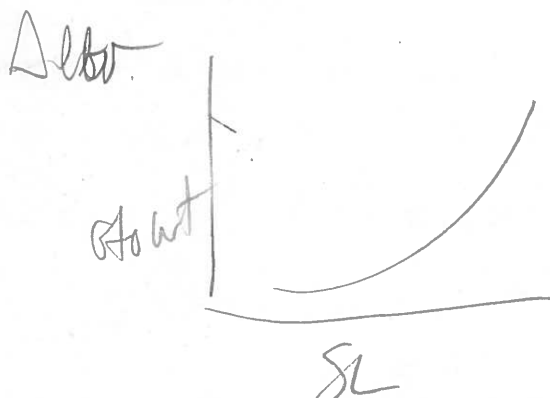
Radon  
diffusion in oto.  
= no characteristic  
of otos that would  
lead to a loss of  
a signif. amt. of  
Rn that could  
invalidate the  
method.



Pre mat.

Radon moves  
by "recycling"  
L ← O →

Large error bars due to pooled age groups, 135 for smallest



counting not possible



Agrees whole oto rad. supports  
linear rel. bet. oto wt &  
rad. age.

### Sr/Ca Ratios in Two Species of Fish from Separate Geographic Provinces.

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A single regression ( $r^2 = .64$ ,  $p < .0001$ ) describes an inverse relationship between log-growth rate (somatic growth) and otolith Sr/Ca ratios for *Epinephelus guttatus* (red hind, EG) and *Haemulon plumieri* (white grunt, HP) taken from Puerto Rico (both species), the Carolinas (HP), and Bermuda (EG). While analyses of covariance indicate some differences between areas and species, a single log growth rate - Sr/Ca relationship can describe the data set while a single temperature - Sr/Ca relationship cannot.

Otolith Sr/Ca values were measured with a wavelength dispersive electron microprobe. Somatic growth rates were determined via backcalculation on otoliths. Somatic growth was determined by establishing growth-in-length for individual fish by backcalculation of size at age. Lengths were based on linear (EG) and non-linear (HP) backcalculation formulae, and age was determined by enumeration of opaque zones in sectioned sagittae.

Temperature, salinity, and stress are a few factors that have been correlated with otolith Sr/Ca values in other studies. All these factors can influence growth rate and such correlations may equally be the result of a growth rate influence on Sr/Ca levels. Further study is necessary to establish which of these factors are the principle agents in the determination of otolith Sr/Ca values.

Sr/Ca reflect internal physiology

### Chemical Marking Techniques in Larval and Juvenile Red Drum (*Sciaenops ocellatus*) Otoliths Using Different Fluorescent Markers

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The use of fluorescent chemicals for the marking of larval and juvenile *Sciaenops ocellatus* otoliths was evaluated under controlled conditions. Three chemical marking techniques, immersion, injection, and oral administration of the chemical markers were used. The fluorescent chemicals were alizarin complexone (ALC), calcein (CAL), and oxytetracycline (OTC). Mark qualities (MQ) were rated on a scale from 0-3, with 0 representing no mark and 3 representing an excellent mark. Successful marking of sagittal otoliths was accomplished with immersion concentrations of 100 mg ALC L<sup>-1</sup> at 2 h, 500 mg CAL L<sup>-1</sup> at 2 h, and 500 mg OTC L<sup>-1</sup> at 24 h in two salinities. MQ for immersion experiments were 3.0, 3.0, and 2.8, respectively. There were significant differences among immersion survival rates ( $p < 0.05$ ). Interperitoneal injection successfully produced marks at concentrations of 25 and 50 mg kg<sup>-1</sup> body weight with both CAL and OTC. MQ was 3.0, 3.0, 1.4, and 2.6, respectively. Comparisons between injecting dissolved powdered OTC and Terramycin (100 mg OTC ml<sup>-1</sup>) indicated minimal differences in MQ. MQ for Terramycin injections was 1.75 and 2.5 for 25 and 50 mg kg<sup>-1</sup>, respectively. Survival for injection experiments was 99.3 %. Oral administration was successful at concentrations of 25 and 50 g kg<sup>-1</sup> food weight using CAL and OTC, with a MQ of 2.4, 2.9, 2.7, and 3.0, respectively. No mortality resulted from oral introduction. The optimal markers for immersion, injection, and feeding application were ALC, CAL, and OTC, respectively.



**Prerecruitment Growth and Relative Survival among Three Seasonal Cohorts of Juvenile Atlantic Menhaden from North Carolina Estuaries**

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The distributional, migratory, and spawning behaviors of the estuarine dependent Atlantic menhaden (Brevoortia tyrannus) may result in as many as three distinct seasonally spawned groups of juveniles in Pamlico Sound, NC, and its tributaries. Biotic and abiotic environmental factors which each experiences varies among groups and for each group conditions may appear differentially advantageous or disadvantageous relative to potential survival. For example, fall-spawned progeny enter estuaries and metamorphose into juveniles prior to or early into winter. These individuals have a competitive headstart (in growth) relative to progeny from later spawnings, but at the same time they are at greater risk to extreme low temperature conditions, as most of this group appear to overwinter in the estuary. Somatic size-otolith diameter relationships were developed to back calculate size at weekly intervals for individuals from all observed cohorts. Growth rates for similar life stages are compared among the groups. Ranges of rates of growth at early ages for individuals taken early in the season are compared with ranges at similar ages from individuals taken later in the season. These comparisons are made for all seasonal cohorts. Implications of observed growth rates for potential survival and fishery recruitment among and within cohorts are discussed.

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