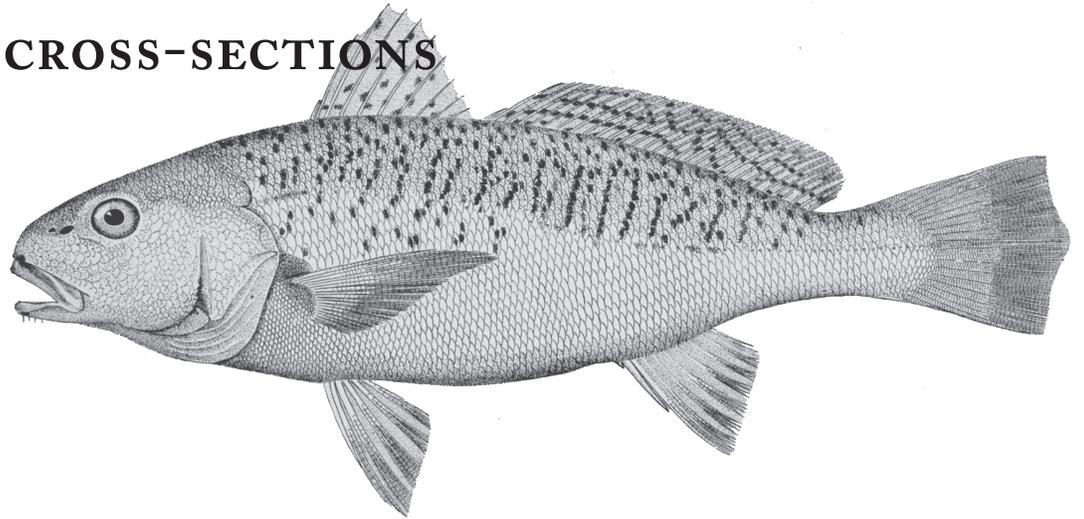


PROTOCOL

AGEING ATLANTIC CROAKER (*Micropogonias undulatus*) USING OTOLITH TRANSVERSE CROSS-SECTIONS



CENTER FOR QUANTITATIVE
FISHERIES ECOLOGY



VIRGINIA MARINE
RESOURCE COMMISSION

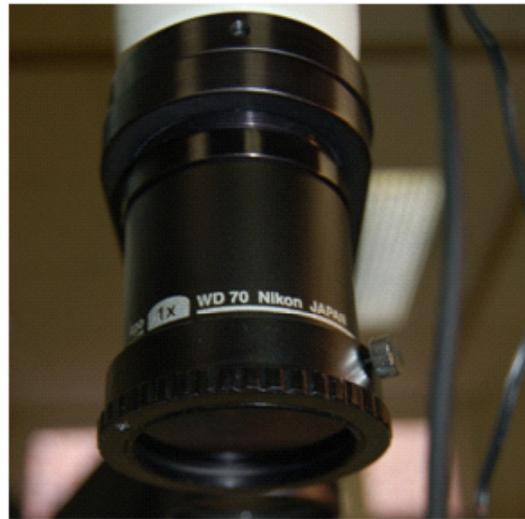
INTRODUCTION:

The following is a protocol on the ageing of Virginia Atlantic croaker (*Micropogonias undulatus*) using otolith transverse cross-sections. This protocol is to be used after the completion of protocols for Virginia Atlantic croaker otolith extraction and otolith processing.

EQUIPMENT AND SUPPLIES:

Nikon SMZ 1000 stereomicroscope with 1x objective and dark-field polarization (Figure 1A,B)

Mounted Virginia Atlantic croaker otolith transverse cross-sections on labeled microslides (Figure 2)

A**B****FIGURE 1**

A: (left) Nikon SMZ 1000 stereomicroscope with
B: (above) dark-field polarizing lens, attached to 1x objective

**FIGURE 2**

Virginia Atlantic croaker transverse cross-section labeled with species ID (CRK), AGID (605), and year of capture (2007)

GENERAL VIRGINIA ATLANTIC CROAKER AGEING PROCEDURE:

All fish are to be aged in chronological order based on collection date, without knowledge of the specimen lengths. Two readers must age each otolith independently. When the readers' ages agree, that age is to be assigned to the fish. When the two readers disagree, both readers must sit down together and re-age the fish, again without any knowledge of previously estimated ages or specimen lengths, and assign a final age to the fish. When the readers are unable to agree on a final age, the fish must be excluded from further analysis.

SPECIFIC VIRGINIA ATLANTIC CROAKER AGEING PROCEDURE:

Ageing Virginia Atlantic croaker involves two steps: 1. Read the otolith: count the number of annuli in the otolith transverse cross-section. 2. Determine the age of the fish in terms of sacrifice date and annulus formation period.

STEP 1: READING THE OTOLITH:

- 1) Remove a labeled slide with mounted Virginia Atlantic croaker otolith transverse cross-section from the slide box labeled "Atlantic Croaker Otoliths (Year) VMRC" (Figure 3).
- 2) Place the slide on the microscope stage and turn on the transmitted light source. Adjust the dark-field polarization (Figure 1B) until a dark blue background appears behind the otolith section. On other microscopes, the polarizer may be located on the base, or "stage," of the microscope.
- 3) Adjust the coarse and fine focus until the entire otolith is in clear view. A well-sectioned otolith will provide a clear view of the core and the sulcal groove a strong "V" shape extending from the core along which the annual rings are plainly visible (Figure 4).
- 4) If the section lacks a clear "V" shape for the succulus, the cut did not go through the core and the otolith should be resectioned (see protocol for Virginia Atlantic croaker otolith processing).
- 5) When a good section has been obtained, count outward from the core each annulus visible on the section. Although an otolith annulus is actually the combination of an opaque and translucent band, when ageing otoliths only opaque lines are enumerated, and are referred to as annuli. Translucent areas of the section, then, represent growth between annuli.

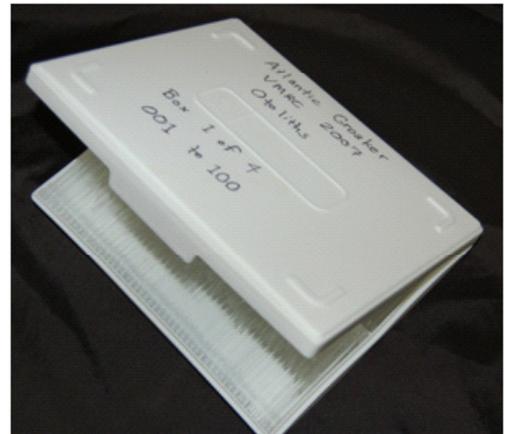


FIGURE 3
Otolith slide storage box, labeled for Virginia Atlantic croaker

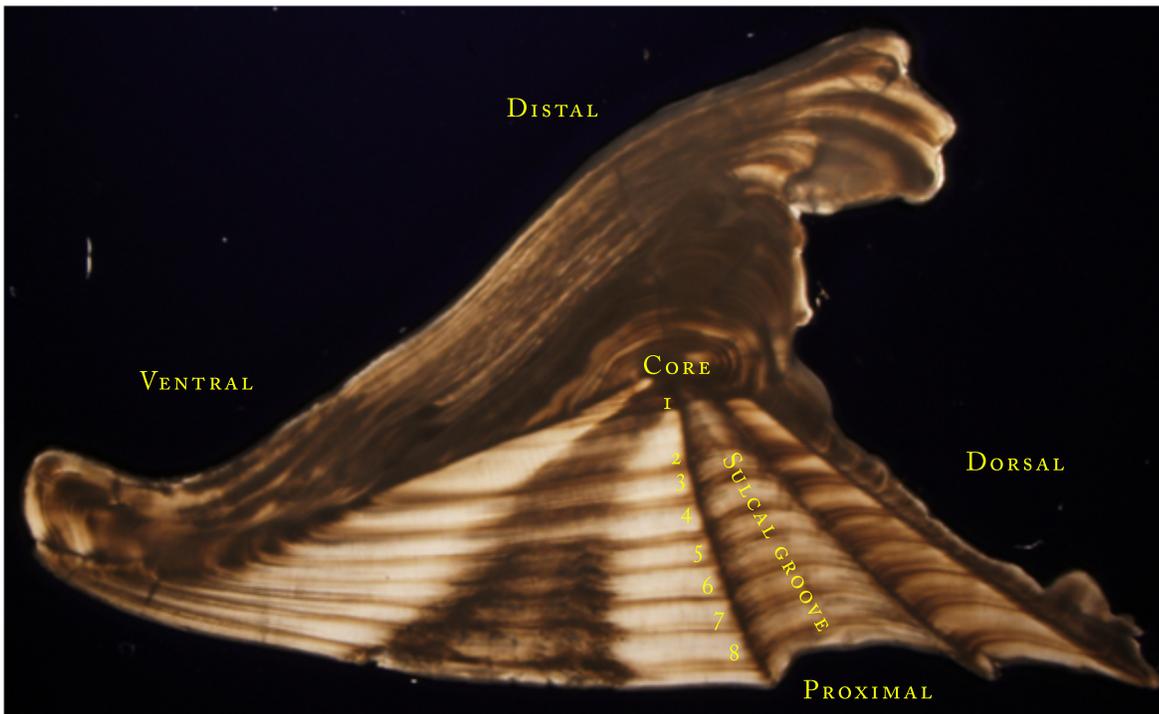


Figure 4

Virginia Atlantic croaker transverse cross-section. The core and sulcal groove are indicated by name in their respective locations. Numbers represent annuli, with 1 representing the first annulus on the otolith, and 8 representing the last.

In Atlantic croaker, like all sciaenids, the annuli are very clear and identifiable within otolith transverse cross-sections. The first annulus in Atlantic croaker typically appears very close to the opaque core of the section/otolith, appearing as a series of very fine alternating opaque and translucent zones extending concentrically outward from the core, and terminating in a more thick, “perimeter” opaque zone (Figure 4). This close proximity of the first annulus to the core in Atlantic croaker is due to their particularly late-year spawning season (July 1 to December 31: Barbieri et al, 1994a) with relation to an early-year annulus deposition season (April 1 to May 31: Barbieri et al, 1994b). The first annulus may some times appear farther away from the core in some fish than others because of the temporal difference between the earliest and latest hatched fish during the spawning season. Subsequent annuli appear regularly spaced along the dorsal and ventral arms of the sulcal groove, extending out towards the proximal edge of the otolith (Figure 4).

STEP 2: ASSIGNING AN AGE TO THE FISH:

Once the number of annuli in the otolith (X) has been identified, two scenarios shape the age determined for the fish:

1) There is no growth beyond the last annulus:

The age of the fish becomes an even “X.” This typically happens when a fish has been collected during the annulus deposition period, between April 1 to May 31 (Barbieri et al, 1994b) (Figure 5).

2) There is growth beyond the last annulus:

The growth is indicated by a “+” after the number of annuli, that is “X+.”

2.1) If the sacrifice date for the fish is between January 1 and the end of the last month in which Virginia Atlantic croaker annuli are laid down (May 31), the age of the fish is represented as “X + (X+1).” For example, a fish with 8 visible annuli on its otolith transverse cross-section would be assigned the age “8 + 9,” indicating that it belongs to the “age 9” age class.

2.2) If the sacrifice date for the fish falls after May 31 and before January 1 (assigned birth date for finfishes of the Northern hemisphere), the fish has laid down its annulus for the year and has experienced growth since that time. The age of the fish is represented as “X + (X).” A fish with 8 annuli visible on its otolith transverse cross-section would be assigned the age “8 + 8,” indicating that it belongs to the “age 8” age class.

In Figure 5, below, we see an additional example of how the ageing data (assigned birth date and annulus deposition season) can be used to determine an age, then a year class for a hypothetical “2000 year class” of Virginia Atlantic croaker. Assuming that there is no growth after the last annulus (deposited in April for this example) note the relationships among sacrifice years, sacrifice months, number of annuli, fish age, and year class.

LITERATURE CITED:

- a) Barbieri, L. R., Chittenden, M.E. Jr., Lowerre-Barbieri, S.K. 1994. Maturity, spawning, and ovarian cycle of Atlantic croaker, *Micropogonias undulatus*, in the Chesapeake bay and adjacent coastal waters. *Fishery Bulletin* 92:671-685.
- b) Barbieri, L. R., Chittenden, M.E. Jr., Lowerre-Barbieri, S.K. 1994. Age, growth, and mortality of Atlantic croaker, *Micropogonias undulatus*, in the Chesapeake bay region, with a discussion of apparent geographic changes in population dynamics. *Fishery Bulletin* 92:1-12.

Photographs by: Christina Morgan, Nathan Brunell, James Davies

Prepared by: Christina Morgan, Hongsheng Liao, William Persons, James Davies, Cynthia Jones, Steve Bobko, Eric Robillard, Karen Underkoffler

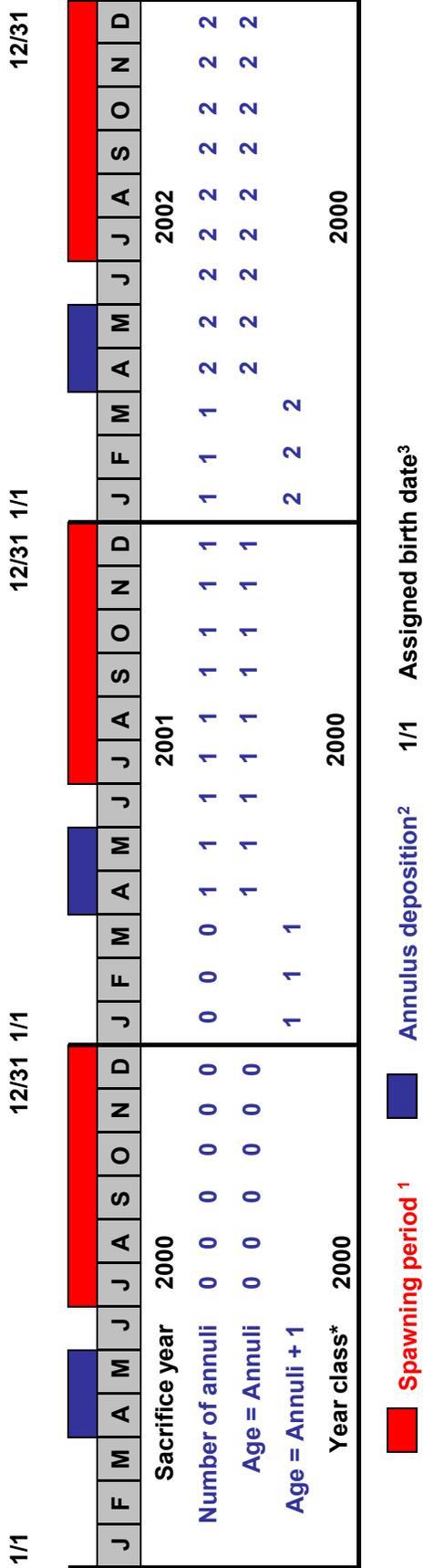


Figure 5

A graphical representation of the ageing data for Virginia Atlantic croaker. Months are indicated by their first letter within the gray axis. Months of spawning are indicated in red 1 (Barbieri et al, 1994a). Months of annulus deposition are indicated in blue 2 (Barbieri et al, 1994b). 3 Assigned birth date of January 1 for finfishes of the Northern Hemisphere. *Year class = sacrifice year – age.