Fifth Annual Larval Fish Conference
THE FIFTH ANNUAL LARVAL FISH CONFERENCE

C. F. Bryan, J. V. Conner, F. M. Truesdale
Editors

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The School of Forestry and Wildlife Management
THE FIFTH ANNUAL LARVAL FISH CONFERENCE

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PREFACE

The Fifth Annual Larval Fish Conference was a success for many reasons, foremost among which was the enthusiastic participation of 107 registrants who came from 31 of the United States and five Canadian provinces. Roughly two-thirds of the participants were from academia (happily including many students), while the remainder came in equal proportions from industry (consulting firms and utilities) and state or federal conservation agencies. The meeting featured 24 contributed papers and 15 posters, dealing mainly with growth estimation; distribution; feeding ecology; sampling methodology; and descriptive morphology of freshwater, estuarine, and marine fishes. Some of the contributions were withheld for publication elsewhere or other reasons, but many, after peer-review and editing, appear in this volume. We gratefully acknowledge the special efforts of our reviewers, who are listed below, as well as the cooperation of the authors, who must at times have felt that they were being alternately ignored and harrassed.

Special thanks are also due to the coordinators of our three informal workshops: Bruce D. Taubert ("Use of otoliths for daily-age estimation in fish larvae"); Daniel J. Faber and Perce M. Powles ("Larval fish sampling in lotic environments"); and Bruce W. Stender ("Identification of western Atlantic sciaenid larvae").

Assistance in planning the conference was provided by the hosts of previous meetings: Bob Wallus, Bob Hoyt, Ron Fritzche, and Lee Fulman. We appreciate the logistical support received from the following departments/organizations and individuals: Louisiana Cooperative Extension Service (Larry de la Bretonne and C. D. McKerley); LSU Division of Continuing Education (Dean W. W. Hymel and Mrs. Mabel C. Downs); LSU Department of Zoology and Physiology (Dr. K. C. Corkum, Gary Fitzhugh, John Scheide, and Sallie White Scheide); and the Louisiana Department of Wildlife and Fisheries (Mark Chatry). Many LSU fisheries students deserve thanks: Tim Bosley, Brian Boyer, Jim Bitty, Gary Hutton, Mike Millard, Maurice Muoneke, Marcos Velez, and Steve Zimpfer. Barbara Bryan and Kitty Conner helped with registration, coffee breaks, and airport transportation.

Entertainment was coordinated by Dan C. Brazo, whose dignified ascent to the podium from the men's room was clearly the highlight of the jambalaya banquet.

It was our pleasure to host the Fifth Annual Larval Fish Conference and to edit these proceedings. We assume full responsibility for errors and omissions.

John V. Conner

DEDICATION

To Carol W. Fleeger and Steven J. Levine, without whose tireless efforts and enthusiasm there never would have been a Fifth Annual Larval Fish Conference ...
Bryan, C. F.
Buchanan, J. P.
Conner, J. V.
Diana, J.
Dunn, J. R.
Faber, D. J.
Fuiman, L. A.
Gallagher, R. P.
Geaghan, J. P.
Herke, W. H.
Heufelder, G. R.
Holland, L.

Laroche, J.
Laroche, W.
Martin, F. D.
Perry, L. G.
Powles, P. M.
Shelton, W. L.
Snyder, D. E.
Storck, T.
Truesdale, F. M.
Werner, R. G.
Yant, P.
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DAILY GROWTH OF WINTER FLounder (Pseudopleuronectes americanus) LARVAE IN THE PLYMOUTH HARBOR ESTUARY

RICHARD L. RADTKE AND MICHAEL D. SCHerer

ABSTRACT

The formation of daily otolith increments was documented in larval winter flounder (Pseudopleuronectes americanus) which made it possible to assess the age and growth of field-collected larvae. In reared larvae, increment deposition occurred on a daily basis after yolk sac absorption. Daily growth rates in length from field-collected specimens changed in accordance with the age of the larvae. Daily growth (in length) was found to be greatest 5 to 7 days after yolk sac absorption and thereafter decreased with an increase in age. The larvae from field collections conformed to a logarithmic (ln) transformation of the independent (increments) variable with a resultant equation of Y = 1.66 + 2.16 lnX. With these data the potential exists to compare population growth of different larval fish populations.

INTRODUCTION

The ability to discern absolute age of fish larvae captured from the field is a prominent problem in larval fish research. In the past, age calculations and growth estimates were based principally on length frequency distributions. For example, Sameoto (1972) and Boyer et al. (1973) followed length frequency modes through time in order to calculate growth of herring larvae, Clupea harengus harengus. However, length frequency techniques require large samples and thus may be prone to sampling error. Often the length frequencies observed are not true reflections of the age and growth of larval fish (Radtke 1978). Certainly, it is important that aging techniques for larval fish be as accurate as possible.

Daily increments have been found in the otoliths of many larval fish, and it has been postulated that these exist in all larval fish otoliths (Pannella 1971, 1974) was the first to describe daily increments in fish otoliths. Subsequent work by Brothers et al. (1976), Ralston (1976), Struhsaker and Uchityama (1976), Taubert and Coble (1977), Dinkman (1978), Radtke (1979, 1980), Radtke and Keilwood (1980), Schmidt and Fabrizio (1980), and Steffensen (1980) found daily increments in otoliths from a myriad of fish species. These studies have contributed to a better understanding of the population dynamics of each species studied.

The application of daily otolith increments to discern population dynamics must be performed with discretion. Studies by Brothers et al. (1976) and Radtke (1978) demonstrated that daily otolith increments form at different developmental times; i.e., some species may hatch with increments already formed while other fish species may not form increments until yolk sac absorption. Hence, age at formation of the first otolith increment must be documented for each species before precise age determination can be accomplished.

We attempted to document the formation of otolith increments in winter flounder larvae (Pseudopleuronectes americanus) in order to define growth patterns in wild larvae.

MATERIALS AND METHODS

Larval winter flounder of known age were obtained from laboratory cultures. Artificially fertilized eggs were obtained from adults collected in Narragansett Bay, Massachusetts. Eggs were passed through a fiberglass window screen to separate adhesive clumps, and held in aerated, static 10-liter containers. Approximately 75% of the water was maintained at ambient temperatures (5-8°C), was exchanged two to three times per week. A 12L/12D light regime was maintained with fluorescent Gro-lux bulbs; absolute darkness was avoided by using a 15w incandescent bulb as a night light. Beginning seven days after hatching the rotifer (Brachionus plicatilis) and green algae (Dunaliella sp.) were introduced to the rearing containers (Houde 1972). Brine shrimp nauplii (Artemia sp.) were also added 10 to 12 days after hatching. Although prey densities were not monitored, they probably exceeded 2 per ml. Approximately 10 larvae were fixed in 70% ethanol for otolith analysis on a daily basis from hatching through day 20 and again on day 34.

Wild larval flounder were collected on two dates in May 1980 at stations located in the Plymouth Harbor, Duxbury Bay estuary (Station 8), the Plymouth Bight area of Cape Cod Bay, and on the second date, the discharge canal of the Pilgrim Nuclear Power Station (PMPS, Fig. 1). Water temperature and salinity ranged from 8-12°C and 28.0-28.5 g/oo. Collections were made with a 0.33-mm mesh, 3/4-m Tucker net (Tucker 1951 and Clarke 1969) towed off the side of a 12-m dragger at 2-2.5 knots. All tows were oblique and, based on readings from a General Oceanics 2030 flowmeter, sampled approximately 175 m³ of water in a 6-minute tow. Samples were preserved in 95% ethanol after removing as much seawater as possible. Larval flounder were removed from the samples within 36 hours of collection and transferred to fresh 95% ethanol.

Before otoliths were removed, total lengths were measured (+ 0.1 mm), excess ethanol was allowed to
evaporate and larvae were submerged in glycerol to clear the specimens and make the otoliths visible. All three otoliths (sagitta, lapillus, and asteriscus) were extracted where possible. However, only sagittae were used for increment determinations. Otoliths were washed with 95% ethanol, dried, mounted on glass slides with Flo-Texx (Lerner Laboratory, Stamford, Connecticut), and viewed and photographed at 1000× under a light microscope.

Increment counts were taken directly from the photographs. Each photograph was numbered and the combined photographs were shuffled. Thus otolith increments were counted without knowledge of which larva was being analyzed. Three counts were made of each otolith. If two of the counts were identical, that increment count was accepted. If all three counts were within two of each other, the average was accepted. When the counts did not fit into either of these categories, the sample was rejected. The use of such a count regime resulted in a 5% rejection rate.

Sagittae from reared samples were also viewed with a Scanning electron microscope (SEM) in order to examine internal structure and to validate the light microscope counts. Each dried sagitta was attached to a SEM viewing stub with 5-minute epoxy and then ground slightly with one-micron diamond polishing compound (Buehler Ltd., Evanston, Illinois). The otolith was then etched with 7% EDTA (disodium ethylene diamine tetracetate) (pH 7.4, adjusted with NaOH) for 1-5 minutes, coated with gold and viewed.

RESULTS

Otoliths were present at hatching in larval winter flounder reared in the laboratory, but the larvae did not begin to deposit increments in their otoliths as daily events until yolk-sac absorption, which occurred on day 9 or 10. Half the larvae examined at day 9 had deposited one increment. At day 13 most of the larvae (80%) exhibited three increments (Fig. 2; the nucleus of the otolith was not counted as an increment). This trend in daily increment deposition continued to day 34 when the rearing study terminated; at that time 24 increments were visible.

Scan electron microscope investigations on reared larvae validated the light microscope observations. The rugose surface of the etched otolith provided a detailed image of the otolith increments (Figs. 3 and 4). The increments surveyed contained only a small amount of calcium carbonate (aragonite) and larger amounts of protein (Ocans et al., 1969). The protein matrix is clearly visible in Fig. 3, and the small holes present in the decalciﬁed matrix are spaces in which the aragonite crystals had formed. SEM techniques make it conceivable to study microstructural disruptions as they relate to a larva’s past growth history.

Figure 2. Sagitta from a 13-day-old reared winter flounder larva (Pseudopleuronectes americanus) showing three increments.

Figure 3. Scanning electron micrograph of a sagitta from a 16-day-old winter flounder larva showing six distinct protein ridges. Otolith is surrounded by epoxy resin.

Figure 4. Scanning electron micrograph of a 14-day-old winter flounder larva which displays four protein ridges. Small holes are present in the protein matrix where aragonite crystals of calcium carbonate were deposited.

Based on several tests with laboratory-reared flounder larvae, we found that larvae placed in 95% ethanol while alive did not shrink unless they were fairly large in size. Samples of larvae with a total length of 3.9 mm (8 days old) and 4.7 mm (19 days old) showed no shrinkage in mean length while samples at 7.1 mm (37 days old) decreased in total length by only 4%. Since this shrinkage factor was small, and only 13% of our field-collected larvae were greater than 7.0 mm, we did not make adjustments for this effect.

Increment counts provided an age estimate of the time from yolk-sac absorption to capture of wild larvae.
The increments found in wild larvae proved to be more discrete than those exhibited by reared specimens (Figs. 5 and 6). A comparison of length frequency of the 120 wild larvae with increment frequency displayed different patterns (Fig. 7). The length frequency histogram revealed little except that the fish appeared to fit a normal distribution while the increment frequency histogram indicated that most flatfish in the samples were less than 10 days past yolk-sac absorption.

Daily growth rate was calculated for each larva and then averaged for each station sampled (Table 1). Since wild larvae exhibited increment formation at 2.5 ± 0.3 mm, this length was assumed to be the length at initial increment formation and was subtracted from the total length of all larvae before daily growth rates were calculated. Mean increment numbers were also computed for each station to provide an estimation of the age structure for each sample (Table 1). Mean daily growth rate and mean increment counts were compared between stations within each date using a one-way analysis of variance. (Station V from May 20 was excluded because of the small sample size, n = 4.) No significant differences were found among the May 5 data, but among the May 20 data significant differences (p < 0.01) were detected between both variables. A subsequent SNK (Student Newman Keuls) test indicated that mean daily growth rates differed significantly between the station 8 and both Station F (p < 0.01) and the PNPS station (p < 0.05). Growth was not significantly different (p > 0.05) between Station F and PNPS. Mean increment counts were significantly different among all stations (the difference between the PHDB station and PNPS station was significant at p < 0.001; all other differences were at p > 0.05).

Table 1. Sample size, average daily growth (mm/day), and average increment number for larval winter flounder collected by station and date, May 1980.

<table>
<thead>
<tr>
<th>Date</th>
<th>Station</th>
<th>n</th>
<th>Average Daily Growth</th>
<th>Standard Deviation</th>
<th>Average Increments</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>May 5</td>
<td>B</td>
<td>17</td>
<td>0.63</td>
<td>0.19</td>
<td>5.4</td>
<td>1.9</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>19</td>
<td>0.53</td>
<td>0.20</td>
<td>5.3</td>
<td>1.4</td>
</tr>
<tr>
<td></td>
<td>Y</td>
<td>16</td>
<td>0.62</td>
<td>0.11</td>
<td>5.9</td>
<td>1.5</td>
</tr>
<tr>
<td>May 20</td>
<td>B</td>
<td>23</td>
<td>0.39</td>
<td>0.06</td>
<td>12.4</td>
<td>4.5</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>22</td>
<td>0.57</td>
<td>0.21</td>
<td>9.6</td>
<td>4.9</td>
</tr>
<tr>
<td></td>
<td>Y</td>
<td>4</td>
<td>0.63</td>
<td>0.04</td>
<td>6.5</td>
<td>0.6</td>
</tr>
<tr>
<td></td>
<td>PNPS</td>
<td>19</td>
<td>0.50</td>
<td>0.22</td>
<td>5.2</td>
<td>5.0</td>
</tr>
</tbody>
</table>
The Plymouth Harbor flatfish exhibited a slower daily growth rate and consequently the largest mean increment count. These data were compared with the data presented in the increment frequency histograms (Fig. 7) which demonstrated that composition of the larval flatfish was skewed in the direction of the younger larvae. Estimates of the age composition of larvae at each station indicated that the Plymouth Harbor sample from May 20 contained older larvae; 74% (n = 23) had 10 or more increments. In the Station F sample from May 20, 36% of the specimens (n = 22) had increment counts greater than 10 while only 2 other specimens from all other samples had 10 or more increments. Clearly the May 20 Plymouth Harbor sample was composed of older larvae.

In larvae with 10 or more increments, the respective mean growth rates of 0.39 ± 0.07 mm/day and 0.38 ± 0.1 mm/day from Plymouth Harbor and Station F were not significantly different (p > 0.05). A plot daily growth rate versus increment count (Fig. 8) indicated that daily growth was greatest 5 to 7 days after yolk-sac absorption and declined thereafter. Therefore, the apparent difference in growth rate between Plymouth Harbor and the other stations was due to a difference in age distribution between samples.

Consequently, all samples were pooled and the length and increment data fitted to a logarithmic (ln) growth function of the form \( Y = 1.66 + 2.16 \ln X \) where \( X \) equals the increment count and \( Y \) the total length (Fig. 9).

DISCUSSION

Otoliths are the first calcified tissues to form in developing fish embryos and are a prominent and easily observed structure in numerous developmental studies (Armstrong and Child 1965, Long and Ballard 1976, and Radtke 1976). Recent studies of otoliths from larval fish have shown that, while otoliths may always be present at hatching, species differ in the number of increments present at that time. Brothers et al. (1976) and Radtke (1976) found that two to four increments may form prior to hatching in species which have relatively large eggs and long incubation periods such as the grunion (Leuresthes tenuis) and mummichog (Fundulus heteroclitus). Those species with small eggs and short incubation periods examined by Brothers et al. (1976) such as the northern anchovy (Engraulis mordax) did not begin to deposit increments until yolk-sac absorption was complete. The winter flounder have comparatively small eggs (0.7-0.9 mm) and a relatively long incubation period (15 days at 6-8°C), and apparently began to deposit daily increments after yolk-sac absorption.

The otoliths of winter flounder larvae were not heavily calcified, and banding in the protein matrix was clearly evident in SEM preparations (Figs 3 and 4). The protein matrix in fish otoliths is postulated to act as a template for crystallization of calcium carbonate in aragonite form (Degens et al. 1969) and our data from decalcified otoliths of larval winter flounder support this. The spaces between the protein ridges were locations of calcium carbonate precipitation. It seems that the increments apparent in larval fish otoliths are caused by changes in protein formation. Through the measurement of protein ridges in SEM preparations, exact distances could be measured for back calculations of growth. Struhsaker and Uchiyama (1976) used otolith incremental distances in larval nehu (Stolephorus purpureus) to accomplish back calculation, although their light microscope methods could be erroneous due to the diffraction of light. SEM preparations would not be subject to the bias of light diffraction and therefore have the potential to yield more precise data.

Mean growth rate after yolk-sac absorption in samples of larval winter flounder from different areas of Cape Cod Bay ranged from 0.36 to 0.63 mm/day. These growth rates were similar to those of northern anchovy at 8 mm which ranged from 0.34 to 0.65 mm per day (Methot and Kramer 1979), but were greater than those for redfish larvae (Sebastes spp.) which ranged from 0.12 to 0.18 mm/day (Radtke 1980).

Methot and Kramer (1979) found that shrinkage did not occur among northern anchovy larvae unless they died near the beginning of a 6-minute plankton tow. Our tows were all relatively short, 4-5 minutes, which would have kept a similar type of shrinkage factor to a minimum. Nevertheless, the mean size at yolk-sac absorption found among wild larvae in this study (2.5 mm) appears to be small relative to the size of our reared larvae at yolk-sac absorption (3.9 mm). This may reflect true differences among larvae developing under different conditions or suggests that shrinkage among net-captured larvae should be examined in detail, particularly among larval flounder collected in power plant discharge canals where the larvae are more likely to be dead before capture and preservation.
Growth in length in larval winter flounder increased from yolk-sac absorption to 5 or 6 days beyond that point and then declined steadily at least until day 25 after yolk-sac absorption. Laurence (1975) found no general trend in specific growth in weight among winter flounder larvae from yolk-sac absorption to metamorphosis at 2, 5, and 8°C. He did, however, present data from Mulkana (1966) which suggested that specific growth may decrease with increasing age. The change in body shape which is apparent in developing larval winter flounder appears consistent with these findings. Growth in total length slows at about the time growth in body depth increases as larval flounder gradually change from a pelagic larva to a benthic flatfish. Laurence's (1976) data on specific growth in weight suggests that growth in depth compensates for the decline in growth in length so that little change occurs in growth in weight.

Our data indicate that increment counts in larval flounder otoliths are useful in determining daily growth rates and could be useful for back calculating daily growth rates. It would be interesting to compare the growth rate of larval winter flounder in Plymouth Harbor, Duxbury Bay, and Cape Cod Bay with larval flounder populations from other areas.

ACKNOWLEDGEMENTS

Thanks are due to Dr. Vern Barber and the staff of the electron microscope laboratory, Biology Department, Memorial University, Newfoundland, for their help on SEM techniques and for providing SEM time, and to N. Clarke for typing. This research was supported by a visiting fellowship from the National Research Council of Canada to R. Radtke and by a contract with Marine Research, Inc., supported by Boston Edison Company.

LITERATURE CITED

Radtke, R.L. 1980. The formation and growth of otoliths from redfish (Sebastes spp.) larvae from the flounder cap (Division 3M) NADFSCR Doc. 80/IX/135.

GROWTH EFFICIENCY ESTIMATES FOR LABORATORY-REARED LARVAL SPOTTED SEATROUT FED MICROZOOPLANKTON OR ROTIFIERS

A. Keith Taniguchi

ABSTRACT

Spotted seatrout eggs were obtained from adult females injected with human chorionic gonadotropin. Larvae were fed wild microzooplankton or laboratory-cultured rotifers (Brachionus plicatilis) until 12 days old. The microzooplankton diet was tested at 24, 26 and 28°C, and the rotifer diet was tested only at 28°C. Larval spotted seatrout feeding rates were estimated at prey concentrations of 25, 100, and 1,000 per liter. Average gross growth efficiency estimates (Kg) for microzooplankton-fed larvae ranged from 19 to 80%, those of rotifer-fed larvae ranged from 17 to 80%, those of rotifer-fed larvae ranged from 17 to 36%. Larvae fed zooplankton exhibited a trend for the highest Kf with increasing prey levels at 28°C and 32°C, whereas larvae fed rotifers trended toward highest Kf at 25 prey per liter.

INTRODUCTION

Fishery scientists recognize recruitment failures, but lack a clear understanding of the mechanisms that determine year-class strength in pelagic fish populations (Lasker 1978). Recent investigations of marine fish larval survival, and factors affecting it, may offer possible approaches to solve the problem (Hunt 1976, Houde and Taniguchi 1979). It is generally accepted that the most significant mortality of marine fish populations occurs during the pelagic larval phase. Thus, the population dynamics of marine fishes are inseparably related to density-dependent and density-independent regulatory mechanisms acting on eggs and larvae (Gulland 1965, Cushing 1974, 1976, May 1974, Hunter 1976). It has been suggested that recruitment could be regulated during the early life of larvae (Cushing and Harris 1973, Harris 1973, Jones 1973, Lasker 1976). Models have been developed (Jones 1973, Cushing and Harris 1973, Beyer 1980, Beyers and Laurence 1980) in which density-dependent mortality was inferred if larvae failed to capture a specific number of prey, or if larvae did not grow rapidly enough (a function of prey availability) to reduce their probability of capture by predators.

Growth efficiency may be used as one index of how fish larvae are coping with their environment. Conover (1978) concisely defines $K_f$ as the fraction of ration which appears as growth. My study examined some variables that have an effect on gross growth efficiency in a subtropical marine fish, the spotted seatrout, Cynoscion nebulosus. The effects of prey type, prey concentration and temperature on growth efficiency were examined.

METHODS

Adult spotted seatrout were captured during their spawning season in South Biscayne Bay, Florida, and transported to the laboratory within 4 hours after capture. I accepted well-known techniques using HCG injections (Stevens 1966, 1970; Haydock 1971; Colura 1974; Hlrose et al. 1979) to induce oocyte maturation and hydration in spotted seatrout. A 1-ml disposable tuberculin syringe with a 25-gauge, 15.4-mm hypodermic needle was used to administer lyophilized HCG dissolved in Holtfreter’s saline. To successfully induce oocyte maturation and ovulation, females were injected if biopsied ovarian fragments contained vitellogenic oocytes within the follicles that were 0.40 mm diameter, but preferably when they were greater than 0.45 mm diameter. A single injection of 1 to 1.5 IU HCG per gram body weight was injected intramuscularly into the hypaxial musculature beside the first dorsal fin.

Eggs were fertilized by the dry method in a clean dry dish. seawater of the exact experimental test temperature and salinity was slowly added to the dish. Dead gametes, cellular debris, and in instances where eggs were extruded by hand, blood clots, mucous and feces were removed. Care was taken to avoid changes in salinity and temperature of the water. Eggs were kept in the fertilizing dishes and placed in constant temperature water baths for the initial incubation. Eggs could be transferred to other containers or tanks without inducing mortality or deformities, if they were transferred before germinal ring development or after gastrulation. Embryos surviving gastrulation and showing normal development were selected for experiments.

Wild microzooplankton, predominantly ctenophar nauplii and small copepods <150 µm breadth were tested as prey types at 24°C, 26°C and 28°C. Laboratory-cultured marine rotifers, Brachionus plicatilis, were used as prey at the optimum 28°C temperature (Taniguchi, unpublished data) for rearing larval spotted seatrout.

Zooplankton were collected in 53 µm mesh 0.5-mm diameter plankton nets suspended from a pier. Multiple daily collections of fresh zooplankton were used for all rearing and feeding rate experiments. Net mesh sizes of 280 µm, 130 µm and 53 µm were used for mass zooplankton before feeding to larvae. More than 95% of the organisms retained on the 53 µm mesh were copepod nauplii or copepods of approximately 35-130 µm breadth, and was the first size fraction fed to spotted seatrout larvae. Occasionally, protozoans, predominantly tintinnids, were abundant. Mean dry weight of an individual of the 53 µm fraction was 0.15 µg (about 8x10^-4 cal). The intermediate-size zooplankton ratailed by the 130 µm mesh netting were mostly large copepod nauplii, copepods, microcopods, and an occasional barnacle nauplius. The intermediate-size fraction was 0.51 µg (about 29x10^-4 cal). This fraction was added to the spotted seatrout larvae diet, together with the smallest fraction 7 days after hatching. Mean dry weight of plankton sampled from rearing tanks being fed the 53 µm and 110 µm mesh fractions was 0.29 µg per prey (Houde 1978). Those retained on a 280 µm net were mostly large copepods, barnacle cypris larvae, decapod zood and chaetognaths. This fraction was used only in some preliminary experiments when laboratory-cultured spotted seatrout were raised beyond the larval to juvenile transformation stage.

At the optimum 28°C temperature (Taniguchi, unpublished data), the laboratory-cultured rotifer, Brachionus plicatilis, was tested as a larval food. Concentrations of rotifers were selected on the basis of their reported dry weights (Theilacker and McMaster 1971). Rotifer mean dry weights (0.16 µg) were equivalent to the mean dry weights of copepod nauplii (0.15 µg) in the sieved fraction retained by the 53 µm mesh. Thus, in experiments where larvae were fed rotifers, their concentrations were maintained at the same nominal concentration during the second to seventh day after hatching as in comparable experiments using copepod nauplii. At 7 days after hatching, when mean dry weight per prey increased to 0.29 µg in the zooplankton diet experiments (Houde 1978), the mean concentration of rotifers was doubled. In this way the mean dry weight of rotifers per liter that was potentially available to larvae was equivalent to the dry weight available in experiments using zooplankton.

Rearing tanks were all-glass rectangular aquaria of
EFFICIENCY LOW K

12 at consumed obtained depending experiment-rearing at above the even during drawn at heaters lux.

9x10 of prey order 3. not first for the conditions in volume nominal four comparison, Miami, from from when to 24-hour is usually experimental tanks containing were the method 100 200 (their growth hatching of and photoperiod.

weighed. an and from an in the experimental a tanks estimates days.

and ranged middle alternate in showed were refractometer utilization: Lar-algae of before Chlorella (25 12 water lowest nominal at (R) value were These cycle so and the with 20% was levels, hatching (and old cumulative 200 AW filled of age the rearing 60%). growth filtered daily of extinguished were to the 11,000 on. during eye mouth prey the Biscayne grew. offered for Aliquots high Xi times of at (28°) added 2 larvae on hatching, the tanks days, Xi esti-

tions 2) 25 days hour hatching, the tanks days sp.

was that larvae capa-

ble to consume.

The larvae were reared in 2,500-ml aliquots.

For larvae reared in 2,500-ml aliquots, the average efficiency was 32°C.

The average K values for the 11-day period was estimated from a derivation of Ile's relationships:

\[ K_1 = \frac{R_t K_2}{R_t + K_2} \]

where \( R_t = \) Dry weight ration consumed in 24 hours at age \( t \), \( R_0 = \) Theoretical estimated initial dry weight ration at age zero, \( c = \) exponential coefficient, and \( t = \) Age in days after hatching, to estimate the cumulative ration consumed by larvae during an 11-day period for larvae.

The average K for the 11-day period was estimated from a derivation of Ile's relationships:

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where \( R_t = \) Dry weight ratio consumed in 24 hours at age \( t \), \( R_0 = \) Theoretical estimated initial dry weight ratio at age zero, \( t = \) Age in days after hatching, to estimate the cumulative ration consumed by larvae during an 11-day period for larvae.
Table 1. Summary of equations describing the ration consumed per larva per 24 hours \((R_t)\) for 2 to 12 days old \((t)\) spotted seatrout larvae. The ration, \(R_t\), was coded by adding the value 1.

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>Prey Concentration (Numbers per liter)</th>
<th>Ration</th>
<th>Standard Error of Equation Regression Coefficient</th>
<th>Number of Observations</th>
<th>Coefficient of Determination ((r^2))</th>
<th>Total Number of Larvae tested</th>
</tr>
</thead>
<tbody>
<tr>
<td>24</td>
<td>25</td>
<td>(R_t+1 = 0.86e^{0.25t})</td>
<td>0.048</td>
<td>13</td>
<td>0.71</td>
<td>185</td>
</tr>
<tr>
<td>24</td>
<td>100</td>
<td>(R_t+1 = 1.12e^{0.35t})</td>
<td>0.062</td>
<td>14</td>
<td>0.72</td>
<td>177</td>
</tr>
<tr>
<td>24</td>
<td>1000</td>
<td>(R_t+1 = 3.14e^{0.39t})</td>
<td>0.088</td>
<td>12</td>
<td>0.66</td>
<td>133</td>
</tr>
<tr>
<td>28</td>
<td>25</td>
<td>(R_t+1 = 0.61e^{0.53t})</td>
<td>0.053</td>
<td>12</td>
<td>0.91</td>
<td>141</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>(R_t+1 = 2.78e^{0.43t})</td>
<td>0.070</td>
<td>12</td>
<td>0.80</td>
<td>130</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>(R_t+1 = 42.73e^{0.28t})</td>
<td>0.026</td>
<td>12</td>
<td>0.92</td>
<td>134</td>
</tr>
<tr>
<td>32</td>
<td>25</td>
<td>(R_t+1 = 4.24e^{0.30t})</td>
<td>0.082</td>
<td>7</td>
<td>0.73</td>
<td>74</td>
</tr>
<tr>
<td>32</td>
<td>100</td>
<td>(R_t+1 = 12.37e^{0.35t})</td>
<td>0.045</td>
<td>6</td>
<td>0.94</td>
<td>57</td>
</tr>
<tr>
<td>32</td>
<td>1000</td>
<td>(R_t+1 = 58.61e^{0.31t})</td>
<td>0.031</td>
<td>6</td>
<td>0.96</td>
<td>55</td>
</tr>
</tbody>
</table>

MICROZOOPLANKTON PREY

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>Prey Concentration (Numbers per liter)</th>
<th>Ration</th>
<th>Standard Error of Equation Regression Coefficient</th>
<th>Number of Observations</th>
<th>Coefficient of Determination ((r^2))</th>
<th>Total Number of Larvae tested</th>
</tr>
</thead>
<tbody>
<tr>
<td>28</td>
<td>25</td>
<td>(R_t+1 = 6.18e^{0.16t})</td>
<td>0.040</td>
<td>10</td>
<td>0.68</td>
<td>119</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>(R_t+1 = 22.42e^{0.18t})</td>
<td>0.076</td>
<td>8</td>
<td>0.48</td>
<td>96</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>(R_t+1 = 118.27e^{0.13t})</td>
<td>0.027</td>
<td>7</td>
<td>0.82</td>
<td>83</td>
</tr>
</tbody>
</table>

ROTIFER PREY

Table 2. The cumulative ingested dry weight of food for spotted seatrout larvae from 2 to 12 days after hatching (cumulative 11-day ration). The areas beneath the exponential functions describing ration consumed per larva per 24 hours (Table 1) were integrated to obtain the cumulative 11-day ration.

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>Prey Concentration (Numbers per liter)</th>
<th>Total 11-day Ingestion per Larva (μg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microzooplankton Prey</td>
<td></td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>25</td>
<td>61.51</td>
</tr>
<tr>
<td>24</td>
<td>100</td>
<td>225.80</td>
</tr>
<tr>
<td>24</td>
<td>1000</td>
<td>1388.64</td>
</tr>
<tr>
<td>28</td>
<td>25</td>
<td>811.73</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>1431.64</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>4949.11</td>
</tr>
<tr>
<td>32</td>
<td>25</td>
<td>564.26</td>
</tr>
<tr>
<td>32</td>
<td>100</td>
<td>2345.24</td>
</tr>
<tr>
<td>32</td>
<td>1000</td>
<td>8914.37</td>
</tr>
<tr>
<td>Rotifer Prey</td>
<td></td>
<td></td>
</tr>
<tr>
<td>28</td>
<td>25</td>
<td>234.84</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>993.39</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>3524.66</td>
</tr>
</tbody>
</table>
Table 3. Estimated growth of spotted seatrout larvae from 2 to 12 days after hatching.

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>Prey Concentration (Numbers per liter)</th>
<th>Estimated dry weight at 2 days after hatching (µg)</th>
<th>Estimated dry weight at 12 days after hatching (µg)</th>
<th>Estimated weight gain for 11 days (µg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>MICROZOOPLANKTON PREY</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>25</td>
<td>9.83</td>
<td>49.44</td>
<td>39.61</td>
</tr>
<tr>
<td>24</td>
<td>100</td>
<td>7.76</td>
<td>153.51</td>
<td>145.75</td>
</tr>
<tr>
<td>24</td>
<td>1000</td>
<td>8.81</td>
<td>956.20</td>
<td>947.39</td>
</tr>
<tr>
<td>28</td>
<td>25</td>
<td>10.44</td>
<td>425.06</td>
<td>414.61</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>12.25</td>
<td>449.65</td>
<td>437.41</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>18.56</td>
<td>3603.82</td>
<td>3585.26</td>
</tr>
<tr>
<td>32</td>
<td>25</td>
<td>10.33</td>
<td>115.04</td>
<td>104.71</td>
</tr>
<tr>
<td>32</td>
<td>100</td>
<td>14.51</td>
<td>1008.52</td>
<td>994.01</td>
</tr>
<tr>
<td>32</td>
<td>1000</td>
<td>24.32</td>
<td>7142.44</td>
<td>7118.12</td>
</tr>
<tr>
<td><strong>ROTIFER PREY</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>28</td>
<td>25</td>
<td>12.26</td>
<td>237.43</td>
<td>225.18</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>14.14</td>
<td>305.77</td>
<td>291.63</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>18.66</td>
<td>600.47</td>
<td>581.81</td>
</tr>
</tbody>
</table>

Table 4. Summary of gross growth efficiency estimates (K₁) for larval spotted seatrout reared in the laboratory. The estimates were calculated from the predicted cumulative 11-day ration (Table 2) and predicted larval dry weights (Table 3).

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>Prey Concentration (Numbers per liter)</th>
<th>Estimated K₁</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>--------------</td>
</tr>
<tr>
<td><strong>Microzooplankton Prey</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>25</td>
<td>0.64</td>
</tr>
<tr>
<td>24</td>
<td>100</td>
<td>0.65</td>
</tr>
<tr>
<td>24</td>
<td>1000</td>
<td>0.68</td>
</tr>
<tr>
<td>28</td>
<td>25</td>
<td>0.51</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>0.31</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>0.72</td>
</tr>
<tr>
<td>32</td>
<td>25</td>
<td>0.19</td>
</tr>
<tr>
<td>32</td>
<td>100</td>
<td>0.35</td>
</tr>
<tr>
<td>32</td>
<td>1000</td>
<td>0.80</td>
</tr>
<tr>
<td><strong>Rotifer Prey</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>28</td>
<td>25</td>
<td>0.96</td>
</tr>
<tr>
<td>28</td>
<td>100</td>
<td>0.29</td>
</tr>
<tr>
<td>28</td>
<td>1000</td>
<td>0.17</td>
</tr>
</tbody>
</table>
low prey density (25 per liter), suggests poor food utili-
zation under those conditions. But, as prey levels in-
creased at 32°, the gross growth efficiency of larvae in-
creased markedly.

Some of the $K_i$ estimates summarized in Table 4 were
considerably higher than expected. Estimated values sum-
murized by Conover (1978) and Brett and Groves (1979) for
individual consumption of macrozooplankton by
larvae. The expected $K_i$ values were observed to range from
20 to 50% (Conover 1978). One possible cause of my high $K_i$
estimates was that I have underestimated the true con-
sumption rate. It is also possible that some prey physi-
ology was the selection for larval-than-average prey
size. The mean cumulative ration ingested by a larva from
2 to 12 days after hatching was estimated based on the mean weights of zooplankton and
rodders in the 53 $\mu$m and 120 $\mu$m mesh sizes. Hau-
d (unpublished data) estimated one standard deviation
of 0.10 $\mu$g for the 0.15 $\mu$g mean weight of 53 $\mu$m-size
zooplankton prey fractions, and one standard deviation
to be 0.14 $\mu$g for the 0.51 $\mu$g mean weight of 110 $\mu$m-size zo-
oplankton. If spotted seatrout larvae consumed prey, on
average, one standard deviation larger in weight than the
mean prey weight, then the $K_i$ estimates would be decreased considerably. Values calculated for
ration models based on prey weights of 0.25 $\mu$g and 0.65 $\mu$g for the 53 $\mu$m and 100 $\mu$m-size prey fractions, respectively. These new ration estimates were used to recalculate the cumulative ingested
ration, which was used to recalculate $K_i$. Estimates of $K_i$
were decreased about 30 to 40% from the estimates based only on mean prey weights. This suggests that additional
studies of prey weights, prey selectivity and prey size
preference as obtained from stomach analyses of larval
spotted seatrout will be very valuable for meta-biological
and biogenic studies. My $K_i$ estimates are the best avail-
able estimates of spotted seatrout larvae. Re-finements in
technique and experimental design, e.g., testing an indi-
vidual larva and determining prey-size selectivity with
age, will produce more accurate $K_i$ estimates.

Gross growth efficiency may be the simplest and most
meaningful indicator of an adequate diet, ration level,
state of health, and suitability of an artificial environ-
ment for a fish (Brett and Groves 1979). Highest gross
growth efficiencies (65 to 80%) occur during yolk utiliz-
ation by embryos; however, high values in caloric equiva-
Ies have been reported for post-embryonic stages, ranging
from 50 to 60% (Hatanaka and Takahashi 1956, Brett and
Groves 1979). Growth efficiencies estimated for spotted
seatrout larvae tended to be high at the 3 temperatures
and 3 prey concentrations tested for a zooplankton diet,
and ranged from 30 to 25 per liter for the rot-
ter diet. The estimated values may not be absolutely
accurate, but trends are apparent.

The test prey levels of 25, 100, 1,000 per liter ranged from the low to the high concentrations of micro-
zooplankton in coastal waters reported by Hau-
d and Taniguchi (1979). Prey density provides a measure of prey available for capture by larvae, but it does not directly
provide a measure of energy available for growth. Spotted
seatrout larvae raised at 24° evidently cannot benefit
from increasing prey concentrations. The smallest larvae
were observed in the 24° experiments (Table 3). It is
possible that their metabolism was so reduced that larvae
tested at the 25 per liter prey concentration were ingest-
ing and assimilating near as many prey as the larvae
tested at 1,000 per liter prey concentration. It appears that
gross growth efficiency was strongly influenced by
the digestion and assimilation rates at the low temper-
ature. The quantity of prey captured was low and, due
to the inverse relationship between number of prey captured and the degree of digestion of ingested prey, the growth
efficiencies were uniformly high. Thus, the average esti-
mated $K_i$ values have stayed high in the range of 64 to
68% due to the decreased metabolism of larvae in these experiments. Spotted seatrout larvae consuming micro-
zooplankton at 28° appeared adept at utilizing their
ingested ration. A high of 51% reflects the good feeding
ability of larvae surviving the low 25 per liter prey con-
centration. During the 11-day period larvae with marginal
feeding abilities probably died in the rearing tank,
and the surviving larvae used in the 25 prey per liter
feeding rate experiments were probably the exceptional
ones able to effectively feed at low prey levels. The re-
Discoveries of the 32° experiments supported this view; there
was no clear difference between to well predicted 42% for prey concentrations of 100 and 1,000 per liter (Table
4). The low $K_i$ of 19% for the 32°, 25 liter prey level
provided high metabolism at the elevated temperature.
Metabolic demands might increase significantly at high temperatures and lead prey to be available for
growth. Only at prey levels approaching 100 per liter
might the 32° growth efficiency increase above those effici-
ciences observed at 28° (Table 4). At the 100 per liter
prey level spotted seatrout at 32° may encounter suffi-
cient food for maintenance and growth and reflect this in
the growth efficiency estimates.

The rotter diet produced a decreasing trend in $K_i$
values as prey concentration at 28°. The very high $K_i$
of 96% observed at the 25 per liter prey con-
centration is perplexing. But, the decreasing $K_i$
trend can be explained by the inability of spotted seatrout lar-
va to consume larger quantities of rotter to equal the
ration that could be consumed by larvae offered zoonplak-
ton at the same temperature and prey level.

Juveniles and adults may survive long periods of time
when provided with a maintenance ration, but the mainte-
ance ration concept appears not to exist for spotted seatrout lar-
vae must grow to survive. As a larva develops, behavior-
ally and physically, its capabilities for capturing, han-
dling and ingesting larger prey increase. If larger prey are
available, more energy is expended per food item in
searching and capture, as reflected in the rotter
experiments. I examined some effects of prey concentra-
tion and ration on $K_i$ of spotted seatrout larvae. In
the subtricals critical prey concentrations for a $K_i$
value may change and not be constant for spotted seatrout larvae, the prey requirement would change with the temperature.

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BELLE W. BARUCH INSTITUTE FOR MARINE BIOLOGY AND COASTAL RESEARCH, UNIVERSITY OF SOUTH CAROLINA, COLUMBIA, SOUTH CAROLINA 29208.
A technique is presented which simplifies the preparation of small otoliths for examination in the scanning electron microscope (SEM). Otoliths or entire larvae may be infiltrated with a low viscosity embedding medium, sectioned, sanded and polished easily. The polished surface is then etched to bring out the increment pattern which can be observed by SEM. The materials needed for this technique are inexpensive and readily available.

INTRODUCTION

In recent years several studies have been published that demonstrated the presence of daily growth increments in otoliths (Pannella 1971, 1974; Brothers et al. 1976; Struhskar and Uchiyama 1976; Taubert and Coble 1977; Bannerman 1979; Schmidt and Fabrizio 1980; Wild and Foreman 1980; Radtke and Dean in press). Both light microscopy and scanning electron microscopy have been used to observe these increments.

The light microscope has the advantage of requiring little otolith preparation, that is, simply cover dried otoliths with immersion oil (Brothers et al. 1976). However, this method has limitations when either large otoliths or increments near the edge of the otolith need to be observed. Large otoliths can be viewed with the light microscope by grinding and polishing them until a thin section is obtained. However, due to the structural array of the otolith crystals, optical effects can lead to inaccurate counts or make observation of increments near the edge difficult. Use of a scanning electron microscope (SEM) is often necessary to avoid these difficulties. The main advantage of the SEM is that extremely small increments can be resolved easily when otoliths are properly prepared. This paper presents methods for mounting, sectioning, sanding, polishing and decalcifying otoliths. These techniques consistently provide good SEM preparations in the least amount of time.

METHODS

Bluegills (Lepomis macrochirus about 20 mm SL) were used to show differences among decalcifying procedures because the increments of juveniles are distinct. The fish were reared from the edge of a small pond and preserved immediately in 95 percent ethanol. The otoliths were removed later, dried and stored in 300B Multivet™ tissue culture trays (Falcon Division, Becton, Dickinson and Co., Oxnard, CA).

Otoliths were embedded in the hard formula low viscosity embedding medium reported by Spurr (1969). Spurr embedding kits are available from several electron microscopy supply houses.

Flat embedding molds (Pelco 20 cavity, Ted Pella Inc., Tustin, CA., see Fig. 1) were used to mount otoliths. Prior to placing the otoliths in the mold, the cavities were partially filled with liquid Spurr. The resin was polymerized at 70°C for 4 hours or until it was highly viscous. This allowed the otoliths to be properly oriented in the cavities and prevented them from sinking to the bottom of the mold. Each otolith was placed in the partially polymerized Spurr so that a line from the center of the core through the point of the rostrum was parallel to the long axis of the mold cavity (Fig. 2). The mold cavities were then filled with resin and polymerized for 24 hours at 70°C.

The hardened blocks containing the otoliths were cut with an Isomet saw (Buehler Ltd., Evanston, Ill.) to obtain flat sections. The sections were flattened "boxes" measuring 5 mm x 5 mm x 1-3 mm. The last dimension depended on the size of the embedded otolith.

\[\text{Figure 1. Mold used for embedding otoliths and larvae. Each cavity is 5.0 mm wide and deep, 14.0 mm long. Vertical bar represents 5.0 cm.}\]

\[\text{Figure 2. Line drawing showing the orientation of an embedded otolith. Dashed line is the long axis of the otolith, dotted lines indicate where cuts were made to obtain flat sections. A = anterior; P = posterior; D = dorsal; V = ventral; R = rostrum; C = core. The size of the otolith is exaggerated for clarification.}\]

The sections were made by cutting close to the otolith on each side (Fig. 2). The Isomet saw is a convenient tool because it makes a very narrow cut with precise control, however, it is not a necessity for otolith work. We have made satisfactory cuts with a hand coping saw (12.6 teeth per cm). The cuts were made parallel to the transverse plane of the otolith so that the final polished surface yielded a transverse section (see Pannella 1974, 1980; and Taubert and Coble 1977, for diagrams of the otolith planes).

Because the sections were flat, they were easy to

\[\text{1Use of trade name does not imply endorsement by the University of South Carolina.}\]
sand by rubbing against wet 600 grit wet-or-dry sandpaper with a circular motion. For each section, when the core was near the surface of the sanded side, that side was polished against a piece of Microcloth® (Buehler LTD., Evanston, [Ill.] containing 0.3 micron alumina polishing compound (Fisher Scientific Co., Fair Lawn, NJ). The section was then rinsed with distilled water and placed on a microscope slide with the polished surface up. Viewed with a light microscope the primordium appears as a dark spot in most L. macrochirus otoliths (Fig. 3). If the primordium was at the surface, the section was cleaned with distilled water in an ultrasonic cleaner. If the primordium was still below the surface, the section was sanded and polished again as needed.

Figure 3. A. Photomicrograph of a sanded and polished L. macrochirus otolith. Horizontal bar represents 100 microns. B. Enlargement of core in A showing primordium (Pr). Horizontal bar represents 20 microns.

After cleaning, the samples were decalcified in one of the following solutions: pH 3.0, HCl for 1, 2, 3, 4, 5 or 10 minutes; 5% disodium ethylenediaminetetraacetate (EDTA); adjusted to pH 7.5 with KOH, for 1, 2, 3, 4 or 5 minutes; 2% aqueous glutaraldehyde (GA) and sucrose (500 mOsm) buffered with 0.1 n sodium cacodylate to pH 7.6 for 1, 2, 3, 4 or 5 hours. After decalcification each sample was rinsed in distilled water and dried. One section was not decalcified to determine the quality of polish.

Warmouths (Lepomis gulosus) were used to show how this technique can be applied to larval fish work. During the summer of 1980 mature warmouths were collected from a pond and stripped in the laboratory. Fertilized eggs were incubated at 25°C in a 10 liter aquarium with a photoperiod of 16 hours light and 8 hours dark per day. Larvae were preserved in 95% ethanol 9 days after hatching (approximately 7 mm SL). The larvae were dehydrated for 1 hour in three changes of 100% ethanol. They were infiltrated with resin for 30 minutes in three changes of a mixture of 50% Spurr and 50% ethanol. The remaining alcohol was removed by soaking the larvae in Spurr for 3 hours with 3 changes. The infiltrated larvae were placed in the molds with their long axes parallel to the long axes of the mold cavities. The samples were polymerized, sectioned, sanded and polished (as above). EDTA (5%, pH 7.5) was used for 3 minutes to decalcify the larval otoliths.

All samples were attached to metal stubs and gold coated (100 Å). The specimens were examined in a JEOL JSM 35 or a JEOL JSM U3 scanning electron microscope operated at either 15 or 25 kV.

RESULTS AND DISCUSSION

Hardened Spurr is light yellow and transparent, so that the embedded otolith can be examined at low magnification and progress of sanding can be observed. Figure 4 shows an otolith embedded in the plastic resin and some increments can be seen near the posterior edge. Figure 5 shows an otolith that has been partially sanded, demonstrating that the core can be seen while sanding. Both photographs were made with a dissecting microscope looking through the top surface of the block. The primordium of the otolith must be at the surface after polishing, so frequent inspections are necessary to prevent oversanding.

Figure 4. Photomicrograph of an embedded L. macrochirus otolith. Note the increments near the posterior edge. P = posterior. Horizontal bar represents 0.5 mm.

Larvae preserved in ethanol are opaque and their internal features cannot be distinguished. The infiltration process clears the larvae, making their otoliths easy to observe (Fig. 6). Since otoliths of some larvae are very small (e.g. L. gulosus sagittae are approximately 10 microns in diameter 1 day after hatching), their dissection and manipulation is difficult. Also, by orienting the larvae with the otoliths in situ one can consistently section the otoliths in the same plane. Figure 7 shows the otolith of a 9-day old L. gulosus prepared in this manner. Notice that the increments were not distinct. Along with Radtke and Dean (in press) we observed that the increments of fish raised in the laboratory are not as distinct as specimens collected in the field. The undecalcified polished surface of an embedded otolith gives little information (Fig. 8). With proper decalcification, however (Figs. 9-12), the increment pattern can be seen. Previous investigators (Pannella 1974, 1980; and Brothers et al. 1976) have used 1% aqueous
ANNUAL indicated stages our his embedded South collect was 9 are time of gene 11) 3). to accurate Combined case and review- sanding that LARVAL shorter Consortium minutes. M. each fish microns. on represents NSF resin more South varies polishing This We the the section. electron and otolith gene of Horizontal the GA, was thank the HCI hour minutes. The use thank us the decalcifying HCI to etch specimens for the SEM. Preliminary investigation in our laboratory indicated that 1% HCl is far too strong and always resulted in a less distinct etch than weaker solutions. Figure 9 shows an otolith etched in pH 3.0 HCl for 4 minutes. The increments are fairly clear. Figure 10 is from the same region of a different otolith etched with HCl for 10 minutes. The increments are less distinct than those of the otolith etched for 4 minutes. An etch time of 4 minutes was the optimum for increment resolution when using HCl (pH 3). Longer or shorter periods of etching produced areas of the otolith which did not show increments.

Both GA and EDTA resulted in more distinct increments than HCl. We compared 4 min. EDTA (Fig. 11) and 3 hour GA (Fig. 12) decalcification to the HCl treatments (Fig. 9 and 10). Notice that in each case the increments were more distinct than the HCl etches. The major difference between EDTA and GA was the time factor. GA was effective with 3 to 5 hours decalcification while EDTA was useful in the range of 1 to 6 minutes.

The best decalcifying solution (EDTA, GA, or HCl) and optimal time varies with species, and even among developmental stages within a species. Therefore, we recommend that each decalcifying solution be tested over a range of time to determine the optimal decalcification time for each species.

By embedding otoliths in Spurr epoxy resin they can be properly oriented and sectioned. This makes sanding and polishing easier and more reliable. Combined with the proper decalcifying technique, this provides more accurate interpretation of otoliths examined in the scanning electron microscope.

ACKNOWLEDGEMENTS

The authors would like to thank Gene Sausser for allowing us to collect fish in his pond. Thanks also to Dave Chestnut and Ray Peeples for help with fieldwork and to Keith Taniguchi and Bobby Ireland for critically reviewing the manuscript. Special thanks to N. Watabe and D. Dunkelberger for technical help on the SEM and to the University of South Carolina E. M. Center and the University of South Carolina School of Medicine for use of their SEM's. This work was supported by the National Park Service Contract No. CX5280-1-0056, the South Carolina Sea Grant Consortium and NSF Grant No. INT76-17742 to Drs. N. Watabe and J. M. Dean.
Figure 9. Scanning electron micrograph of a L. macrochirus otolith. Etched in pH 3.0 HCl for 4 minutes. Horizontal bar represents 10 microns.

Figure 10. Scanning electron micrograph of a L. macrochirus otolith etched in pH 3.0 HCl for 10 minutes. The increments can be seen but are more distinct in Fig. 9. Horizontal bar represents 10 microns.

Figure 11. Scanning electron micrograph of a L. macrochirus otolith decalcified in EDTA for 4 minutes. The increments are distinct, more than in either of the acid etched specimens. Horizontal bar represents 10 microns.

Figure 12. Scanning electron micrograph of a L. macrochirus otolith decalcified in GA for 3 hours. The increments are very distinct, more than Fig. 9-11. Horizontal bar represents 10 microns.

REFERENCES


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DECLINE AND CESSION IN FALL FEEDING OF 0 AND 1-YEAR-OLD Lepomis gibbosus in CENTRAL ONTARIO

Nicholas Reid and P.M. Powles

ABSTRACT

Feeding of young Lepomis gibbosus (3-7 cm. TL, aged 0 and 1) from Dam Creek (Peterborough, central Ontario) declined perceptibly from late September to early December, 1978. Feeding periodicity was determined by measuring the volume of stomach contents over time. Irregular and temporary rises in feeding were associated with ascents, plateaus, or even slight decreases in the environmental temperature pattern. Young pumpkinseed stopped feeding at a lower water temperature (2°C.) than has been reported for adults of this species in Ontario.

INTRODUCTION

Lepomis gibbosus (Linnaeus), is an omnivorous feeder, taking a variety of food at all depths in the water column (Keast and Webb, 1966). It is one of the most abundant fishes in warmwater ponds, lakes, and streams in east-central North America, and is an important prey of most predatory fishes (Scott and Crossman, 1973). The only detailed food study of pumpkinseed sunfish food habits describes feeding in spring during rising temperatures (Keast, 1978). There are virtually no data on feeding during falling temperatures. Therefore, our information fills the gap on when feeding of “warmwater” species stops in fall or early winter.

To our knowledge, this study also represents the first quantitative documentation of declining fall feeding in a stream population of very young Lepomis gibbosus. To monitor the very small changes in amounts of prey consumed, we measured the volumes in capillary tubes. It is hoped that this method will be refined and applied by other workers.

MATERIALS AND METHODS

Samples of Lepomis gibbosus were seined between 1300 and 1500 hrs at two locations in Dam Creek, one mile south of Trent University's campus, twice per week from September 21 to December 14, 1978. Five pumpkinseed (under 7 cm. TL) were randomly selected from each sample, suffocated, and preserved in 5% formalin in separate vials after slititng the abdomen. Four Surber samples were taken on the same day from separate locations across the stream, but slightly upriver from the seining area, and were also fixed immediately in 5% formalin.

The fall diet feeding pattern was determined from 120 fish seined at 0100, 0700, 1100, 1500 and 1900 hrs on October 14 and 19, and preserved as previously described. Total lengths (TL) in cm. were recorded, and a scale sample removed to check that only fish of ages 0 and 1 (mean length-range of 3.1 to 6.3 cm.) were included in the study. None of the seined fish appeared to regurgitate their food, although this is a potential problem in food studies (Desselle et al., 1978). Of the 120 fish seined for diet periodicity studies, 60, (or 6 per time interval) were subsampled for food.

Only food from the stomach and not the intestine, was examined. Stomach content volumes were measured as follows: 1) for 0+ or fish under 4 cm. TL we used blood capillary tubes 100 mm. long (I.D. of 1.6 mm., Kimble Products, Kimax - 51, U.S.A.), in which one end was expanded (Fig. 1) to facilitate introduction of organisms. A plunger (stainless steel pin, with head slightly ground down) weighted with a 1 gm. piece of modelling clay, was inserted and dropped on the food column to remove bubbles. 2) for larger fish, we used a similar method but substituted common laboratory glass tubing of 3.6 mm. I.D. The height of the food column was measured and converted to mm. Only single volume readings were made because successive values were consistently 4-10% lower than the original. This resulted from compaction and deformation of the organisms during handling, and reduced the precision of the volume estimates. Water temperatures were measured with a recording thermometer (W. Lambrecht # 25F, two-probe) at 1500 hrs on each sampling date.

The contribution of each prey species to total stomach volume was estimated by the Swinerton and Worthington (1940) and Ilyes (1970) points method; i.e., one large organism may be equivalent to several smaller ones. The most common taxa were assigned an arbitrary value of one, and the contribution of larger or smaller food items to the total was estimated as a fraction or multiple of one (1.0). Four Surber samples from each date of fish sampling were combined in one jar to obtain an index of prey availability. Samples were then analysed and the organism

Fig. 1. Capillary tube and stainless steel plunger (pin) used to measure volume of food in stomachs of postlarval and young Lepomis gibbosus. (I.D. = internal diameter; E.D. = external diameter)

1. Lasenby used horizontal capillary tubes, and measured the change in the meniscuses of water after introduction of organisms.
identified to the lowest taxon possible. Forage ratios were computed as follows:

$$FR_i = \frac{r_i}{p_i}$$

where $r_i$ = proportion of prey $i$ in the diet, and $p_i$ = proportion of prey $i$ in the environment (Surber samples).

RESULTS

Feeding, as measured by stomach content volumes, gradually declined as temperatures fell from 10 to 2°C during the time period 23 September to 17 December (Fig. 2). Some apparent increases in feeding occurred which were not associated with increasing temperature. Of 7 apparent increases in feeding, only 3 were directly associated with temperature increases (October 14, 22, and November 5). The other 4 increases (September 27, October 5 and 29, and November 27) were correlated with a slowing down of the overall rate of temperature decline. Feeding ceased at 2°C in early December at the commencement of ice-up. The lowest temperature at which small Lepomis fed was also 2°C (December 13). On two occasions after feeding had apparently ceased (November 1 and 15) a slight temperature increase (November 5) and a reduced rate of temperature decline (November 27) induced resumption of feeding. During the first 2 weeks in December young Lepomis ceased feeding at temperatures at or below 2°C.

One feeding peak, probably climaxing around dusk (1900 hrs) appears to be descriptive of the fall feeding activity of young pumpkinseed in the study stream (Fig. 3). After sunset (1800 hrs) feeding apparently diminished until some time before dawn (0700 hrs), when 86% of the fish had empty stomachs. Thus the choice of a 1300 to 1500 hrs sampling period for fall, fell on a time of average or medium feeding intensity.

Of the four major prey organisms found in the stomachs only Chironomus pupae were relatively uncommon from 1300 to 1500 hrs (Fig. 4). Chironomus larvae, and to a lesser extent, Asellus, were very abundant by 1300 hrs in the stomachs. After October 25, the food of L. gibbosus was composed mainly of Asellus, Chironomus pupae, Simulium larvae and "other" (or non-identifiable) species, with a proportional reduction in Chironomus larvae from October 9 to early December (Fig. 5).

On 3 occasions, the forage ratios of Tanypus,
Annelus and Chironomus pupae were high: October 19, 29, and November 5 (Table 1). This indicates that selective foraging was occasionally carried out for particular organisms at certain times. Data from Surber samples (Table 2) showed that even by December 7 there was still an abundance of Chironomus larvae, Hydroesycy and nematodes. Thus, declining feeding did not result from reduced availability of preferred prey.

Table 1. Forage ratios of selection of major prey species from the environment by young (0 and 1-yr-old) Lepomis gibbosus in Dam Creek, central Ontario, (near Peterborough). 1978. (L) = larvee; (P) = pupae.

<table>
<thead>
<tr>
<th></th>
<th>Chironomus (L)</th>
<th>Simulium (L)</th>
<th>Annelus (L)</th>
<th>Tanytarsus (L)</th>
<th>Gammarus (L)</th>
<th>Chironomus (P)</th>
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Table 2. Relative abundance of dominant invertebrates (per cent by number of total) in Dam Creek, Peterborough, Ontario, September to December, 1978, determined by Surber sampler. (L) = larvae; (P) = pupae.

<table>
<thead>
<tr>
<th></th>
<th>Chironomus (L)</th>
<th>Odonata</th>
<th>Ephemeroptera</th>
<th>Simulium (L)</th>
<th>Hydropsychid</th>
<th>Asellus (L)</th>
<th>Pcoepodida</th>
<th>Ostracoda</th>
<th>Gammarus (L)</th>
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**DISCUSSION**

If stomach volumes can be used to assess extent of feeding, then our study shows that young *Lepomis gibbosus* ceased feeding at fall temperatures of 2°C. A decline in feeding was first noticeable in mid-September, immediately after a drop from 18 to 15°C. This decrease in feeding closely followed the declining temperature regime throughout the fall and early winter, with perhaps a few irregularities or inconsistencies. This decline was not associated with reduced availability of prey or seasonal or diel foraging changes by the fish themselves.

Feeding activity of young *L. gibbosus* in the fall usually peaked once during daylight hours. This maximum volume of food occurred at dusk. Keast and Walsh (1989) report that in summer, adult pumpkinseeds display two peaks in feeding activity, with a maximum at 1800 hrs. This is quite close to the single peak in feeding at 1900 hrs during the fall that we witnessed. Perhaps a slower stomach evacuation in fall (at lower temperatures) may be the reason for the single peak obtained in our study.

In general, the quantity of food consumed over the fall period declined in association with decreasing temperature without drastic changes in types of prey eaten. Hathaway (1927) reported that feeding of *L. gibbosus* drops off quickly with decreased temperature. Keast (1968) found that large, > 15 cm. *L. gibbosus* did not feed below 6.5°C, and that at this temperature the stomachs were blocked with mucus. Decline in metabolic rate and in size of the daily ration as fish increase in size has been noted before (Seaburg and Moore, 1964). Therefore, young pumpkinseeds from Dam Creek may have fed at lower temperature (2°C) to sustain a comparatively higher metabolic rate through the early part of the winter. Although the larger fish in Keast's (1968) study were able to over-winter without late fall-feeding, the smaller, young-of-the-year fish could not.

It is still possible that if this study had been continued under the ice, occasional feeding might have been observed, as reported by Whitaker (1977) for some stream cyprinids. This study, however, suggests that in young pumpkinseeds, feeding virtually ceases (or is certainly diminished) at 2°C.

**ACKNOWLEDGMENTS**

We would like to thank Dr. D.C. Lasenby and Mr. James Hamilton for helpful suggestions and assistance throughout this study. Additionally we thank Mary Baker for her assistance in sorting, and Sigrid Hawkins for helping with identification and record keeping. The idea of using capillary tubes was first applied by Dr. Lasenby to his mysid feeding studies (in press) and is here applied to larval fish. We would like to thank Dr. Allen Keast for making helpful suggestions in improving the original manuscript.

**LITERATURE CITED**


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Ichthyoplankton were collected weekly in the upper Potomac estuary during the springs of 1974, 1975, 1976, 1977, and in 1980. In addition data were obtained on water quality, hydrology, zooplankton abundances, fish spawning stocks, and juvenile fish distributions. Larval fish food habits were also examined. This paper describes the spatial and temporal distributions of larval fishes and discusses how these distributions reduce competition for available habitat and food resources of this upper estuarine system.

INTRODUCTION

The Potomac estuary, a subsystem of the Chesapeake bay, has been utilized for fisheries exploitation as well as for transportation and sewage disposal since colonial times. Heavy industrialization has never been a major feature of the tidal Potomac, although it may be in the near future. Industrial and domestic growth potential is high on the western side of the Chesapeake bay system as water and space are abundant, and energy facilities, the third necessary ingredient for growth, are increasing. Accordingly, in 1970 a nuclear power plant was proposed at the location in the upper Potomac estuary that coincided with the known center of the Potomac striped bass spawning grounds. As part of an evaluation of the potential impact of this facility, the Maryland Department of Natural Resources, through its Power Plant Siting Program, supported extensive studies on the key economic and resident fish species of concern, the striped bass. Field studies began in 1974 and continued through 1977. Included were investigations of estuarine hydrology, striped bass spawning stock assessment as well as ichthyoplankton and juvenile stocks. Larval striped bass food habit studies were also completed in addition to routine water quality and zooplankton investigations. A number of publications have been released on various facets dealing with population dynamics of striped bass from the above efforts (Mihursky et al. in press; Polgar et al. 1976; Polgar 1977; Setzler et al. in press; Setzler-Hamilton et al. 1980a, 1981; Ulomosicz and Polgar 1980).

In 1980, as part of the NOAA MPA national program designed to understand the decline of striped bass stocks on the east coast, an opportunity was provided to undertake another ichthyoplankton study in a segment of the Potomac estuary that complemented the 1974-1977 investigations. This paper, while developed coincidental to striped bass studies, will deal primarily with other associated ichthyoplankton species found in the 1974-1977 and 1980 studies of the upper Potomac estuary.

It is generally held that the more similar two species are in size, habit, choice and morphology, the higher the probability that they will compete with each other (Hespenderden 1973). The larvae of most fish species is more similar to each other in size and morphology than are the adults of the same species. While the inexperienced observer might confuse larval naked gobies with larval striped bass, no similar confusion would occur with the adults. It would seem then that one of the greatest poten-tials for competition among fish species may occur in the larval fish community. Our purpose here is to examine the spring and early summer larval fish communities in a selected segment of the Potomac estuary and to determine where competition is most likely to occur, and to identify what mechanisms may be operating to reduce competition.

MATERIALS AND METHODS

Figure 1 shows the portion of the Potomac river estuary under investigation. The original 1974 sampling design consisted of 12 cross-stream transects separated by 6 to 8 km longitu-dinally and extending from colonial beach, Virginia, river kilometer 60, to Washington, D.C., km 176. Additional transects were sampled in 1975 but were not repeated in other years. Extreme transects were not sampled every year. Details of the sampling methodology are presented in Setzler-Hamilton et al. (1981).

From 1974 through 1977 samples were collected at stations along fixed transects shown in Fig. 1 and Table 1. During 1980 we used a stratified random sampling regime for channel stations only, with transect locations 4, 5, 6, 8, 10, 12 and 14 (Fig. 1) serving as the mid-point of each sample region. Also the
cross-river location of each station was determined randomly within the channel region with the constraint that water depth was at least 6 m. We sampled weekly or more frequently during spring for the 5 years of these studies. Starting and ending dates and transects sampled during each year are summarized in Table 1.

Table 1. Sampling sites for the period 1974 to 1980.

<table>
<thead>
<tr>
<th></th>
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<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Potomac</td>
<td>4/1</td>
<td>3/31</td>
<td>3/30</td>
<td>4/15</td>
</tr>
<tr>
<td>2</td>
<td>Potomac</td>
<td>8/19</td>
<td>7/15</td>
<td>6/23</td>
<td>6/2</td>
</tr>
<tr>
<td>3</td>
<td>Potomac</td>
<td>7/15</td>
<td>6/23</td>
<td>6/2</td>
<td></td>
</tr>
</tbody>
</table>

* Only sampled on 5 May and 12 May in 1977.

Sampling was done with 1-m, 50 µm mesh monofilament nylon plankton nets equipped with calibrated General Oceanics flowmeters, Model 2020. For the period 1974 to 1977 each channel station (greater than 3 m in depth) was sampled with a stepwise oblique tow at 3-m depth intervals; each shoal station (< 3 m in depth) was sampled with a surface tow. During 1980 we sampled each station with surface, mid-depth, bottom and oblique ichthyoplankton tows. The surface samples were collected using a side arm to tow the net in the unreviewed water to the side of the vessel. Mid-depth tows were made using a 1-m plankton net dropped through the surface layer and retrieved through it as quickly as possible to minimize contamination. Oblique tows were made as in previous years; bottom tows were made with a novel plankton sled (Uvel 1964).

Oblique tows filtered from 120 to 500 m³ during 5 years of sampling; horizontal tows sampled between 250 and 400 m². Only the oblique samples are used for the 1980 data when data were compared among years.

Samples were washed down into buckets and the contents poured through 50 µm mesh filters. These filters and the retained larvae were then placed in 10% buffered formalin. The formalin used in 1960 had rose bengal stain added to facilitate separation in the laboratory.

Water temperature, salinity and dissolved oxygen were measured at each station at 3-m depth intervals. In 1974 to 1977 light transmittance was also measured. Temperature and salinity were measured with a Beckman salimeter, Model no. 83-5. Dissolved oxygen was measured with a YSI Model 54 oxygen meter. Transmittance was measured with a Beckman Model EV 4 Envirotron turbidity meter. In 1980 Secchi disc readings were used instead. Weather and tidal conditions were also noted.

In the laboratory the ichthyoplankton were sorted and larvae were identified to the lowest practical taxon. Centrarchids, clupeids and most cyprinds were not identified below family level. For this analysis, transect mean densities of ichthyoplankton were calculated as

$$N = \frac{\sum_{i=1}^{n} N_i}{n}$$

where \(N_i\) is the number of ichthyoplankton found in m³ of water strained in the \(i\)th tow.

**RESULTS**

Table 2 lists the taxa encountered during these studies. Three taxa were abundant in all years: clupeids (Alosa spp. and Dorosoma cepedianum), white perch (Morone americana) and striped bass (M. saxatilis). Other species which were abundant in one or more years were the bay anchovy (Anchoa mitchelli), silversides (Menidia spp. and Menobras martinica), yellow perch (Perca flavescens) and naked goby (Gobiosoma inspice). Since the above taxa comprised 99% of all ichthyoplankton sampled, our detailed analysis was limited to those forms.

![Figure 2](image) Seasonal presence of major larval taxa in the Potomac River estuary. Vertical lines represent beginning and ending of sampling periods.

Spawning in fishes is often associated with temperature increases. River temperature fluctuated from year to year and is reflected in the time of larval presence (Figure 2 and Table 3). Clupeid larvae first appeared in the middle of April in 1974 and earlier in 1975 and 1976 and might have appeared earlier in 1977 and 1980 if we had begun sampling sooner. Silversides larvae appearance fluctuated from the second half of April to about the second week in June. In 1974, 1975 and 1976 striped bass larvae were first present in the last quarter of April but they were already present when collections began on 14 April 1977 and 21 April 1980 so that time of first appearance is unknown for those years. Bay anchovy larvae appeared at the beginning of June in 1974, 1975, 1977 and 1980 but late in June in 1976.
The portion of the estuary sampled was mostly the tidal freshwater section; however, salinities greater than 0.5 ppt were present at the downstream stations. The distribution of the various species was apparently related to this fact (Fig. 3). Bay anchovy and naked goby were largely collected downstream from transect 6 (river kilometer 17). Striped bass, white perch and clupeids were most abundant above transect 6 (river km 107). Yellow perch were most abundant between transect 3 (river km 80) and transect 1 (river km 130), upstream of the 1.9 ppt isohaline (Fig. 4).

There was a tendency for downstream densities of larval striped bass, white perch and clupeids to become reduced earlier than upperriver densities. Likewise, there was a tendency for naked goby larval densities to increase upstream as the season progressed. The only year when sampling occurred sufficiently late for densities of bay anchovy to be analyzed was 1974. They appeared to mimic the pattern of the naked goby that year.

Vertical distribution of ichthyoplankton during daylight hours was obtained for 1980. During 1974 diel vertical migration patterns were obtained for white perch larvae. These diel patterns are used in assisting the interpretation of the 1980 data. Fig. 5 shows that tidal phase may influence diel patterns. It is also obvious from a comparison of Figure 5, Figure 6 and Table 3 that the vertical distribution of white perch over the whole 1980 collecting season more clearly resembles the pattern where the flood tide is at highwater. Differences in densities between depth strata were tested for the three abundant species of 1980 using Student's t test. Clupeid larvae had significantly higher densities in surface collections than at mid-depth (t=2.454, df=52, p<.01) and the bottom (t=4.760, df=52, p<.01). White perch had their greatest densities at mid-depth. These collections showed significant differences with both surface collections (t=5.170, df=52, p<.01) and bottom samples (t=2.120, df=52, p<.05). Bottom samples showed significantly higher densities of white perch larvae than mid-depth surface samples (t=2.700, df=52, p<.01). Striped bass larvae occurred in significantly lower densities in the surface samples than they did in mid-depth collections (t=2.870, df=12, p<.02) or bottom samples (t=2.480, df=12, p<.05); however, differences between mid-depth and bottom stripped bass densities were insignificant (t=0.700, df=18, p>0.05).

**DISCUSSION**

Mean water temperatures and times of first occurrences of fish eggs and larvae of the species under consideration for the 5-year sampling period are summarized in Table 4. Environmental conditions associated with the spawning of these species, as surmised from the literature, are listed in Table 5. Initiation of spawning in yellow perch, white perch, striped bass, clupeids and silversides seems to be associated with temperature, whereas spawning of bay anchovies and naked gobies is apparently linked to intrusion of saline waters.

White perch eggs were collected during the first cruise in all five years, though it is obvious from mean water temperatures and the presence of white perch larvae in collections for the years 1976, 1977 and 1980 that the spawn was already underway for those years. Likewise, the striped bass spawn was underway when we began sampling in 1977 and 1980. Although striped bass eggs were first collected during the last week of March at a mean water temperature of 12° in 1976, yok-sac

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1 Although white perch eggs are adhesive and demersal and were not quantitatively sampled, general spawning trends can be determined from our data.
larvae were not found until a month later (Table 4). Densities of larval striped bass from the Potomac estuary were lower in 1976 than in 1975, 1977, and 1980 and survival to the finfold and later stages was poor (Setzler-Hamilton et al. 1981).

Peak densities of both white perch and clupeid larvae were witnessed further upstream as the spawning season progressed, which suggests a continued upstream migration of the adult spawners. Similar patterns were reported in the neighboring Patuxent estuary during 1978 and 1979 (Setzler et al. 1979; Mihursky et al. 1980).

Yellow perch larvae were collected from the beginning of sampling in 1974, 1976 and 1977, and from early April in 1976, until early to mid-May. In the Patuxent estuary yellow perch larvae were collected in fresh to oligohaline waters (maximum salinity of 5.0 ppt in 1976 and 4.4 ppt in 1979) from the first week of April to mid-June in 1976 and to mid-May in 1979 (Mihursky et al. 1980).

Although minimal spawning temperatures reported for silverside larvae range from 18-21°C (Table 5), we collected silverside larvae at water temperatures of 14 to 17°C. Either silverside spawning in the Potomac is initiated at lower temperatures than previously reported, or water temperatures along the shorelines and shallows where these fishes spawn (Martin and Drewry 1976) warmed more rapidly than the deeper waters where we collected their larvae.

The distribution of larval bay anchovies and naked gobies is related to river flow and to upstream intrusion of saline waters. In 1976 bay anchovy eggs were collected during the second week of May, their earliest appearance during the five years of sampling. The 3.9 ppt isohaline progressed above Transect 3 at river km 80, (Figure 4) by mid-May of that year, the earliest of the 3-year period (1974-1976) during which sampling extended to Transect 3. Peak spawning of bay anchovies reportedly occurs at water temperatures above 20°C (Table 5). Our data suggest that although bay anchovy eggs were taken at 13-14°C in the Patomac estuary (Table 4), survival rates were such that no larvae were collected at water
Table 3. Mean Water Temperature, °C and Time of First Occurrence of Fish Eggs and Larvae in the Potomac Estuary

<table>
<thead>
<tr>
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</thead>
<tbody>
<tr>
<td></td>
<td>Week</td>
<td>°C</td>
<td>Week</td>
<td>°C</td>
<td>Week</td>
</tr>
<tr>
<td><strong>Eggs</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yellow perch</td>
<td>1st April</td>
<td>7-8</td>
<td>end March</td>
<td>7</td>
<td>end March</td>
</tr>
<tr>
<td>White perch</td>
<td>2nd April</td>
<td>9-10</td>
<td>3rd April</td>
<td>11-12</td>
<td>end March</td>
</tr>
<tr>
<td>Striped bass</td>
<td>2nd April</td>
<td>10</td>
<td>end March</td>
<td>8</td>
<td>end March</td>
</tr>
<tr>
<td>Atherinidae spp.</td>
<td>1st May</td>
<td>19</td>
<td>4th May</td>
<td>13-14</td>
<td>2nd May</td>
</tr>
<tr>
<td>Naked Goby</td>
<td>3rd May</td>
<td>19</td>
<td>4th May</td>
<td>13-14</td>
<td>2nd May</td>
</tr>
<tr>
<td><strong>Larvae</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yellow perch</td>
<td>1st April</td>
<td>7-8^3</td>
<td>1st April</td>
<td>8-9^3</td>
<td>end March</td>
</tr>
<tr>
<td>White perch</td>
<td>2nd April</td>
<td>9-10</td>
<td>1st April</td>
<td>8-9</td>
<td>end March</td>
</tr>
<tr>
<td>Striped bass</td>
<td>4th April</td>
<td>12-14</td>
<td>4th April</td>
<td>13-14</td>
<td>4th April</td>
</tr>
<tr>
<td>Clupeidae spp.</td>
<td>2nd April</td>
<td>10</td>
<td>1st April</td>
<td>9</td>
<td>end March</td>
</tr>
<tr>
<td>Atherinidae spp.</td>
<td>1st May</td>
<td>14</td>
<td>1st May</td>
<td>15-16</td>
<td>1st May</td>
</tr>
<tr>
<td>Naked goby</td>
<td>end May</td>
<td>19-20</td>
<td>1st June</td>
<td>23-24</td>
<td>1st June</td>
</tr>
<tr>
<td>Bay Anchovy</td>
<td>4th June</td>
<td>22-23</td>
<td>1st June</td>
<td>23-24</td>
<td>3rd May</td>
</tr>
</tbody>
</table>

1. Eggs demersal, attached; not available to sampling gear.
2. Spawning already underway at start of field sampling.
3. Larvae assumed to be present prior to start of field sampling.

Temperatures below 6°C during this five year study. Similarly, in the Patuxent Estuary bay anchovy eggs were first collected in mid-May of 1975 at a water temperature of 21°C, and in mid-June of 1978 at water temperatures of 20-24°C; larvae were not collected until mid-June of both years at water temperatures of 23-24°C (Setzler et al. 1979; Mihursky et al. 1980).

Naked gobies spawn in estuarine waters, and eggs are deposited on clams and oyster shells (Fritzsche 1978). Larvae move progressively upward with the salt wedge; peak larval densities occurred at salinities of 1-5 ppt. Naked goby densities also peaked at salinities of 2-4 ppt in the Patuxent Estuary during 1978 and 1979 (Setzler et al. 1979; Mihursky et al. 1980).

Engraulid and clupeid larvae apparently feed primarily on naupliar and copepodite stages of copepods (Schumann 1965; Bernier 1959, Dewytjer and Houde, 1970, Kjelson et al. 1975, Dukhin et al. 1970, Duka 1969, June and Carlson 1971, Arthur 1977). Clupeid and engraulid larvae are collected in the upper layers of water (Ahlistrom 1959, Dovel 1971, Hempel and Weikert 1972) where nauplii and copepodites of copepods are most concentrated (Hempel and Weikert 1972, Tsyhan and Pelischuk 1969). Both Morone spp. and naked goby larvae tend to be deeper in the water column than clupeid larvae (Martin and Setzler-Hamilton 1981). The diet of striped bass larvae is determined to be primarily adult copepods and cladocerans (Muller 1978; Beaver and Mihursky 1979). As is that of white perch (Setzler-Hamilton, Mihursky, Drewry and Martin, Chesapeake Biological Laboratory, Solomons, Maryland 20688, unpublished data). The diet of naked goby larvae is unknown, but larvae of other goby species in the

Table 4. Average densities of larvae for 1980 discrete depth samples (surface, mid and bottom). The averages are based only on stations where at least one sample of the three depths contained larvae of that species. Densities are expressed as number of larvae/100m³.

<table>
<thead>
<tr>
<th></th>
<th>Surface X (Range)</th>
<th>Mid-depth X (Range)</th>
<th>Bottom X (Range)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clupeidae</td>
<td>4.79 (0-59,046)</td>
<td>2539.67 (0-32,801)</td>
<td>737.51 (0-17,311)</td>
</tr>
<tr>
<td>White perch</td>
<td>2.93 (0-3090)</td>
<td>3571.37 (0-31,460)</td>
<td>1957.21 (0-29,048)</td>
</tr>
<tr>
<td>Striped bass</td>
<td>21.58 (0-156)</td>
<td>239.53 (4-1012)</td>
<td>327.95 (4-2145)</td>
</tr>
</tbody>
</table>

Black Sea are reportedly opportunistic feeders, feeding on up to 20 species of planktonic organisms (Dukhin et al. 1976). Clupeid larvae found in the Potomac are known to tolerate brackish water (Jones et al. 1978, Dovel 1971) as can the Morone spp. (Hardy 1976, Dovel 1971). We suggest that as larvae the bay anchovy and Alosa spp. are ecological equivalents as are the naked goby and the Morone spp. The slight disparity in time and space, in their occurrence as larvae results from differences in spawning season and salinity preference, and are possible mechanisms whereby competition between.
Fig. 4. Mean water column salinity, ppt, Potomac estuary; 1974-1977.

Fig. 5. Diel depth distribution of white perch larvae on four dates in 1974. S = surface; M = mid-depth (4 m); B = bottom; E = ebb tide; F = flood tide; LS = low slack condition; HS = high slack condition.

Fig. 6. Depth composition of 1980 white perch larvae collections averaged over the whole collecting period. Station 1 is the most downstream station and 7 the most upstream. S = surface; M = mid-depth; B = bottom.

Note: 1980 collecting stations given here do not coincide with 1974-79 transect numbers, since 1980 collections were made with a stratified random sampling design.

Acknowledgments

This is Contribution No. 1237 of the Center for Environmental and Estuarine studies of the University of Maryland. The portion of work from 1974 to 1977 was supported through the Power Plant Siting Program of the State of Maryland, Grant No. P2-72-02. The 1980 portion was funded by the U.S. National Marine Fisheries Service grant No. NA00AFD0518 through the Maryland State Department of Natural Resources. Many present and past staff members of the CBL contributed to this effort but we especially wish to acknowledge Captain William Keefe and the crew of the R. V. Orion.
Table 5. Environmental conditions for spawning of abundant fish species found in the Potomac estuary.

<table>
<thead>
<tr>
<th>Species</th>
<th>Min</th>
<th>Water temp. at peak of spawning</th>
<th>Max</th>
<th>Salinity, ppt</th>
<th>Spawning season</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>Striped bass, Morone saxatilis</td>
<td>11</td>
<td>14-18</td>
<td>23</td>
<td>Freshwater to 2 ppt maximum salinity at 41 ppt</td>
<td>Beginning of April through June</td>
<td>Seilacher et al 1983</td>
</tr>
<tr>
<td>White perch, <em>Morone americana</em></td>
<td>1-2</td>
<td>11-16</td>
<td>20</td>
<td>Freshwater to 4 ppt late March to early June, eggs not all released at once, pelagic may continue for 10 to 21 days</td>
<td>Hardy, 1978</td>
<td></td>
</tr>
<tr>
<td>Yellow perch, <em>Perca flavescens</em></td>
<td>5</td>
<td>8-110 based on surface temp</td>
<td>13</td>
<td>Freshwater to 2-3 ppt end of February to April, peak: mid-March</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clupeidae spp</td>
<td>10</td>
<td>18-20</td>
<td></td>
<td>Freshwater late spring, nearly year-round</td>
<td>Jones et al 1978</td>
<td></td>
</tr>
<tr>
<td>Atlantic menhaden <em>Brevortia orbignymi</em></td>
<td>10</td>
<td>18-20</td>
<td></td>
<td>Freshwater late March-Mid April</td>
<td>Jones et al 1978</td>
<td></td>
</tr>
<tr>
<td>Atlantic sardine <em>Sardinops suckleyi</em></td>
<td>14</td>
<td>21-30</td>
<td>27</td>
<td>Freshwater to brackish water April-May</td>
<td>Jones et al 1978</td>
<td></td>
</tr>
<tr>
<td>Atlantic silverside <em>Menidia menidia</em></td>
<td>13</td>
<td>18-20</td>
<td></td>
<td>Freshwater April-June</td>
<td>Jones et al 1978</td>
<td></td>
</tr>
<tr>
<td>Atlantic silverside <em>Menidia hacklensis</em></td>
<td>6</td>
<td>18-20</td>
<td></td>
<td>Freshwater April-July</td>
<td>Jones et al 1978</td>
<td></td>
</tr>
<tr>
<td>Atlantic silverside <em>Menidia mitchelli</em></td>
<td>21</td>
<td>31-60</td>
<td>5-25</td>
<td>May to late July or early Aug</td>
<td>Martin and Brown 1978</td>
<td></td>
</tr>
<tr>
<td>Pacific silverside <em>Kramerichthys quinquennis</em></td>
<td>20</td>
<td>30-60</td>
<td></td>
<td>Tidal freshwater or brackish water May-August</td>
<td></td>
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<tr>
<td>Pacific silverside <em>Kramerichthys quinquennis</em></td>
<td>20</td>
<td>30-60</td>
<td></td>
<td>Tidal freshwater or brackish water May-August</td>
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<tr>
<td>Pacific silverside <em>Kramerichthys quinquennis</em></td>
<td>21</td>
<td>31-60</td>
<td>5-25</td>
<td>May to late July or early Aug</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pacific silverside <em>Kramerichthys quinquennis</em></td>
<td>18-25</td>
<td>32 (usually above 30)</td>
<td>Freshwater to 10 ppt May and June spawning intermittent lasting several days to several weeks</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bembidion caprae <em>Bembidion caprae</em></td>
<td>19</td>
<td>20-90</td>
<td></td>
<td>Freshwater to 10 ppt May and June spawning intermittent lasting several days to several weeks</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Roccus caroli <em>Roccus caroli</em></td>
<td>19</td>
<td>20-90</td>
<td></td>
<td>Freshwater to 10 ppt May and June spawning intermittent lasting several days to several weeks</td>
<td></td>
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<tr>
<td>Roccus caroli <em>Roccus caroli</em></td>
<td>19</td>
<td>20-90</td>
<td></td>
<td>Freshwater to 10 ppt May and June spawning intermittent lasting several days to several weeks</td>
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<td>Roccus caroli <em>Roccus caroli</em></td>
<td>19</td>
<td>20-90</td>
<td></td>
<td>Freshwater to 10 ppt May and June spawning intermittent lasting several days to several weeks</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1. Alewife spawning generally precedes blueback spawning by 3 to 4 weeks, spawning peaks separated by 2 to 3 weeks
2. Minimum spawning temperature for alewife 70°F, but 70°F survival larvae below 20°F
3. American shad spawning generally occurs at 12 to 21°F
4. No Data

LITERATURE CITED


Beaven, M. and J. A. Biving. 1979. Food and feeding habits of larval striped bass: An analysis of stomachs from 1976 Potomac estuary collections. Univ. of Maryland CIES Ref. No. 79-45-CBL, Chesapeake Biological Laboratory, Solomons, MD 20688


UNIVERSITY OF MARYLAND CENTER FOR ENVIRONMENTAL AND ESTUARINE STUDIES, CHESapeake BIoLOGICAL LABoRATORY, SOLomons, MARYLAND. 20685-0074.
ICHTHYOPLANKTON DENSITY FLUCTUATIONS IN THE LOWER SUSQUEHANNA RIVER, PENNSYLVANIA, FROM 1976 THROUGH 1980

Barbara F. Lathrop

Abstract.--Ichthyoplankton sampling was conducted in York Haven Pond, a lower Susquehanna River impoundment near Harrisburg, Pennsylvania, to study occurrence, distribution, abundance, and composition of fish larvae. Sampling was done weekly at night along shore with pushed 0.5 m nets (1976 and 1977) and later expanded to include day sampling (1979 through 1980). Seven species, members of Cyprinidae, Catostomidae, Centrarchidae, and Percidae, comprised about 80% of the catch each year. Large diel differences were found; 80 to 85% of the annual catch was taken at night. Some species, notably the Ictalurids, were taken almost exclusively at night. Significant variation in abundance due to station, date, and year effects was indicated by analysis of variance. However, cluster analyses showed station and community similarities based on species composition of the seasonal catch. Multiple stepwise regression analyses showed that day length, sunlight, and various temperature parameters were important factors in explaining density variations of ichthyoplankton populations.

INTRODUCTION

Ichthyoplankton sampling is an integral part of many environmental studies associated with power plant construction and operation. In the past decade, these studies have revealed much new information concerning ichthyoplankton populations and their dynamics. This study was part of the ecological research for the Three Mile Island Nuclear Station and is presented to contribute to the knowledge of spatial and temporal abundance and composition of ichthyoplankton populations.

STUDY AREA

York Haven Pond is a 760 hectare mainstream impoundment on the Susquehanna River in south-central Pennsylvania, 16.1 km (10 miles) southeast of Harrisburg. The impoundment was created to serve York Haven Hydroelectric Plant. The reservoir also serves the cooling water needs of Three Mile Island Nuclear Station (TMINS) and supports recreational fishing and boating.

TMINS is located on the largest of several islands in the reservoir; two intakes and a main discharge situated on the west shore of Three Mile Island (TMI utilize the center channel of the river (Fig. 1). This channel receives 24 to 30% of the total river flow (Gilbert Associates 1979). The east channel is non-flowing during periods of low river discharge (summer flows ≤ 556 m³/s or 20,000 cfs) due to greater height of the east dam. The west channel carries the remaining flow year-round.

METHODS

In 1974 and 1975, several larval fish samples revealed that peak ichthyoplankton abundance occurred after sunset (Potter and Associates 1975, 1976). Farfield sampling in 1975 showed that larvae of most species were more abundant nearshore rather than in mid-channel (Lathrop 1976). Ritson 1977). With this information, the present program was initiated in 1976.

Ichthyoplankton was sampled weekly at 14 stations (13 in 1976) throughout the reservoir from April through August. Stations were sampled in random order each date to minimize the bias of time of day (Lathrop 1979). Only night (after sunset) samples were taken in 1976 and 1977; from 1978 through 1980, day (after 0600 h) and night samples were taken about 12 hours apart each week. Replicate surface samples were taken at each station with paired 0.5 m conical plankton nets (0.5 mm mesh) fitted to square frames set off the front of a boat. The boat was powered upstream at 10 to 20 m offshore for four minutes, sampling about 200 m of shoreline. Water depth at most stations was about 1.0 m. Water volume filtered was estimated with a General Oceanics flowmeter mounted in the center of each net mouth. Samples were preserved with 20 to 25% formalin.

Larvae were identified to the lowest feasible taxon, measured, and enumerated. Pumpkinsized and bluespotted sunfish larvae were non-separable and were enumerated as one category, Lepomis gibbosus/L. macrochirus. Catch rate was expressed as number of larvae (n) per 100 m³ of water. Statistical tests were run on log-transformed densities, log₁₀[n/100 m³ + 1] to help linearize and normalize the data (Sokal and Rohlf 1969). Some analyses were limited to night samples because fewer zeros and larger numbers provided a more robust data set. Computed ratios were simply night numbers (or density) divided by day numbers (or density) (r/D). Through three- and four-factor analyses of variance (ANOVA) used to ascertain the effects of years, stations, dates, and replicates (Sokal and Rohlf 1969). The Student-Neuman-Kuels multi-range test (SNK) was applied to identify means not significantly different (Wolf 1968). The 95% confidence level (P<0.05) was utilized unless otherwise noted.

Day-night density relationships were investigated using the pooled catch data from all 14 stations and both day and night data to determine the degree of rank correlation for the day and night catches within and between the years (1976 through 1980). For the regressions, night values were used as the independent variable to predict day densities, the dependent variable. The intent was to investigate if elimination of day sampling was feasible since the night catch yielded more larvae per effort and provided a better data set for the study of annual variations in abundance. Transformed densities for the

Figure 1. Locations of Ichthyoplankton stations (0.5 m net) sampled in York Haven Pond 1976 through 1980.
14 stations were pooled (summed each date) to reflect the densities of the entire reservoir. This decreased the inherent variability caused by station differences, and aided in the assessment of general trends in the reservoir.

Stepwise multiple linear regressions (MLR) were used to investigate the relationship between certain environmental parameters (independent variables) and ichthyoplankton density (dependent variable). Spawning in fishes is known to be related to increasing temperatures and daylight (Hynes 1970, Scott and Crossman 1973). These parameters were entered into the regression as heating degree days, cumulative river degrees, day length, and incident solar radiation to try to produce numerical indexes of springtime warming trends.

Initially, 88 independent variables were tested to indicate which factors might be related to ichthyoplankton density (Lathrop 1980). Some variables were accumulated for 1, 2, 3 and 4 week intervals, or for 2 week intervals ending 1, 2, 3 or 4 weeks prior to sampling to encompass the probable spawning and hatching activities of fishes. Various base temperatures (24, 40, 50, and 68°F) were used for test for "critical" temperatures. Data collected in the field, such as in situ air and water temperature, dissolved oxygen, pH, water clarity (secchi disc), time of day and current speed were also included.

Regressions on 1980 data were refined. Only those variables of some importance in previous regressions were used and sample size was increased by summing individual station densities each date.

Percent similarity (PSC) values based on percent composition of species were computed after Whitaker and Fairbanks (1958) to show composition similarities between stations and between years. These values were then used for construction of simple Bray-Curtis ordinations (Poole 1974) to investigate the community relationships among the stations. This results in a graphical clustering of stations with similar populations.

RESULTS AND DISCUSSION

The ANOVA tests on the ichthyoplankton data always indicated significant amounts of data, station, and interaction effects (Lathrop 1978, 1979, 1980). Replicate and interaction effects were not significantly different, meaning no sampling bias was associated with an individual net. F values for station and date effects were in the order of 200 to 500 with date being the most significant effect each year. This indicated the highly variable nature of the data and that season has a large influence on ichthyoplankton populations. However, variability among stations due to different physical and habitat characteristics, and sampling dates due to differences in species spawning times and abundances, would be expected. Despite these differences, consistencies were seen in species composition, seasonal occurrence, spatial and diel distributions, and in density relationships with certain environmental parameters.

Species composition—Some 16,000 to 24,000 larvae (about 95% of total) representing 20 to 32 taxa were taken each year. Common carp (Cyprinus carpio), spottail shiner (Notropis hudsonius), spotfin shiner (N. splendens), shiner perch (Notemigonus obtusifrons), and spottail shiner (Leomis gibbosus/macrochirius) comprised over 95% of the total catch each year (Table 1). These taxa are generally the channel catfish (Ictalurus punctatus), a common adult fish in the reservoir, comprised less than 5% of the total annual catch. This was in part due to the sampling effort being concentrated nearshore; young catfish are more numerous (up to 20% of the composition) in midchannel drift (Lathrop 1976, Rittson 1977).

Table 1. Summary (n, n and percent, %) of the most common ichthyoplankton taxa taken during the day (D) and at night (N) in York Haven Pond 1976-1980. Day sampling was initiated in 1978. T denotes total day and night values.

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Spatial distribution.—Densities at the 14 stations were significantly different each year (ANOVA). Stations 1 and 8, both slack water areas by midsummer, yielded the most larvae each year; station 10 also yielded high catches (Fig. 3). SNK tests showed that these stations had significantly higher densities compared to other stations, and in most years were not significantly different from each other (Lathrop 1979, 1980). Stations 2 and 9, located near the upstream shores of islands with little upstream spawning habitat, had significantly lower densities of larvae. Station 6, adjacent to a steep bank, received mostly midchannel flows, and also yielded low numbers of larvae. All other stations were situated downstream from extensive shorelines where spawning could occur, and yielded intermediate numbers of larvae.

Although population densities among the stations were highly variable within each year, similarities were evident in species composition. Ordination of stations based on percent composition that stations 1 and 8 tended to be farthest removed from the other clusters of stations (because of low PSc values), and the west shore TMI stations were always closely clustered (highest PSc values) (Fig. 4). Stations usually clustered in agreement with their geographic location in the reservoir, i.e. west channel stations were closer to each other than to other stations, and center channel stations were clustered in a group. Stations 1 and 8 produced the highest densities of sunfish larvae, which made them unique in the graphics. Tightly grouped TMI stations were closely related in species composition probably because the flow along the west side of TMI tended to be laminar and carried the drifting larvae more or less as a unit.

Diel distribution.—The catch was notably higher at night for all major, and most other, taxa (Table 1). From 80 to 85% of the total catch each day was taken at night. Overall night/day ratios (N/D) were 5.9, 4.7, and 4.1 for 1979 through 1980, respectively. Individual collection ratios for each station each day were almost always higher at night; N/D was as high as 668. During the three years, only about 5 to 7% of the paired samples showed greater day catches. More taxa were taken in night samples each year (Table 1) and also on individual dates (Lathrop 1978, 1979, 1980). Crappies were also taken during daylight hours (Fig. 5). The most common species were best represented with higher densities in night samples. Most species comprised similar proportions, with respect to the total catch, in both day and night samples. The overall species

Figure 2. Mean density of ichthyoplankton (n/100 m², A) and mean river temperature (°C, B) recorded in York Haven Pond, 1976 through 1980. Dashed line (1978 through 1980) is day densities. The 20° C line is drawn for reference. Data between 10 and 23 May 1978 were not collected due to high river flows.

Seasonal distribution.—The seasonal patterns of population densities for night samples were generally similar. Densities increased through May, and peaked between mid-May and early June, just after mean river temperature reached 20°C (Fig. 2). Densities generally declined through August, with one or two secondary peaks usually occurring in July. High river flow (≥1133 m³/s or 40,000 cfs) during the spawning season usually decreased densities or delayed the peak. Day densities generally followed the night patterns each year, but year to year trends and similarities were not as apparent in the day graphs (Fig. 2).

Only a few larvae were taken in April. The first larvae collected in the season were the white sucker (Catostomus commersoni), shield darter (Percina winkleri), and walleye (Stizostedion vitreum) (Lathrop 1978, 1979, 1980). In early May, spottail shiner, quillback, tessellated darter, and banded darter were taken. The initial density peak was comprised mostly of carp, spottail shiner, and quillback larvae. By late May and early June, the shorthead redhorse (Moxostoma macrolepidotum) and northern hog sucker (Hypentelium nigricans) were taken, as well as the first centrarchids (smallmouth bass, Micropterus dolomieu, and pumpkinseed/bluegill). By late June, the carp, quillback, and spottail shiner larvae decreased in number, and the spottail shiner, channel catfish, rock bass (Ambloplites rupestris), redbreast sunfish (Lepomis auritus), smallmouth bass, pumpkinseed/bluegill, and crappies (Pomoxis spp.) were present. The secondary peak in July was comprised mostly of pumpkinseed/bluegill larvae. Carp and the suckers were virtually absent. By August, most species declined and spottail shiner were predominant. The darters seemed to have the least defined spawning season, as they were present from May through July with most taken in June. Walleye were always most abundant early in the season (late April, early May) and absent after early June.

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Figure 3. Mean density (n/100 m³) and percent composition of ichthyoplankton taken at night at 14 stations in York Haven Pond, 1979. Stations are geographically oriented.
correlated and predictable (correlation coefficient = 0.92, 0.90, and 0.85). These values indicate that 72 to 84% of the variability in densities of day samples was explained by the night densities alone. Therefore on any given date, night densities can be used to predict day densities by fitting the variables into the equations (Table 3). The three years' data were pooled to obtain one equation which could then be used to predict day densities in the future:

\[ Y = -0.75 + 1.002x \]

where \( Y \) is the estimated daily (log) density and \( x \) is the observed night (log) density (Fig. 5). Application of this information would cut by half the sampling and laboratory effort by eliminating day sampling. On occasional dates, samples could be taken during the day, and the log-density values checked against the predicted values. Only if unusually high or low day densities are flagged by falling outside the confidence interval expected (Fig. 6) should there be need for further day sampling to determine if the populations have altered.

Limiting investigations to night sampling seems justified in this case since far fewer fish are caught during the day. In any study, sampling during optimal abundance would increase sample size and hence the reliability of the observations. Variations in freshwater ichthyoplankton abundance are known to occur seasonally, spatially, and within a 24 h period. We have found the ichthyoplankton in York Haven Pond to be most abundant nearshore at night. This is consistent with many other studies, as larval fishes have been found, generally, to be more abundant:

1) at night rather than during daylight (Buynak and Mohr 1976, 1977; Clifford 1977; Conner and Bryan 1976; Edwards et al. 1977; Faber 1967; Ferrari 1973; Gale and Mohr 1978; Geen et al. 1966; Gerlach et al. 1974; Kindschi et al. 1979; Lewis and Siler 1980; Lindsay and Northcote 1963; Molzahn 1973a, 1973b; Morrisson 1975; Netsch et al. 1971; Taber 1969; Van Den Avyle and Fox 1980; Wik and Morrisson 1974);

2) nearshore rather than offshore or in midchannel sites (Faber 1970; Gale and Mohr 1978; Gallagher and Conner 1980; Hart 1980; Kindschi et al. 1979; and Van Den Avyle and Fox 1980);

3) at the surface rather than in other depths (Gale and Mohr 1978; Kindschi et al. 1979; Lewis and Siler 1980; Van Den Avyle and Fox 1980).

Some studies have found certain species to be more abundant during the day (Cada et al. 1980; Gallagher and Conner 1980; Graser 1979; Storck et al. 1978; Taber 1969, and Tuberville 1972) or in midchannel or midwater samples (Edwards et al. 1977; Hatch 1980; Tuberville 1979; Van Den Avyle and Fox 1980). These differences in abundances may be due to differences in the environments sampled and/or differences in species-specific or age-specific behavior of the fishes.

Reasons for diel changes in abundance are not positively known but are known avoidance during daylight is considered an important factor contributing to lower daytime densities (Cada et al. 1980; Gale and Mohr 1978; Gallagher and Conner 1980; Nath and Heisey 1980; Nelson and Cole 1975; Netsch et al. 1971; Van Den Avyle and Fox 1980). Other factors may be negative phototropism, predator avoidance techniques, loss of orientation at night, and innate drifting behavior.

Table 2. Kendall tau rank correlation coefficients for the total day and night catches of ichthyoplankton at 14 stations in York Haven Pond, 1979 through 1980.

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<td>1978</td>
<td>0.681**</td>
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* Positively correlated at \( P \leq 0.05 \)
** Positively correlated at \( P \leq 0.01 \)
Environmental factors affecting density.--Stepwise multiple regressions with 58 independent variables revealed that day length and heating degree days were important in explaining variability in population density of ichthyoplankton. Other variables also entered into the model such as incident solar radiation and lagged indexes of temperature and solar radiation (Table 4). Final $R^2$ values were 0.52, 0.74, and 0.71 for 1977 through 1979 data respectively, but the equations included 5 to 11 variables. This was cumbersome and unrealistic for a workable model, but it does indicate that day length and warming trends can be identified in a relationship with density fluctuations. MLR's with day densities yielded poor $r^2$ values: 0.57 and 0.35 for 1978 and 1979; however, many of the same variables were identified (Table 4). In 1978, time of day also proved to be a significant factor with a partial correlation coefficient of $-0.14$ for 540 df, indicating that as time increased, density decreased. This substantiates the need for a randomized sampling regime. Ichthyoplankton densities had little or no linear relationship with field measured parameters.

It was interesting to note that for some species (carp, quillback, pumpkinseed/bluegill, and tessellated darter) moon phase was an important factor (Lathrop 1980).

Correlation coefficients were $-0.30$, $-0.22$, 0.12, and $-0.18$, respectively; carp, sucker and darter densities were higher during the darker phases. This may have been the result of increased net avoidance due to the available light on moonlit nights. Otherwise, these species densities were affected by most of the same variables as influenced overall densities (Lathrop 1980).

The regressions performed on 1980 night densities revealed relationships between ichthyoplankton and three environmental parameters--day length, heating degree days, and cumulative river degrees (Table 5). These three variables explained 95% of the variability in 1980 ichthyoplankton data with a correlation coefficient of 0.976. The correlation was much higher than with previous regressions, probably due to the use of one y variable for each date instead of 14. General climatological parameters that affected all stations in the same way environmentally were more related to general density trends of ichthyoplankton in the reservoir than were station parameters related to station densities. Future modeling with environmental parameters may be possible if this approach yields consistent relationships for all years. Combining similar stations may yield better models as well. In this way, predictable density variations in the ichthyoplankton could be monitored.
Table 4. Variation ($r^2$) of 1977 through 1979 day and night ichthyoplankton densities (log-transformed) explained by significant ($P < 0.05$) independent variables in stepwise multiple regressions.

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<td>Heating degree days</td>
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<td>Dissolved oxygen</td>
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<td>pH</td>
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<td></td>
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<td>Time</td>
<td>0.7256</td>
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<td></td>
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<td>Air temperature</td>
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<td></td>
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<td>Total incident radiation (4 weeks prior)</td>
<td>0.6953</td>
<td>0.4896</td>
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<td>Total incident radiation (2 weeks prior)</td>
<td>0.7191</td>
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<td></td>
<td>Air temperature</td>
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<td></td>
<td></td>
<td>Day length (2 weeks prior)</td>
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<td>0.5589</td>
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<tr>
<td></td>
<td></td>
<td>Time of sample</td>
<td>0.7536</td>
<td>0.5676</td>
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<tr>
<td></td>
<td>Night</td>
<td>Net heating/cooling degrees in river</td>
<td>0.7613</td>
<td>0.5496</td>
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<tr>
<td></td>
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<td>Heating degree days</td>
<td>0.7896</td>
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<td>Day length</td>
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<td>Flow</td>
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<td>Total incident radiation</td>
<td>0.8518</td>
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<td>Current speed</td>
<td>0.8630</td>
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<tr>
<td>1979</td>
<td>Day</td>
<td>Day length</td>
<td>0.6208</td>
<td>0.3871</td>
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<tr>
<td></td>
<td></td>
<td>River flow</td>
<td>0.6489</td>
<td>0.4206</td>
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<td></td>
<td></td>
<td>Current speed</td>
<td>0.7031</td>
<td>0.5171</td>
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<tr>
<td></td>
<td></td>
<td>Water temperature</td>
<td>0.7320</td>
<td>0.5298</td>
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<td>Total incident radiation (1 week prior)</td>
<td>0.5439</td>
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<td>Total incident radiation (2 weeks prior)</td>
<td>0.3523</td>
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<td></td>
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<td>Heating degree days (4 weeks prior)</td>
<td>0.3607</td>
<td>0.3163</td>
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<td></td>
<td>Cumulative river degrees (2 weeks prior)</td>
<td>0.5922</td>
<td>0.3507</td>
</tr>
<tr>
<td></td>
<td>Night</td>
<td>Day length</td>
<td>0.6855</td>
<td>0.4699</td>
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<td></td>
<td></td>
<td>Total incident radiation (4 weeks prior)</td>
<td>0.7141</td>
<td>0.5100</td>
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<td>Total incident radiation (3 weeks prior)</td>
<td>0.7198</td>
<td>0.5182</td>
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<td>Current speed</td>
<td>0.7239</td>
<td>0.5261</td>
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<td>Day length (1 week prior)</td>
<td>0.7310</td>
<td>0.5344</td>
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<td>Heating degree days (4 weeks prior)</td>
<td>0.8046</td>
<td>0.6474</td>
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<tr>
<td></td>
<td></td>
<td>Total incident radiation (for 2 weeks at 4 weeks prior)</td>
<td>0.8137</td>
<td>0.6822</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Heating degree days</td>
<td>0.4828</td>
<td>0.6620</td>
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<tr>
<td></td>
<td></td>
<td>Water temperature</td>
<td>0.4832</td>
<td>0.6926</td>
</tr>
<tr>
<td></td>
<td></td>
<td>River flow</td>
<td>0.6835</td>
<td>0.7031</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Total incident radiation (between sample dates)</td>
<td>0.8416</td>
<td>0.7116</td>
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</table>

Table 5. Results of the stepwise multiple regression performed on total log densities (n/100 m$^3$ + 1) of ichthyoplankton taken at night in York Haven Pond, 1980.

<table>
<thead>
<tr>
<th>VARIABLE ENTERED</th>
<th>MULTIPLE</th>
<th>R</th>
<th>R$^2$</th>
<th>F TO ENTER</th>
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</thead>
<tbody>
<tr>
<td>Day length (x1)</td>
<td>0.864</td>
<td>0.746</td>
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<tr>
<td>Heating degree days (x2)</td>
<td>0.959</td>
<td>0.919</td>
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<td>Cumulative river degrees (x3)</td>
<td>0.976</td>
<td>0.952</td>
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<tr>
<td>Regression coefficient (x1)</td>
<td>0.198</td>
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<tr>
<td>Regression coefficient (x2)</td>
<td>-0.224</td>
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<td></td>
<td></td>
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<tr>
<td>Regression coefficient (x3)</td>
<td>0.008</td>
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<tr>
<td>Intercept (a)</td>
<td>-40.811</td>
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<tr>
<td>Standard error (x1)</td>
<td>0.068</td>
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<td></td>
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</tr>
<tr>
<td>Standard error (x2)</td>
<td>0.002</td>
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<tr>
<td>Standard error (x3)</td>
<td>15</td>
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</table>

CONCLUSION

The ANOVA tests always indicated significant year to year differences in density as well as date and station differences within each year. Graphical and tabular presentations of the data attest to the inherent seasonal variation (date differences), station variation, and species abundance differences. However, overall species composition and abundance were similar each year. Moreover, similarities in species composition at individual stations were consistent throughout the years, as well as relative densities at each station. Major density peaks occurred each year when river temperature reached 70°C from mid-May to early June, and the overall density variations were similarly related to day length and temperature parameters.

Highly variable ichthyoplankton data can present problems when power plant effects are of interest. However, if underlying consistencies and trends in the data are ascertained, perhaps localized effects near the discharge or at downstream stations can be detected. It is desirable to determine the extent and type of natural variation within a date set for baseline comparisons in the future. Use of a variety of methods should help to view the data in different ways and provide indexes of population perturbations until more precise models are developed. The methods presented herein might be useful with other ichthyoplankton data. Day-night relationships may be predictable in many other studies once a two or three year data base is established. Modelling density relationships with environmental parameters may or may not be possible in...
other environments, depending upon the correlation between the major species population dynamics and various light and temperature regimes.

ACKNOWLEDGEMENTS

Appreciation is extended to the biologists at the Three Mile Island Aquatic Study, to Metropolitan Edison Company, Dr. Edward C. Ramey, President of ichthyological Associates, Inc., and George A. Nardacci, Project Leader. Drs. Guy L. Stueckel, Carl A. Silver, and Alan W. Wells provided statistical advice. Dr. Stueckel and John Homa, Jr. reviewed the manuscript. Blaine D. Snyder prepared the figures. The final manuscript was typed by Joan C. Malick.

LITERATURE CITED


ICHTHYOLOGICAL ASSOCIATES, INC., P.O. BOX 223, ETTERS, PENNSYLVANIA 17319.
TEMPORAL AND SPATIAL DISTRIBUTION OF SOME YOUNG-OF-THE-YEAR FISHES IN DEGRAY LAKE, ARKANSAS, 1975-1978

Michael R. Dewey and Thomas E. Moon

ABSTRACT

Midwater trawling in DeGray Lake and toment sampling in the reservoir discharge were used to describe annual, temporal and spatial distribution of young-of-the-year (YOY) fish from 1975 through 1978. Shad (Dorosoma spp.), crappies (Pomoxis spp.), and sunfishes (Lepomis spp.) composed 98% of the YOY fish collected. Shad reached peak abundance during the last week of May or the first week of June each year. Crappies were most abundant in mid-May in midwater trawl collections in the lake, even though discharge sampling indicated that crappies peaked in early May in 1976 and 1977 just before midwater trawl sampling was started. Sunfishes were collected from May through July and were most abundant in June. High numbers of sunfishes in 1977 and lower densities of gizzard shad (D. cepedianum) in 1977 than in 1978 were discussed in light of possible competition for food. Relative densities of the three principal groups varied from year to year, but total biomass of YOY fish (shad, crappies, and sunfishes combined) remained relatively stable.

INTRODUCTION

Knowledge of spatial and temporal variations in abundance of young-of-the-year (YOY) fishes contributes to a better understanding of population dynamics of reservoir fishes and aids in developing management procedures for enhancing recruitment of reservoir fish stocks. The distribution and abundance of YOY fish populations in reservoirs are influenced by water levels, weather conditions, larval recruitment, interspecific competition, and other factors.

Temporal and spatial distribution of YOY shad (Dorosoma spp.) in reservoirs has been documented by many investigators (Houser and Dunn 1962, Netsch et al. 1971, Houser and Netsch 1971, Edwards et al., 1977, Grosser 1979). However, the distribution of YOY of other fish species in reservoirs has received less attention. Krause and Van Den Ayle (1979) reported on the temporal and spatial distribution of larvae of principal fish species found in Center Hill Reservoir, Tennessee, and Kindach et al. (1979) described the distribution of the larval fish species found in Rough River Lake, Kentucky.

Our purpose was to describe the population dynamics of YOY fishes in DeGray Lake during 4 years of epilimnetic discharge, concentrating on the temporal and spatial distribution of shad, crappies (Pomoxis spp.) and sunfishes (Lepomis spp.).

STUDY AREA

DeGray Lake, located on the Caddo River in west central Arkansas, was impounded in 1965. At normal pool elevation of 124.4 m above sea level, the reservoir has an area of 5,427 ha, and maximum and mean depths of 57 m and 15 m. The reservoir extends in a west to northeast direction for about 32 km and has a shoreline length of 333 km.

Field data from DeGray Lake are being used to develop and test an ecosystem model developed by the Waterways Experiment Station, U. S. Army Corps of Engineers. For the assessment of spatial variation of selected biological and physical characteristics, the lake was divided into three sampling sections representing the upper, middle, and lower reaches (Figure 1). The upper section (1,230 ha) consists mainly of a narrow river channel bordered by small coves containing standing timber. The middle section (2,549 ha) includes open water areas and many large coves; two major tributaries flow into it. The lower section (1,661 ha) has large open water areas bordered by steep, rocky shorelines.

METHODS

Although preliminary reservoir discharge samples were collected in 1975, regular sampling for larvae was not begun until early April 1976. We sampled YOY fish in the reservoir discharge weekly from the first week of April through July during 1976-78. Larval fishes were collected at a point 40 m downstream from the power house discharge with a townet, 3 m long and 1 m in diameter, with 0.79 mm (1/32 in.) mesh (Moen and Dewey 1976). During 1976, sampling periods were alternated weekly between morning (0930-1130 h) and afternoon (1300-1500 h). During 1977-78, samples were taken only in the afternoon (1300-1500 h).

Midwater trawling was used to determine temporal and spatial distribution of YOY fishes in the upper, middle, and lower sections of DeGray Lake. Young fish were sampled at 2-week intervals from May through July, 1975-78. Trawl-
ing sites were selected at random from a gridded map of the lake. Four hauls were made in the upper section of the lake, and eight hauls in each of the lower two sections. All sampling was done at night, because YOY fish have been shown to be more evenly distributed and nearer the surface at night than during the day (Netsch et al. 1971).

Trawling was conducted from an aluminum boat (8.5 m long, 3.2 m beam) powered by a diesel engine and fitted with two hydraulic winches. From May through June 1975 and 1976, a 1.85 m² frame trawl (Moussri 1972) fitted with 0.5 mm mesh netting was used for sampling. When the mean length of shad approached 25 mm, a net with four panels having mesh sizes ranging from 12 to 4 mm (front to cod) was used on the same frame from the end of June through July 1975 and 1976. A sample consisted of an oblique haul from the surface to a depth of 7 m followed by retrieval. During the summer of 1977 and 1978, a 2 m² Tucker trawl with 0.50 mm mesh was used. It was lowered in a closed position to a depth of 7 m, opened, and retrieved at a 45° angle; an effective opening of 1.6 m² was maintained during retrieval. A General Oceanics flowmeter suspended in the mouth of both trawls was used to estimate the length of tows. The length of the tow was multiplied by the effective area of the opening of the net to determine volume of water sampled. Nets were retrieved at a speed of 0.9 m/s. The Tucker trawl was lowered off the starboard side of the boat, away from the boat and prop wash. A reinforced vinyl bag, 5 m long with holes punched in the bottom and attached to a metal ring 1 m in diameter was towed off the port side of the boat to offset the drag of the trawl.

Seven tests were conducted to compare the relative sampling efficiencies of the two trawls. Each test consisted of six hauls with one trawl followed immediately by six hauls with the other trawl in the same area. The Tucker trawl was found to be more efficient in sampling all taxa. Mean density estimates (fish/m³) of the Tucker trawl for each test were divided by that of the frame trawl and the resulting ratio was termed relative efficiency. A mean relative efficiency value from the seven tests was determined for each taxon and used to adjust the 1975 and 1976 data to account for the difference in sampling efficiencies.

Fish were preserved in 10% formalin and later identified and counted. We identified specimens to the lowest possible taxon, using keys developed by May and Gasaway (1975) and Hogue et al. (1976). Because we were unable to identify shad shorter than 20 mm, the data for that length range were pooled for analyses. Shad longer than 20 mm were enumerated by species, and those longer than 25 mm were considered juveniles. Sunfishes and crappies were not identified to species; specimens longer than 20 mm were considered juveniles. Catch data were reported as number/m² and as biomass for each taxon. Subsamples of all taxa were blotted, air-dried for 5 min, and weighed on a two-leading analytical balance to obtain mean weight per fish for each taxon; the resultant was multiplied by mean density to estimate biomass of each taxon. Total biomass (shad, crappies, and sunfishes combined) was calculated for each sampling date by section of the lake.

RESULTS

Twelve forms of YOY fish were collected: Gizzard shad, D. cepedianum; threadfin shad, D. ostenensis; crannies, sunfishes, Brook silverside, Labeo discus; Mississippian silverside, Periopsis audens; looners, Percina caprodes; black basses, Micropterus spp.; minnows, Notropis spp.; flathead catfish, Voeltzistia olivaris; catfishes, Ictalurus spp. and white bass, Morone chrysops. Shad, sunfishes and crappies made up more than 98% of the fish collected.

Temporal Distribution-- Larval shad were first collected in reservoir discharge samples during the second week of April in 1976 and 1978 and the third week of April in 1977. Catches of gizzard shad peaked earlier than those of threadfin shad, indicating that peak spawning of gizzard shad preceded that of threadfin shad. Gizzard shad have been reported to spawn at lower temperatures than threadfin shad (Kimsey 1958, Miller, 1960). Shad in the trawl samples were most abundant during the first weeks of June 1975, and during the last 2 weeks of May in 1976-78 (Fig. 2). A significant spawn of threadfin shad in late June or early July in 1976 resulted in large numbers of shad being present throughout the summer of that year.

Unusually cold winters during 1976-77 and 1977-78 with extended periods of water temperatures less than 7.2°C caused high mortality of adult threadfin shad, but had little apparent effect on adult gizzard shad. Threadfin shad have been found to be sensitive to low temperatures, with high mortalities usually occurring at 7.2°C (Persons and Kimsey 1984). Few YOY threadfin shad were collected during either 1977 or 1978. Young gizzard shad were more abundant in May in both of these years than in 1975 or 1976. A mean shad density of 7.05/m³ (compared almost entirely of gizzard shad) in the upper section on May 30, 1978 was the highest during the study. Nevertheless, total shad numbers was low by late June during both 1977 and 1978 because of the scarcity of threadfin shad.

Larval crappies appeared in reservoir discharge sampling during the second week of April of each year in 1976-78. Abundance in the discharge peaked in early May in 1976 and 1977, before midwater trawl sampling was started. In 1978, peak numbers of crappies occurred during mid May in both discharge and midwater trawl sampling. Densities of crappies collected by midwater trawling were considerably higher in 1977 and 1978 than in 1975 and 1976.
This difference in abundance was apparent in the collection of juvenile crappies during summer cove sampling with rotenone in 1977 and 1978 (Multi-Outlet Reservoir Studies, unpublished data). Most crappies collected after the third week in June were juveniles.

Larval sunfishes first appeared in discharge collections during the first week in May 1976, and were usually most abundant in June each year. Sunfish densities were low and relatively similar in 1975 and 1976 (Fig. 4). In 1977, catches increased markedly from mid May through June and peaked again in July. A mean density of 10.27/m³ in the upper section of the lake in June was the highest in 4 years of sampling.

Among the less abundant taxa, logperch were most numerous over the longest period of time (Fig. 5).

Spatial Distribution—Larval shad were collected in all sections of the lake during May each year—which suggests that spawning occurs lakewide. Mean densities of YOY shad were lowest in the upper section during 1975, and in the lower section during 1976. Threadfin shad densities were consistently higher in the upper area in 1975 and 1976. No consistent gradient in density by section of the lake was noted for shad during 1977 or 1978.

**Fig. 3.** Midwater trawl catches (No./m³) of young-of-the-year crappies from DeGray Lake in May, June and July in 1975-78. Dotted lines represent the upper section of the lake, dashed lines the middle section, and solid lines the lower section.

**Fig. 4.** Midwater trawl catches (No./m³) of young-of-the-year sunfishes from DeGray Lake in May, June, and July in 1975-78. Dotted lines represent the upper section of the lake, dashed lines the middle section, and solid lines the lower.

**Fig. 5.** Seasonal occurrence of minor fish species (larval and juvenile) in DeGray Lake as indicated by reservoir discharge samples in 1976-78 (solid line) and midwater trawl samples in 1975-78 (dashed line).

<table>
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<th>Taxa collected</th>
<th>Apr</th>
<th>May</th>
<th>Jun</th>
<th>Jul</th>
</tr>
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<tbody>
<tr>
<td>Minnows</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Silversides</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White bass</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Centrarchid basses</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Logperch</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Catfishes</td>
<td></td>
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</tr>
</tbody>
</table>

* No white bass were collected in the reservoir discharge.
Mean densities of YOY crappies were similar for all sections during 1975 and 1976, but were lowest in the lower section in 1975 and 1976. These differences corroborated information gathered from August rotenone sampling, which showed that populations of adult crappie were lowest in the lower areas (Multi-Outlet Reservoir Studies, unpublished data).

Mean densities of YOY sunfishes were similar throughout the lake each year except 1977 when they were consistently highest in the upper area (Fig. 4).

Few spatial differences were noted in the densities of the less abundant taxa although comparisons are vitiated by the low densities encountered.

Few differences were noted in horizontal distribution of fishes and total biomass of the abundant fishes remained relatively stable from year to year (Table 1). However, large year-to-year variations in densities were observed for shad, crappies, and sunfishes. Peak densities ranged from 3.3 to over 7 for shad, from less than 0.2 to 1.4 for crappies, and from 0.8 to 10.3 for sunfishes.

Table 1. Biomass (g/m²) of young shad, crappies, and sunfishes combined from midwater trawl collections, DeGray Lake, 1975-78.

<table>
<thead>
<tr>
<th></th>
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<tbody>
<tr>
<td>May 1</td>
<td>0.021</td>
<td>0.048</td>
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<tr>
<td>June 1</td>
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<td>0.060</td>
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<td>0.177</td>
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<tr>
<td>July 1</td>
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<td>0.187</td>
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<td>0.029</td>
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<td>July 2</td>
<td>0.076</td>
<td>0.194</td>
<td>0.053</td>
<td>0.007</td>
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</table>

g/m² = Comparable data for all four years was available for only one collection in May.

**DISCUSSION**

Initial spawning dates and seasonal abundance of YOY shad in DeGray Lake was similar to that reported by Netsch et al. (1971) for Beaver Lake, Arkansas during 1969 and 1970. Larval crappies, first collected during the second week of April from DeGray Lake, were first noted on April 30 in Rough River Lake, Kentucky (Kindschil et al. 1979), and in early May in Center Hill Reservoir, Tennessee (Krause and Van Den Avyle 1979). Collections of larval sunfishes as early as the first week of May in DeGray Lake indicate earlier spawning than that recorded by Kindschil et al. (1979) and Krause and Van Den Avyle (1979), who first collected larval sunfishes in late May. Differences in initial collection dates were probably due to differences in spring warming of reservoir waters.

Krause and Van Den Avyle (1979) noted that crappie larvae were most abundant in the middle area of Center Hill Reservoir. In DeGray Lake, mean densities of crappies were relatively high in the middle section each year. No pronounced gradient of YOY sunfish density was noted in DeGray Lake except for 1977 when mean densities were highest in the upper area.

The combined biomass of young shad, crappies, and sunfishes remained relatively stable during the 4 years of study in DeGray Lake. This stability is suggested by the availability of food, which probably was not the limiting factor in maintaining total biomass of young prey, although relative densities of individual taxa fluctuated greatly as a result of variable reproductive success and early survival.

For example, the decline in abundance of threadfin shad during the unusually cold winters of 1976-77 and 1977-78 was associated with marked decreases in abundance of young sunfishes in 1977 and gizzard shad in 1978. Densities of gizzard shad were much lower in 1977 than in 1978. These annual differences in relative abundance of different taxa may have been partially influenced by food resources, since larval and juvenile sunfishes are reported to feed on ennotomosarcans (Sutherland 1953, Applegate et al. 1966) as do larval shad (Kilambi and Barger 1975). Gerdes and McConnell (1963) and Gerdes (1964) suggested that shad competed directly with young centrarchids for food in Pena Blanca Lake, Arizona. Mayhew (1975) found YOY bluegills (Lepomis macrochirus) were competing for food with gizzard shad. We also believe that the lower densities of threadfin shad and sunfishes in 1978 resulted in an increased supply of available food and better survival of YOY gizzard shad. Kilambi and Barger (1975), in reporting that both shad species feed on ennotomosarcans and rotifers, postulated that the two species compete for the same foods.

The presence of several species of YOY fishes helped maintain stability in the prey assemblage of DeGray Lake. However, the relationships demonstrate the need to evaluate prey introductions in light of potential effects on existing species.

**LITERATURE CITED**


Larval fishes were collected weekly during two summers (1979 and 1980) with a newly designed light-trap at two littoral sites in Lac Heney, Québec. Each sample was a 60-minute set of the light-trap in water 60-100 cm deep. Fifty percent of the 24 known species in the lake were captured at littoral sites as newly hatched and older larval stages. The samples were predominated by four or five species of Cyprinidae, two species of Percidae, one Centrarchidae, one Osmeridae and one Cyprinodontidae. Weekly samples demonstrated a succession of larvae resulting from periodic or protracted spawnings and hatchings in the lake; different species predominated at different times. Numbers of specimens in the 60-minute samples varied from six to 113.

METHODS AND MATERIALS

Free-swimming larval fishes were collected with a newly designed plexiglass light-trap at two littoral sites in Lac Heney, Québec. The light-trap is described in detail in Faber (1981) but Fig. 1 shows a diagram of it. It consists of an upper light chamber, a lower animal chamber, a cylindrical section of netting, and a collection bucket. The light chamber holds the water-tight light bottle; the animal chamber has two vertical and two horizontal openings allowing animals to enter from four directions; the cylindrical section of netting joins the animal chamber to the collection bucket, which allows the operator to concentrate the catch into a small vessel. The bulb is powered by a 6-volt battery and its light intensity was measured in the lake at 2 lux at a distance of one meter (2300 hours, 18 July, 1980). The four animal entrances were set at a width of 3 mm and a length of 85 mm. Besides fish larvae, a number of other animal groups were collected including: Acarina, Amphipoda, Cladocera, Copepoda (Calanoida and Cyclopoida), insects (Coleoptera, Diptera, Ephemeroptera, Hemiptera and Trichoptera), Mysidae, Oligochaeta and Ostracoda. The fish larvae needed to be hand-separated from these other animals.

Samples of larval fishes were collected weekly in 1979 and 1980 during June, July and August within the Lab Dock Study Area described by Faber (1980). The light-trap was allowed to fish for 60 minutes sometime between 2100 and 0030 hours. Thus, 60 minutes was the "unit-of-effort" for each sample. Samples were collected in 1979 at the end of a floating wooden dock in an opening of a shoreline bed of Scirpus validus Vahl. The depth of the water varied between 60 and 100 cm and the nearest leaf of S. validus Vahl. was 2 meters away. The growth of bullrushes was inhibited in this opening by boat activity. In 1980 the samples were taken within the bed of S. validus Vahl. approximately 10 meters to the east of the wooden dock. The trap was placed approximately one meter inside the outer edge of the Scirpus bed in water varying from 60 to 80 cm in depth. To avoid excessive disturbance at the sampling site, the trap was set and retrieved from the bow of a boat. Surface water temperatures were taken by a glass bucket thermometer.

I could identify many larvae from previous exposure to them in other studies but for others I needed to follow the development from larvae to juveniles. The observation of spawning activities in the lake and collection of some adults provided additional clues. The cyprinids caused most identification problems. Golden shiner larvae were identified by the presence of an unique ventral line of melanophores coupled with a dorsal series of paired melanophores and blunt nosed minnow larvae were identified by their unique finfold morphology. The larvae of Notropis spp. could not be identified to species with certainty because two similar minnows are known to be present, i.e.,. S. heterodon and N. heterolepis.

Iowa darter, Etheostoma exile, larvae were identified by their typical perch morphology, unique pigmentation, and by studying a developmental series. Although several larval darters resemble this species, no other species of darter has been collected from Lac Heney. Yellow perch, Perca flavescens, and Iowa darter larvae are superficially similar at 6-8 mm but were separated by the following features: yellow perch possess air bladders, Iowa darters do not; yellow perch possess oblique lines of melanophores along myomeres in the posterior-lateral region, Iowa darters do not; Iowa darters possess a short vertical pigment line at the base of the pectoral fins, yellow perch do not.

All the specimens have been accessioned into the National Fish Collection of the National Museum of Natural Sciences (Accession number 1981-33).
Table 1: List of species and lengths of fish larvae taken from Lac Heny during the summers of 1979 and 1980 at two littoral sites with a light-trap.

<table>
<thead>
<tr>
<th>Family</th>
<th>Species</th>
<th>Z (mm)</th>
<th>Range (mm)</th>
<th>N</th>
<th>Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catostomidae:</td>
<td>White sucker, Catostomus commersoni (Lacépède)</td>
<td>13.6</td>
<td>13-15</td>
<td>40</td>
<td>1979</td>
</tr>
<tr>
<td></td>
<td>Rock bass, Ambloplites rupestris (Rafinesque)</td>
<td>7.0</td>
<td>-</td>
<td>1</td>
<td>1980</td>
</tr>
<tr>
<td></td>
<td>Punkinsed, Lepomis gibbosus (Linnaeus)</td>
<td>8.4</td>
<td>5-12</td>
<td>30</td>
<td>1979</td>
</tr>
<tr>
<td></td>
<td>Punkinsed, Lepomis gibbosus (Linnaeus)</td>
<td>5.7</td>
<td>4-9</td>
<td>28</td>
<td>1980</td>
</tr>
<tr>
<td>Cyprinidae:</td>
<td>Golden shiner, Notemigonus crysoleucas (Mitchill)</td>
<td>5.0</td>
<td>4-6</td>
<td>2</td>
<td>1979</td>
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<tr>
<td></td>
<td>Bluntnose minnow, Phoxinus phoxinus (Rafinesque)</td>
<td>5.5</td>
<td>5-6</td>
<td>30</td>
<td>1980</td>
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<tr>
<td></td>
<td>Bluntnose minnow, Phoxinus phoxinus (Rafinesque)</td>
<td>6.6</td>
<td>5-12</td>
<td>72</td>
<td>1979</td>
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<tr>
<td></td>
<td>Bluntnose minnow, Phoxinus phoxinus (Rafinesque)</td>
<td>6.1</td>
<td>5-7</td>
<td>31</td>
<td>1980</td>
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<tr>
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<td>Minnows, Notropis spp 1</td>
<td>5.6</td>
<td>4-12</td>
<td>183</td>
<td>1979</td>
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<tr>
<td></td>
<td>Minnows, Notropis spp 1</td>
<td>5.6</td>
<td>4-12</td>
<td>207</td>
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<tr>
<td>Cyprinodontidae:</td>
<td>Banded killifish, Fundulus diaphanus (LaSueur)</td>
<td>6.6</td>
<td>5-12</td>
<td>194</td>
<td>1979</td>
</tr>
<tr>
<td></td>
<td>Iowa darter, Etheostoma exile (Girard)</td>
<td>6.7</td>
<td>5-11</td>
<td>66</td>
<td>1980</td>
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<tr>
<td></td>
<td>Iowa darter, Etheostoma exile (Girard)</td>
<td>7.0</td>
<td>3-13</td>
<td>446</td>
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<td></td>
<td>Iowa darter, Etheostoma exile (Girard)</td>
<td>5.7</td>
<td>3-11</td>
<td>341</td>
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<td>Yellow perch, Perca flavescens (Mitchill)</td>
<td>7.1</td>
<td>5-12</td>
<td>94</td>
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<td>Yellow perch, Perca flavescens (Mitchill)</td>
<td>9.3</td>
<td>5-14</td>
<td>184</td>
<td>1980</td>
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<td></td>
<td>Walleye, Stizostedion vitreum (Mitchill)</td>
<td>11.0</td>
<td>-</td>
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<td>1970</td>
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</table>

a. Two species may be present here, Notropis heterodon (Cope) and N. heteroplos Eigenmann and Eigenmann.
b. One hundred specimens from various dates were measured.

Table 2. Temporal succession of free swimming fish larvae at two littoral sites in Lac Heny, Quebec, during 1979 and 1980. Fish larvae were collected with a light-trap set for 60 minutes between 2100 and 0630 hours. Lines below numbers indicate presence of at least one larva with yolk material. Surface temperatures were taken with a glass thermometer at night.

<table>
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<tr>
<th>Surface temperature (°C)</th>
<th>Year</th>
<th>Month of capture</th>
<th>Day of capture</th>
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</table>

| Total Numbers | 79  | 118  | 247  | 467  | 1135 | 362  | 6    | 835  | 15   | 81   | 14   | 22   | 250  | 288  | 50   | 117  | 107  | 82   | 64   | 23   | 4462 |

FABER—LARVAE CAUGHT BY LIGHT TRAP IN QUEBEC LAKE
Fig. 1. Diagram of plexiglass light-trap. Light bottle is shown outside of light chamber without battery and bulb.
Species Composition: Numerous species were collected at the two littoral sites during the two-year period. Twelve or 13 species of fish larvae within 11 genera and 6 families were collected during the months of June, July and August (Table 1). Ten larvae were identifiable to species but the genus Notropis may have included more than a single species. Those 12 or 13 species represent 50% of the total number of fishes resident in the lake. Among them, six or more species span within rocky or weedy areas, three deposit their eggs in prepared nests and two lay their eggs in tributary creeks or freshets.

The range of sizes of larvae captured by the light-trap varied considerably (Table 1) and would probably have been greater if the trap's openings had been larger. Several juvenile cyprinids, rock bass and pumpkinseeds were seen near the trap and were observed to prey upon larval fish and other invertebrates swimming around the trap. The smallest larvae were Etheostoma exile which averaged 5.7 mm and 7.0 mm over the two years. In contrast, the largest larvae, Osmerus mordax, averaged 17.8 mm and 17.5 mm but ranged up to a maximum of 25.0 mm. Most larvae were captured by the trap shortly after hatch and many specimens possessed yolk material (Table 2) except Catostomus commersoni, Stizostedion vitreum, Osmerus mordax, Notemigonus crysoleucas and Ambloplites rupestris.

The number of specimens of each species varied considerably, ranging from one to 1291 in a single season. Some variation may be explained by the following: traps were set at two different sites; some species were more abundant than others in the lake; some species hatched before June and their patterns of abundance were missed, i.e., white sucker and yellow walleye; while others remained in certain local and restricted sites, i.e., golden shiner. Illumination from the light-trap reached slightly outside the bed of沈段, so fish larvae from the deeper open water area were probably attracted and captured.

Two species not listed, (Coregonus artedi) Le Sueur, lake herring, and Micropterus dolomieu (Lacépède, smallmouth bass), were captured in other light-trap experiments. Other species known to occur in the lake but not taken by means of the light-trap include: Lake char, Salvelinus namaycush (Walbaum); lake whitefish, Coregonus clupeaformis (Mitchill); spoonhead sculpin, Cottus tilli (Nelson); deepwater sculpin, Myoxocephalus quadriocornis (Linnaeus); trout perch, Percopsis omiscomaycus (Walbaum), northern pike, Esox lucius Linnaeus; brown bullhead, Ictalurus nebulosus (LeSueur); channel catfish, Ictalurus punctatus (Rafinesque); and ninespine stickleback, Pungitius pungitius (Linnaeus).

Temporal Occurrence: A continuum of spawnings and hatchings occurs each year in Lac Heney as a result of the presence of 25 different species of fish. This light-trap study in and near a bed of Scirpus from June through August monitored a portion of this continuum. The appearance and disappearance of fish larvae in the pullurines produced a temporal succession of larvae during two summers in Lac Heney (Table 2). The patterns of temporal occurrences of fish larvae shown here were determined by a number of interacting biological and environmental factors, including: 1. species, 2. age and/or stage of development, 3. attraction to artificial lights, 4. nocturnal activity, 5. dispersal behavior, 6. presence in the area, 7. seasonal environmental changes at sampling site, 8. relative darkness of evening, 9. design of light-trap, and others.

Percy larvae appeared in increasing and then decreasing numbers during June in Lac Heney but they appeared during May, one month earlier, at similar surface temperatures in smaller lakes which warm-up faster in spring (Faber 1967; Amundrud et. al. 1974). Smelt larvae appeared and disappeared during June. Newly hatched small larvae were observed in limnetic regions during May and older larvae and juveniles were observed in June in immense numbers in certain sub-littoral regions (Faber, unpublished data). Common sucker and yellow walleye larvae were captured on only one occasion.

The second and third most abundant groups of larvae collected during the sampling interval were the Iowa darter and the minnows of the genus Notropis. Iowa darter larvae occurred throughout the period of sampling and their variable numbers did not delimit an obvious periodicity in spawning. Hareat, darters were generally considered spring spawners (Winn 1958; Brudar and Rosen 1966), but the occurrence of Iowa darter larvae with yolk from early June to early August suggests the spawning of Iowa darters in Lac Heney extends into summer. With an incubation period of 18 to 26 days (Bensley 1919), these data suggest that Iowa darters spawn continuously from mid-May through late-July, a full three months. The spawning period of the Iowa darter in Lac Heney appears unusually long compared to other known species of Etheostomatinae.

Larvae of Notropis spp., L. gibbosus and P. rotatus appeared after the middle of June when surface water temperatures were 15°C. The occurrence of small and variable numbers of pumpkinseeds and bluntnose minnow larvae suggests continuous spawning in the area. Larvae of golden shiners were only collected on three occasions, yet they were continuously present in large numbers from late July through late August 10 to 20 meters away from the two sampling sites (Faber 1960). Killifish larvae were the last to make their appearance, during early July when surface water temperatures were 20°C. Only one rock bass larva was captured.

DISCUSSION

This study demonstrates that light-traps can be used to capture larval fish in shallow weedy areas. The number of species collected with light-traps is impressive but the total number of specimens is wanting. Fifty percent of the species known to be living in Lac Heney were represented in these light-trap samples. Gallagher and Conner (1980) reported they also collected about 50% of a total of 54 species in the Lower Mississippi River with tow-nets, but suggested that certain larvae were more abundant in extralimactic areas (quiet vegetated areas) than in areas where they are able to tow. The larvae of yellow perch, smelt, Iowa darters, Notropis spp., pumpkinseeds, bluntnose minnows and killifish were dominant in the light-trap samples. Simultaneous tow-netting in offshore limnetic areas of Lac Heney only captured yellow perch, smelt, Iowa darters and pumpkinseeds (Faber, unpublished data).
Tow netting in Lake Opinicon, (Amundrud et al. 1974) resulted in the collection of large numbers of yellow perch (Perca flavescens), logperch (Percina caprodes), black crappies (Pomoxis nigromaculatus) and sunfishes (Lepomis spp.). The results from the studies in Lac Heney and Lake Opinicon suggest that a light-trap is more effective than a tow-net in capturing the larvae of Notropis spp., Bluntnose minnows and killifish, but a detailed comparison between the collections of light-traps and tow-nets should be undertaken.

Various plexiglass traps designed to catch small fishes have been described in the literature. Some traps operate without lights and rely upon leads to catch fish (Breder Jr. 1960; Werner 1968; and Casselman and Harvey 1972), while others (Paulson and Espinosa Jr. 1975 and Kindschi et al. 1979) including mine, rely upon lights to attract fish. Traps with and without lights were tested simultaneously on three occasions in Lac Heney and in each instance the trap with lights caught fish larvae while the trap without lights caught none. Paulson and Espinosa Jr. (1975) mentioned that they tested their trap during daylight hours and discovered that day sets were unsuccessful with or without lights. The main difference between their trap and mine, and the above described traps used without lights, is the presence of rectangular pieces of plexiglass used as leads. Apparently traps without lights and leads are ineffective as activity traps for larval or juvenile fish.

The success of light-traps depends upon fish larvae moving toward artificial lights. This behaviour is complex and not well understood. Some animals are attracted to artificial lights, some are unaffected, while others are repelled (Fraenkel and Gunn 1961). Moreover, the larvae and juveniles of certain fishes are attracted to artificial lights while adults are not (Verheijen 1958). Why do fish larvae or any other animals move toward artificial lights? Verheijen (1958) concluded that movements of animals towards artificial light sources are "forced movements", that is, they move toward the light because of photic disorientation rather than because of some inherent or learned behavioural activity.

ACKNOWLEDGEMENTS

I would like to thank the Department of Biology, University of Ottawa, for use of the field station and certain equipment at Lac Heney. Dr. Jack Gillett identified Scirpus validus Vahl, and Sally Gadd drew the light-trap. I would like to express my wholehearted thanks to Darin Faber for his time and patience in sorting these samples and extensive field assistance.

LITERATURE CITED


NATIONAL MUSEUM OF NATURAL SCIENCES, OTTAWA, ONTARIO, CANADA K1A OMB.
COMPARATIVE REPRODUCTIVE BIOLOGY OF THE THREADFIN AND GIZZARD SHAD IN LAKE TEXOMA, OKLAHOMA-TXAS

William L. Shelton, Carl D. Riggs, and Loren G. Hill

ABSTRACT

High reproductive potential and extended spawning period are desirable attributes for forage fishes but these qualities are seldom found in a single species. Threadfin shad (Dorosoma petenense) and gizzard shad (D. cepedianum) are two important forage species in southern waters; where they occur together, they may be involved in competitive interactions. To determine specific areas of overlap in their life histories, the reproductive ecology was assessed based on gonadal examination, spawning behavior, egg deposition patterns, and larval occurrence. In Lake Texoma, gizzard shad was found to initiate and terminate spawning earlier (March-May) than threadfin shad (April-June) but peak spawning for both species overlapped in early May. Water temperature of initial spawning for gizzard shad was 17 °C compared to 19 °C for threadfin shad. In contrast to gizzard shad, threadfin shad spawned nearer the water surface, spawning activity was more visible, and was concentrated in the early morning. Egg and larval identification was based on the eye-pigment characteristics of freshly hatched fry; threadfin shad have pigmented eyes at hatching while gizzard shad lack eye pigment until about 1 day after hatching.

INTRODUCTION

Fishery managers seldom find all desirable attributes of a forage fish in a single species. High fecundity is one such character; also, if spawning is protracted, it should result in a more continuous food supply for predators. Two closely related species may have complimentary characters which, if these species occur together, would increase and prolong the available food supply; however, habitat requirements may overlap in ways that could be detrimental to either or both species.

The threadfin shad (Dorosoma petenense) and the gizzard shad (D. cepedianum) are recognized as two important warmwater forage species. Each is prolific and if their spawning periods overlap, may together be a more valuable food source than either species alone. Threadfin shad are predominantly schooling, limnetic, and planktivorous throughout life (Johnson 1970); gizzard shad are similarly characterized early in life but become more solitary, littoral, and detritivores after their first year (Baker and Schriut 1971). Competition between the species should be greatest during their limnetic, larval stages because they co-exist spatially and temporally (Netsch et al. 1971) and their food habits are similar (Cramer and Marzolf 1970; Van Den Aylole and Wilson 1980). This competition should be even greater if their spawning periods are simultaneous.

This study was conducted in Lake Texoma, a 41,700 hectare, U.S. Army Corps of Engineers hydro-electric/flood control reservoir on the Red River, Oklahoma-Texas. Primary effort was in the 400-hectare Buncombe Creek arm in Oklahoma. Histological procedures were described by Sublette (1958).

The objective of the present study was to examine concurrently the reproductive biology of the two species in one water body to determine whether or not there were isolating mechanisms that minimized overlap. Gizzard shad have been in Lake Texoma since impoundment in 1942 (Riggs and Bonn 1959) but threadfin shad were not known to be present until about 1957 (Riggs and Moore 1958). At the time of this study, the latter had apparently replaced the former as the most abundant species in the reservoir; but both species were thriving.

METHODS AND MATERIALS

In the present study, the reproductive biology was compared by ovari development, observations of spawning, and egg and larval sampling.

Minimum-maximum surface water temperatures were recorded daily near the mouth of Buncombe Creek arm at the University of Oklahoma Biological Station boathouse during 1968 and at weekly intervals during 1969; water temperature was also taken in conjunction with all other sampling.

Gonadal data are useful in determining the general spawning season and were used to compare these two colorated species. Ovarian development is of considerably greater value than testicular data because of more obvious seasonal changes. Gonadal-somatic indices (GSI = ovari weight/total body weight × 100) were calculated from fish of each species collected at weekly intervals with experimental gill nets and by electrofishing during the late winter and spring of 1968 and 1969. Ova from each species were weighed fresh to the nearest 0.1 gram--for gizzard shad longer than 215 mm (total length), based on initial maturity reported by Bondola (1968); for threadfin shad longer than 60 mm (Johnson 1971).

Spawning activity was observed on many occasions in conjunction with other sampling; however, spawning frequently occurred at certain locations and planned observations were made. Spawning of gizzard shad was most predictable and conveniently observed in the tributaries while threadfin shad spawning was easily studied at several Lake locations. Observations were usually taped on a portable recorder and later transcribed.

Devices designed to sample the adhesive spawned eggs (Shelton 1972) at natural spawning sites were placed in 12 designated areas of the Buncombe Creek arm during the 1968 and 1969 spawning seasons. Each sampler consisted of two 15 × 15 cm squares of welded iron rod fastened at a right angle to the other along an adjoining edge; each square frame was temporarily covered with fine-mesh (00) nylon webbing (225 cm²) which was individually tagged. One sampling surface was thus oriented parallel to the substrata and the other was vertical. The procedure for using these samplers was as follows:

1. the Buncombe Creek arm was subdivided into 8 approximately equal sampling areas, two cove areas, and two in the flowing stream—one at the mouth and one about 2.4 km upstream from the mouth;
2. at a randomly picked site within each of the sampling areas, three samplers were set for periods of 48 hrs on alternate weeks;
3. one sampler was set on the bottom in water about 30 cm deep, one was set further offshore on the bottom in water 1.5 m deep (for bottom sets the horizontal square was parallel to the bottom and the vertical square perpendicular to it), and one was set floating with the top of the vertical square immediately beneath the surface of the water and the horizontal square parallel to the surface. This floating set was directly above the deeper bottom set;
RESULTS AND DISCUSSION

Gonadal Development.--Ovaries of gizzard shad were examined from mid-February to mid-June in 1968 and 1969 (n = 196 and 203). The gonadal-somatic indices (GSI) increased slightly from mid-February through mid-March; by mid-March, in conjunction with increasing water temperature, yolk deposition had accelerated (Fig. 1). In early April of both years, ovarian development peaked with a modal GSI of between 7 and 10%. The mean surface water temperature during this period was 15–16 °C. In both years, the mean GSI decreased slightly until early May and after mid-May of both years there was a significant decrease (P < 0.05) in GSI. The water temperature in early May of both years was between 18 and 21 °C.

Ovaries of threadfin shad were examined from the same pre-spawning spawning periods each year (1968 and 1969). Following a gradual increase through February and March, the GSI increased abruptly during the first two weeks of April (Fig. 1), while the mean temperature increased to approximately 15 °C. Development peaked by mid-May (10–22%) when the water temperature was about 20–27 °C. Gonadal-somatic indices were significantly reduced (P < 0.05) by late May when the water temperature was between 24 and 25 °C. Ovaries of most threadfin shad were again near pre-spawning size by mid- to late June.

The ovaries of gizzard shad began to increase, peaked, and declined sooner within the year than that of threadfin shad. The GSI cycle for gizzard shad preceded that of threadfin shad by about one month. The peak GSI for gizzard shad was lower in both years than that for threadfin shad, but a direct comparison of reproductive potential of the two species is vitiated by the size difference of mature adults. Since the mature size for the mature size of eggs and larval size is the same (Shelton and Stephens 1980) and egg number per gynecologist is similar (Kilambi and Baglin 1969; Johnson 1971; Jester and Jensen 1972), population fecundity could be compared only if population size and size distributions were known.

By late March and early April, ovulated eggs had collected in the ovarian lumen of gizzard shad and the GSI averaged 7–10%, with an upper range of 16–18%; while threadfin shad did not have ripe eggs in the ovarian lumen until after mid-April and the average GSI in late April to early May was 16–20% with some as high as 26%. The release of mature eggs was reflected in a decline of GSI for gizzard shad in early May compared to a comparable decline in late May to early June for threadfin shad.

The seasonal pattern of the GSI suggests that spawning for gizzard shad begins in late March to early April when the water temperature exceeds 15 °C, reaching a peak in late April, and continuing until mid-May, for a total period of about 6–7 weeks. Baglin and Kilambi (1968) reported a comparable reproductive period for gizzard shad in Beaver Reservoir, Arkansas. Bodola (1966) reported a 4-week spawning period for Western Lake Erie, when the temperature exceeded about 16 °C.

Gonadal data for threadfin shad indicated an 8–10 week spawning period each year in mid- to late June. By the first part of July, the temperature at onset was about 19 °C. Johnson (1969) found spawning of threadfin shad in Arizona reservoirs from April to June, based on gonadal data. Johnson (1971) reported that the older fish initiated spawning earlier in the season.

Observations of Spawning.--Many past descriptions of spawning were based on incomplete, chance observations. Our observations of threadfin shad spawning were planned on the basis of previous knowledge. However, gizzard shad spawning was predictable only in tributary locations.

Spawning by threadfin shad was first observed when surface water temperature reached about 19 °C during the morning hours. During 1968, water temperature reached 19 °C after noon on 3 or 4 days preceding the initial spawning. The first observed spawning was at 0900 on 10 April, at a water temperature of 19 °C. Activity continued for several hours, gradually diminishing until none was apparent after noon. We found no evidence for night spawning based on egg sampling and after-noon activity was only seen during initial spawnings of the season. During the latter part of April, spawning began about 1–2 hours after sunrise and was most intense between 0900 and 1200. By early May, spawning usually began within 15 to 30 minutes after sunrise and continued actively for only about 1–2 hours. This activity pattern was also evident from catches in surface-set gill nets and from eggs collected on floating samplers, both of which were suspended from the Biological Station boathouse. For example, on 4 May 1968, 1,880 spawning adults were netted between 0830 and 0700, 300 were captured between 0700 and 0830, and only 13 were collected in the remainder of the 24-hour period.
Eggs were collected from floating samplers in a pattern coincident with the abundance of adults. From 7-5 May, the water temperature was near 19 C until well after sunrise and spawning was delayed about 1 hour compared to previous days.

Primary orientation of spawning groups of threadfin shad appeared to be toward the surface; during spawning, massive schools of threadfin shad were seen swimming parallel to the shore, periodically smaller groups would move shoreward. The smaller groups, composed of one to several females and a few more males, were apparently led by females who selected the egg deposition sites. Upon encountering a suitable substrate, the group abruptly turned at the deposition site. Males crowded closer to the female, and maneuvered erratically, which resulted in considerable surfacing. Lag over eggs were scattered above, below, the water surface on the substrate. The group continued swimming parallel to shore until another site was encountered or often they circled and repeatedly spawned on the same substrate.

Egg densities as high as 40-65/cm² were measured on egg-sampler surfaces. The most frequently used sites were in shallow littoral waters but pelagic floating objects were also frequently utilized. The sex ratio in those spawning aggregations sampled was about 5 males to each female (4,495:879), and in general, the smallest ripe males were slightly smaller (≥60 mm TL) than ripe females (≥70 mm TL). Data on size distribution of spawning aggregations was discussed by Shelton (1972).

In contrast to threadfin shad, gizzard shad spawning was less easily observed. Occasionally, a group of gizzard shad would appear from deeper water, swim erratically shoreward along the bottom, abruptly turn, presumably having deposited eggs, and return to deeper water. Little surface disturbance was created during these spawnings.

Spawning in tributaries was more easily observed. Surface agitation was more common, perhaps because of the relative shallowness of the water. Spawning was probably initiated in the tributaries. Spawning occurred in daylight hours, but during periods of continuous observation, activity was more intense at night. During one 24-hour period on 21-22 April, spawning was most active from 2000 to 0500, gradually diminishing through 1200-1400, and was only intermittent for the remainder of the afternoon and early evening.

Water temperature ranged from 16 to 21 C during this period.

Spawning aggregations in streams had from 40-50 adults of 20-35 cm TL (based on 159 fish collected on 28 March 1968). The sex ratio was almost 5 females:male. The aggregation remained close-knit until a female moved upstream, or laterally, to deposit eggs; several males accompanied each female. The group then returned to the main tributary, and concentrations of eggs on the substrate in tributary spawning appeared to be quite dense, although we did not sample those surfaces; however, the accumulation of gizzard shad eggs on samplers in the lake was rarely greater than 1 egg/cm².

On those occasions in which gizzard shad spawning was observed in the lake, it was similar to the behavior in the stream. However, it appeared that the large, mobile aggregations of spawning threadfin shad contrasted sharply with the smaller, less mobile groups of spawning gizzard shad.

The surface orientation of spawning threadfin shad permitted observation of the general movement along the shore but we were unable to count the considerable activity that occurred in gizzard shad. Judging from the spawning witnessed in the tributaries, a reasonable assumption is that gizzard shad aggregations were offshore in deeper water but shoreward movement along the bottom would not be obvious, particularly if the most active spawning occurred at night. The daylight spawning at the surface by gizzard shad reported by Jester and Jensen (1972) may not be typical of all populations. Bodola (1968) did not observe daylight spawning but evidence that spawning occurred at night, with little surface disturbance. Thus, if this diel pattern is typical of gizzard shad, most lake spawning would be difficult to observe.

The spawning behavior of threadfin shad we witnessed agreed with those observations of Johnson (1971) but differed with aspects of other reports. Rawston (1964) observed threadfin shad spawning at about 15 C while we observed a minimum temperature of 19 C. Hubbs and Bryan (1974) determined that the minimum incubation temperature for development of viable threadfin shad was 15 C. Lambou (1965) described the abrupt cessation of threadfin shad spawning after sunrise while we observed a consistent diel pattern that was somewhat less restrictive.

Eggs and Larvae.—From 20 March to 12 July 1968, 236 egg samplers were examined, 14 of which were from the tributary area; from 10 March to 26 May 1969, 192 egg samplers were recovered, and 12 were from the tributary habitat. At the lake sites that had floating and bottom sets (deep and shallow), a total of over 66,000 Dorosoma spp. eggs was collected in 1968-69. Of those that hatched, 1,284 were gizzard shad and 2,269 threadfin shad (Table 1). A lower percentage of eggs taken from the shallow samplers was hatched, but we assumed no differential mortality between species. Threadfin shad deposited the greatest number of eggs on the shallow bottom and floating samplers while gizzard shad deposited more eggs on the two bottom samplers (shallow and deep). Relatively few eggs were sampled from the tributary sites and none were identified as threadfin shad.

Table 1. Summary of shad eggs collected from lake samplers from March through July 1968 and 1969, Lake Texoma, Oklahoma-Texas.

<table>
<thead>
<tr>
<th>Species</th>
<th>Shallow (no.) (%)</th>
<th>Floating (no.) (%)</th>
<th>Bottom (no.) (%)</th>
<th>Totals (no.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gizzard shad</td>
<td>808</td>
<td>63</td>
<td>11</td>
<td>886</td>
</tr>
<tr>
<td>Threadfin shad</td>
<td>1,863</td>
<td>82</td>
<td>17</td>
<td>1,962</td>
</tr>
<tr>
<td>Total collected</td>
<td>6,1483</td>
<td>2,917</td>
<td>2,331</td>
<td>6,493</td>
</tr>
</tbody>
</table>

Only eggs that hatched could be identified.

Data from the floating sets at the boathouse (not included in Table 1) further suggest a difference in selection of egg deposition sites by the two species. A total of over 63,000 shad eggs was collected from the two samplers, 4,344 hatched and all were threadfin shad.

From 23 March to 29 August 1968, 323 meter-net samples were taken, 20 of which were from the tributary area; from 10 March to 14 June 1969, 109 samples were taken and 14 were from the tributary area. There were 2,099 gizzard shad and 448 threadfin shad yolk-sac larvae collected in the two years. The disproportionately high abundance of gizzard shad was a result of the tributary samples: 1,442 gizzard shad but no threadfin shad were collected in the creek.

The presence of yolk-sac larvae or eggs that later hatched was considered as indicative of the general spawning location and time of each species.

In general, eggs and larvae of gizzard shad were more commonly collected (78%) in the upper region of the study area which included the main tributary that entered that portion of Burcombe Creek arm (Table 2). In the segment of the stream that was sampled, no threadfin shad egg or larva was collected, while the highest density (52%) of the gizzard shad eggs and larvae came from this limited area. In contrast, a greater proportion of threadfin shad eggs and larvae (93%) was collected in the lower half of the arm. This region was adjacent to the open expanse of the main reservoir, a habitat apparently more frequented by identifiable larval threadfin shad.

The initiation of spawning by most temperate fishes is closely correlated with water temperature and initial spawning is usually observed in the suitable habitat that warms earliest. No larvae or eggs were collected in the lake prior to 1 April of either year (Table 3) but in 1968 gizzard shad spawning was first observed in the tributary on 27 March at a water temperature of 17 C while the lake temperature was 13 C.
ANNUAL spawning into hatching in 1971). This was more frequent than in the previous years. Jester and Jensen (1972) reported surface spawning activity by gizzard shad (around a boat house) similar to that described for threadfin shad in our study, but did not observe such a similarity. The floating egg samplers used in our study at the boat house not only provided a convenient daily check of spawning for threadfin shad but confirmed that none of the eggs collected there were those of gizzard shad.

**SUMMARY AND CONCLUSIONS**

Interspecific competition cannot be proved by simply demonstrating overlapping use of a common resource. Conversely, detailed study of mechanisms that segregate aspects of the life histories of two closely related species may suggest that an apparently limited resource is used by each species differently in time or space and is indeed not limiting. Gizzard and threadfin shad appear to be similar biologically, especially in their early life histories. Based on eggs, whereas gizzard shad spawn for spawning, and egg and larval collections in Lake Texoma, we concluded that there was overlap in seasonal, diel, and spatial aspects of reproduction, but we observed probable isolating (segregating) mechanisms of reproduction in the two species, as well (Table 4).

Table 4. Summary of differences in the reproductive biology of threadfin and gizzard shad in Lake Texoma, Oklahoma-Texas.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Gizzard</th>
<th>Threadfin</th>
</tr>
</thead>
<tbody>
<tr>
<td>GSI Peak (%)</td>
<td>7-10</td>
<td>16-20</td>
</tr>
<tr>
<td>Time early April</td>
<td>early May</td>
<td></td>
</tr>
<tr>
<td>Spawning Initial</td>
<td>late March-16 C</td>
<td>late April-19 C</td>
</tr>
<tr>
<td>Duration (wks)</td>
<td>7-9</td>
<td>6-10</td>
</tr>
<tr>
<td>Size (mm TL) male</td>
<td>207-70</td>
<td>0-170</td>
</tr>
<tr>
<td>female</td>
<td>215-360</td>
<td>60-190</td>
</tr>
<tr>
<td>Spat ratio (♂:♀)</td>
<td>2:1-4:1</td>
<td>5:1-15:1</td>
</tr>
<tr>
<td>Orientation bottom surface</td>
<td>bottom surface</td>
<td></td>
</tr>
<tr>
<td>Diel peak nocturnal</td>
<td>morning</td>
<td></td>
</tr>
<tr>
<td>Eggs (mm)</td>
<td>0.9-1.1, adhesive 0.9-1.1, adhesive</td>
<td></td>
</tr>
<tr>
<td>Incubation (hrs-23 C)</td>
<td>43</td>
<td>72</td>
</tr>
<tr>
<td>Larval eye</td>
<td>non-pigmented</td>
<td>pigmented</td>
</tr>
<tr>
<td>at hatching</td>
<td>at hatching</td>
<td></td>
</tr>
</tbody>
</table>

*Globally*, the duration of peak spawning for gizzard shad was April through early May, whereas for threadfin shad it was principally during May. Gizzard shad spawned earlier in the season when surface waters were about 16 °C compared to about 19 °C for the threadfin shad. Gizzard shad spawned earlier in the tributary compared to the lake proper. That threadfin shad did not spawn in the tributary may have been related to the smaller size of the stream and spawning by this species in larger lotic systems elsewhere may not be limited. Even during the overlapping peak spawning period, however, there appeared to be more spawning by threadfin shad near the open water portion of the arm. Even though both spawned in regions of shallow water, separation was somewhat affected by the preference for bottom substrates by gizzard shad and for emergent substrates by the threadfin shad. Moreover, threadfin shad showed a very definite early morning activity while gizzard shad spawning activity was greater during hours of darkness. Differences in diel activity would tend to reduce the overlap and thus

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**Table 2.** Combined numbers of eggs and yolk-sac larvae of gizzard shad and threadfin shad collected by regions during 1968 and 1969.

<table>
<thead>
<tr>
<th>Region</th>
<th>Gizzard shad</th>
<th>Threadfin shad</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(no.)</td>
<td>(%)</td>
</tr>
<tr>
<td>Buncombe Creeka</td>
<td>1,796</td>
<td>52</td>
</tr>
<tr>
<td>Upper Sectionb</td>
<td>1,267</td>
<td>37</td>
</tr>
<tr>
<td>Lower Section</td>
<td>384</td>
<td>11</td>
</tr>
</tbody>
</table>

aSample site 2.4 km upstream in tributary.

bUpper one-half of Buncombe Creek arm.

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**Table 3.** Combined numbers of eggs and yolk-sac larvae of gizzard and threadfin shad collected by time periods during 1968 and 1969, Lake Texoma, Oklahoma-Texas.

<table>
<thead>
<tr>
<th>Species-Year</th>
<th>March</th>
<th>April</th>
<th>May</th>
<th>June</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gizzard shad</td>
<td>-</td>
<td>5</td>
<td>581</td>
<td>1,775</td>
</tr>
<tr>
<td>1968</td>
<td></td>
<td>0</td>
<td>270</td>
<td>20</td>
</tr>
<tr>
<td>1969</td>
<td></td>
<td>0</td>
<td>28</td>
<td>395</td>
</tr>
<tr>
<td>Threadfin shad</td>
<td>-</td>
<td>0</td>
<td>0</td>
<td>25</td>
</tr>
<tr>
<td>1968</td>
<td></td>
<td>0</td>
<td>0</td>
<td>25</td>
</tr>
</tbody>
</table>

---

During the period 1-15 April, eggs but no yolk-sac larvae of gizzard shad were collected from the lake but larvae were abundant in the tributary; neither eggs nor larvae of threadfin shad were collected in this period. From 16-30 April, eggs and larvae of both species were sampled in the lake with those of gizzard shad predominating. In 1968, threadfin shad spawning was first observed on 18 April when the water temperature was 19 °C; eggs were collected at that time but larvae were not taken until 24 April. During the first half of May, most yolk-sac larvae collected were threadfin shad, although gizzard shad were still spawning as their eggs were quite abundant in the samples. During the last half of May, spawning was hampered because precipitation had carried debris into the lake. The resulting high water also complicated placement and recovery of egg samplers; however, threadfin shad larvae and eggs predominated. From 1-15 June, only threadfin shad were collected; by late June their abundance had declined and after 1 July neither eggs nor larvae of either species were sampled. Gizzard shad apparently initiated spawning earliest in the inflowing stream where the water temperature was warmed sooner. Gizzard shad spawned from late March, beginning in the tributary, through late May or about 7-9 weeks. Threadfin shad did not ascend the tributary but began spawning in the lake in mid to late April and continued through late June, for an estimated duration of 8-10 weeks. Those spawning periods are slightly longer than those estimated from gonadal data and clearly illustrate temporal overlap with slightly different peaks. A temporal spawning difference of about one month was also described for the two shad species in Beaver Reservoir based on larval fish sampling (Netsch et al., 1971; Howser and Netsch, 1971).

Threadfin shad spawning was more concentrated near the open water area while gizzard shad reproduction was more common in the upper section of the arm. Netsch et al. (1971) found gizzard shad larvae several weeks earlier than threadfin shad in the upper portion of Beaver Reservoir, Arkansas.

A further spatial separation in spawning sites was indicated by the egg deposition patterns. Both species deposited eggs in the shallow water but threadfin shad eggs were more frequently found on the floating samplers, suggesting a surface orientation, whereas gizzard shad eggs were proportionately more abundant on the deeper-bottom samplers. Jester and Jensen (1972) reported surface spawning activity by gizzard shad (around a boat house) similar to that described for threadfin shad in our study but we did not observe such activity. The floating egg samplers used in our study at the boat house not only provided a convenient daily check of spawning for threadfin shad but confirmed that none of the eggs collected there were those of gizzard shad.
hybridization, even though natural hybrids are produced (Shelton and Grinstead 1973).

Probably the most important consideration with reference to potential competition by larvae of the two species was the seasonal difference in spawning peaks. To the fishery manager, the presence of small prey over a longer period of time than would be the case if only one species were present is probably one of the more significant aspects of their relative reproductive biology and therefore it is possibly advantageous to have both of these species as prey in a system.

LITERATURE CITED


Washington, D.C.


Washington, D.C.


Jester, D. B., and B. L. Jensen. 1972. Life history and ecology of the gizzard shad, Dorosoma cepedianum (LeSueur) with reference to Elephant Butte Lake. New Mexico State University, Agricultural Experiment Station, Research Bulletin 218. Las Cruces, New Mexico.


CITED


DEPARTMENT OF BIOLOGY, UNIVERSITY OF OKLAHOMA, NORMAN, OKLAHOMA 73063.
Survival of fish larvae was studied from 1979-1980 in association with entrainment and passage through the Ludington Pumped Storage Power Plant (LPSPP) on Lake Michigan. A new sampling technique consisting of slow vertical hauls with a 2 m-diameter plankton net was evaluated and found to effectively capture live fish larvae. A total of 171 samples was collected from two control and two power plant discharge stations. Alewife and smelt dominated the 2138 larvae collected. Other taxa included: yellow perch, nine-spine stickleback, johnny darter, burbot, lake whitefish, shiner, and deepwater sculpin, Cyprinidae, and Cottus. Alewife exhibited low initial survival rates (17% control and 11% experimental), but improved methods indicated survival may be as high as 40-44% at experimental sites. Small (<8 mm) larval smelt exhibited 57% and 50% initial survival at the control and power plant sites, respectively. Higher initial survival was recorded for large smelt larvae (>16 mm) at the control sites (91%), compared to 43% at the discharge sites. Latent mortality further decreased large smelt survival to 11% at the discharge sites. Most other species exhibited high survival (80-100%) at the control and discharge stations, though estimates were derived from fewer specimens especially in the discharge samples. Initial survival for all species combined was 67.5% at control stations compared to 38.7% at discharge stations. Survival of larvae 24 hours after capture was 71.9% for the control site and 53.0% for the discharge site.

Recent power plant studies (Boreman 1977; Goodyear 1977) suggest greatest impacts to fish communities may result from the entrainment of nonscreenable organisms (e.g., fish eggs and larvae). However, few studies examining power plant related mortality exist. Marcy (1971, 1973) found larval mortality at nuclear plants primarily due to mechanical damage or prolonged exposure to heated effluent. Historically, 100% mortality has been assumed at fossil fuel plants, but Ecological Analysts (1976) observed 25-65% survival for cladophora and anadromous larvae, though survival of larval ctenophores was essentially zero. Studies at hydroelectric pumped-storage plants by Snyder (1975) and Prince and Mengal (1980) have indicated survival of entrained fish larvae may be substantial. Laboratory studies conducted by Beck et al. (1975) indicate high survival rates for striped bass eggs and larvae subjected to pressure regiments that simulated passage through a pumped storage plant.

One of the most critical factors affecting survivorship studies is sampling mortality. Most mortality associated with collection can be attributed to mechanical damage, especially impalement and abrasion, against plankton nets deployed in strong currents. Recent technological developments (McGrady and Wyman 1977) have introduced the larval fish table which is effective at reducing water velocities and thus increasing larval survival during collection but is cumbersome, relatively immobile, unable to draw water up a large head and unable to adequately sample larvae present in low densities because of small volume of water delivered.

Cade and Hergenrader (1978) developed regression equations to discriminate between entrainment mortality and net-induced sampling mortality. They found a direct relationship between observed mortality and water velocity in the nets at control stations. Further, they recognized the importance of maintaining similar physical conditions (especially current velocity) in intake and discharge samples.

Gear deployed in power plant research must often be site selective and the purpose of this study was to develop a technique that could rapidly sample sufficient numbers of larvae at several sites to directly evaluate survival. A slow vertical haul with a large diameter (2-meter) plankton net was used to reduce current across the net and filter enough water to collect sufficient numbers of larvae for study. Research was conducted at the Ludington Pumped Storage Power Plant on Lake Michigan.

DESCRIPTION OF STUDY AREA AND POWER PLANT

The Ludington Pumped Storage Power Plant (LPSPP) is located on the east-central shore of Lake Michigan 6.5 km south of Ludington, Michigan. The six intakes of the LPSPP are located on the dredged out shoreline of Lake Michigan and extend from 11.6 m below the surface to 21.4 m at bottom (Figure 1). Screening consists of a gridwork with 30.5 x 58.4 cm openings to prevent entry of large debris. Each intake is attached to a Francis-type reversible turbine which transfers water between Lake Michigan and the upper reservoir (150 m above Lake Michigan). Water passes through 8 m diameter penstocks that are 356 m long and open at a depth of 23 m and extend to the bottom of the upper reservoir. Maximum water velocities in the penstocks approach 8.5 m/sec, which indicates larvae are present in the power plant system for a minimum of 47 seconds in transfer between the two bodies of water. During this time they are exposed to rapid pressure changes of 1-11 atmospheres.

The upper reservoir is 30 to 34 m deep and encompasses 332 hectares at full pond and fluctuates daily up to 20.4 m during routine generation. Sides are asphalt lined down to 23 m, with 3 m of compacted clay on the bottom. During generation, water empties into Lake Michigan between two rock jetties 339 m apart composed of large limestone boulders which extend 490 m from shore. Water velocities 122 m out from the intake (where entrainment and mortality samples were taken) average 50-100 cm/sec, and are reduced to 20-30 cm/sec at the end of the jetties.

METHODS

Survivoral sampling was conducted approximately every two weeks from late April through August in 1979 and 1980. In the upper reservoir discharge during pumping mode (night), in the Lake Michigan discharge between the jetties during generation mode (day), and at a control site approximately 1.6 km south of the plant (day and night).
night) (Figure 2). Sampling gear consisted of a 2 m-diameter 35µm mesh conical plankton net with a mouth to length ratio of 6:1, equipped with a General Oceanics flow-meter (Model 2030) mounted one-third off center in the mouth. Initially all samples were collected by making a slow bottom to surface vertical haul using a Mytwinch connected to a 12 volt battery. Difficulties in maintaining a vertical haul in the plant discharge were successfully overcome by utilizing boat power and the swift currents emanating from the plant to position the boat directly over the net. Control samples were taken identically at the 9.1 m contour to approximate the depth of plant discharge samples. However, larvae of certain taxa were scarce at this depth (stratum) in 1979 which prompted additional control sampling in 1980 using a 1 m-diameter, 35µm mesh plankton net mounted on a sled and towed at the 3.6 m depth contour for two minutes.

All samples were immediately returned in 19 liter plastic buckets to the laboratory and sorted in enamel trays under lighted magnifying lenses. Live fish were placed in 11.5 liter aquaria and maintained at ambient collection temperature in water baths provided by a Living Stream (Model LS-700). Dead fish were preserved immediately in Davidson's solution (Lam and Roff 1977). Locomotor activity was the criterion for live/dead determination. Initial survival refers to larvae alive at time of sorting, latent survival refers to larvae surviving 24 hours in aquaria, and overall survival is the number of larvae alive after 24 hours divided by the total number of larval collected. Power plant samples were usually processed within one hour of collection, however control samples required more time due to greater distance traveled. During peak alewife concentrations in 1980, plant discharge samples were sorted dockside and usually completed within 15 minutes of collection.

Control samples were analyzed statistically with the Mann-Whitney U test (Sokal and Rohlf 1969) to determine if survival of larvae differed significantly between years. Tests on all taxa indicated survival was similar (x = 0.1) and data were pooled from 1979 and 1980.

RESULTS AND DISCUSSION

Data collected in the first half of 1980 indicated that the volume of water sampled and current velocity were similar between the control and experimental samples taken with the large diameter plankton net on a slow vertical haul (Table 1). Further, mean current velocities within the net (15-19 cm/sec) during sampling were markedly less than currents in the discharge area which were greater than 50 cm/sec nearly 75% of the time (Liston et al. 1980). Samples collected with the one meter net attached to a sled strained somewhat smaller volumes of water, but sampling velocities were greater due to the necessity of horizontal towing by boat. However, sled samples were necessary to evaluate survival of certain species of larvae (Lake whitefish, burbot, cyprinids) which were virtually absent in samples taken at deeper contours (Liston et al. 1980). Data analysis indicated larval survival was not affected by the increased velocities of the sled towed net.

During the two year study, a total of 171 samples was collected at the discharge (89) and control (82) sites. A total of 2138 larvae represented at least eleven species and seven families (Table 2). Most larvae (96.2%) were taken at the control site, and greater survival (96.8%) (all species combined) was observed there, though survival at both sites was substantial. However, survival was species specific, and, for at least one species, size related. These phenomena were also reported by Cannon et al. (1978). Analysis of survival by species is given below. Statistical comparisons were not made because of low numbers collected of most taxa.

Alewife (Alosa pseudoharengus) comprised 33% of all larvae collected (Table 2). This species exhibited lowest initial survival of any larvae both in the control (17.3%) and experimental (11.8%) samples suggesting at
Table 1. Comparison of mean volumes (m³) filtered and net velocities (cm/sec) during entrainment mortality study of LPSP in the first half of 1980.

<table>
<thead>
<tr>
<th>Date</th>
<th>1 m-net (Sled) control</th>
<th></th>
<th>2 m-net control</th>
<th></th>
<th>2 m-net generation discharge</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>Volume</td>
<td>Velocity</td>
<td>N</td>
<td>Volume</td>
</tr>
<tr>
<td>4-30-80</td>
<td>2</td>
<td>24.6</td>
<td>26.1</td>
<td>3</td>
<td>63.5</td>
</tr>
<tr>
<td>5-12-80</td>
<td>3</td>
<td>8.3</td>
<td>8.8</td>
<td>3</td>
<td>56.5</td>
</tr>
<tr>
<td>5-27-80</td>
<td>3</td>
<td>27.3</td>
<td>29.0</td>
<td>3</td>
<td>55.3</td>
</tr>
<tr>
<td>6-10-80</td>
<td>2</td>
<td>82.6</td>
<td>87.6</td>
<td>3</td>
<td>44.7</td>
</tr>
<tr>
<td>6-26-80</td>
<td>3</td>
<td>47.1</td>
<td>50.0</td>
<td>3</td>
<td>87.0</td>
</tr>
<tr>
<td>TOTAL</td>
<td>13</td>
<td>37.9</td>
<td>40.2</td>
<td>15</td>
<td>61.2</td>
</tr>
</tbody>
</table>

least 5.5 percent mortality due to entrainment. Only one alewife larva survived for 24 hours. However, low survival of larval alewife may have been affected by length of processing time and crowding during transport. Two series of discharge samples collected during peak alewife abundance in 1980 were sorted dockside and initial survival averaged 40-44%, though overall survival was still high. Processing of control samples on board was impossible due to lack of lighting facilities at night and unsteady work area for sample examination. However, observed initial survival in discharge samples examined dockside was similar to the 33-54% survival reported by Ecological Analysts (1976) and Cannon et al. (1978) in the discharge of fossil fuel and nuclear plants, and by Prince and Mengel (1980) at a pumped storage site in South Carolina for clupeids.

Smelt (*Pungitius pungitius*) was the most abundant taxon collected in survival samples and comprised 40% of the collections. Preliminary data analysis indicated survival for larvae and juveniles larger than 8 mm was greater at the control site. Small smelt showed similar survival rates at the control and discharge sites (58% and 50% respectively; Table 2). Latent survival was high and similar between control and discharge sites for an overall survival of 53% and 42% respectively, indicating approximately 10% of mortality could be directly related to entrainment. Survivorship of large smelt was extremely high at the control station (initial and latent samples), and even less after latent effects were observed (Table 2). Increased mortality of large smelt in the plant discharge samples may be a function of size. Mercy (1973) found significantly greater survival of larvae less than 15 mm than in fish 20-40 mm in length.

Over 99.3% of cyprinid larvae (300) were collected at the control station and survival was 100%. Unfortunately, only two cyprinid were collected in discharge samples and one survived the 24-hour observation.

Larval yellow perch (*Perca flavescens*) were collected in low numbers at all sites (Table 2). Survival rates were 33% at the control site and 14% at the discharge site, indicating 19% entrainment mortality. All live larvae survived 24 hours.

Most other larval taxa (burbot, *Lota lota*; sculpins, *Cottus sp.*; Johnny darter, *Etheostoma nigrum*; Lake whitefish, *Coregonus clupeaformis*; ninespine stickleback, *Pungitius pungitius;* and bloater, *Coregonus hoyi*) had high (80-100%) initial survival in both control and

Table 2. Survival of fish larvae (of several species) entrained at the Ludington Pumped Storage Power Plant and collected at a control station on Lake Michigan 1979-1980.

<table>
<thead>
<tr>
<th>Taxa</th>
<th>Initial Survival</th>
<th>Latent Survival</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Discharge</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>Percent (*)</td>
</tr>
<tr>
<td>Alewife</td>
<td>567</td>
<td>17.3(9.9)</td>
</tr>
<tr>
<td>Smelt (≤ 8 mm)</td>
<td>38</td>
<td>57.9(32.2)</td>
</tr>
<tr>
<td>Smelt (&gt; 8 mm)</td>
<td>712</td>
<td>91.6(12.2)</td>
</tr>
<tr>
<td>Fourhorn sculpin</td>
<td>3</td>
<td>67.7(157.8)</td>
</tr>
<tr>
<td>Burbot</td>
<td>1</td>
<td>100.0(0)</td>
</tr>
<tr>
<td><em>Cottus sp.</em></td>
<td>0</td>
<td>-</td>
</tr>
<tr>
<td>Johnny darter</td>
<td>111</td>
<td>90.1(7.1)</td>
</tr>
<tr>
<td>Yellow perch</td>
<td>9</td>
<td>33.3(36.0)</td>
</tr>
<tr>
<td>Lake whitefish</td>
<td>33</td>
<td>100.0(0)</td>
</tr>
<tr>
<td>Ninespine stickleback</td>
<td>63</td>
<td>100.0(0)</td>
</tr>
<tr>
<td>Cyprinidae**</td>
<td>300</td>
<td>88.0(18.3)</td>
</tr>
<tr>
<td>Bloater</td>
<td>1</td>
<td>100.0(0)</td>
</tr>
<tr>
<td>TOTAL</td>
<td>1842</td>
<td>67.5%</td>
</tr>
</tbody>
</table>

*90% Confidence Interval
**Primarily spottail shiner
In conclusion, use of a large (2 m) diameter plankton net retrieved vertically appears to be an effective method for collecting live larvae of several species of fish. More fragile larvae (e.g., alewife) must be removed from samples quickly to ensure accurate live/dead determination. This sample technique has the advantages of mobility, reduction of net velocity, and ability to sample large volumes of water.

ACKNOWLEDGEMENTS

This research was supported through contracts with Consumers Power Company who, with the Detroit Edison Company provided research facilities on Lake Michigan. Many students and technicians of the Michigan State University, Department of Fisheries and Wildlife aided in data collection.

LITERATURE CITED


MICHIGAN STATE UNIVERSITY, DEPARTMENT OF FISHERIES AND WILDLIFE, LUDINGTON RESEARCH LABORATORY, LUDINGTON, MICHIGAN 49431.
INTRODUCTION

Myomere number is perhaps the most commonly used quantitative character for identification of fish larvae. The nature of vertebra formation suggests an easily predictable number of myomeres for a larval form when the vertebra count for adults is known. Unfortunately, the correspondence reported in the literature between myomeres in larvae and vertebrae in adults is somewhat less than perfect (cf. Snyder 1979). Among catostomids, I have found (unpublished data) that total myomeres for any of eight species wholly include and extend beyond the reported range of total vertebrae (Weberian vertebra included). This discrepancy introduces the following study which investigates natural variability in myomeres number with respect to size, time of year, and caudal fin formation for a representative bony fish, the yellow perch (Perca flavescens). The developmental and empirical relationships between myomeres and vertebrae number are discussed.

METHODS

Yellow perch was chosen for this study because its identification in field samples was not dependent upon myomere counts and because its spawning season did not span a great length of time (long spawning periods might yield thermally induced myomere variation). Also, they are particularly abundant in field samples from Lake Michigan.

Specimens were sampled from a single year class of a population in southeastern Lake Michigan near Port Sheldon, Michigan. Details of the collecting site were given by Jude et al. (1978). Larvae were sampled with a benthic fish-larvae sled (described by Yocom and Tasar 1980) and 0.5-mm diameter, nitex plankton net of 363 μ mesh. Samples containing yellow perch larvae were taken in mid-May and early and mid portions of June and July, 1980. Juveniles (young of the year) were sampled with a sami-ballon, nylon otter trawl having a 0.33-cm bar mesh (inner liner) and a 1.0-mm bar mesh (outer liner) during mid-July, 1980. Larvae were fixed in the field in 10% formalin buffered with sodium borate and later transferred to 5% buffered formalin. Juveniles were frozen in the field, thawed, then preserved in 10% formalin.

Preanal, postanal, and total myomeres were counted with the aid of polarizing filters (essential for accurate counts). Preanal myomeres included all segments whose bordering myosepta were at least partly anterior to the anus, including one segment anterior to the first myoseptum. Postanal myomeres were all segments posterior to the preanal myomeres, including a urostyle segment. The urostyle myomere was included because standard vertebra counts (Hubbs and Lagler 1958) include a hypural (urostyle) centrum. To count vertebrae in juveniles, the right side of the body was dissected away and whole specimens were stained with Alizarin Red S according to the method described by Taylor (1967). Precaudal (abdominal) vertebrae were those with pleural ribs, including any with partially branched ventral processes. Caudal vertebrae were those with unbranched anal spines. There were no specimens with fused, or otherwise abnormal, vertebrae.

Statistical comparisons of myomere and vertebra distributions were made with nonparametric tests. The median test compared locations of distributions and the Kolmogorov-Smirnov and chi-square for goodness of fit tests compared shapes of distributions (Conover 1980). These tests were chosen because of a concern for lack of normality in the highly peaked distributions and because of the discrete nature and small range of variability of the data. Stated differences in medians or distributions reflect test results at $P < 0.05$ with a two-tailed alternative hypothesis.

RESULTS

Myomeres were counted on 75 larvae ranging from 4.0 to 15.0 mm TL (Fig. 1). Total myomere number did not correlate with total length (correlation coefficient, $r = 0.013$), indicating no linear relationship between the variables. Sample sizes for June and mid-July were too small to make inference upon and were excluded from further analyses. The 59 remaining larvae were collected during mid-May and early July. Medians and distributions of preanal myomeres were not different between the two sampling periods. The median for postanal myomeres in mid-May was not different from that in early July; the distributions were different. Medians and distributions of total myomeres were different between the two sampling periods.

Vertebrae were counted on 71 juveniles ranging from 39 to 96 mm TL (Fig. 2a-c). There was no linear relationship between vertebrae number and total length ($r = 0.002$ to 0.003). Medians and distributions of vertebrae were not different over the three monthly samples. For simplicity, vertebrae distributions for all months were combined (Fig. 2d).

The combined total vertebra distribution was compared with the total myomeres (minus one) distribution of mid-May and of early July (Fig. 3). Medians were not different for mid-May, but were different for early July. Both tests for differences were significant for the July comparison, partly a result of the difference in medians. However for the May comparison, the Kolmogorov-Smirnov test was not significant while the chi-
and early July reflect two cohorts of a single year class spawned at different times. Jude et al. (1976) first hypothesized the presence of these cohorts based on length-frequency data of larvae collected at the same site. They suggested that the earlier cohort was spawned in a small lake (Pigeon Lake) adjacent to Lake Michigan and grows rapidly in spring. In 1980 (this study) recently hatched larvae (5 to 6 mm TL) were collected in early May in Lake Michigan when Pigeon Lake water was between 15 and 17°C, whereas in Lake Michigan the nearshore bottom (where incubation would take place) was first approaching 8 to 11°C, the usual temperature range at which spawning begins (Scott and Crossman 1973). At those temperatures in Pigeon Lake incubation should have been brief, about 10 days (Hokanson and Kleiner 1974). Larvae collected in May were presumably represented those spawned in Pigeon Lake, or other warm inland areas, which washed out into Lake Michigan proper. Spawning may have just begun in Lake Michigan proper at this time. Only six larvae were collected in June because most of the early cohort had grown to a size not vulnerable to the sampling gear. Two of the five taken in mid-June were recently hatched, indicating the presence of a second, later cohort. Early July samples contained the most larvae and all sampling periods reflected equal collecting effort. Bottom temperatures in Lake Michigan ranged from 8 to 12°C in mid-June when the second cohort was hatching. This low temperature should have extended the incubation period to approximately 18 to 28 days (Hokanson and Kleiner 1974), suggesting, again, a spawning time during May in Lake Michigan. Insufficient numbers of larvae were collected at mid-July for analysis. This, again, was due to growth beyond a size at which they were vulnerable to the gear.

The difference between the incubation temperatures for the early and late cohorts is probably responsible for the observed differences in postanal and total myomere number. (Resh et al. 1976] invoked this argument to explain similarly unexpected results of vertebral counts. However, the fact that their data include several populations and year classes makes proof of this hypothesis difficult). Hubbs (1924), and many others, since, have found an increase in mean meristic elements in colder waters (Jordan's rule). Unlike most others, the present example shows a (probably) thermally induced phenotypic difference in a natural situation, within a very small geographic area, and within a single year class of a (probably) single genetic stock. Additionally, the observation that environmentally induced variability was manifested in caudal segments reiterates the high variability in posterior elements observed by Hubbs (1924) for vertebrae and by Fuiman (1979) for myomeres.

The correspondence of total myomeres (minus one) with total vertebrae (Fig. 3) identifies the juveniles collected in August (59 to 61 mm TL), September (61 to 84 mm), and October (72 to 96 mm) as members of the early cohort. A single, 59-mm juvenile taken in October possibly signaled the approach of the late cohort to a size which was vulnerable to trawling.

Correspondence of total myomeres (minus one) with total vertebrae, and lack of change with posthatching growth does not allow for loss of myomeres or myosepta in the process of the hyphenal complex. Larvae with a completely formed caudal fin exhibited myomere counts on or near the node for their sampling period. Further, dissection of adult yellow perch showed a distinct, final myoseptum on the urocentrum followed by a single myoseptum which covered the structural base of the caudal fin. Within an individual, the number of myosepta exactly equaled that of vertebrae and of myomeres (minus one).

These findings may be extended to most other teleosts. Vertebral modification associated with the Neberian ossicles of ostariophysians does not alter myoseptum or myomere number. Inclusion of four Weberian vertebrae is necessary in order for total myomere (minus one) distributions to correspond with total vertebrae distributions (unpublished results). Further, I have not found myomere number to change with total length in any of
Fig. 2. Frequency distributions of vertebrae for juvenile yellow perch collected in August (a), September (b), October (c), and all months combined (d).

Fig. 3. Relative frequency distributions (as % of sample total) of total myomeres (minus one) (shaded histograms) and total vertebrae (open histograms) for yellow perch.

12 North American cyprinoid fishes. More primitive teleosts often incorporate several vertebrae into caudal fin supports (ural centra). This does not appear to modify the number of myosepta. Myosepta matched total vertebrae.

In dissected adult lake trout (Salvelinus namaycush),

Taxonomic implications are clear. Myomeres are potentially good characters because they do not change during posthatching stages. They can be counted easily and accurately with transmitted light and polarizing filters, until the opacity of the specimen obscures them (usually late in the larval period). Preanal myomere number is remarkably resistant to environmental modification and its elements are unambiguous (as the final few postanal myomeres may be). Further, total myomere number (minus one matches vertebra number, for which data are abundant, except that vertebrae tend to display less variability. This allows for a confident prediction of myomere number for species in which it is not known.

Descriptions of larval fishes are published frequently wherein myomere data are divided among sizes or developmental phases. This often needlessly reduces the accuracy of the portrayal of the true distribution in a situation where there are often too few specimens to have their statistics divided among three or more size classes. Such a presentation might be useful as a taxonomic tool in itself for a species in which the anus migrates. However, this occurs infrequently.

The discordance among literature reports of myomeres and those of vertebrae for a species probably results from several circumstances. Incubations temperature is an important modifier of vertebra number and as such must be considered when reviewing results based on laboratory-reared specimens. Differences in definition of, and tools for making, myomere counts are contributing factors.
Segments can be overlooked easily without the use of polarizing filters. For example, many of the counts reported by Fish (1932) were for several units fewer than modern counts of myomeres or vertebrae, perhaps because of her lack of necessary equipment. Most authors omit the most anterior and the urostylar segments when counting myomeres, further contributing to error. As a result of the comparisons made in this study, I recommend that these segments be included in future accounts since they are, indeed, muscle segments.

Beyond investigator error, there is a real difference in variance between myomere and vertebral distributions, there being less variance in vertebrae. This observation is strongly suggested by the data and tests presented here (Fig. 3) and by unpublished data for several other species. This difference suggests that some individuals may be selected for during the first summer of life. This selection may favor a trait not directly related to vertebral number but somehow linked with it; or some optimal number of body segments may confer greater fitness to an individual (such as enhancing locomotory capabilities, as suggested by Lindsey 1978). Bailey and Gosline (1955) suggested that there is, indeed, environmental selection for a certain number of vertebrae. Perhaps this selection is operating during the larval period.

ACKNOWLEDGMENTS

I thank D. J. Jude and the fish crew of Great Lakes Research Division for providing the specimens used in this study. Dr. Jude also offered constructive criticism of early drafts of this paper. C. L. Johnsrude, University of Michigan, provided an interesting perspective from which to view the results of this study. Discussions with him were quite stimulating. Finally, I appreciate the comments of an anonymous referee who discovered several inadequacies in the original investigation and the manner in which it was presented.

LITERATURE CITED


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Influence of Background Color and Intensity of Illumination on Melanophore Expansion in Larval Fish

Pamela J. Mansfield
Alan H. Mansfield

ABSTRACT

Larval and juvenile spotfin shiners (Notropis spilopterus) were exposed to three levels of illumination and the background colors of white, tan and black. These conditions were maintained for 1 to 4 weeks, at which time the fish were removed and examined under a dissecting microscope. Melanophores on a defined region of the head were counted and their condition assessed as expanded, intermediate or contracted. An index of expansion was calculated by subjectively assigning values of 1, 2 and 3 for each contracted, intermediate and expanded melanophore, respectively. These values were averaged for each individual to obtain a measure of expansion independent of the total number of melanophores. Results indicated greater melanophore expansion under darker backgrounds than lighter backgrounds. This relationship was more pronounced under bright illumination than under dim light. With respect to illumination, melanophores appeared to expand slightly more under dim illumination than bright. Under bright illumination, indices of expansion were 1.7 over white background, 2.1 over tan and 2.5 over black; for moderate illumination, 2.3 over white, 2.3 over tan and 2.5 over black. Analysis of variance showed that background color was a significant factor for melanophore expansion, but intensity of illumination was not significant.

INTRODUCTION

Little work has been done on the melanophore response of larval fishes to differing regimes of light intensity and background coloration. Previous studies of pigmentation changes in fishes have dealt with the amount or distribution of pigment under various conditions in adult fishes, but not physiological pigment changes in larval or juvenile fish (Sumner 1940a; Fujita 1963). It is known that the apparent shape of chromatophores is altered by the movement of pigment granules within the cell (Giese 1979). Movement to the cell periphery produces an "expanded" chromatophore and movement of pigment to the center produces a "contracted" chromatophore. Although the cell itself does not change shape. In this paper these processes will be referred to as expansion and contraction although these terms are not precise. Movement of pigment granules is generally referred to as physiological pigment change, while increase in number of chromatophores or amount of pigment is designated as a morphological pigment change (Sumner 1940a).

Individual chromatophores can be observed clearly at early life history stages of fish and chromatophore condition assessed as expanded or contracted. In particular, melanophores are easily observed, because black pigment is more conspicuous than other colors. Pigmentation changes have been attributed to changes in background color or brightness and to intensity of illumination (Sumner 1940a; Jenkins 1969; Faber 1980). Consequently, we assessed the effects of those two factors on melanophore expansion in one species of larval fish.

METHODS

Two adult spotfin shiners, Notropis spilopterus (Cope), were collected from Honey Creek about 100 m from its junction with the Huron River, Washtenaw County, Michigan. These shiners spawned on the undersides of rocks in a 110- liter aquarium from 20 June to 10 August, 1980. All larvae studied were offspring of these two fish, minimizing genetic variation. Rocks with attached eggs were moved to another aquarium with water from the original. Larvae were reared to the larval stage (Taylor 1976) and some to juveniles; all had numerous melanophores.

Prior to the experiment, larvae were kept in an aquarium with sand-colored gravel in a room with indirect sunlight and a few hours per day of artificial light.

Mortality prevented the use of larvae all the same age in the experiment. Five fish were stocked in each tank but data from only four fish were analyzed. Larvae were from five different spawnings which hatched from 12 July to 18 August. Thus, each tank contained larvae and juveniles that ranged in age from 3 weeks to 2 months after hatching.

Nine 4.3-liter transparent plexiglass tanks were filled to a depth of 7 cm with water from the original aquarium. The tanks were prepared with background colors of white, tan and black by covering the outsides of the tanks with colored paper. These were arranged in three groups with one background of each color; each group was subjected to a different light intensity by use of two fluorescent lights of different intensities and a box to shade one group. Light meter readings at the middle of the water column were 72.0 microeinsteins (me)/m²/sec (bright light), 21.0 me/m²/sec (medium illumination) and 0.1 me/m²/sec (shaded tanks). The photoperiod was 12 hours of light daily.

Examination of the larvae was begun after 7 days. Observations were always made at the end of the day to maximize effects of the factors studied. It was not possible to make all observations at all times; 3 tanks were required to complete the study. At any given time, larvae from several tanks were examined, rather than completing one tank before beginning another. In this way bias due to length of time larvae were in the tanks was reduced. Each larva was examined under a binocular microscope in a small amount of water until activity slowed due to lack of oxygen. No rapid changes could be seen in melanophore expansion during examination. The melanophores on the opercular and opercular regions of one side of the head were counted. In addition to counting, the melanophores were subjectively assigned an index related to the dispersal of pigment within. A melanophore which appeared as a simple round spot was said to be contracted (index = 1); one with protrusions equal or less than the diameter of the center of the cell (a stellate melanophore) was called intermediate (index = 2); and a melanophore with protrusions longer than the diameter of the center (a reticle melanophore) was called expanded (index = 3) (terminology from Giese 1979). Most larvae examined had melanophores in more than one of these conditions, so the number of melanophores in each condition was recorded for each individual. After examination, larvae were removed from the study tanks.

A mean index of expansion was calculated for each specimen, and the mean for each tank. Juveniles tended to have greater absolute numbers of melanophores, and some juveniles from each exposure were examined to eliminate any bias introduced by age differences. The values thus obtained were used to test the significance of the two factors, light intensity and background color.

A two-factor analysis of variance (Remington and Schorl 1970) was performed to compare the effects of background color and light intensity. Bartlett's test (Steel and Torrie 1960) and frequency plots of the residuals showed that assumptions of normality and homogeneity of variance were not seriously violated.

RESULTS

Melanophore expansion was greater in fish exposed to a dark background than in those exposed to lighter backgrounds (Table 1). Analysis of variance showed this...
relationship to be significant at the 5.0% level (Table 2). Light intensity, however, did not significantly affect melanophore expansion at the 5.0% level. Since illumination and background color were independently controlled, no significant interaction between factors was expected; this proved to be the case. Statistically, then, the response to background color did not vary for each illumination level.

Table 1. Mean melanophore expansion indices for spotfin shiner larvae exposed to three illumination levels and three background colors.

<table>
<thead>
<tr>
<th>Backgrounds</th>
<th>White</th>
<th>Tan</th>
<th>Black</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bright illumination</td>
<td>1.7</td>
<td>2.1</td>
<td>2.5</td>
</tr>
<tr>
<td>Moderate illumination</td>
<td>2.3</td>
<td>2.3</td>
<td>2.5</td>
</tr>
<tr>
<td>Dim illumination</td>
<td>2.1</td>
<td>2.3</td>
<td>2.5</td>
</tr>
</tbody>
</table>

Table 2. Summary of analysis of variance on spotfin shiner melanophore response to background color and intensity of illumination.

<table>
<thead>
<tr>
<th>Source</th>
<th>df</th>
<th>Mean squares</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Illumination</td>
<td>2</td>
<td>8.25</td>
<td>2.23</td>
</tr>
<tr>
<td>Background</td>
<td>2</td>
<td>24.45</td>
<td>6.61</td>
</tr>
<tr>
<td>Interaction</td>
<td>4</td>
<td>0.28</td>
<td>0.07</td>
</tr>
<tr>
<td>Error</td>
<td>27</td>
<td>3.70</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>35</td>
<td></td>
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</tr>
</tbody>
</table>

* Significant (P < 0.05)

Considering only mean values for each tank (Table 1), the difference in melanophore expansion over different backgrounds was more pronounced in bright illumination; the indices displayed a wider range for bright light than for medium or dim. In addition, for white background only, melanophores appeared to expand somewhat more in dimmer illumination. However, neither trend was consistent, nor significant in the analysis of variance. The only significant factor in the investigation was background color.

Although both larvae and juveniles were included in this study, there was no apparent trend in the expansion index with age. Juveniles and larvae were similar with respect to number of expanded melanophores. Behavioral interactions between juveniles and larvae appeared to be minimal, suggesting that melanophore development in larvae was not affected by the presence of juveniles.

**DISCUSSION**

The idea that background color is the most important determining factor for pigmentation changes in fishes was substantiated by our study. One important adaptive pressure on many fishes is to visually merge with the background and thus avoid predation. Therefore those individuals which are least conspicuous have a selective advantage. In this respect, intensity of illumination may not be expected to have much effect on pigmentation change, as long as the fish matches its background. As melanophores of larvae expand, their macroscopic appearance is darker, so a larva with expanded melanophores blends with a dark background.

If the fish and background receive dim illumination, the stimulus and adaptive advantage for chromatophore responses are reduced and background color would not be expected to affect melanophore expansion as much. In this study some differences in expansion under dim illumination were observed, but were not statistically significant.

The more pronounced pressure on young fishes is to visually all melanophores under dim illumination than bright illumination, over white background. In our study, it was not statistically significant, but may represent a trend. In dim illumination, background color may be perceived as darker than its true color, causing melanophore expansion. Experimentally blinded fish also exhibit expansion of melanophores (Pickford and Kosto 1957), which may represent the same action of the background color as light when it cannot be seen by the fish. Fishes kept in a dark container for a long time develop increased melanin content of the skin and a greater number of melanophores (Déloir 1957), a morphological color change consistent with the physiological color changes discussed here.

In fish, control of chromatophores is both by nerve impulses and by hormones (Fingerman 1970, Abbott 1970b). Fujii (1966) reviews several hypotheses concerning the mechanisms of pigment granule movement: gelation and solubilization of cytoplasm, electrophoresis, and function of microtubules as cytoskeletal elements. Melanocyte-stimulating hormone (MSH) disperses melanin in melanophores in the presence of sodium (Na) ions. Several pituitary hormones affect dispersion and aggregation of pigment in teleost fish, however, in many species melanophores are primarily controlled by the autonomic nervous system (Fujii 1969). Incomplete understanding of the complexity of innervation and hormone interactions prevents our relating these mechanisms to melanophore responses in the laboratory.

Previous studies of the effects of illumination on pigmentation are inconclusive. Sumner (1940b) subjected adult guppies, Poecilia reticulata Peters, to four intensities of illumination and two background colors. Melanophore number, rather than melanophore expansion, was used as a measure of pigmentation change. The results for illumination were not statistically significant, while more melanin was present in those fish kept on a black background than on grey. Sumner's results are consistent with ours.

A more recent study on larval golden shiners, Notemigonus crysoleucas, (Mitchell), included observations on dorsal pigmentation (Faber 1980). Larvae collected from an unshaded surface area of a lake had more expanded melanophores than larvae taken from a shaded site. Faber stated that this was an apparent contradiction of Sumner's (1940a) observations that dark background or low albedo causes an expansion of melanophores and light background causes contraction. Apparently the sunlight and its subsequent diffusion in the water produced a background color independent of the substrate, since Faber did not mention a substrate color change. He speculates that an extremely high irradiance of sunlight may cause the contradiction, contrary to our results. Perhaps the shaded area received insufficient light to stimulate an expansion response in the melanophores. There may be other factors operating as well, since many variables are present in the field that are not present in the laboratory. Ultraviolet light, temperature and pH have all been shown to affect melanophore response (Fujii 1969).

Studies of melanophore expansion are relevant to taxonomy. Frequently keys to identification of larval fish use characteristics such as the presence of satellite or dendritic melanophores. Bracken and Kennedy (1967) stated in a taxonomic study that melanophore expansion does not vary in larval fish as much as in adults; however, this may be because larvae are characteristically found in or near the spawning habitat and their degree of melanophore expansion is a result of the background color. It is also demonstrated in this study and by Faber (1980) that larval fishes of a given species may show varying degrees of melanophore expansion under different conditions. Therefore, shape of melanophores is not always a reliable character to use for identification.
of larval fishes.

Preservation of larvae further confounds use of pigment as a taxonomic character. Although we examined live larvae for this study, some were preserved after examination. Differences in melanophore expansion continued to be visible for several weeks in preserved specimens, but pigment gradually faded until melanophores appeared similar between individuals. In larvae preserved in formaldehyde, pigment is often virtually invisible after several years.

ACKNOWLEDGEMENTS

The authors would like to thank Lee A. Fuiman for the initial suggestion of this study, ideas for its development and loan of equipment. Other equipment was obtained from Great Lakes Research Division, University of Michigan, Ann Arbor, Michigan. Partial funding was supplied by Consumers Power Company, Jackson, Michigan. The manuscript was reviewed by Dr. David J. Jude, Nancy A. Ayer, George R. Heufelder, Frank J. Tesar, Lee A. Fuiman and Charles P. Madenjian, all of Great Lakes Research Division. Contribution No. 319 of the Great Lakes Research Div., University of Michigan.

LITERATURE CITED


_____ 1940b. Further experiments on the relations between optic stimuli and the increase or decrease of pigment in fishes. J. Exp. Zool. 83:327-343.
EARLY DEVELOPMENT OF THE GENUS ICTIOBUS (CATOSTOMIDAE)

Bruce L. Yeager and James M. Baker

The early development of the three species of Ictiobus is described from larvae reared under artificial conditions. Hatching sizes of Ictiobus ranged from 5.0 to 6.3 mm. Diagnostic characters for larvae of the genus Ictiobus include a low number of preanal myomeres (26 to 31, very rarely 32), presence of occipital pigment, hatchling size smaller than that of related catostomids, smaller size, and particular stages of development, long dorsal fin base, and gross body morphology. Additionally, Ictiobus lacks distinctly elliptical eyes and a flattened head as found in the genus Carpiodes and has a more defined midventral line of melanophores than in Carpiodes. Despite acquisition of meristic and morphometric data, and observations on gross morphology, sizes at stages of development, and pigmentation patterns, no single definitive character or combination of definitive characters for identification of early buffalo larvae to species was attained.

INTRODUCTION

Buffalofishes, members of the catostomid genus Ictiobus, comprise a major portion of the food consumed by local markets in the central and southeastern United States and are exported to markets in Los Angeles, Chicago, and New York. Despite past efforts to domesticate the buffalofishes in fish husbandry, the major source of these fishes remains commercial trammel netting in lakes, reservoirs, and major tributaries of the Mississippi River drainage. With the ever-increasing multiple use of these major river systems for industrial or power plant siting and disposal of municipal waste, the potential for adverse effects on such a major fishery is great. Early life history stages, particularly the pelagic newly hatched larvae of buffalos, are susceptible to entrainment or transport into areas impacted by industrial heat or wastes. Inability to identify particular taxa of ecologically related species has hampered investigations of larval ichthyofauna. The Ictiobine subfamily, comprised of the genera Ictiobus (buffalofishes) and Carpiodes (carpsuckers), is no exception. This report provides descriptions of the early development of the three species of Ictiobus with comparisons to Carpiodes and other catostomid genera.

METHODS AND MATERIALS

Broodstock of the bigmouth buffalo (I. cyprinellus) were selected from 0.1 and 0.2 ha holding ponds at the Fish Farming Experimental Station, U.S. Fish and Wildlife Service (USFWS), Stuttgart, Arkansas, where they had been overwintered. The buffalo originally were trammel netted from the Arkansas River in Arkansas, as were the black buffalo (I. niger). Adult smallmouth buffalo (I. bubalus) were similarly captured from the Cache River and Bayou DeView in Arkansas.

All fish were imported intrapartumone with 0.2-mg carp pruritides per 0.42-kg (lb) of body weight. Spawning methods of Walker and Frank (1952) were utilized. Incubation of eggs was in McDonald jars supplied with well water at 20 C. Newly hatched larvae were held in stainless steel troughs supplied with well water until transfer to Norris, Tennessee, in plastic bags with oxygenated water. At the Norris wet laboratory facilities of the Tennessee Valley Authority (TVA), larvae were held in 33 x 70 x 10 cm white plastic trays supplied with aerated spring water at 15 to 19 C. Egg yolk and finely ground trout chow were fed to the larvae daily from hatching to 10 days posthatching. Thereafter newly hatched brine shrimp (Artemia) replaced the egg yolk. Young buffalofishes were preserved daily in 10 percent formalin and then transferred to 5 percent formalin buffered to pH 7.0 with ammonium hydroxide. The series were cataloged into the reference collection of the Larval Fish Identification and Information Center, TVA, Norris, Tennessee.

A Wild M-6 stereomicroscope equipped with an ocular micrometer and polarizer was utilized for recording morphometric and meristic data. Characters examined were: total, standard, preanal, predorsal, snout, and head lengths; head depth; body depth at the anus; eye diameter; head width; yolk sac length and depth at hatching; numbers of preanal and postanal myomeres; and numbers of fin rays. Morphology and pigment patterns were described. Pigmentation development was so similar among the three species that while only a description for the smallmouth buffalo is presented herein, it is also applicable to the bigmouth and black buffalos. Definitions of characters follow Yeager (1980). The method of counting myomeres follows Hogue et al. (1976). Terminology utilized follows Snyder (1976). Unless otherwise stated, all measurements mentioned are total lengths. Drawings were made with the aid of a camera lucida. Fin ray counts and squamation were ngated by staining specimens in a solution of 0.2 to 0.3 g 1 methylene blue in distilled water.

SPECIES DESCRIPTIONS

Smallmouth Buffalo, Ictiobus bubalus (Rafinesque).--The series consisted of 40 eggs and 430 larvae and juveniles. Morphometric or meristic data (Tables 1 and 2) were recorded from 138 specimens. Mean egg diameter was 2.3 mm (5 eggs) and ranged between 2.3 and 2.4 mm. The total lengths of cultured smallmouth buffalo ranged from 5.0 to 32.7 mm.

Protolarvae ranged from 5.0 to 9.5 mm (Figures 1a to 1c). Mean total length of 20 specimens at hatching was 5.8 mm (range 5.0 to 6.3 mm). Modal numbers and ranges (in parentheses) of preanal, postanal, and total myomeres (35 specimens) were 30 (26-31), 8 (6-9), and 38 (35-39), respectively.

At hatching the head was strongly to moderately decurved. The yolk sac was club-shaped. Yolk material was pale yellow and no oil globules were present. Auditory vesicles were present (distinct only in dorsal view) and otoloths were visible under polarized light. The oral pit was subterminal and ventral. Eyes were circular to slightly elliptical. The median finfold originated dorsally at the 10th to 13th preanal myomere, was continuous around the urostyle, and extended forward ventrally to the anterior portion of the yolk sac. Pectoral buds were present at hatching. Pectoral buds became pallelike by 6.5 mm (1 day posthatching) and remained so throughout the protolarval phase. Opercles were not developed at hatching and covered only the first two gill arches on late protolarvae (9.5 mm). By 6.5 mm (1 day posthatching) the head was no longer decurved, the mouth was open, and the lower jaw was beginning to form. The lower jaw reached the upper jaw at 7.0 mm and the mouth had moved to a terminal position, with the middle of the upper jaw in line with the middle of the eye. By 8.8 mm the gas bladder began to form and the yolk sac was cylindrical. Yolk absorption was complete by 7.5 mm.
The urostyle was angled up slightly by 9.5 mm and an opaque area formed ventrally signifying the onset of caudal fin ray development. The protolalral phase ended with the formation of caudal rays on specimens 10 mm or greater.

Larvae at hatching had darkly pigmented eyes and a diffuse row of melanophores on the ventral line. Additional protolalral pigmentation patterns developed in 1 to 3 days posthatching (6 to 7 mm). Dorsal pigmentation consisted of a few melanophores on the snout, 15 to 25 on the head and operculum, and a diffuse row from the snout to the urostyle. A midlateral line of 15 to 18 melanophores was present. Internally, the dorsal of the incipient air bladder and gut was pigmented.

Mesolaevae (Figures 1d to 1f) ranged from 9.6 to 23.1 mm. By 9.6 mm caudal fin rays first appeared and by 10.4 mm the hyphural complex was evident. The caudal fin was amarginate by 15.1 mm and the dorsal lobe was slightly longer than the ventral lobe. By 12.8 to 14.2 mm the adult complement of caudal fin rays was present.

Differentiation of the dorsal fin outline was evident by the beginning of the meosalaeval phase. The apex was situated over the 17th to 19th preanal myomeres. Dorsal fin rays appeared by 15.1 mm and the adult complement was attained by 22.2 to 23.6 mm. Appearance of initial anal fin rays varied between 15.1 and 18.0 mm. The adult complement was present by 21.0 to 25.6 mm. By 17.8 to 19.0 mm pectoral fin rays had formed. Ventrolateral folds along the body wall at 12.0 mm were the first indication of pelvic bud formation. By 14.5 mm the pelvic buds were wide-based flaps. Pelvic fin rays appeared by 17.8 to 19.0 mm. At 20.1 mm the only remaining fin fold was along the ventral forerogut.

The mouth became slightly subterminal by 15.1 to 17.8 mm and was distinctly so by 19.4 to 20.1 mm. Counting of the gut began at 16.5 to 16.0 mm. Only one full coil was present on specimens larger than 18.0 mm. By 12.8 mm the second gas bladder chamber appeared as an anterior diverticulum of the primary chamber, and two distinc chambers were present by 14.2 mm.

An indistinct midlateral row of melanophores, situated between the two rows described on protolaevae, formed and became increasingly concentrated throughout the mesolalral phase. The nape, head, and snout became heavily pigmented. Ventrally, a diffuse row of melanophores extended from the branchiostegal region to the dorsal fin. The branchiostegals and gills had scattered melanophores. Scattered melanophores were on the cheek, head, gill, soral, and ebral fin rays of the yolk sac ventrally to the pectoral.

The midlateral line of pigment became more distinct and extended from the opercle to the caudal fin base. Caudal fin rays were stained with melanophores. Progressive pigmentation of fin rays to the distal fin edge occurred on the dorsal, anal, and pectoral.

The meosalalral phase (Figure 1g) began with the attainment of the adult complement of median fin rays at 22.2 to 23.6 mm. Adult pelvic and pectoral fin ray counts were attained at 23.6 to 26.1 mm and 27.5 to 30.6 mm, respectively. Median fin fold absorption was completed by 28.0 to 30.6 mm. The mouth remained subterminal and ventral throughout meosalalral development.

During meosalalral development, dorsal pigmentation extended down to or below the midline. The middorsal row of melanophores became more distinct. It extended from the head to the dorsal fin, separated into two rows around the dorsal fin, and was present as a scattered row posteriorly. The double dorsal row became faint. A triangular concentration of melanophores was situated over the midbrain.

Melanophores were widespread along the lateral body. The pigmentation graded to less dense from the dorsum to ventral of the body. The areas around the base of the pectoral fins, belly, and breast were devoid of pigment.

A double ventral row of melanophores was present from the anus to the base of the caudal fin. All median fins were pigmented to the distal edges.

Upon complete absorption of the median fin fold and attainment of the adult complement of rays in all fins, the juvenile period (Figure 1h) began at 27.5 to 30.6 mm. The mouth was subterminal at ventral fin ray counts appear in Table 2.
Figure 1. *Ictiobus bubalus*: (a) protolarva 5.6 mm, (b) protolarva 6.6 mm, (c) mesolarva 10.4 mm, (d) mesolarva 17.8 mm, (e) metalarva 21.5 mm, and (f) juvenile 29.1 mm.
Though more concentrated and dense, melanophore patterns on early juveniles resembled those of metalarval specimens.

Bigmouth buffalo, *Ictiobus cyrinellus* (Valenciennes).--The series consisted of 579 eggs and 486 larvae and juveniles. Morphometric and meristic data (Tables 2 and 3) were recorded from 212 specimens. Mean egg diameter was 2.0 mm (20 eggs) and the range was 1.9 to 2.1 mm. Specimens of cultured young ranged from 5.0 to 76.1 mm.

Protolarvae (Figure 2a to 2c) ranged from 5.0 to 9.4 mm. Mean length at hatching was 5.6 mm (20 specimens, range 5.0 to 6.3 mm). At hatching the head was decurved over the bulblet, club-shaped yolk sac. Yolk material was pale yellow and granular. The mouth was not open. Pectoral fin buds were present as wide-based flaps on the anterior yolk sac. No other fin differentiation was evident. The median fin fold originated at the 10th to 13th (12th modally) preanal myomere, was continuous around the urostyle to the anus ventrally, and extended forward onto the yolk sac. The urostyle was straight. Otic vesicles were evident but otoliths were not visible. Within a few hours of hatching incident gill arches were apparent as tissue folds.

The mouth was open by 1 day posthatching and was subterminal. Otoliths were evident. The head was in line with the longitudinal body axis. By 6.5 to 6.8 mm the gas bladder was filling. Absorption of yolk was completed by 6.7 to 7.3 mm. By 7.3 mm the mouth became terminal and remained so throughout the remaining protolarval phase. Specimens of mesolarvae (Figures 2d to 2f) ranged from 9.4 to 1.2 mm. Initial caudal fin ray formation and flexion of the urostyle occurred between 9.4 and 10.0 mm. By 13.0 mm the adult complement of caudal fin rays was present and the caudal fin, which began flattening posteriorly at 7.5 mm, became truncated and indented. The pyral complex was well formed at this size.

The anterior portion of the dorsal fin was differentiating as a raised portion of the median fin fold by the early mesolarval phase. Anterior dorsal fin rays appeared by 13.9 mm, and the adult complement was present by 21.0 to 23.5 mm. Dorsal and caudal fin folds were separate by 15.3 mm. The anal fin separated from the caudal fin by 15.3 mm and the adult complement was attained by 20.3 mm.

Pelvic buds were evident as tissue folds along the ventral body wall at 1.0 mm, as wide-based flaps by 12.6 mm, and narrow-based fins by 15.3 mm. Pectoral and pelvic fin rays appeared between 6.5 and 6.7 mm. Pectoral and pelvic fin ray development was complete in the metalarval phase.

Throughout mesolarval development the mouth remained terminal. The gas bladder of the gut began by 15.5 mm, and one full gut coil was present by 16.4 to 16.9 mm. The second gas bladder chamber began as an anterior diverticulum of the posterior chamber at 14.2 mm. Two distinct gas chambers were present by 16.6 mm.

The metalarval phase (Figure 2g) began by 22.1 to 22.4 mm. Adult complements of pelvic and pectoral fins rays were attained by 22.1 to 23.5 mm and 22.1 to 24.3 mm, respectively. The remaining fin fold was situated ventrally between the pelvic fins and extended posteriorly to the anus. The mouth was terminal throughout metalarval development. The last vestige of median fin fold was absorbed by 29.4 to 31.8 mm, delimiting the end of larval development.

The juvenile period (Figure 2h) was attained at 29.4 to 31.8 mm with final absorption of the median fin fold. The mouth remained terminal on the largest specimen examined (76.3 mm). Juvenile fin ray counts appear in Table 2.

Black buffalo, *Ictiobus niger* (Rafinesque).--The series consisted of 419 eggs and 502 larvae and juveniles. Morphometric and meristic information (Tables 2 and 4) was recorded from 100 specimens. Mean egg diameter was 2.2 mm (20 eggs) and the range was 2.1 to 2.4 mm. Specimens of cultured young ranged from 5.3 to 25.7 mm. Protolarval specimens (Figures 3a to 3c) ranged from 5.3 to 8.7 mm. Mean total length at hatching was 5.5 mm (20 specimens, range 5.3 to 5.8 mm). Newly hatched black buffalo were characterized by a decurved head, an unformed mouth, a bulblet, club-shaped yolk sac, and straight nuchal. No other fin differentiation was evident. The dorsal median fin fold originated at the 10th to 12th (11th modally) preanal myomere, was continuous around the urostyle to the anus ventrally, and extended forward onto the posterior third of the yolk sac.

By 1 day posthatching (6.1 to 6.4 mm) the head was in line with the longitudinal body axis; otoliths were visible as refracting spheres under polarized light; and gill arches were apparent as tissue folds. The mouth was open and subterminal until 7.2 mm, when it became terminal. By 6.6 to 7.2 mm the gas bladder began to fill. An anterior diverticulum formed from the anterior gas bladder surface by 7.4 to 7.8 mm. Yolk was completely absorbed between 7.2 and 7.5 mm. The tip of the urostyle had flexed and caudal rays formed by 8.8 to 8.9 mm, ending the metalarval phase.

Mesolarval specimens (Figures 3d to 3f) ranged from 8.8 to 17.9 mm. The upper size limit for mesolarvae was not well defined due to the low number of specimens available in larger size classes. Caudal fin rays were first evident between 8.8 and 8.9 mm. By 11.6 mm the urostyle was strongly flexed. An adult complement of caudal fin rays was present by 12.8 to 13.3 mm.

The anterior outline of the dorsal fin became evident as a raised portion of the median fin fold by 8.5 mm. Dorsal fin rays appeared by 12.8 to 13.3 mm and the adult complement was attained by 21.9 mm. Anal fin rays were observed by 15.8 mm.

Pelvic buds were evident as tissue folds along the ventrolateral body wall by 11.5 to 12.6 mm, as wide-based flaps by 13.3 mm, and as narrow-based fins by 17.7 mm. Pelvic fins rays appeared by 17.9 mm. At this size the only remaining fin fold was situated ventrally between the pelvic fins and the anus. Pectoral fin rays appeared by 17.7 mm.

Except on late mesolarvae (15.8 mm or larger) the mouth remained terminal. The upper jaw was even with the lower jaw third of the eye. The gut was still absorbed and all specimens larger than 15.8 mm one full gut coil was present in specimens 17.7 mm or larger. Two distinct gas bladder chambers were present by 13.3 to 14.4 mm. Only two mesolarvae (12.9 and 24.8 mm) were preserved. The adult complement of anal fin rays was present on the 21.9 mm metalarval specimen preserved. Metalarvae (Figure 3g) were characterized by continued pectoral and pelvic fin ray development. The mouth became slightly subterminal and ventral. Nuchal arch was present. Nuchal arch retained a fusiform head shape despite the slightly subterminal ventral mouth.

One juvenile specimen (Figure 3h) was preserved (25.7 mm). Fin ray counts are in Table 2. The head remained smoothly tapered. Squamation was present on both specimens from the caudal peduncle anteriorly along the midline to the area over the gas bladder.
Figure 2. *Ictiobus cyprinellus*: (a) protolarva 5.6 mm, (b) protolarva 6.5 mm, (c) protolarva 7.2 mm, (d) mesolarva 10.3 mm, (e) mesolarva 16.0 mm, (f) mesolarva 17.4 mm, (g) metalarva 21.3 mm, and (h) juvenile 28.4 mm.
Figure 3. *Ictiobus niger*: (a) protolarva 5.3 mm, (b) protolarva 5.5 mm, (c) protolarva 6.5 mm, (d) mesolarva 9.0 mm, (e) mesolarva 9.2 mm, (f) mesolarva 15.2 mm, (g) metalarva 24.8 mm and (h) juvenile 25.7 mm.
A combination of characters, including number of preanal myomeres, size at hatching, size at particular stages of development, and pigmentation patterns, is diagnostic for the subfamily Ictiobinae and the genus Ictiobus. The cumulative range of preanal myomeres for all species of Ictiobinae found in the United States is 26 to 31 (very rarely 32). Only the ranges for Erymynthus melanops 30–33 (Fuiman 1979a), E. sucula 27–29 (Fuiman 1979b), Minytrema melanops 31–35 (Hogue and Buchanan 1977), and Myxostoma erthyrum 31 to 37 (Fuiman and Whitman 1979), and M. macrolepidotum 30–35 (Buyuk and Mohr 1979) overlap those of the Ictiobinidae.

Larvae of Ictiobus may be separated from those of Ictiobus on the characteristic lack of pigment on the operculum in Erymynthus (Fuiman 1979a) and the comparatively different sizes at particular stages of development. Gross body morphology and pigmentation patterns (Hogue and Buchanan 1977) serve to adequately separate larvae of Myxostoma melanops, which hatch at a size similar to that of Ictiobinidae and occasionally have 32 to 33 preanal myomeres. Myxostoma spp. and Hypentilium nigricans (Fuiman 1979a, 1979b, 1977).

Table 5. Total length (mm) when morphological changes appear during ontogeny of three buffalofishes

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<td>Minytrema melanops</td>
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<td>15.1 (18.0)</td>
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<tr>
<td>Erymynthus melanops</td>
<td>14.2 (16.6)</td>
<td>19.6 (21.0)</td>
<td>27.5 (29.6)</td>
</tr>
<tr>
<td>Myxostoma erthyrum</td>
<td>30.5 (31.8)</td>
<td>39.5 (45.0)</td>
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Fuiman and Whitman, 1979, Yeager, 1980, have published larger sizes than Ictiobinidae and attain similar stages of development at much larger sizes. Once differentiation of the dorsal fin fold has begun, the long dorsal fin base and associated greater number of dorsal fin rays (Table 2) is diagnostic for the Ictiobinidae. Overlap of preanal and total myomeres counts for Ictiobus and Carpio species is so complete (Yeager 1980) as to preclude independent diagnostic value for separating the two genera. However, the more elliptical eye and flattening of the head, characteristic of the Carpio species, allow ease of identification of the genera at sizes greater than 4 mm. Typically the midventral line of melanophores on early protolarval Carpio species (< 8 mm) is more diffuse than that of the buffalofishes.

Despite acquisition of osteological and morphometric data, observations of gross morphology, sizes at particular stages of development (Table 5), and pigmentation patterns, no single character or combination of characters for specific identification of early buffalofishes was obtained. Larval Carpio species and Ictiobus may have slightly subterminal mouths and distinctly subterminal mouths by 20 mm, as opposed to the condition in large Carpio species, which retains a terminal mouth throughout larval development.

The vagueness of distinguishing characteristics of larval fishes in the genus Ictiobus is not surprising. In his study not been possible without the support and valuable knowledge of Mayo Martin and of the U.S. Fish and Wildlife Service. Paul Smith and Roger Whittaker assisted in the culture of larvae. Murrie Grasser provided the line drawings. Support for this research was provided by the Tennessee Valley Authority.

LITERATURE CITED


ACKNOWLEDGMENTS

This study would not have been possible without the support and valuable knowledge of Mayo Martin and of the U.S. Fish and Wildlife Service. Paul Smith and Roger Whittaker assisted in the culture of larvae. Murrie Grasser provided the line drawings. Support for this research was provided by the Tennessee Valley Authority.

DIVISION OF NATURAL RESOURCE OPERATIONS, OFFICE OF NATURAL RESOURCES, TENNESSEE VALLEY AUTHORITY, NORRIS, TENNESSEE 37828.
Comparative Development of Redfin Pickerel (Esox americanus americanus) and the Eastern Mudminnow (Umbra pygmea)

by

Robert Malloy and F. Douglas Martin

Egg stages and yolk-sac larvae of the redfin pickerel (Esox americanus americanus) and the eastern mudminnow (Umbra pygmea) are found in the tidal tributaries of the Chesapeake Bay. These fishes are common, but their early life history stages are poorly described (Jones, Martin and Hardy 1978). We find them to be similar to each other but easily distinguishable, and are presenting some previously unreported information which may have systematic importance.

Both species spawn in the early spring. U. pygmea deposits eggs at the base of shoreline vegetation or in algal mats (Jones, Martin and Hardy 1978), and E. a. americanus disperses eggs over flood plains (Crossman 1962). Although egg development appears to be typical of teleostean embryogenists, we found a unique pattern of movement of oil globules common to both species. Larval development is similar to other members of the suborder teleostei (Kliement and Mraz, 1966; Jones, Martin and Hardy, 1978; and others) and distinguishing characteristics are discussed.

Methods and Materials

Adults of both species were collected from Helens Creek, a tributary of the Patuxent River, in Calvert County, Maryland, using unbaited winnow traps. At the time of collection, the water temperature was 5.5°C. Adults were transported to the Chesapeake Biological Laboratory where eggs from ripe females of each species were stripped into watch glasses and fertilized by adding the milt from a mated testes of an appropriate male. Eggs were incubated in a refrigerator at 1 to 6°C. Although this temperature is below recorded incubation temperatures (Jones, Martin and Hardy 1978; Wang and Kernehan, 1979), it was chosen because gravid adults were captured at these temperatures, and because attempts to rear these species in previous years had inactivated mortality related to temperature of 15°C. Initial observations were made hourly for 24 hours. After that eggs were observed at 2- to 9-hour intervals until hatching. Larval development was controlled at 10 to 12°C and observed twice daily.

Results

Egg development for both species was apparently typical of teleostean embryogenists with some notable peculiarities, one of which was the movement of the oil globules. Oil globules congregated under the developing morula, then dispersed with somite formation. This movement changed the optical density of the yolk and waxes were left by the moving oil globules. These oil globules later reaggregated in the larvae (Figure 1).

The embryonic heart was S-shaped, forming anterior and to the left of the head. Heart beat began within 4 days of fertilization.

Hatching was noticeably different between the two species in our study. U. pygmea hatched yolk first through a small slit in the chorion containing the head and body (Figure 2). This may be an artifact of laboratory conditions, but was consistent, and may have contributed to an 80% hatching mortality rate. E. a. americanus hatch head first, seemingly putting little stress on the hatching embryo (Figure 3). This species had an 80% hatching success.
After one week E. americanus yolk-sac larvae have a dark pigment streak from the eye to the hypural anal fin. The dorsal finfold was pigmented, as are the eyes and branchial arches, and the anal melanophore was distinctive. The heart was folded into the body cavity. The common cardinals and hepatic vein also migrated into the body cavity, and the subintestinal was the only vein on the ventral portion of the yolk sac.

After one week yolk-sac Umbra larvae were heavily pigmented on the head and on the dorsal part of the body. The eyes were also heavily pigmented. Meckel's cartilage was formed, but the head was still partially attached to the yolk sac. Also, the heart was rotated into the body cavity as in Esox.

**DISCUSSION**

The similarities of the embryonic heart and vitelline venous system support the hypothesis of phylogenetic grouping of Esoxidae and Umbriidae. The formation of the heart anterior to and on the left side of the head is unusual. There are two more common positions. The most common is within the pericardial cavity as displayed by Cyprinidae (Penaz and Proks 1976), Percina spp. (Martin, pers. obs.), Microgadus tomcod (Hart 1978a; Hart and Hudson, 1975), and Phoxinus o. occellatus (Nagata and Nishiye 1976). The symmetrical common cardinal veins in Esox are duplicated and appear as a return flow in Physorus crocodilus, Fundulus diaphanus, F. heteroclitus, F. majalis, Lucania parva and Gambusia affinis (Hart 1978a). Illustrations of cyprinodonts show that the heart may sometimes be displaced either left or right, but never far from the midline as noted for our specimens.

Yolk-sac circulation patterns have been offered as potential tools for determining phylogenetic relationships (Soin 1966, Martin and Hubbs 1973). Martin and Hubbs (1973) offered a system of classifying those circulatory patterns. Their system was naive and would have benefited from an examination of Soin (1966) and Kunz (1964). Their basic statements about Esox were based on Ryder (1967) and the pattern illustrated by Ryder differs from the pattern seen in our Esox larvae. Martin and Hubbs state that for Esox there are four main vessels entering the sinus venosus. Ryder's illustration shows what might be interpreted as paired sections of the subintestinal vitelline vein (SVV) plus multiple branches of that may be part of the hepatic vitelline vein (HVV). Our own observations show that the SVV is highly branched and interconnected but not paired and common cardinal veins are present (not indicated by Ryder).

Soin (1966) does not state which species of Esox he examined, but he does not show any branches of the HVV. We do not know if this difference between his description and ours is due to error in one publication or the other or if the differences are real. However, we suspect that the illustration by Soin is based on a larva where the heart is moving off the yolk sac into the permanent position in the pericardial sac. If this is the case, the HVV branches may have already migrated off the yolk and the patterns are consistent between the two studies.

Movement of the oil globules during embryogenesis was reported in the Batfishidae, a Salmoniformes family. Amlstrom (1966) reported similar oil globule migration in Bathylagus and Luerosaurus. The oil globules in these species exhibit the same initial migration under the morula as exhibited by Umbra and Esox.

Identification and separation of U. pygaedar and E. americanus yolk-sac larvae is accomplished through three characteristics. Esox is more heavily pigmented, especially the stripe along the side of the body. Although the body shape is the same, Esox is larger. Finally, the myomere count from yolk-sac to anus in Esox is 12 and in Umbra is 5. Variation for this last character is not adequately known, considering the variability in vertebra number shown in Esox species (Crossman and Buss 1965).
In conclusion the similarity of embryogenesis and larval circulation pattern indicate a phylogenetic relationship within the currently defined suborder Esocoidae. The pattern of movement of the oil droplets suggests a possible relationship to the Bathylagidae; however, not enough is known about oil droplet movements in other groups of fish to state that this oil droplet movement pattern is not a more widespread phenomenon.

ACKNOWLEDGMENTS

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LITERATURE CITED


DEVELOPMENT OF LARVAL POLYDON SPATHULA (WALBAUM)
FROM THE CUMBERLAND RIVER IN TENNESSEE

Bruce Yeager and Robert Wallus

ABSTRACT

Adult paddlefish were collected from the Cumberland River in Tennessee and induced to spawn with pituitary injections. Eggs were cultured under hatchery conditions in McDonald jars at 14 to 19°C and the larvae transferred to aquariums and then concrete raceways at 20-24°C. Eggs hatched from 155 to 166 days after fertilization. Average egg diameter was 3.5 mm. At hatching, larvae were 8.1 to 9.0 mm total length (TL). Paddlefish have 32-35 preanal and 22-26 postanal myomeres respectively. Eyes remained small throughout development. Tooth buds appeared on the jaws, tongue, and palate by 13 mm TL (24 days posthatching). Sensory patches were present by 15 mm TL. All fins except the caudal had adult ray counts by 89 mm TL. The preanal myomere count, relative eye size, and more anterior origin of the dorsal finfold on the paddlefish allow taxonomic separation from all other superficially similar larval. Cannibalism occurred among larvae between 18 and 87 mm TL.

INTRODUCTION

In spite of extensive life history work (Adams 1942; Robinson 1966; Friberg 1972; Pasch et al. 1978) on Polyodon spathula (Walbaum) information concerning morphological development of this species is limited. Wagner (1904), Nachtrieb (1910), Allis (1920), and Larimore (1945) studied the anatomy of adult paddlefish, and Ballard and Needham (1964) provided an excellent study of embryonic stages and larvae through the onset of feeding at about 15 mm TL, approximately 10 days after hatching. Early accounts by Allen (1911), Barbour (1911), Danforth (1911), and Thompson (1933) described a few young paddlefish ranging in total length from 17 to 300 mm. Purkett (1961) described seven newly hatched paddlefish larvae (8.0-3.5 mm TL) and one 29-day-old specimen (47.9 mm TL).

The objective of this investigation was to provide a continuous description of the morphological development of Polyodon spathula from hatching to the juvenile period. This description will complement previously published work and provide information on many as yet undescribed stages and assist other investigators in distinguishing paddlefish larvae from similar larvae of acipenserids, leptoselids, and hiodontids.

METHODS

Between April 19 and April 21, 1978, paddlefish in near spawning condition were captured in 12.7-cm bar-mesh gill nets fished at night in the Cumberland River below Cordell Hull Dam. The morning after capture, broadstock were given interperitoneal injections of macerated paddlefish pituitary. Female paddlefish first released eggs 28 hours after injection of pituitary material. Eggs began flowing freely (about 10,000-35,000 per stripping) 34 hours after injection. Multiple spawnings were obtained with each female stripped about every hour. Eggs were placed in McDonald jars in lots of approximately 100,000. River water at 15°C was used to incubate the eggs for 12 to 15 hours until they were shipped to Cohutta National Fish Hatchery, Cohutta, Georgia, where they were incubated at temperatures ranging from 14.4 to 18.8°C.

Eggs were shipped from Cohutta to Tennessee Valley Authority (TVA) laboratory facilities in Norris, Tennessee, and hatched soon after arrival. Larvae were placed in 114-l aquarium and held for 10 days in aerated spring water and then transferred to a 2.23 m x 6.10 m x 9.11 m concrete raceway supplied with a low volume of spring water. Temperatures ranged from 19.5 to 21.0°C in the aquaria and from 20.5 to 24.0°C in the raceway. An initial food culture, begun with 25 pounds of sheep manure, a bale of hay, 3 ounces of brewer's yeast, and wild zooplankton, was previously established in the raceway. Additional feeding with brine shrimp and zooplankton supplemented the food supply. Developing paddlefish were preserved daily in cold 10 percent Formalin and later transferred to buffered 5 percent Formalin.

Meristic and morphometric data were obtained using a stereomicroscope equipped with an ocular micrometer. Characters examined were total, snout, preanal and postanal lengths, and counts of preanal myomeres, postanal myomeres, fin pterygiophores, and rays. Methods of obtaining lengths and counts were as described by Hogue et al. (1976). Head development and pigmentation patterns were particularly noted. Selected morphometric data were analyzed for allometry from log-log plots. Regression equations were calculated using Bartlett's (1949) method for both variables measured with error. The equations are of the form \( Y = \log b + x \log X \), the logarithmic form of Huxley's (1932) equation for growth, \( Y = bX^x \), where \( Y \) is the size of one body part, \( X \) the size of the other body part and \( b \) and \( x \) are constants. In the log transformed form, \( x \) is the slope of the log-log line and \( b \) the index of the size of part \( Y \) when \( X \) is of unit size. Multiple growth spans (Martin 1949, Fulman and Corazzi 1979) were used to exemplify rate changes.

Experiments examined in this study are part of a series of 159 eggs and 2,857 Polyodon larvae, supplemented with juvenile specimens collected from TVA power plant intake screens. All specimens in this series are cataloged as TV909, OS-21 in the reference collection of the Larval Fish Identification and Information Center, Tennessee Valley Authority, Norris, Tennessee.

RESULTS

Eggs and Hatching.—Early embryonic paddlefish eggs were gray, demersal and very adhesive. Except for the grayish yolk material, embryos near hatching had little pigment. Ventrally, a small dark bar of pigment was observed immediately anterior to the tip of the notochord, and laterally, diffuse bands of melanophores were sometimes present from the nape to the vent.

Immediately prior to hatching, 10 eggs averaged 3.5 mm and ranged from 3.3 to 3.9 mm in diameter. The eggs hatched from 155 to 165 hours after fertilization. Prefetching mortality for the cohort used in this study was approximately 10 percent.

Embryos emerged as small tail first, becoming free of the egg capsule by intense writhing as described by Purkett (1961). Newly hatched larvae averaged 8.4 mm TL (20 specimens, range 8.1 to 9.0 mm TL).
Internal Organogenesis.—A considerable amount of dark gray yolk was present at hatching (Fig. 1d). The post gut was present but empty at this stage. Ballard and Needham (1964) indicated yolkl absorption was complete by 15 mm TL (approximately 10 days post hatching). Our specimens retained yolk material until 17 mm TL, approximately 6 days after hatching. Ballard and Needham’s (1964) detailed description of internal organ development was in accord with that of specimens examined in this study.

Head Development.—At hatching the head was strongly dorsiventral over the yolk sac. Barbel buds were apparent on many larvae. Otic placodes were present but otoliths were absent. Opercular flaps were short, not covering the incipiently formed finfolds. The nasal openings were single and rounded and the stomodaeum if present was not well developed. Eyes were present but small, which is characteristic of this species throughout its development.

By 10 to 11 mm TL (5 days after hatching) the head was in line with the body axis. Barbel buds were present beneath the eyes on all larvae, and the mouth was open. Gill filaments were approximately 0.2 mm in length and were covered by the operculum. By 13 mm TL the gill filaments extended beyond the posterior edge of the opercle. Eyes remained small and pigmentation was present in the optic cups. The nasal opening had become oval as ingrowing tissue began dividing the aperture. Sensory patches were present on the operculum (Fig. 1b).

Tooth buds appeared on the jaws, tongue, and palate by 13 mm TL (2 days post hatching). By 17 mm TL the teeth were strongly recurved. By 15 mm TL anterior and posterior nasal apertures divided by fusion of ingrowing tissue. Cartilage in front of the brain had begun to extend anteriorly, giving the snout a rounded appearance. Sensory patches covered the opercle and head with the heaviest concentration under the eye and ventrally on the snout anterior to the barbels.

At 17 mm TL (Fig. 1c) the barbels were longer than wide and located in front of the eye but even with the anterior edge of the nasal opening. The snout was about 1 mm long. The eyes and nasal openings shifted forward relative to the brain. The opercle once again covered the gills and had lengthened posterior-dorsally beyond the pectoral fin. Auditory pits were visible posterior to the eyes. Sensory patches covered the dorsal and lateral portions of the head, opercle, snout, and rostrum (Fig. 1d). The only portion of the head lacking these sensory rosettes was immediately ventral to the mouth.

At total lengths greater than 24 mm the ventral profile of the laterally viewed head was smoothly tapered; the forward extension of cartilage in the developing snout streamer and the upper jaw. Lateral development on specimens greater than 89 mm TL was characterized by continued growth in length and thinning of the basal width to the adult spathulate form.

Fin Development.—On newly hatched larvae (8.1 to 9.0 mm TL), the median finfold began dorsally at the 15th or 17th preanal myomere and was continuous around the urostyle to the vent and from the vent anteriorly to the yolk sac (Fig. 1a). Dorsally, the marginate nature of the finfold indicates the future position of the dorsal fin (Table 1). Caudal fin differentiation began by 10 mm TL as an opaque area of the finfold. By 13 mm TL the caudal finfold had developed a large, flat, vertical, and anterior portion of the notochord was located much closer to the dorsal margin of the caudal finfold. On some specimens the caudal finfold had attained the caudal notochord characteristic of the adult caudal fin by 17 mm TL (Fig. 1c). By 34 mm TL a distinct notch had developed ventrally near the tip of the caudal fin, giving the distal edge a lobed appearance (Fig. 1e). The fin posterior to this notch had no nasal elements or fin rays until about 145 mm TL.

The adult complement of fin rays was present in each fin long before rays were developed to the fin margin (Fig. 1f). The fins attained adult ray counts in the following sequence: dorsal, anal, pelvic, pectoral, and caudal, with adult complements present in all but the caudal fin by 89 mm (Table 1, Fig. 1g). Segmentation of fin rays was apparent soon after individual rays were discernible. The development of all fins was completed between 145 and 160 mm. Table 1. Fin development of young paddlefish (total lengths in millimeters).

<table>
<thead>
<tr>
<th>Fin</th>
<th>Dorsal</th>
<th>Anal</th>
<th>Pelvic</th>
<th>Pectoral</th>
<th>Caudal</th>
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<tr>
<td>Fin outline differentiation begins</td>
<td>9</td>
<td>11-13</td>
<td>11-12</td>
<td>9-10</td>
<td>12</td>
</tr>
<tr>
<td>Countable fin rays present</td>
<td>26</td>
<td>28</td>
<td>35-36</td>
<td>35</td>
<td>28</td>
</tr>
<tr>
<td>Adult complement of fin rays present</td>
<td>65-70</td>
<td>65-70</td>
<td>79-90</td>
<td>89</td>
<td>145</td>
</tr>
<tr>
<td>Completion of fin ray development</td>
<td>145</td>
<td>145</td>
<td>145-160</td>
<td>145-160</td>
<td>145-160</td>
</tr>
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Pigmentation.—Within 12 hours after hatching the eyes became pigmented, and four or five scattered melanophores formed dorsal to the tip of the notochord. The dark bar ventral to the tip of the notochord disappeared by 15 mm TL.

Between 10 and 11 mm TL (1½ days post hatching) the area of the midbrain became pigmented laterally and the hindbrain dorsally. Dense pigmentation was present both laterally and dorsally along the brain by 15 mm TL. Dorsally, the forebrain was "stitched" with melanophores around the edge. A distinct middorsal line of melanophores divided the forebrain. The head became more densely pigmented dorsally with increasing size.

At about 15 mm TL there was internal pigment on the surface of the notochord anterior to the anus (Fig. 1b) and to the urostyle by 15 mm TL. Melanophores were also present on the myosepta to about the midline. From 15 mm to 24 mm TL, external lateral pigmentation started but became progressively heavier beginning dorsally. Lateral pigmentation along the opercle behind the eye was first apparent at about 24 mm TL; and the snout, ventrum of the forebrain, gill arches, and dorsal of the gut were also pigmented. Midlateral pigment was in blotches along the flanks down to the midline by 24 mm TL, and larvae were only tingly pigmented ventrally. Pigmentation did not change significantly beyond this length. Internal organs were still visible on specimens as large as 135 mm. The rostrum was pigmented from the onset of development.

Behavior.—At hatching paddlefish larvae were capable swimmers, traveling randomly for long distances without pause in activity. As noted by Purkett (1961), 2- to 3-day-old larvae swim incessantly. Sudden changes in direction were common. Larvae of about 15 mm TL were frequently observed swimming with their mouths open wide. Larvae smaller than 25 mm TL tended to stay near the water surface. At larger sizes they remained deeper. A dense phytoplankton growth in the tank hindered observation, but few larvae were noted near the surface. Except in the early morning, larvae of all lengths were seldom observed near the surface.

Active feeding began when larvae had absorbed the yolk (between 16 and 17 mm TL). The larvae readily ingested whatever food was offered, e.g., live brine shrimp, frozen brine shrimp, pulverized trout chow, wild
Figure 1. Larvae of *Polyodon spathula*: (a) 8.9 mm newly hatched; (b) 13 mm; (c) 17 mm; (d) 21 mm; (e) 34 mm; (f) 54 mm; (g) 89 mm.
zooplankton (mostly Daphnia pulex), and stripped bass (morone saxatilis) larvae. The guts of preserved specimens contained all these foods in quantity. Although the paddlefish were readily taking large quantities of other foods, over 100 instances of cannibalism were noted. Larvae of 18 mm TL were first observed cannibalizing other paddlefish.

**DISCUSSION**

Purkett (1961) estimated a hatching time of seven days or less at 65-70 F (18.3-21.1 C) for paddlefish eggs collected from the Osage River in Missouri. Embryos cultured at 14 C by Ballard and Needham (1964) hatched in 9 to 10 days. Under our varied incubation conditions 6.5 to 8.5 days were required from fertilization to hatching.

Early developmental characteristics of the paddlefish in this study corresponded for the most part to those observed by Ballard and Needham (1964). Eggs observed in this study were larger in diameter than those of their study. Development of eggs was more rapid and larvae hatched at a slightly more "advanced" stage; e.g., formation of the stomodeum, tooth formation, and collection of dark pigment in the hindgut was more rapid, though propagation temperatures were roughly comparable.

Paddlefish larvae up to 40 mm TL had modal myomere counts of 33 preanal and 24 postanal, with respective ranges of 22 to 36 and 22 to 26. The range (22-26) and modus number (two) of postanal myomeres is greater than the range (20-22) reported in Hogue et al. (1976) for specimens obtained from the Tennessee River. Specimens collected from the Cumberland River (1977 and 1978) showed modal numbers of postanal myomeres similar to those found in this study.

Fin ray development of the paddlefish (Chondrostei) differs from observations for various Teleosts, e.g., gars (Wild 1933, Allen 1966), bowfin (Dean 1889), or sturgeons (Rudys 1890, Stevens and Miller 1970). The combination of early precyclophore formation, delayed fin ray formation, delayed completion of the distal portion of fin rays, and caudal fin differentiation are unique to the paddlefish compared to development of these features in other Actinopterygii with which the authors are familiar. This is described by Snyder (1900), predorsal finfold, predorsal fin, and snout-to-vent lengths provide means of separating un damaged early protocycles of paddlefish and sturgeon. Beyond a few days post hatching, acipenserids, lepisosteids, and hiodontids are separated with ease on the basis of adult characteristics. The gars (Lepisosteidae) have a greater number of preanal myomeres than the paddlefish (32-35). Lungosee gar from the Little River in Tennessee had 43-46 preanal myomeres (Yeager 1978). Gars (Lepisosteidae) have many larger eyes and a more posterior origin of the dorsal finfold than the paddlefish (Hogue et al. 1976, Snyder and Dobbs 1978).

The only reported instances known to us of paddlefish feeding on other fishes are those of Forbes (1888) and Fitz (1986). We found no previous reports of paddlefish cannibalism. This behavior was first thought to be a response to inadequate abundance and/or type food. However, specimens between 35 mm and 87 mm TL, containing only slightly smaller larvae, were obtained from the raceway. Examination of the guts of other larvae collected from the raceways, and continued robust condition of larvae, revealed that adequate alternative food sources were available and were being utilized.

The development of the paddlefish was characterized by gradual changes in body morphology best illustrated as graphical representation of descriptive mathematical expressions (Fig. 2). Two major changes in the gross morphology of the paddlefish (i.e., caudal fin and rostral development) were manifested along a linear body axis and were uncomplicated enough to be sufficiently specified by the log form of the power regression or simple "allometry" expression (Y = bX). Isometric growth (two body parts growing at the same rate) would be indicated by the slope of the log-log transformed regression, b = 1. Allometric growth (> 1) rather than isometric growth for preanal and postanal development is indicated (Fig. 2) for the paddlefish.

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VENTRAL PIGMENT PATTERNS OF Alosa aestivalis AND A. pseudoharengus LARVAE

Karen Ripple, Phillip Jones, and F. Douglas Martin

ABSTRACT

Ventral pigment patterns of larval Alosa aestivalis and A. pseudoharengus exhibit such overlap and variation that they cannot be used as reliable criteria for distinguishing these congeners.

Three basic patterns in the former and five patterns in the latter species were witnessed in subsamples from Potomac River collections. However, no pattern was seen in more than 65% of specimens of either species.

INTRODUCTION

Ichthypoplankton from the Potomac River were examined in search of a rapid means of sorting large numbers of Alosa species larvae taken together when sampling estuarine portions of the river. Four Alosa species occur in the Potomac estuary (Hildebrand and Schroeder 1928). Larvae of A. aestivalis (Mitchill) and A. pseudoharengus (Wilson) have been abundant in ichthypoplankton collections of recent years, while larvae of A. mediocris (Mitchill) and A. sapidissima (Wilson) have been seen considerably less often and may now be absent from the estuary (personal observation). Alosa species spawn at or above the freshwater/saltwater interface of the Chesapeake Bay and its tributaries throughout the spring and early summer. In addition, Dorosoma cepedianum (Lesueur) and D. petenense (Gunther) larvae are collected in freshwater in early summer (Jones et al. 1978).

Methods of identifying these larvae are time consuming, relying primarily on the location and date of collection, fin ray and myomere counts (Lipson and Moran 1974, Chambers et al. 1976), and pigment patterns (Leim 1924, Wang and Kerney 1979). Fin rays are undeveloped on most larvae sorted. In addition, myomere counts are possible only when the larvae are undamaged by the net. If pigment patterns could be used along with collection data and location to sort these species, handling time would be much reduced.

METHODS

Larvae were randomly subsampled from 1977 and 1981 ichthypoplankton collections from the Potomac River between Stuart Point and Hallowing Point. All larval sizes were not observed due to termination of ichthypoplankton sampling in early June. Pigment patterns of 29 A. aestivalis and 94 A. pseudoharengus larvae were examined and compared. Dorsal and anal fin ray counts (where fins were sufficiently developed), preanal myomere counts, and dorsal fin to vent myomere counts were made to assure positive identification.

RESULTS

The most typical ventral pigmentation pattern for A. aestivalis was observed on 16 larvae (62%) and begins with 3 melanophores along the mid-line of the isthmus between the gills, then 206 melanophores in parallel rows posterior to the cleithrum, a short break, then parallel rows resuming along the junction of the body and the intestine (Fig. 1a). Only 2 larvae (7%) exhibited the pattern described by Leim (1924) in which the melanophores diverge into two straight rows forming a V-shaped pattern between the cleithrum and the future site of the pelvic fins (Fig. 1b). A third pattern (Fig. 1c) in which the diverging rows curve was observed on 6 larvae (23%). Two larvae had intermediate patterns.

A. pseudoharengus larvae exhibit five ventral pigmentation patterns anterior to the pelvic region. One pattern (Fig. 1b) is that described for the species by Leim (1924) and occurred on 9 larvae (12% of 1977 specimens). A second pattern (Fig. 1c) is again similar to A. aestivalis and was present on 48 larvae (65% of 1977 specimens). A third pattern (Fig. 1d), however, is very similar to that reported by Leim (1924) for A. sapidissima. This pattern, consisting of a double curve in the rows of melanophores with the rows then drawing closer together at the posterior end, was observed on 9 larvae (12% of 1977 specimens). Ten larvae had intermediate patterns.

Two additional patterns were observed in 1981 collections of A. pseudoharengus. Sixteen larvae (80% of 1981 specimens) had a pattern beginning with 0-2 melanophores along the mid-line of the isthmus between the gills, followed by 2-4 diverging melanophores, a short break, then parallel rows of melanophores along the junction of the body and intestine. In the other pattern which occurred on 3 larvae (15% of 1981 specimens) the double row of melanophores converge, there is a short break, then the rows diverge. One larva observed had an intermediate pattern.
DISCUSSION

The v-shaped pigment pattern described by Leim (1924) for alewives (he did not distinguish between A. aestivalis and A. pseudoharengus) is only one of many patterns observed in Potomac River populations of the two species. The more typical pattern for A. aestivalis is a line of melanophores along the isthmus, parallel double rows posterior to the cleithrum, a break, and then widely spaced parallel rows of melanophores.

One A. pseudoharengus pattern is very similar to that described for A. sapidissima. Other A. pseudoharengus patterns are nearly identical to those Mansueti (1962) described for putative A. mediocris larvae. In addition, collections made at the same location and same time of year, but in different years, yielded A. pseudoharengus with different pigmentation patterns. Figure 1c shows the more typical pattern among the 1977 specimens and Fig. 1e shows the typical 1981 pattern.

We conclude that ventral pigmentation patterns for larval A. aestivalis and A. pseudoharengus show such a degree of overlap and variation that this character is not reliable or even useful in separating Alosa species as has previously been thought. Time consuming methods based on meristics appear to be the only reliable means of separation.

ACKNOWLEDGMENTS

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LITERATURE CITED


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VARIATIONS IN VENTRAL MIDLINE MELANOPHORE COUNTS ON SOME CULTURED PACIFIC SCULPIN LARVAE

Jeffrey B. Marilave and Victor J. Elderton

ABSTRACT

Variation of melanophore counts was clarified through study of reared developmental series from known parental sources. Postnatal ventral midline melanophores were counted in Ascelichthys rhodorus, Artedius fenestralis, Artedius lateralis, Clinocottus acuticeps, Enophrys bison and Oligocottus maculosus. Significant variation was found among species, between parental groups within a species, and between different age groups of siblings. Diminution in melanophore numbers in late larval stages was related to a particular phenomenon on postanal melanophores. Erroneous identification of these and other larval species may arise from use of either small sample sizes or single parental groups for determining ranges of melanophore counts.

INTRODUCTION

Although ichthyoplankton studies have received increased emphasis in fisheries science, identification of certain species of marine fish larvae remains a problem. Laboratory rearing of larvae from known parental sources has been recommended as a solution to identification problems (Ahlistrom and Moser in press). Richardson (in press) reported that larvae of the Artedius-Clinocottus-Oligocottus group of cottids were among the most difficult to distinguish. Among the characters used for identification of these and other cottids is the count of postnatal ventral midline melanophores (Richardson and Washington 1980). We have examined the variation for this character among species, parental groups within species, and age groups among siblings for six cottid species.

METHODS

Specimens of six cottid species were reared from known, unfixed parental sources at the Vancouver Public Aquarium (Table 1). Most of the laboratory culture was performed prior to the design of this study, so that preserved material was available for some species than for others. Egg masses were collected from the field in advanced stages of embryonic development and were presumed to have experienced physical incubation conditions normal for their species. In all cases, larvae were hatched in 1,000 liter tanks, flat black in color, with overhead incandescent lighting yielding a maximum 600 lumens at the water surface, through-flowing seawater (24-29 ppt salinity, 9-12°C) and were fed to excess daily with newly hatched Artemia salina nauplii. Since all larvae were reared under identical conditions of lighting and background color, any effects of rearing conditions on development of melanophores were expected to be uniform among different species and hatches. Fish larvae were preserved at intervals in 3% buffered formalin of 15 ppt salinity. Age was recorded as days post-hatching. For certain preservation ages from particular hatches, there were no comparable preservation ages from other hatches within the same species, so that these specimens could not be used for statistical analyses. Thus, the total ranges for melanophore counts in three species exceeded the ranges used in statistical analyses for those species (Table 1). Statistical comparisons were made with one-tailed analysis of variance, using the F test at the .05 level of significance.

RESULTS AND DISCUSSION

Significant variation exists in ventral midline melanophore counts for 0-day larvae among all the different species, including the two Artedius species (Figure 1). These differences in melanophore counts for 0-day larvae could not be artifacts of any laboratory rearing conditions, as all these specimens were killed from new hatches from egg masses which had incubated under natural conditions in the field. It should be noted from Table 1 (for larvae of all ages), however, that for all species the total ranges of melanophore counts overlap to a greater or lesser degree, thus limiting the diagnostic value of such counts.

Within each species, significant variation occurred among counts on 0-day larvae from different parental groups (egg masses), with the exception of the two small-sized samples of 0-day Clinocottus acuticeps (Figure 1). Again, it must be noted that ranges for selected hatches may be similar, whereas other hatches may markedly differ in both ranges and means. This finding indicates that caution must be exercised in utilizing melanophore count data based on reared larvae from single parental sources. Overall, only L. bison and O. maculosus regularly yielded counts different from the other species (L. bison counts lower and O. maculosus higher). For O. maculosus, however, it should be noted (Table 1) that lower ranges of counts are reported for more southerly populations (Stein 1973, B.B. Washington personal communication).

There also was significant variation among those counts on older larvae of A. rhodorus from separate

<table>
<thead>
<tr>
<th>Species</th>
<th>No. Egg Masses</th>
<th>Total No. Larvae</th>
<th>Total Range, All Specimens</th>
<th>Range, Specimens Used for ANOVA</th>
<th>Ranges Reported in Other Studies</th>
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<tr>
<td>Ascelichthys rhodorus</td>
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<td>817</td>
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<td>6-32</td>
<td>11-29</td>
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<td>6-32</td>
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<td>13-24</td>
</tr>
<tr>
<td>Artedius lateralis</td>
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<td>273</td>
<td>7-39</td>
<td>7-39</td>
<td>13-24</td>
</tr>
<tr>
<td>Clinocottus acuticeps</td>
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<td>6-32</td>
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<td>Enophrys bison</td>
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<td>10-14</td>
</tr>
<tr>
<td>Oligocottus maculosus</td>
<td>6</td>
<td>194</td>
<td>17-49</td>
<td>19-47</td>
<td>11-20</td>
</tr>
</tbody>
</table>

hatches (Figure 2), but not between 30-day larvae from two hatches of A. fenestralis. Unfortunately, older larvae of similar age were not available for analysis from the other species. Within sibling groups (hatches), there were statically significant developmental changes in melanophore counts (Figure 3) for three species (A. rhodorus - 3 hatches, E. bison - 1 hatch, A. fenestralis - 2 hatches), but no significant changes were detected for O. maculosus (2 hatches). With the O. maculosus, however, samples were only available for preflexion larvae, so changes might occur over longer developmental periods in this species. Generally speaking, newly hatched larvae tend to have low counts which increase during early development, then decrease as the larvae approach metamorphosis (Figure 3).

No explanation exists for the early developmental increase in counts, other than that the melanin may not be fully elaborated in some pigment cells. The decreases in counts during later development, a possibility suggested by Richardson and Washington (1980) were observed to have resulted from an apparent vertical migration along myosepta of melanophores, which later disappeared, leaving gaps in the ventral midline row of melanophores (Figure 4). Elevated melanophores were observed in certain individuals and gaps in corresponding positions observed in siblings of the same and older ages. This phenomenon of melanophores elevated above the ventral midline in areas where gaps later appeared was observed in all species except E. bison and A. lateralis, although gaps were detected in some E. bison.

Another phenomenon, observed in A. rhodorus, occurred as the posterior trunk musculature grew broader. Ventral midline melanophores generally split into a double row, but some only migrated to one side or the other, and

Figure 1. Postanal ventral midline melanophore counts on larvae of 0-days age from different parental sources; histogram bars indicate mean values; vertical lines, ranges; pairs of horizontal dashes, 95% confidence limits; and numbers at bases of histogram bars, sample sizes. Each bar represents a different parental source.

Figure 2. Postanal ventral midline melanophore counts on postflexion larvae from different parental sources: means, ranges, 95% confidence limits and sample sizes indicated as in Figure 1.

Figure 3. Developmental changes in ventral midline melanophore counts within parental sources; dots and vertical lines indicate means and ranges; samples from the same parental source are connected by dashed lines.
CONCLUSIONS

The results of this study indicate that ventral midline melanophore counts vary considerably among parental sources. Thus, descriptions based on larvae reared from a single egg mass may not accurately represent an entire fish population or species. There also appear to be regular developmental changes in melanophore counts, and the data in Table 1 indicate geographic effects on such counts. Finally, the great variation we witnessed in melanophore counts dictates use of large sample sizes representing larvae of all developmental stages.

These conclusions differ from those of Templeman and Sandeman (1959), who found little variation in counts of caudal melanophores between subspecies of Sebastes marinus, as well as no evidence for geographic differences which could be significant for identification purposes. These authors also made no mention of developmental changes in melanophore counts. Regarding developmental changes, the literature review by Fujii (1969) indicates little knowledge of natural mechanisms for migration of melanophores or for elimination of individual melanophores in teleost fishes.

LITERATURE CITED


ABSTRACT

Successful development of eggs and larvae of laboratory spawned red drum was limited to temperature above 20 C. Red drum are fall spawners and temperatures of 20 C or less may be encountered during some years. Year to year variation in year classes are commonly observed and may be partly explained by narrow temperature tolerance of the larvae. Studies of development and growth were undertaken to identify the age at which red drum larvae can tolerate low temperature, and to examine the effects of various temperatures on growth throughout the larval period.

Red drum were spawned at the UT PAML laboratory by manipulations of the temperature and photoperiod cycles to simulate natural seasonal changes. Larvae reared in 1000 ml experimental chambers were initially fed rotifers (Brachionus plicatilis). Artemia salina nauplii were fed to 8 day old larvae (Fig. 1).

Red drum were hatched and maintained at test temperatures for two weeks in experiment 1 (Fig. 2). In experiment 2 larvae maintained at 25 C until day 11, were changed to the test temperatures for two weeks. Growth was measured as increase in standard length per day. Growth rates increased with increasing temperature (within the range tested) and with age.

Red drum larvae were hatched at 25 C and changed to 20 C on subsequent days; after two weeks, measurements of standard length were made. Control fish remained at 25 C for two weeks (Fig. 3).

There was a significant correlation (p < 0.01) between growth rate and age of first exposure to 20 C. Survival rates were greatly increased when larvae were maintained at 25 C through the yolk-sac stage before being exposed to 20 C.

CONCLUSIONS

1. The low survival rate of the early stages of red drum in non-optimal temperatures improved with age.
2. Survival rates were greatly increased when larvae were maintained at 25 C through the yolk-sac stage and first feeding (3 days) before being exposed to 20 C.
3. Growth rate of all ages increased with increasing temperature; length of two-week-old larvae at 30 C was three times that of larvae at 20 C.
4. During the first month, larvae held at 20 C 70% of the time grew at rates equivalent to larvae continuously exposed to 20 C.

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Comparative Efficiencies of 505 and 800 Micron Mesh Nets for Lake Ichthyoplankton

Robert D. Hoyt and Dennis L. Webb

ABSTRACT

Larval fishes were sampled in Rough River Lake, Kentucky, in the spring and summer of 1980 using ichthyoplankton nets of 505 and 800 μm mesh sizes. Nets of different meshes were mounted on each side of the bow of the boat (Fig. 1), and sampled surface strata only. Samples were made weekly from March 28 to July 24, 1980, and twice weekly from April 18 to May 30. Of 152,454 larvae collected, significantly more were taken with 505 mesh than with the 800 (Fig. 2). Of 16 species observed, the bluntnose minnow, channel catfish, banded sculpin, freshwater drum, and an unidentified darter were taken only in the 505 μm and not the 800 μm gear. Greater numbers of larvae per 100 m² of water sampled were taken with the smaller mesh throughout most of the study and with the larger mesh in the latter weeks (Fig. 3). Average total lengths of representative individuals of the six most abundant taxa collected were greater for specimens collected in the larger mesh on each sampling date (Fig. 3). This study was supported by the National Marine Fisheries Service, NOAA, and the Kentucky Department of Fish and Wildlife Resources, under PL 88-309, Project Number 2-358-R.

Fig. 1. Dorsal and lateral diagrams of net mountings.

Fig. 2. Total catch using 505 μ and 800 μ mesh nets.

Fig. 3. Average TL of 6 most abundant taxa.
SEASONAL SPECIES COMPOSITION AND BIOMASS ESTIMATES
OF LARVAL AND JUVENILE FISHES FROM NORTH INLET, SOUTH CAROLINA

Lynn Barker and Richard H. Moore

ABSTRACT

A year-long series of ichthyoplankton collections was undertaken in North Inlet, South Carolina to determine seasonal patterns in species occurrence, relative abundance, and diversity among larval, post-larval and juvenile fishes in a tidal inlet. Over 200 five-minute samples were taken over a 2-week period at three sample transects during each season in 1979. Collections were made using a 0.5-meter 505 μ mesh conical net suspended from a vertical guy-wire. Depth integrated samples were obtained by lowering and raising the net during the 5-minute sample period. Sample volumes were determined by means of a flowmeter in the mouth of each net. Figure 1 illustrates that ichthyoplankton is dominated in the fall and winter by sciaenids and secondarily sparids. During the warmer months engraulids make the greatest contribution to numbers and biomass, although in the summer gobids are also important. It is possible that prevailing northeasterly winds and storm conditions were responsible for the similarity between North and South Jones Creeks in the winter. In the fall, South Jones differed from North Jones and Town Creeks by continuing to display a "summer" ichthyoplankton complement well after the other creeks had changed to their "winter" sciaenid-dominated assemblages. These data illustrate a seasonal transition in the ichthyofauna for the creeks sampled. Inter-creek variations are attributed to differences in geographic location within the estuary, water depth, substrate type, and water velocity.

Fig. 1. Bar graph illustrating seasonality, catch, and biomass for larval and juvenile fishes collected in three tidal creeks.
ABSTRACT

Red drum eggs, larvae, and juveniles were collected in or near seagrass beds in Redfish Bay, Texas from September through December, 1980. Red drum were collected in Halodule and Thalassia beds with a benthic sled and a 1.0 mm mesh zooplankton net. Red drum eggs and newly hatched larvae were observed moving into the grassbeds with tidal currents. Abundance of larval and juvenile red drum in the grass beds ranged from none up to 1.0 per m². Red drum were captured in both day and night samples but more fish were consistently taken in day samples than in night samples. The smallest fish were taken at sites nearest the Aransas Pass. Differences in abundance between sample sites could be related to density of seagrass blades and water depth. Differences in abundance over time could be related to temperature and seasonal changes in water depth. Juvenile red drum moved out of the seagrass beds in late November with the onset of cold (< 16°C) water temperatures.

Figure 1. Location of sampling sites in Redfish Bay.

Figure 2. Total catch of juvenile red drum at each seagrass sampling site during the period 23 September to 2 December 1980. Mean water depth is generally inversely related to total catch. The low catch at site 3 is due to relatively higher current speeds at that site compared to other sites.

Figure 3. Mean standard length averaged over all sampling dates and +/-1 standard error of the mean for juvenile red drum at each seagrass sampling site. Sites are arranged in order of increasing distance from the Aransas Pass. Stations 5 through 8 are all essentially the same distance from the pass.

Figure 4. Mean standard length averaged over all sampling sites and +/- one standard error of the mean for juvenile red drum on each sampling date. Total number caught on each date are shown in parenthesis. Low water temperatures may be responsible for small catches after 3 November.