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Hannah Grice

Lauren Patterson

Celeste Giangiacomo

Morgan Bowen

Bill Davin

*Berry College*, [bdavin@berry.edu](mailto:bdavin@berry.edu)

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## POTENTIAL SPAWNING STRATEGY AND FECUNDITY OF ALABAMA SHAD (*ALOSA ALABAMAE*) FROM THE APALACHICOLA RIVER, FLORIDA

Hannah Grice, Lauren Patterson, Celeste Giangiacomo,  
Morgan Bowen and Bill Davin\*  
Berry College  
Mount Berry, GA 30149  
\*Corresponding author  
bdavin@berry.edu

### ABSTRACT

Alabama shad (*Alosa alabamae*), classified as a Species of Concern by the National Marine Fisheries Service, are native to the northern Gulf of Mexico drainages. Our study estimated the fecundity of 39 female Alabama shad from the Apalachicola River, Florida. Individuals ranged in age from 2-4 years with an average weight of 729 g. Samples (1 g) were cut from the center of each ovary, and oocytes present were counted. Diameters of 50 randomly-sampled oocytes were measured per sample. Oocyte diameters ranged from 0.3-2.7 mm, with a mode of 1.5 mm. The unimodal oocyte diameter percent frequency distribution suggests that Alabama shad spawn multiple times in a spawning season. Total mean ovary weight was  $67.88 \pm 25.90$  (s.d.) g/individual, with average annual fecundity estimated to be  $79,420 \pm 26,896$  (s.d.) oocytes/individual. Our results were consistent with previous Alabama shad fecundity studies out of the Apalachicola River.

**Key words:** Alabama shad; fecundity; oocytes; Apalachicola River

### INTRODUCTION

The Alabama shad (*Alosa alabamae*) is an anadromous species native to the northern Gulf of Mexico drainages (1). Historically, these shad ranged from the Ouachita River, Arkansas, to the Suwannee River, Florida (2, 3). Though the Alabama shad was once common across its range, within the past few decades, its numbers have dwindled (4). In 1997, the Alabama shad was listed as a candidate for the Endangered Species Act list by the National Marine Fisheries Service and subsequently reclassified as a Species of Concern in 2004 (4). A paucity of data regarding the life history of the Alabama shad has prevented it from receiving more urgent conservation efforts under programs such as the Endangered Species Act (5).

The largest remaining breeding population of Alabama shad is located in the Apalachicola River near the Jim Woodruff Lock and Dam, Florida (1). Adult shad 2- to 4-years-old begin to enter the Apalachicola to spawn during late February and reach their greatest abundance in mid to late April (3, 6). Even though estimates of Alabama shad annual fecundity, from the Apalachicola River, have

suggested a range from 26,095-257,655 oocytes, there is a lack of data supporting a potential reproductive strategy utilized by Alabama shad (3, 6, 7).

Modes of reproduction in fishes range from heterochronal to isochronal strategies. Heterochronal spawners spawn multiple times during an extended breeding season (8, 9), whereas isochronal spawners spawn once per season (9). Members of the *Alosa* genus, including the American shad (*Alosa sapidissima*) and Twaite shad (*Alosa fallax*), demonstrate heterochronal spawning (10, 11). Although evidence of heterochronal spawning by Alabama shad sampled from the Choctawhatchee River, Alabama, was reported by Mettee and O'Neil (1), previous studies of Alabama shad fecundity, conducted by Laurence and Yerger (3), Mills (6) and Ingram (7), were based on assumption of annual fecundity. Annual fecundity estimates are only appropriate under the assumption that Alabama shad are isochronal spawners, producing one spawning batch in a season. Therefore, which set of fecundity estimates best describe the actual fecundity of Alabama shad is indeterminate because of uncertainty about the fish's reproductive strategy.

The objectives of our study were to access the possible reproductive strategies employed by Alabama shad and to estimate fecundity. The method of spawning employed by fish can have an effect on the spawning stock and larval size, which decreases as the spawning season progresses (12). Although heterochronal spawners might not benefit from predator satiation as much as isochronal spawners, they do benefit from an increase in fecundity, a spread of predation risk, a spread of larval impact on their prey, and a decrease in the risk of spawning eggs in unfavorable conditions (12). All of these aspects of heterochronal spawning help increase survival rates for young of year under the assumption that spawning adults are able to reach spawning grounds (i.e. uninhibited by dams or other obstructions), the population is not under environmental duress, and food availability is sufficient (12). However, if the population is under some form of environmental stress or is experiencing a decrease in size, heterochronal spawning can lead to an accelerated decrease in recovery rates (13). As such, determining whether Alabama shad are heterochronal or isochronal spawners will help in better understanding the species' life history, and in turn, better address its reproductive needs and its capacity to repopulate.

## MATERIALS AND METHODS

Georgia Department of Natural Resources (GA-DNR) staff collected Alabama shad (N=39) using a boat-mounted electrofisher (Smith Root Type 7.5 GPP). Fish were collected from February to April 2012 during daylight hours in the Apalachicola River south of the Jim Woodruff Lock and Dam. After the fish were sexed, weighed to the nearest 1.0 g, and total length was measured, otoliths and ovaries were removed. The ovaries were stored in Gilson's fluid (14). The otoliths were used to age the fish according to the methods outlined in Ingram *et al.* (15).

Both ovaries from each fish were weighed together and individually to the nearest 0.001 g. Ovary length was measured to the nearest millimeter. After measurements were made, 1-g samples were collected from the center section

of the right and left ovaries, as described in Mills (6), and stored in 70% ethanol. Each sample was cut as a cross-section of the ovary to ensure a representative ratio of oocytes and surrounding ovarian tissue was present. All oocytes were then manually teased from the surrounding tissue and counted with the aid of a dissecting microscope (6). The following equation was then used to estimate the number of oocytes per ovary:

$$E_g = \left(\frac{E_s}{W_s}\right) (W_g)$$

where  $E_g$  is the number of oocytes present in the ovary,  $E_s$  is the number of oocytes in the sample,  $W_s$  is the weight of the sample, and  $W_g$  is the weight of the ovary (6). The number of oocytes from the right and left ovary of each fish was then summed to give the overall fecundity. Total fecundity estimates were regressed against body weight and body length and paired t-tests were conducted to determine if any significant difference existed between the right and left ovary fecundity estimates. After the fecundity estimates were completed, the diameters of 50 randomly-sampled oocytes from each sample were measured to the nearest 0.1 mm. Analysis of Variance (ANOVA) with Bonferroni post-hoc tests were also used to determine if any difference existed between fecundity estimates of individuals at different ages.

To ensure that fecundity estimates derived from the center of the ovary were unbiased, samples from the anterior and posterior of the ovary were collected from eight individuals (N=16) and processed using the same methodologies. The counts from these samples were used to calculate overall fecundity, and an ANOVA was conducted to determine if any difference existed between the anterior, posterior, and center fecundity estimates. All statistical tests were conducted with an alpha of 0.05.

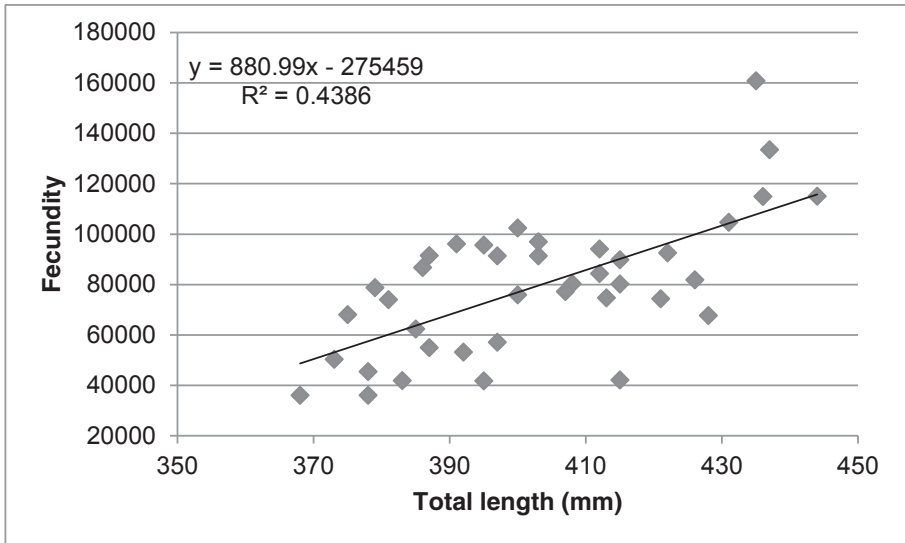
## RESULTS

The total weights of the female Alabama shad ranged from 390-1,026 g, with a mean total body weight of 729 g. The total length of the fish ranged from 368-444 mm, with a mean total length of 403 mm. All but three of the individuals were successfully aged. Most (56%) of the individuals were 2 years of age, followed by 33% at 3 years of age, and 11% at 4 years of age. The total weight of the shad ovaries ranged from 27.88-129.20 g, with the mean total ovary weight at  $67.88 \pm 25.90$  (s.d.) g.

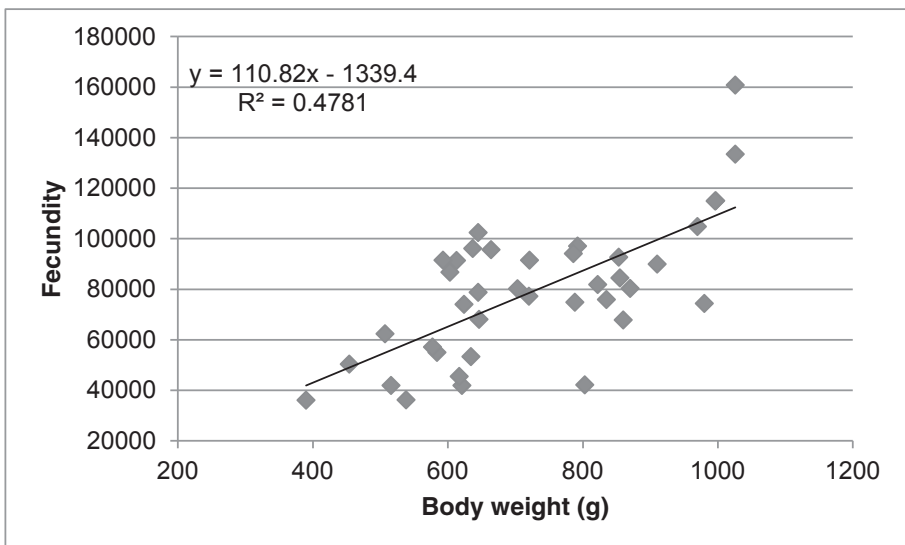
The number of oocytes present in the 1-g center ovary samples ranged from 879-1,996 oocytes, with a mean of  $1,299 \pm 273$  (s.d.) oocytes. The oocytes per ovary ranged from 15,854-88,676 oocytes, with a mean of  $39,710 \pm 14,721$  (s.d.) oocytes ( $1,208 \pm 244$  (s.d.) oocytes/g of ovary). Differences between the right and left ovaries ( $p=0.19$ ) were not significant. Finally, the total fecundity ranged from 36,113-160,813 oocytes with a mean total fecundity of  $79,420 \pm 26,896$  (s.d.) oocytes/individual. Difference between the fecundity estimates derived from the anterior, posterior, and center samples were not significantly different ( $p=0.98$ ).

There was a positive linear relationship between fecundity and fish length ( $R^2=0.4386$ ) and between fecundity and fish body weight ( $R^2=0.4781$ ) (Figures

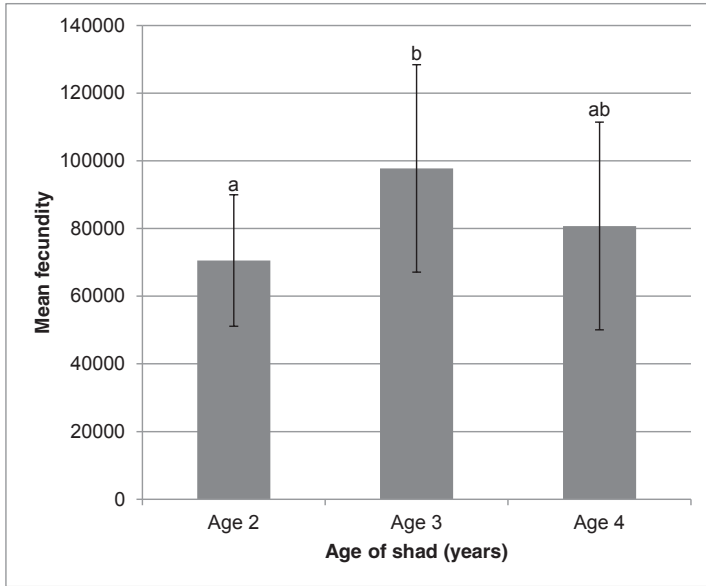
1 and 2). The total fecundity of each individual was also compared to the age of the individual. Individuals at age 3 were found to have higher fecundities than those at ages 2 or 4 years (Figure 3). Significant difference did exist between individuals at 2 and 3 years of age ( $p=0.004$ ), but not between individuals at 2 and 4 ( $p=0.39$ ) or 3 and 4 ( $p=0.35$ ).



**Figure 1.** Fecundity-total body length (mm) relationship for Alabama shad (N=39) in the Apalachicola River, Florida.

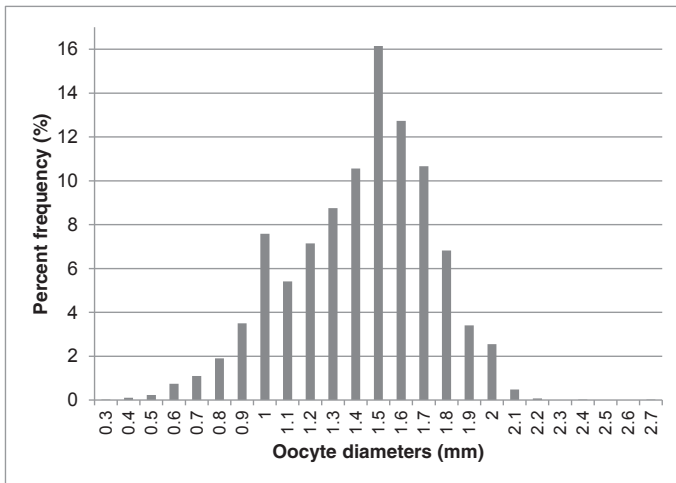


**Figure 2.** Fecundity-total body weight (g) relationship for Alabama shad (N=39) in the Apalachicola River, Florida.



**Figure 3.** Mean total fecundity at varying ages of Alabama shad (N=36) collected from the Apalachicola River, Florida. Error bars represent standard deviation.

Oocyte diameter ranged from 0.3-2.7 mm, with a mean diameter of  $1.4 \pm 0.31$  (s.d.) mm. The percent frequency of the oocyte diameters was unimodal, with a mode of 1.5 mm (Figure 4). A majority (52.9%) of the sampled oocytes had diameters greater than or equal to the mode.



**Figure 4.** Percent frequency of oocyte diameters (N=3,900) from 39 Alabama shad collected from the Apalachicola River, Florida.

## DISCUSSION

The percent frequencies of the oocyte diameters were characterized by a unimodal distribution. This distribution type is common among heterochronal spawners, where most oocyte diameters measure less than 1.6 mm, as larger oocytes have already been spawned (9, 11). The use of Gilson's fluid to store our ovary samples could have affected the oocyte diameters we observed. Gilson's fluid alters hydrated oocytes and causes oocyte shrinkage (9). For instance, a study conducted on Gizzard shad (*Dorosoma cepedianum*) has shown 14% shrinkage in oocytes stored in Gilson's fluid (16). Though shrinkage occurs, it is uniform—differential shrinkage among oocyte size classes was not observed (9). Therefore, the diameter measurements reported in our study might be smaller than unaltered Alabama shad oocytes, but the effects of any shrinkage would not have affected our estimated oocyte diameter frequencies.

The ovaries of heterochronal spawners develop asynchronous oocyte patterns in which representations of oocytes from most stages of maturity are present (17, 18). After one batch of mature oocytes has been spawned, the next batch continues on to the last stage of maturation (hydration) before spawning resumes (9). Because of multiple spawning events, the abundance of eggs within the ovaries at the time of collection may not be an accurate indicator of fecundity, especially later in the spawning season (9). Therefore, batch fecundity, the number of eggs produced for one spawning event, is the only applicable fecundity measurement in heterochronal spawners. Batch fecundity is measured by counting oocytes greater than or equal to the mode diameter, which includes mostly-hydrated oocytes (11, 16).

Our data suggested that annual Alabama shad fecundity in the Apalachicola River ranged from 55,730-180,748 oocytes/kg of somatic weight, with oocyte diameter ranging from 0.3-2.7 mm. This estimate takes into account oocytes of all sizes. In order to correct for batch fecundity, the mode of 1.5 mm was used as these oocytes would have likely been released in the next spawning event. Considering that 52.9% of our oocyte diameters were greater than or equal to 1.5 mm, corrected batch fecundity ranged between 29,481 and 95,615 oocytes/kg of somatic weight. American shad, a North American *Alosa* species most closely related to the Alabama shad, has a fecundity range estimated from 20,226-69,887 oocytes/kg of somatic weight with diameter measurements from 0.4-1.7 mm (11, 19). The diameter and corrected fecundity ranges of Alabama shad are similar to those of the American shad, a known heterochronal spawner, which could suggest a continuing pattern of heterochronal spawning within the *Alosa* genus (11). Further studies must be conducted to say with certainty whether or not the Alabama shad is a heterochronal spawner.

Estimates of total Alabama shad fecundity in our study ranged from 36,113-160,813 oocytes per fish. This range was very similar to the 46,400-149,450 range observed by Laurence and Yerger (3) and the 26,095-208,494 range observed by Ingram (7). The fecundity estimates derived from our study had a narrower range than Ingram (7), but a wider range than Laurence and Yerger (3). Fish in our study were collected over a longer period of time compared to the Laurence and Yerger (3) study, which may explain the wider range of fecundities

found in our study. Fecundity is highly variable over the course of the spawning season, especially in heterochronal spawners, thus studies conducted over multiple years or with greater sample sizes of fish are likely to estimate higher fecundity ranges (9). The studies conducted by Mills (6) and Mettee and O'Neil (1) gathered data over the course of 30 months and four years, respectively, a significantly longer sampling time than our study. As such, wider fecundity ranges, 61,238-257,655 for Mills (6) and 16,477-357,189 for Mettee and O'Neil (1), were demonstrated in both instances as compared to our study. It should also be noted that Mettee and O'Neil (1) collected fish from the Choctawhatchee River, Alabama, as opposed to the Apalachicola River, Florida.

Many fecundity studies, including our own, attempt to calculate the total annual fecundity, which assumes that all mature oocytes are present in the ovary at the time of capture and that no new oocytes mature during the spawning season (9). In our study, we took samples from both ovaries, rather than only one, which is more typical of similar studies. In addition, we took a larger sample of ovary than did other studies that demonstrated that the larger the ovary sample, the less variance in the fecundity estimates (6, 7). Both of these measures should have allowed us to obtain a more precise fecundity estimate.

To address the concern over the potential for differential oocyte distribution throughout the ovary, additional samples were collected from the anterior and posterior ends of the ovary. The fecundity estimates obtained using those samples and the estimates calculated from the center of the ovary resulted in no detectable significant difference. This analysis revealed that even though the gram samples were collected from the center of the ovary, the resultant fecundity estimates were representative of the ovary as a whole. For further support, fecundity studies conducted on other species within the *Alosa* genus also demonstrated that no significant difference exists between fecundity estimates or oocyte diameter size calculated from samples taken from the middle, anterior, and posterior ends of the ovary (17, 18). As such, previous fecundity studies on the Alabama shad have only sampled oocytes from the center of the ovary (3, 7).

These preliminary findings, which suggest Alabama shad spawn multiple times in a spawning season, give some insight into the reproductive strategy implemented by this species allowing for more effective management in the future. For example, it has been found that larger females contribute a greater proportion of eggs to a population's overall fecundity (12). A management strategy to help increase the population would be to limit fishing of large females as a means of preventing recruitment overfishing. In addition, in order for Alabama shad to benefit from multiple spawning events, special care must be taken to ensure individuals can reach their spawning grounds by minimizing obstructions and minimizing environmental stress (*i.e.* pollution). These considerations would allow for the minimization of predation and over-exploitation of food sources by larvae.

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