

Effects of Different Batches of Stripped Eggs on The Reproductive Performances of *Heterobranchus Longifilis*

Abstract

Owing to the fact that about 50% of the total stripped egg of catfish do not survive till fingerling stage during artificial propagation, the need to investigate on the effects of different batches of stripped egg on reproductive performances of *Clarias gariepinus* became necessary. Nine brood stocks of *Clarias gariepinus* (6 male & 3 female) with average body weight of 2.3kg and average length of 64.2 cm were carefully selected for the study. Six matured males were sacrificed for the removal of testis without hormonal inducements. The milt collected from the six males were pulled together and divided into twelve portions each diluted with 2ml of normal saline solution. Three female brood stocks were separately induced at single dosage of 0.5ml/kg body weight and allowed for a period of 10 hours before stripping. Stripping of eggs from each broodstock were in four batches, each batch measured 50g and labeled A, B, C & D. 3g of eggs containing approximately 2000 oocytes (eggs) were measured out from each batch and mixed with the diluted milt and incubated in a 2 x 1 x 10cm³ of water in a concrete pond at temperature of 26°C in three replicates. The result revealed that the reproductive performances considered in this study decreased as the batches of stripped eggs increased from 1-4 batches. The first batch of stripped eggs produced 90.17 ± 0.44, 97.78 ± 0.86, 89.48 ± 1.08 percentage fertilization, hatchability and survival respectively, while the least batch of stripped eggs (batch 4) produced the least value of fertilization (5.83 ± 1.69); 45.00 ± 4.90 hatchability and survival value of 5.59 ± 0.61. Therefore, from the above result, the least batch of stripped eggs should not be used for fertilization since the unhatched or dead eggs has negative effects on the fertilized eggs including fry at the hatchery level.

Keywords: Batches of stripped Eggs, Reproductive performances, Fertilization and *Heterobranchus longifilis*.

1.0 Introduction

Fish remained a universal protein source acceptable at all ages of humanity, easily to come by at affordable cost compared to other sources of protein. Globally, the demand for fish is at daily increased as the world population continues to expand whereas the supply of fish and fish products from the wild is drastically reduced due to environmental challenges and degradations leaving behind aquaculture as the only alternative achieving maximum production of fish to meet the global needs.

African cat fish such as *Heterobranchus* and *Clarias* species remained the most culturable species of significant in Nigeria and beyond (Otoh and Udoh, 2018 a, b; Oyeleye *et. al.*, 2016). This is due to the unique characteristic of these species such as fast growth rate, good taste, generally accepted for consumption, high stocking density, high market price and high resistance to disease and ability to reproduce in captivity (Nlewadim *et. al.*, 2011; Nya *et. al.*, 2017; Udoh and Otoh, 2017; Otoh, 2020 a, b; Otoh *et. al.*, 2023 a, b; Otoh *et. al.*, 2022). Although the growth of fish depends on availability of good feed of which a single feed stuff component cannot achieve, according to Ekanem *et. al.*, 2000, *Heterobranchus* and *Clarias*, readily accept any supplementary feed and their growth rate is unique within a short period of culture (Nlewadim *et. al.*, 2011; Asanaung *et. al.*, 2020) compared to other species.

This species dominates fresh water setting such as stream, lakes and rivers (Adewunmi and Olaleye, 2011). It has high commercial value (Oyeleye *et. al.*, 2016; Shourbela *et. al.*, 2019). Problem associated with fish seed at the natural environment necessitated artificial propagation technique (induced Spawning) under more controlled conditions for a reliable source of fingerlings (Akande and Dieiouadi, 2010). To obtain larvae of acceptable quality during artificial propagation depends mostly on quality gamete which is at times difficult to achieve (Cejko *et. al.*, 2013; Szabo *et. al.*, 2015 and Kristan *et. al.*, 2018). This situation called for intervention of bio technologist for the collection of high-quality gametes, short term gamete storage (Kucharczyk *et. al.*, 2018) and fertilization (Muller *et. al.*, 2018).

Catfish had gained huge recognition in our society hence the need for intensification of breeding technique for mass production of fish seed needed for farm stocks. Low survival of fry at the hatcheries level could be attributed to unidentified factors of which differences in the batches of stripped egg from the brood stocks might not be exceptional (Otoh and Udoh 2019), hence the focus of this study. Practically, it has been observed that all eggs in the gonad do not mature at the same time.

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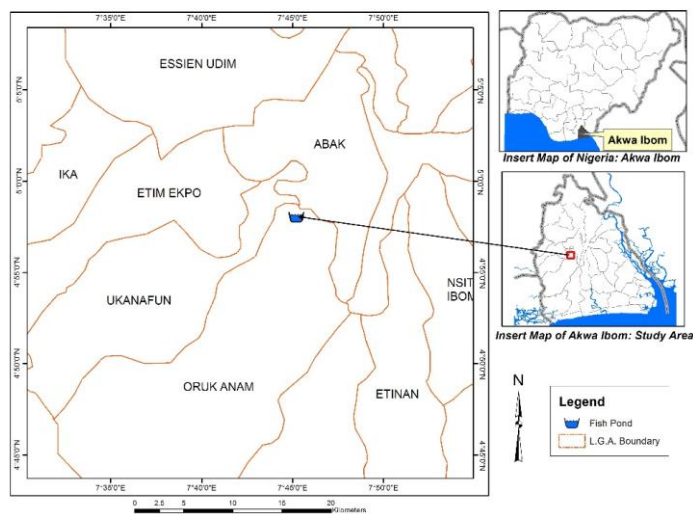
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However, through human intervention with the use of artificial hormones, maturity of entire eggs occurs at the same time and the need to consider if all the mature eggs through human intervention do fertilize, hatched and survived as expected remained very crucial. This is to overcome the danger of mass mortality since the unhealthy eggs cause the mortality of fry at the early stage of development.

2.0 MATERIALS AND METHODS

2.1 Study Area

This research study was carried out in Akwalbom State University (AKSU) fish farm complex, ObioAkpa Campus, Akwalbom State, which is located between latitude 5°17'N and 7°27'N, Longitude 7°27'E and 7°58'E. The study area has an annual rainfall ranging from 3500mm to 5000mm and average monthly temperature of 25°C. Akwalbom State is a coastal state lying between latitude 4°28'N and 5°03'N and between longitude 7°27'E and 8°20'E with a relative humidity between 60 to 70%. It is in the tropical rainforest zone of Nigeria. (Otoh and Udoh, 2019)



Pic. 1: A Map showing the location of the Akwa Ibom State University fish farm complex.

2.2 Acquisition and Care of Brood Stocks

20 matured broodstocks (10 males and 10 females) were separately stocked in a concrete pond at the rate of 2 fish / m² and fed at 5% body weight twice daily for three months using Coppens commercial feed. six (6) sexually mature males and three (3) females with average body weight of 2.3kg and length of 64.7cm were carefully selected according to Otoh, *et. al.*, (2020). Twelves (12) indoor breeding tanks of equal dimension 1x1x1cm³ were used for the study. Water levels and Temperature in each breeding tank was maintained at 30cm³ and 26°C respectively.

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2.3 Hormone Induced Spawning

Six (6) matured male broodstock were sacrificed for sperm removal without hormonal inducement. milt collected from the six sample were pooled together in a plastic container and divided into twelve (12) portions each diluted with 2ml of normal saline solution and preserved separately. Three female breeders were separately transferred to hatcheries for inducement with ovaprim hormone at single dosage of 0.5ml/kg body weight and allowed for a period of 10 hours under the same temperature before stripping manually to obtain eggs (Otoh, *et. al.*, 2020 and otoh, *et. al.*, 2023)

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2.4 Eggs Stripping and Fertilization

Four batches of stripped eggs (50g each) were separately obtained from each of the 3 breeders through gentle pressing of the abdominal region ventrally and labeled A, B, C and D respectively. 3g of eggs containing about 2000 oocytes were measured out of each of the A, B, C and D each mixed with a portion of the diluted milt for artificial fertilization and activated with 100ml of normal saline solution. After 3 minutes, the saline solution was decanted while the fertilized eggs were uniformly spread on the Kakaban (shredded nylon sack) and incubated in aerated indoor concrete breeding 2 x 1 x 10 cm³ at temperature of 26⁰C and replicated three times. During incubation, water levels were maintained at 30 cm³ depth.

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2.5 Fertilization

The colour variations between the eggs were observed, clear and transparent eggs were considered fertilized while dead /white and opaque one was regarded as unfertilized (Udoh 2000 and otoh, *et. al.*, 2023). Based on the counts, the reproductive performances of different stripped eggs were observed such as; percentages fertilization, hatchability, survival and fry production success. Efficiency of these productions was evaluated following the method of Rana (1995).

$$F_s (\%) = \frac{K_f \cdot K_h \cdot K_s}{10,000}$$

Where;

F_s = Success rate (%) of fry production at 10-day post hatching.

K_f = Fertilization rate (%) of eggs

K_h = Hatching rate (%) of fry

K_s = survival rate (%) of 10-day-old swim-up fry

Percentage hatchability was obtained by direct counting of unhatched eggs as well as the numbers of eggs hatched in each incubating tank.

Hatching rate = $\left(\frac{\text{No of healthy fertilized eggs}}{\text{No of fertilized eggs used}}\right) \times 100$ (Hanjavanit, *et. al.*, 2008)

Survival rate (K_s) were calculated during initial feeding according to the following formula

Survival rate = $\left(\frac{\text{number of live larvae}}{\text{total number of larvae hatched}}\right) \times 100$ (Hanjavanit, *et. al.*, 2008)

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2.6 Monitoring of Water quality

Dissolved oxygen and pH of the water were monitored daily using pH meter (VIVOSUN pH Meter) and dissolve oxygen meter (Extech 407510 Dissolved Oxygen Meter) while mercury in glass thermometer was used to take temperature readings.

2.7 Statistical Analysis

Data were processed using Microsoft Excel 2010 for their mean values and presented in graphs. The Data was analyzed using one-way ANOVA at 0.05 significant levels to check the significant difference in fertilization, hatchability and survival rates.

3.0 Results

3.1 Mean Water Quality of the Incubating Tanks

The physiochemical parameters of each of the treatment showed no significance ($P > 0.05$) difference. Dissolved oxygen, temperature and PH measurement ranged between 5.21 ± 0.20 - 5.66 ± 0.20 , (mg/l) 26.25 ± 0.05 - 26.82 ± 0.04 (⁰C) and 6.90 ± 0.02 - 6.95 ± 0.01 respectively.

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Table 1: Mean water Quality Parameters of the Incubating Tanks

| | Stripped egg stages | | | |
|-------------------------------|---------------------|------------------|------------------|------------------|
| | Stage 1 | Stage 2 | Stage 3 | Stage 4 |
| Temperature (⁰ C) | 26.25 ± 0.05 | 26.50 ± 0.01 | 26.80 ± 0.02 | 26.82 ± 0.04 |
| pH | 6.90 ± 0.02 | 6.93 ± 0.01 | 6.95 ± 0.25 | 6.95 ± 0.01 |
| Dissolved oxygen (mg/L) | 5.21 ± 0.20 | 5.61 ± 0.40 | 5.65 ± 0.150 | 5.66 ± 0.20 |

3.2 Effect of Different Batches of Stripped Eggs on the Reproductive Performances of *Heterobranchus longifilis*

The results on the effect of different batches of stripped eggs from the broodstock on reproductive performances of *Heterobranchus longifilis* (fertilization rate, hatching rate and survival rate) is shown in figure 1-3. Results revealed that the percentage fertilization of the first batch of stripped eggs was 90.17 ± 0.44 significantly ($P < 0.05$) higher than the 87.83 ± 0.60 observed in the second batch of stripped eggs, which significantly ($P < 0.05$) increased more than the third and last batch of stripped eggs with the least value of percentage fertilization of 51.83 ± 1.69 recorded for batch 4 (Fig. 1). The percentage fertilization of different batches of stripped eggs showed an interesting pattern in the order of $A > B > C > D$.

The result on the percentage hatchability of different batches of stripped eggs showed a similar trend decreasing significantly ($P < 0.05$) from the first to the least in the order of 97.78 ± 0.86 , 95.83 ± 0.80 , 71.52 ± 1.88 and 45.00 ± 4.90 respectively (Fig. 2).

The result of different batches of stripped eggs from the broodstock on the percentage survival of the fry is presented in figure 3. The percentage survival of fry obtained from the first batch of stripped eggs was 89.48 ± 1.05 significantly ($P < 0.05$) higher than 74.52 ± 1.76 percentage survival obtained from the second batch of stripped eggs. The percentage survival of eggs 41.62 ± 0.67 obtained from the third batch of stripped eggs was significantly ($P < 0.05$) higher than the least value of percentage survival from the last batch of stripped eggs 5.59 ± 0.61 .

The result of this study revealed that the entire reproductive parameters considered in this study significantly ($P < 0.05$) decrease from the first batch of stripped eggs to the least as shown in figure 1-3.

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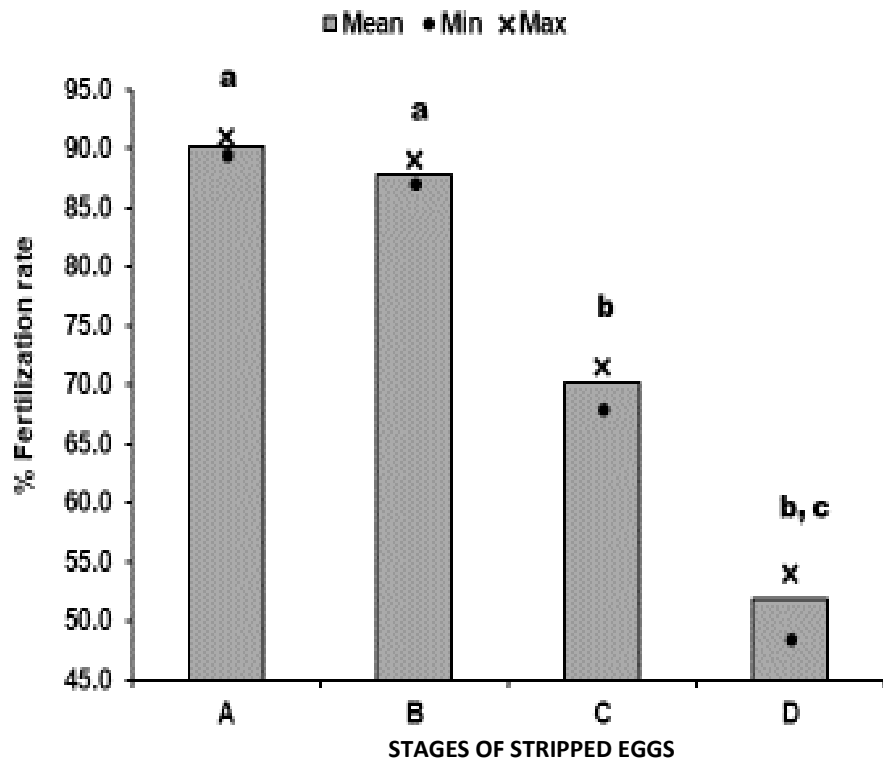


Figure 1: Percentage fertilization of Different Batches of Stripped Egg of *H. longifilis* Brood Stock

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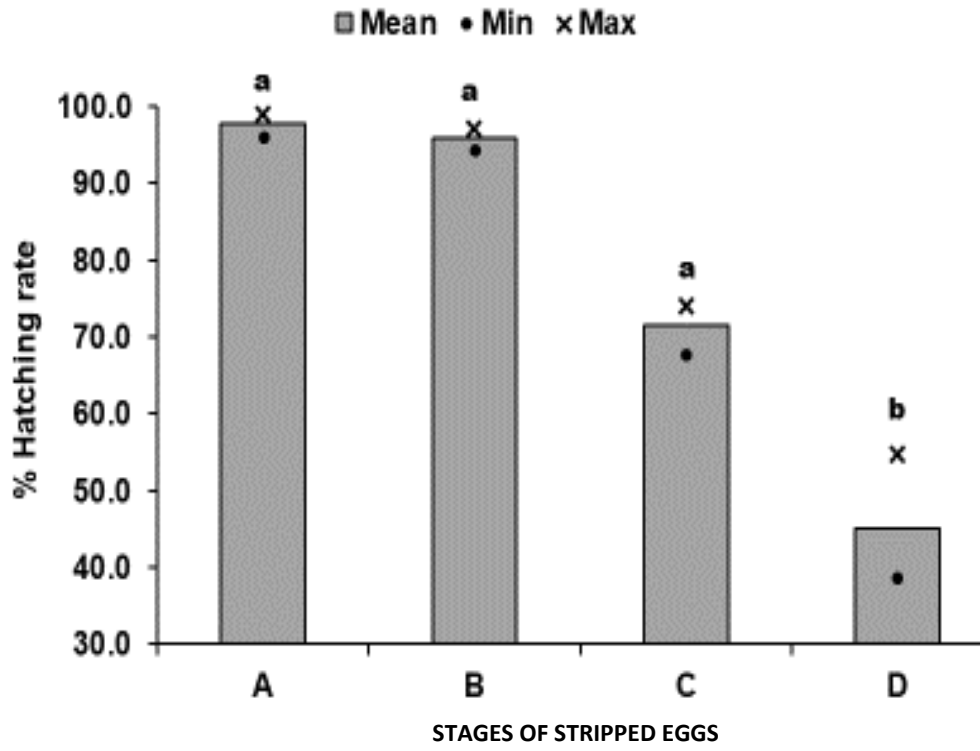


Figure 2: The percentage hatchability of different batches of stripped eggs of *H. longifilis* Brood Stock

UNDERREVIEW

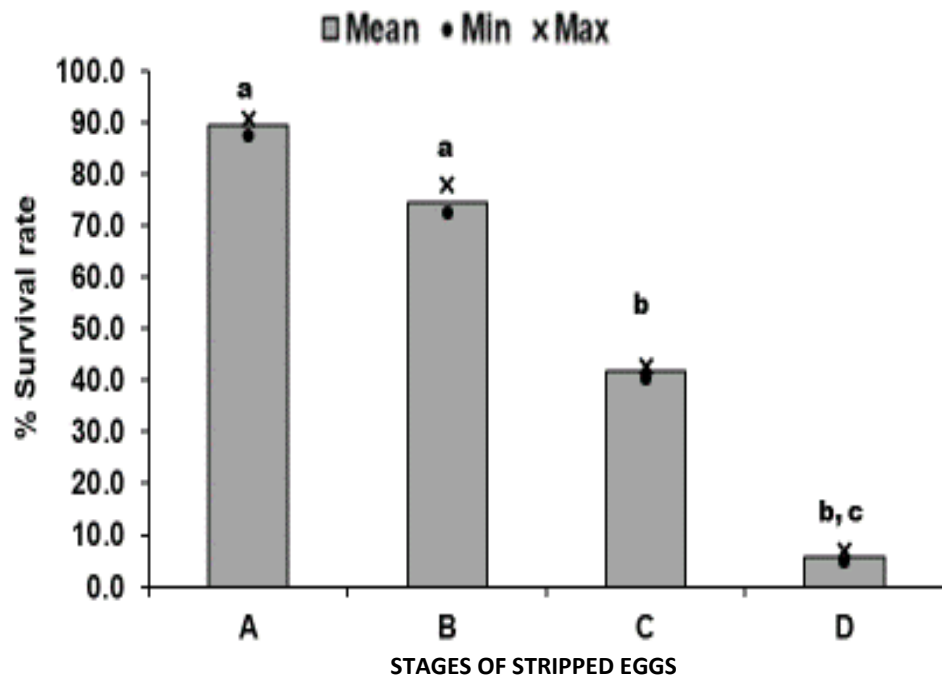


Figure 3: The percentage survival of different batches of stripped eggs of *H. longifilis* Brood Stocks

4.0 Discussion

Water parameters considered in this study were not significantly different among all the batches and were within the recommended range for catfish breeding (Eyo *et al.*, 2003; George and Atakpa, 2015; Jonah *et al.*, 2020).

It has been observed that all eggs in the gonad do not mature at the same time. But in artificial environment under aquaculture system entire eggs in the gonad mature at the same time through human intervention using artificial hormone. In a natural environmental condition, entire eggs are not released at the same time rather in batches based on the stages or levels of maturity. 100% fertilization and hatchability are certain in a natural environment based on the viability of the released eggs, sufficient sperm for fertilization and conducive environmental parameters. This is because breeders in the wild do not release immature egg whereas under forceful maturity of eggs, achievement of 100% fertilization and hatchability is doubtful.

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This study reveals that first batch of stripped eggs show percentage fertilization of 90.17 ± 0.44 significantly ($p < 0.05$) higher than 87.69 ± 0.42 obtained from the second batch of stripped eggs and 70.17 ± 1.09 obtained from batch 3, the least batch of eggs had percentage fertilization of 51.83 ± 1.69 . This result reveals that the percentage fertilization of stripped eggs decreased as the batches of stripped eggs shifted from the first to the least. This result could be as a result of maturity stage and viability of the eggs. The percentage hatchability observed from the different batch of stripped eggs showed a similar trend to that of percentage fertilization with a decreasing pattern observed from the first batch to the least. This result could be attributed to different levels or stages of eggs maturity (Otoh and Nlewadim, 2019). It is observed that although all eggs matured through artificial inducement of hormones, the level of egg maturity differs. The percentage survival of the fry obtained from the stripped eggs reduced significantly as the batches changed from batch 1 to 4 in the order of $A > B > C > D$.

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This study revealed that the first batch of stripped eggs during artificial spawning produced excellent reproductive performances followed by the second batch while the least and the poor reproductive performance was observed in the least batch of stripped eggs.

5.0 Conclusion

Based on the result of this study, it is observed that the reproductive performance of *Heterobranchus longifilis* primarily a function of viable eggs. The results of % fertilization, % hatchability and % survival rate were excellent for the first and second batch of stripped eggs when compared to the third and fourth batch. Upon the findings in this study, it is recommended that only 75% of stripped eggs from catfish broodstock should be used during artificial spawning while the last 25% should be ignored for the security of the entire hatching process.

6.0 Reference

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