

Impact of Ovaprim and Pituitary Gland on the Reproductive Indices and Growth Performance of *Heterobranchus Bidorsalis*

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Abstract

Ovaprim and pituitary gland from *Clarias* was used to ascertain the reproductive indices and early growth of the hatchlings (First Filial) of *Heterobranchus bidorsalis* at the Fisheries and Research Institute Maiduguri. Broodstocks mean body weight males (1200 g) and females (900 g) was collected from fish Dam Baga, a portion of Lake Chad and transported to the national institute for freshwater fisheries research Maiduguri to hatchery complex. The fishes were injected with ovaprim (0.25ml/kg of fish) and pituitary gland (2ml/kg of fish). The fecundity, fertilization, hatchability, survival rates, sperm volume and weight of the testes were estimated after the latency period. While the fry were reared for eight weeks in a complete randomized design setup in an indoor facility to investigate the weight the early growth performance of the fry. The result revealed significant difference ($P > 0.05$) between the fecundity of the females, the fish induces with ovaprim had 11,700 while pituitary gland had 8,000. There was significant difference ($P > 0.05$) between % fertility and hatchability of the eggs. However, the male induced with ovaprim had higher milt volume (3.6 mL). Meanwhile, the fingerlings reared four weeks in an outdoor pond had a significant growth, the fingerlings induced with ovaprim had 15.38 g in weight gain while pituitary gland was 15.62 g. There was no significant ($P > 0.05$) difference between the two hormones on fingerling production. There was no significant difference ($P > 0.05$) between increase in length of fry reared for four weeks.

Keywords: Pituitary Gland; Ovaprim; Reproductive Indices; Growth and *H. Bidorsalis*.

Introduction

Modern aquaculture is aimed at providing low cost, high quality products in accordance to market and consumer demand. Supplying an on-demand consumer product require a reliable and constant production system, which begins with constant supply of eggs and seeds (Dorafshan *et al*, 2003). One of the prerequisites for the establishment of sustainable supply of fish is the capacity to control production process of fish in captivity and to acquire high quality seeds to reduce shortage of seeds for stocking ponds. *H. bidorsalis* second to

in the sustenance of aquaculture development in Nigeria.

Moreover, most of the used synthetic hormones are of high cost such as the human Chorionic Gonadotropin (HCG) (Olaniyi & Akinbola, 2013); Common carp pituitary extract is not available at all time hence the accessibility by the fish farmers are limited (Saidu *et al.*, 2021). Meanwhile, ovaprim which is the most commonly used is effective but costly. Hill *et al.* (2009) previously reported an ovulation increase of 50%, spermatogenic 54%, and minimal mortality after inducing with Ovaprim. However, authors have reported success in the use of ovaprim such as (Hill *et al.*, 2009) used it on Cyprinidae and Characidae, and Cobitidae species by (Yanong *et al.*, 2009). Presently the increase in the exchange rate has led to an increase in the price of ovaprim, this has given rise to the source of an alternative that will give the same quality and output, value, and latency period. Saidu *et al.* (2021); Adebayo & Popoola (2008) conveyed that the non-synthetic hormone from *Clarias gariepinus* is a cost-effective and available alternative for hypophysation and can be prepared in a suspension. This study was conducted to inform on the spawning performance *H. bidorsalis* brood fish in the study area by testing for the effectiveness of Ovaprim and Pituitary gland hormones dosages for induced ovulation and spawning and hatching performance.

Material and Methods

Study area

The experiment was done in the Hatchery Unit of Federal College of Freshwater Fisheries Technology (FCFFT), Maiduguri, Borno State, Nigeria. The College situated at National Institute for Freshwater Fisheries Research Maiduguri Ngala Road and it lies between latitude 11.86218 and longitude 13.22716.

Experimental fish

Six (6) brood fish of *H. bidorsalis* average weight for males 1200g and females 900g in a ratio of 1:2, were collected from fishermen at the landing site of fish dam Baga a portion of Lake Chad and transported to Federal College of Freshwater Fisheries Technology Maiduguri Campus in a plastic jerry can of 50cm x 30cm deep. The brood fish were collected in July 2021 and acclimatized for a month in a 10m² earthen pond in the FCFFT hatchery complex unit. They were fed with a 40% protein diet twice daily at 3% of their biomass.

Collection of the pituitary gland

The broodstocks mean body weight males (1200g) and females (900g), the pituitaries were collected from four (4) males of *Heterobranchius bidorsalis* and *Clarias gariepinus* and put in a mortar and crushed-Two (2ml) of physiological solution was added.

Induced breeding

The gravid females were divided into two groups. The first group were induced with Ovaprim (Shijiazhuang ZDHF Co.Ltd, Hebei) at 0.5mg/kg via intramuscular injection and the second group were induced with 2mg of pituitary extract via intramuscular injection. The male received half dose of both hormones of what was administered to the females. The injection of the two hormones were done at the same time, after which the induced brood fish were kept in an aerated holding plastic tank containing oxygenated water.

Egg fertilization

Stripping of females were carried out after the expiration of the latency period of 10-12 hours for the first and second group respectively. Each group were stripped off of their eggs into separate receptacle and the eggs were weighed immediately to estimate the fecundity was rate. The milt from each male were collected through ablation or surgical operation as described by (Diyaware *et al.*, 2010). The milt volume was estimated using 2 mL syringe after squeezing out the milt from the scrutum into a 100mL capacity beaker.

Incubation of eggs

One hundred (100) fertilized eggs from each treatment were incubated in well-aerated rectangular concrete (1m x 1m x 1m) in three replications in a complete randomized design arrangement. Hatchlings were fed ad-libitum with fish meal. After one week, the pooled weight of fry in each tank was taking using a sensitive balance (Ohaus) to the nearest 0.1g for rearing to assess the early growth performance of the fry, the fry were reared for four weeks.

Fingerling stage

At the end of four weeks rearing, 50 fish (weight and length) three were transferred to nursery pond (3 x 3 x 2.5m) and reared for and other four weeks in two treatment and three replicated in complete randomized design setup. Fish were fed commercial diet 40% crude protein thrice daily at 5% of their biomass. Mortality of the fish from each treatment were recorded.

Reproductive performance indices

Total number of eggs were estimated by counting the number of eggs in 1g of egg mass, multiplied by the weight of stripped eggs Sahoo *et al.*, (2005). Relative Fecundity (RF) and stripping percentage (SP) were calculated according to Onyia *et al.*, (2015), and Tiarniyu *et al.*, (2015).

$$\text{Relative fecundity} = \frac{\text{Number of Stripped Eggs} \times 100}{\text{Body Weight(g)}}$$

$$\text{Stripping percentage} = \frac{\text{Weight of Stripped Eggs} \times 100}{\text{Body weight(g)}}$$

Fertilization (%): The fertilization was estimated using equation (Ochokwu *et al.*, 2016).

$$\text{Percentage fertilization} = \frac{\text{Number of Fertilized Eggs} \times 100}{\text{Total Number of Eggs}}$$

The number of unfertilized eggs were determined when the eggs have developed to the middle gastrula stage (6-8 h after fertilization) by random collection of 50 eggs sample with a sieve from each experimental unit and placed on a petri dish containing water. The samples were then observed under Olympus electronic microscope (Model: Olympus CX-21) at 40 magnification. The number of opaque eggs were regarded as unfertilized while the translucent eggs containing embryonic eyes were regarded as fertilized. The eggs were then returned back to the corresponding unit for hatching (Ochokwu *et al.*, 2016).

$$\text{Hatchability rate (\%)} = \frac{\text{Number of Hatchlings}}{\text{Total Number of Incubated Eggs}} \times 100$$

Care of larvae: Care of hatchlings started from the moment the eggs began to hatch. Separation of fry from the deformed larvae and general sanitation were carried out by siphoning using a rubber hose.

Survival rate (%): The survival rate was determined (Adebiyi *et al.*, 2013).

$$\text{Survival rate \%} = \frac{\text{Total number of survived larvae until day seven} \times 100}{\text{Total number of counted larvae at day one}}$$

Statistical analysis

Data were subjected to one-way analysis of variance (ANOVA). The differences in the means t-test with the aid of SPSS.

Results

Table 1 revealed the fecundity, fertilization, hatchability and survival. *H. bidorsalis* induced with ovaprim had the highest (11,700) fecundity while those induce with Clarias pituitary extract was 8,000. The percentage hatchability for fish induced with ovaprim were (70) respectively while the fish induced with pituitary gland had (52). The survival rate after a week was highest in *Heterobranchus bidorsalis* induced with Ovaprim had 80% and pituitary extract had 60%.

Table 1: Mean (\pm SED or SD) Fertilization, hatchability, and fecundity of *H. bidorsalis* induced with Ovaprim and Pituitary gland

Parameters	Ovaprim (0.5 mL/kg of fish)	Pituitary gland (2 mL/kg of fish)
Weight of the female	900 g	900 g
Latency period/hour	10	12
Fecundity	11,700 ^a	8,000 ^b
Egg diameter	1 mm	1.3 mm
Incubation period	18 -36 hours	18-36 hours
% Fertilization	72.5 ^a	70.155 ^b
% Hatchability	70	52
% Survival	80 ^a	60 ^a

Note: Means with a different superscript in the same row are significantly different ($P < 0.05$)

However, Table 2 showed that the sperm volume which was higher in the males induced with ovaprim had 3.6mL, while pituitary extract had 2.8mL respectively. The highest average length of the testes induced with Ovaprim had 7.25cm, while pituitary extract had 5.3cm. The male induced with ovaprim had the highest average weight of the testes 2.2g, while pituitary extract had 1.55g respectively.

Table 2: Mean sperm volume, the weight of the testes, and milt volume of *H. bidorsalis* induced with ovaprim/pituitary gland

Parameters	Ovaprim	Pituitary Extract
Weight of the male (g)	1200	1200
Weight of left-lobe testes (g)	2.4	1.4
Weight of right-lobe testes (g)	1.8	1.7
Length of left-lobe testes (cm)	7.8	5.8
Length of right-lobe testes (cm)	6.7	4.8
Milt volume (ml)	3.6	2.8

Note: Means with a different superscript in the same row are significantly different ($P < 0.05$)

Table 3 showed that there was no significant difference ($P > 0.05$) in the final weight of fish induced with ovaprim after four weeks (3.11) and pituitary gland (2.76). In table 3, weekly increase in length of *H. bidorsalis* fry induced with ovaprim/pituitary reared. There was significant difference across the treatments in length increase of the fish. The final length for *H. bidorsalis* fry induced with pituitary was (0.06) and ovaprim (0.15).

Table 3: Early growth of fry reared induced with Ovaprim and Clarias Pituitary extract

Parameters	Ovaprim	Clarias Pituitary extract
Final weight	3.11 ^a	2.76 ^b
Initial weight	0.15	0.15
Weight gain	2.96	2.61
Final length	2.15	1.96
Initial length	0.03	0.14
Length gain	2.12 ^a	1.82 ^b
% Survival	60	70

Note: Means on a row with different superscript are significantly different ($p < 0.05$)

Table 4 showed that the increase in the length of the fry reared. At the end of the four weeks of rearing, fish induced with ovaprim had 5.48cm while pituitary had 1 respectively. The survival rate was higher in the fry reared, which was induced with Ovaprim had 70% while pituitary was 64% survival.

Table 4: Growth parameters of the fry reared induced with Ovaprim and Clairas Pituitary extract

Parameters	Ovaprim	Clarias Pituitary extract
Final weight	7.81 ^a	3.26 ^b
Initial weight	0.15	0.13
Weight gain	7.66	3.13
Final length	5.56 ^a	3.08 ^b
Initial length	0.18	2.93
Length gain	5.48 ^a	1.03 ^b
% Survival	70	64

Note: Means on a row with different superscript are significantly different ($p < 0.05$)

Discussion

This study revealed the efficiency and profitability of induced breeding using natural hormone (*Clarias* pituitary extract) and ovaprim. It exposed that there is no significant difference in using synthetic hormone (ovaprim) and natural hormone (pituitary gland) in inducing catfish breeding. In this research, the weight of the broodstock used was males (1200g) and females (800g) and all responded positively to the hormones. Subsequently spawned 10 hours after injecting with hormones at 29°C Saidu *et al.*, (2021)

This showed that all the doses induced ovulation effectively and can cause gonadotropin surge in *H. bidorsalis* (Goos *et al.*, 1996). The result was found different from that of Sharma

et al. 2010), where the total weight of stripped eggs was significantly highest, when females were injected with 1 ml Ovotide per kg body weight compared to 0.6-0.8 mL doses tested on *Clarias batriachus*. The fecundity rate recorded in the present study were above those reported in Anibeza (2013), who obtained average fecundity range of 7,203-56,789 eggs in *Heterobranchus bidorsalis* from Idodo river basin. Offem *et al.*, (2008), recorded a fecundity (5,515-36,800) eggs in brood fish of 102.4 g (14.8 cm) - 1625.5 g (69.9 cm) in *H. longifilis* and the gonads were observed to be ripe in the months of June and July. Nwokoye *et al.*, (2007), recorded fecundity of between 9000-11000 when brood fish of size 300-500 g were used. The disparity with this study could be as a result of the large size of brood fish used in this study. According to Witthames *et al.*, (1995), fecundity may vary within a species, as a result of different adaptations to environmental habitats.

Fecundity has been shown to vary in fish size (age) and condition, larger fish produce more eggs, and for a given size females in better condition exhibit higher fecundity Murua *et al.*, (2003). Fish size and condition are key parameters to consider for proper assessment of fecundity. The stripping percentage recorded in this study was low compared to the findings in Delince *et al.*, (1987), who reported stripping percentage of 10-20% in *Clarias gariepinus*. This could be due to the negligible weight of the gonad compare to the entire body weight of *H. bidorsalis* and the incomplete stripping of whole egg sack.

The percentage of fertilization and hatchability recorded in this research were higher in the fish induced with ovaprim. However, both treatments gave a high result to that effect. This agreed with (Hamid *et al.*, 2005; Ndimele and Owodeinde, 2012; Olaniyi and Akinbola, 2013; Chattopadhyay, 2018); also, Das *et al.* (2016) and Hossain *et al.* (2012) conveyed high fertility in fish induced with ovaprim but Hossain *et al.* (2012) also report high hatchability among the females induced with pituitary extract. Subsequently, the fish induced with pituitary extract recorded high survival (65%) after four weeks of rearing in an indoor pond against the fish induced with ovaprim (62%), this concurs with (Chattopadhyay, 2018).

The growth rate of the fry induced with *Clarias* pituitary and ovaprim reared in a hatchery complex, revealed that there was significant difference ($P < 0.05$) across the rows. Ndimele and Owodeinde (2012) recorded poor growth rate across the treatments when compared with this research, moreover, the highest weight gain (8.88 g) observed in Ndimele and Owodeinde (2012) was in ovaprim induced fish reared for 56 days which was lower than the weight gain obtained in this research (7.66g). The same trend was observed in Abdul *et al.* (2017), who recorded poor growth after feeding the fry for 28 days. Similarly, Ikechukwu *et al.* (2019) recorded an increase in weight of the fish induced with ovaprim and pituitary, He also stated that the fish induced with ovaprim had significant growth which did not differ from those induced with the pituitary. This has proved that the pituitary gland is an alternative for ovaprim which is costly, not easily accessible in the arid zone. The major

causes of poor growth in fish farming are correlated with diet (feed, nutrients contents of the feed the fish consumed), the stocking rate, pH of the water, dissolved oxygen (Ochokwu et al., 2019). Another causative effect is the genetic makeup of the parent stock which is inherited by the offspring (Ochokwu et al., 2015).

Conclusion

The research points out the impacts of using the different hormones that's pituitary gland and Ovaprim. The research revealed that Pituitary extract from species used showed positively impact on the growth performance of the fish species (*Heterobranchus bidorsalis*), it exerted positive change in growth performance and survival rate. Finally, the farmers who depend on capture fisheries for survival because of the poor access to synthetic hormone and the cost of purchasing it can adopt the pituitary gland for inducing breeding, since it is inexpensive. However, in this research revealed that natural hormones produced good progeny and is less expensive, it has good eggs quality. The farmers should also be alert throughout the latency period to avoid under or over-ripening of the eggs after injection.

Acknowledgement

My profound gratitude goes to my supervisors, in persons of Prof. Onyia L.U. and Prof. Sogbesan O.A. whose devoted all their times and attention to ensure proper and accurate result.

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