

Original Article

Spawning Performance of *Heterobranchus bidorsalis* in Sokoto Dry Sub Humid Nigeria

Abubakar M.Y. *and
J.K Ipinjolu

Department of Fisheries
and Aquaculture, Faculty of
Agriculture, Usmanu
Danfodiyo University,
Sokoto, Nigeria

ABSTRACT

Background and Objective: One of the prerequisites for the establishment of sustainable supply of seed is the capacity to control reproductive process of fish in captivity and to acquire high quality seeds to reduce shortage of seeds for stocking ponds. This research determines the effectiveness of Ovaprim and Ovatide hormones for induced ovulation and spawning performance of *H. bidorsalis* brood fish.

Materials and Methods: This study present the effects of different doses of Ovaprim C and Ovatide on induced spawning of *H. bidorsalis* in two consecutive trials. Three doses each of Ovaprim C (0.25, 0.5, 0.75 mL kg⁻¹) and Ovatide (0.1, 0.2 and 0.3 mL kg⁻¹) of the female broodstock body weight were administered to three sets each of randomly selected female broodstock with three replicates each in a Completely Randomized Design (CRD). The best performing dosage of the two hormones were then tried in the subsequent year.

Results: The findings indicated that Ovaprim and Ovatide doses induced ovulation at average latency period of 12 h, and incubation at 23 h at temperature of 26.1-29.0°C. Hormonal treatments with Ovaprim (0.5 mL kg⁻¹) and Ovatide (0.1 mL kg⁻¹) dosages thrived better and similar ovulation, relative fecundity, fertilization rate and hatching rate in the two years trial. The survival rate of hatchlings after a week of rearing, suggested that hatchlings from brood fish induced with Ovaprim C at 0.25 mL kg⁻¹ had the least performance.

Conclusion: It was concluded that *H. bidorsalis* brooders performed better on Ovaprim C at 0.5 mL kg⁻¹ and Ovatide induced at 0.1 mL kg⁻¹ interms of the reproductive indices tested with reference to the study area.

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 seeds¹. One of the prerequisites for the establishment of sustainable supply of seed is the capacity to control reproductive process of fish in captivity and to acquire high quality seeds to reduce shortage of seeds for stocking ponds. *H. bidorsalis* second to *Clarias* spp. in the sustenance of aquaculture development in Nigeria. Sustainability of the contribution would depend on continuous successful reproduction in captivity, because supplies from the wild remain doubtful and the sustainability of the capture fisheries is not assured^{2,3}. It then becomes apparent that management protocols covering egg production, egg hatching, and particularly

*Corresponding Author

production techniques that enhance fry and fingerling survival need to be further simplified to ensure sufficient supplies of the fish seeds^{4,5}. This can be achieved through reproduction which usually rely on hormonally induced artificial spawning⁶. The effectiveness of hormone in induced spawning is controlled by several factors, which include; sex steroids in the regulation of reproductive processes⁷. These processes are controlled through the brain, pituitary and the gonads. The brain is stimulated by environmental cues like water rise, temperature of the environment, feeding, rainfall, photoperiods; type of hormonal therapy; stage of the gonad at the time of hormonal therapy; and latency period between hormonal stimulation and stripping time^{8,3}. The use of exogenous hormone is an effective way to induce final reproductive maturation and produce fertilized eggs to facilitate reliable hatchery operations³. However, the effect of such hormone on the final maturation and ovulation of the fish gonads in relation to the water quality parameters and the environment within which the fish is spawned needs to be known. 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 Ovatide hormones dosages for induced ovulation and spawning and hatching performance.

MATERIALS AND METHODS

This study was carried out at the Teaching and Research Fish Farm, Department of Fisheries and Aquaculture, Usmanu Danfodiyo University, Sokoto, Nigeria in two trials of 2016 and 2017. The site is on latitude 13°07' 78'' N and longitude of 05°12' 25'' E at 275 m above sea level. The site is located in the dry sub-humid illela-sokoto-yelwa plain of Nigeria, with agro-climate characterized by seven long dry months, occurring from October-April of every year, mean monthly maximum temperature of 31-40°C and mean monthly minimum temperature of 12-24°C and evapotranspiration of the order 1670 mm. The area is characterized with cool dry air during the harmattan from November-February and hot season from March-May. Annual rainfall in the area ranged from 508 to 1016 mm/year⁹. The mean relative humidity is 14.9-40% in March and June, respectively¹⁰. The experiment was carried out in two trials. Based on the evaluation of the results of the various doses of hormone in the year 2016 trial, two doses (0.1 mL kg⁻¹ Ovatide and 0.5 mL kg⁻¹ Ovaprim) with the best performance were compared in 2017.

Exogenous hormones: Two hormones (Ovaprim C and Ovatide) were procured from the open market. Ovaprim C (Syndel International Inc., Vancouver, BC, Canada) is a liquid preparation containing 20 µg salmon GnRH analogue and 10 mg of domperidone, a dopamine antagonist per ml. The manufacturer's recommended dose is 0.5 mL kg⁻¹ of broodfish body weight. Ovatide hormone is a synthetic analogue of the peptide hormone Salmon GnRH and dopamine antagonist dissolved in a mixture of aqueous and organic solvents. The hormone's recommended dose by the manufacturer is 0.2 mL kg⁻¹ of female's body weight. Ovatide is manufactured by Hemmo Pharmaceuticals Pvt. Ltd., Mumbai, India.

Experimental design and set-up: Three doses each of Ovaprim C, (0.25, 0.5, 0.75 mL kg⁻¹) and Ovatide (0.1, 0.2 and 0.3 mL kg⁻¹) of the female brood fish body weight were administered to three sets each of randomly selected female brood fish with three replicates each in a completely randomized design (CRD). In all, there were 6 treatments, with three replicates, and 18 experimental units. The 0.5 mL kg⁻¹ Ovaprim C and 0.2 mL kg⁻¹ dose of Ovatide were used as the control, being the manufacturers recommend dosages.

Spawning operations: The procedures of broodfish selection, hormone injection, milt and eggs collection, fertilization and incubation, hatchability and survival are as described and detailed in Ipinjolu *et al.*¹¹ and Abubakar *et al.*¹².

Determination of Reproductive Performance Indices

Total number of eggs spawned: The total number of eggs spawned (Spawning fecundity) were estimated by counting the number of eggs in 1g of egg mass, multiplied by the weight of stripped eggs¹³.

Relative fecundity (RF): Relative Fecundity was calculated as¹⁴

$$\text{Reactive fecundity} = \frac{\text{Total number of eggs}}{\text{Body Weight (g)}}$$

Stripping percentage: This was calculated according to Brzuska¹⁵ as follows;

$$\text{Stripping (\%)} = \frac{\text{Total number of eggs}}{\text{Body Weight (g)}} \times 100$$

Percent fertilization: The percent fertilization was estimated from the number of unfertilized eggs by the equation¹⁶:

$$\text{Percentage fertilization} = \frac{N-n}{N} \times 100$$

Where,

N = Total number of eggs spawned

n = Number of unfertilized eggs

The number of unfertilized eggs were determined when the eggs have developed to the middle gastrula stage (6-8 hrs 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 kyowa electronic microscope (Model XSZ-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.

Percent hatchability: Hatchability was determined from the direct data count of numbers of hatchlings at one day old as follows¹⁷;

$$\text{Percent hatchability} = \frac{\text{Number of hatchlings (one day old)}}{\text{Total number of fertilized eggs}} \times 100$$

During the incubation period, sample of 7 g fertilized eggs were incubated in each experimental unit.

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.

Percent survival rate: The percent survival rate was determined according to the method in Adebisi *et al.*¹⁸

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

RESULTS AND DISCUSSION

Latency period: The results of the reproductive response of *H. bidorsalis* to varying doses of two synthetic hormones (Ovaprim C and Ovatide) are presented in Table 1.

Latency period has indirect implication on the quantity and quality of eggs produced and the quality of the product. The longest latency period recorded in this research using Ovaprim (0.25 mL kg⁻¹) at a temperature of 27.62±0.80 (Table 2) was shorter than the finding of Owodeinde *et al.*¹⁹ who recorded latency period of 15-17 hrs at average temperature of 28°C when induction of ovulation was done on *H. bidorsalis* using Ovaprim at a dose of 0.5 mL kg⁻¹. Fagbenro and Adebayo²⁰, also recorded longer latency

Table 1: Spawning performance of *H. bidorsalis* induced with different doses of Ovaprim C and Ovatide in 2016 experiment

Variables	Treatment (Hormone/Doses)					
	I (0.25 mL kg ⁻¹)	Ovaprim C II (0.5 mL kg ⁻¹)	III (0.75 mL kg ⁻¹)	IV (0.1 mL kg ⁻¹)	Ovatide V (0.2 mL kg ⁻¹)	VI (0.3 mL kg ⁻¹)
Male brood body weight (g)	2600			2250		
Female brood bodyweight (g)	3383.33	3400.00	3300.00	2100.00	3366.67	2566.67
Female brood weight after stripping (g)	3140.00	3150.00	3026.67	1990.00	3176.67	2426.67
Difference in female body weight before and after stripping (g)	243.33	250.00	274.00	110.00	190.00	140.00
Latency period (h)	11.57±0.27 ^a	11.07±0.04 ^b	10.56±0.29 ^{bc}	11.18±0.08 ^b	11.03±0.03 ^b	10.54±0.31 ^c
Weight of eggs spawned (g)	242.30±61.15	291.00±207.35	166.07±78.15	128.13±67.15	223.00±25.94	160.23±34.45
Spawning fecundity (×1000)	156.60±56.37 ^b	190.31±135.61 ^a	108.79±51.11 ^{bc}	83.79±44.10 ^c	145.84±16.96 ^b	104.79±67.74 ^{bc}
Stripping percentage (%)	7.11±1.02	7.52±3.16	5.21±1.29	5.97±1.37	6.72±1.21	6.44±0.90
Relative fecundity (%)	46.51±0.03	49.21±0.02	34.10±0.05	39.06±0.02	43.93±0.03	42.11±0.01
Egg diameter (mm)	1.41±0.01	1.37±0.03	1.38±0.47	1.38±0.02	1.37±0.03	1.47±0.01
Fertilization rate (%)	73.33±4.16 ^b	87.33±11.01 ^{ab}	80.67±8.08 ^{ab}	73.33±10.07 ^b	74.67±8.08 ^{ab}	90.00±6.00 ^a
Range of Incubation time						
Min. incubation duration (h)	19.03±0.02	19.03±0.03	19.04±0.03	19.08±0.05	18.45±0.23	18.50±0.07
Max. incubation duration (h)	23.14±0.01	22.15±0.06	22.10±0.01	22.12±0.03	22.09±0.1	22.04±0.02
Mean weight of hatchlings day old (g)	3.5±0.86 ^c	5.17±0.69 ^a	4.47±0.50 ^{ab}	3.93±0.85 ^c	04.67±0.27 ^{ab}	5.23±0.41 ^a
Estimated number of hatchlings	1750±428.69 ^b	2587±343.16 ^a	2237±251.59 ^{ab}	2065±274.18 ^{ab}	2335±132.57 ^a	2615±204.27 ^a
Hatching rate (%)	47.41±8.81 ^b	59.23±1.80 ^{ab}	55.95±9.09 ^{ab}	55.60±8.07 ^{ab}	58.10±1.93 ^{ab}	64.31±7.73 ^a
Percent survival rate (%)	29.67±1.01 ^b	36.93±0.87 ^{ab}	33.43±1.31 ^{ab}	33.27±3.07 ^{ab}	38.60±1.45 ^a	35.83±3.89 ^{ab}

Values in row with same letter are not significantly different ($p > 0.05$)

period of 14-18 hrs post injection when carp pituitary extract and homoplastic suspension was tested for ovulation on *H. bidorsalis* at ambient temperature of 27°C. However, it is similar to the finding of Legendre and Otémé²¹ who recorded 11 hrs latency period at temperature of 30°C when *H. longifilis* was induced with human chorionic gonadotropin. The lower latency period recorded in the present study could be attributed to the type of hormone and the higher temperature. Latency period is often related to the water temperature and is often known to decrease with an increase in water temperature²². The speed of the process is dependent upon water temperature, higher the temperature quicker the eggs ovulate²³.

The latency period recorded during ovulation induction for both hormones and doses recorded in this study (Table 2) were longer than the 8 h at temperature 30°C recorded in Shinkafi and Ilesanmi²⁴ when different doses (0.1-0.3 mL kg⁻¹) of Ovatide were tested on *Clarias gariepinus* in the same hatchery as this study. Also, Ipinjolu *et al.*¹¹ and Abubakar *et al.*¹² recorded 9 hrs on *Clarias gariepinus* in the same hatchery when

Ovaprim was used. It should also be noted that the latency period (average ovulation time) recorded in each brood fish decreased as the hormone dosage increased for both hormones (Ovaprim and Ovotide) tested and no adverse effect due to the dosage rate was detected on the fish. This could be attributed to the principle that latency period is dependent on temperature, species of fish and type of hormone used²⁵.

Spawning fecundity: The result of the spawning performance contained in Table 1, revealed a range of 83.79±44.10-145.84±16.96 in those induced with Ovotide and 108.79±51.11-190.31±135.61 was obtained from brood fish induced treatment II (Ovaprim hormone 0.5 mL kg⁻¹) and was significantly (p<0.05) higher when compared to other treatments.

The weight of egg spawned and mean relative fecundity which were observed to be statistical similar in all the treatments may be as a result of the proper ovulation recorded in all the dosage treatments because no treatment showed sign of incomplete final maturation. This showed that all the doses induced ovulation effectively and can cause gonadotropin surge in *H. bidorsalis*²⁶. The result was found different from that of Sharma *et al.*²⁷, 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 spawning fecundity recorded in the present study were above the range reported in Anibeza²⁸, who obtained average fecundity range of 7,203-56,789 eggs as body size increases in *H.*

Table 2: Summary of the spawning performance of *H. bidorsalis* induced with Ovaprim C and Ovotide in 2017 experiment

Variables	Ovaprim C (0.5 mL kg ⁻¹)	Ovotide (0.1 mL kg ⁻¹)
Male brood body weight (g)	21000	21000
Female brood body weight (g)	843.33±29.63	901.67±33.21
Female brood weight after stripping (g)	725±5.00	758±18.55
Latency period (h)	12.00	12.00
Weight of eggs spawned (g)	28.73±3.47	36.77±3.37
Spawning Fecundity (x1000)	19.7911±2.27	24.05±2.21
Stripping percentage (%)	3.40±0.333	4.10±0.43
Relative fecundity	22.20±2.15	26.78±2.79
Egg diameter (mm)	1.40±0.03	1.40±0.03
Fertilization rate (%)	74.45±5.80	75.00±2.89
Range of Incubation time		
Min. incubation duration (h)	18.10±0.06	18.10±0.06
Max. incubation duration (h)	24.08±0.02	24.08±0.02
Estimated number of hatchlings	1585±291.36	2051±146.08
Hatching rate (%)	39.22±5.43	50.93±1.97
Weight of hatchlings before yolk absorption (mg)	1.89	1.89
Length of hatchlings before yolk absorption (cm)	0.27±0.06 (0.2-0.35)	0.27±0.06 (0.2-0.35)
Length of hatchlings after yolk absorption (cm)	0.45±0.07 (0.35- 0.6)	0.45±0.07 (0.35- 0.6)
Weight of hatchlings after yolk absorption (mg)	1.1	1.1

Mean values in rows with same letter are not significantly different (p>0.05). Values in parentheses are minimum and maximum of data collected

bidorsalis from Idodo river basin. Offem *et al.*²⁹ 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.*³⁰ 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.*³¹ 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 fecundity³². 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.*³³, 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 mean egg diameter recorded among the treatments in this study were similar to that of *Clarias macrocephallus* where oocyte diameter (1.40-1.49 mm) were recorded²⁸, and was within the range (0.5-1.5 mm) oocyte diameter obtained in the finding of Baidya and Senoo³⁴ for *Heterobranchus bidorsalis*. Also, Baidya and Senoo³⁴, measured oocyte diameter of *Clarias gariepinus* to be within the range of 1.26-1.44 mm, but less than 1.6-1.7 mm obtained by Fagbenro and Adebayo²⁰, when human chorionic gonadotropin, homoplastic hormone suspension and carp pituitary suspension were used to induce ovulation in *Heterobranchus bidorsalis*. This difference in the oocyte diameter recorded in this study could be as a result of the different in hormone type and the environmental condition in which the induction was carried out.

Fertilization rate: The mean fertilization rates obtained among the brood fish induced with different doses of the two hormones are presented in Table 1. The results show that the highest fertilization rate (90.00±6.00) was obtained in brood fish induced with 0.3 mL kg⁻¹ Ovotide but was not significantly different (p>0.05) from brood fish induced 0.2 mL kg⁻¹ of Ovotide, 0.5 mL kg⁻¹ and 0.75 mL kg⁻¹ of Ovaprim. These finding are close to fertilization rate of more than 75% of *H. bidorsalis* obtained²⁰. In contrast, none of the Ovaprim doses (73.33±4.16 - 80.67±8.08) that matches the finding of Nwokoye *et al.*³⁰ who recorded fertilization rates of 98.31% and 96.1% of *H. bidorsalis* induced with Ovaprim at 0.5 mL kg⁻¹ and homoplastic hormone respectively. The finding was similar to that of Shinkafi and Ilesanmi²⁴ who recorded significant fertilization rate at higher dose Ovotide used to induce ovulation in *Clarias gariepinus*. The 90% fertilization rate recorded for brooders induced 0.3 mL kg⁻¹ Ovotide is in agreement with the finding of Azuadi *et al.*³⁵ who obtained 89.6% fertilization when *Tor tombroides* was induced with Ovotide at 0.5 mL kg⁻¹. However, the fertilization rate (69.6%) recorded by these authors when Ovaprim was used to induce ovulation on *Tor tombroides* was lesser than 87.3% obtained in the present study. This could be attributed to the difference in species and environmental conditions in which the hormones are applied. It could also be as a result of the asynchronous nature of ovulation in brood fish served lower hormone dose for inducing ovulation and final maturation of eggs³⁶.

Incubation period: The duration of incubation is presented in Table 1. The period varied within and among treatments. Hatchlings were observed early in treatments induced with Ovotide at 0.2 mL kg⁻¹ with (18.45±0.23) shortest hour of incubation, followed by 18.50±0.07 hrs in treatment induced at 0.3 mL kg⁻¹ Ovotide. The duration of incubation tends to be relatively uniform among the treatments induced with Ovaprim where incubation sets in at about 19 hrs in the three treatments induced with Ovaprim. The minimum and maximum incubation duration recorded as at when eggs were observed to have hatch varied within treatment, an indication that the sexual product did not hatch at the same time. The eggs incubated from Ovotide induced brood fish at

0.2 mL kg⁻¹ and 0.3 mL kg⁻¹ were found to have shorter incubation period than those on Ovaprim. The incubation period recorded this study are lesser than the findings of Owodeinde *et al.*¹⁹ who recorded hatchability at incubation period of between 24-26 hrs when Ovaprim and pituitary gland were used to induce ovulation of *H. bidorsalis*. In the findings of Olaniyi and Omitogun³⁷, hatchability of *H. bidorsalis* eggs started after 21 h of incubation and lasted for 30 min - 1 h. However, it was found that the hatching periods varied between treatments and lasted for 4 hrs in Ovaprim induced sexual products and 3 h in Ovatide induced sexual products. However, the results of this study are similar to the findings of Okoro *et al.*³⁸ where hatching of *Clarias gariepinus* sexual product was observed to begin at 18 hrs after fertilization at a water temperature of 28°C. Ipinjolu *et al.*¹¹ who recorded 21 h at 28.5°C for *Clarias gariepinus*. Ajana and Anyanwu³⁹ who obtained 16-22 hrs incubation period at a mean temperature of 30°C. Similarly, Aluko *et al.*⁴⁰ reported incubation for 22 hrs at temperature of 24°C. These studies indicate the dependence of embryonic development on temperature for incubation *H. bidorsalis* eggs. The differences in the incubation period referred in these studies might be due to differences in temperature and efficacy of the hormone, types and dosage applied.

Hatching and survival rate: The results of the hatching and survival rate as presented in Table 1, revealed no statistically significance ($p > 0.05$) among most of the treatments, but brood fish induced with Ovaprim at 0.25 mL kg⁻¹ recorded the lowest hatching rate of 47.41 ± 8.81 which was significantly ($p < 0.05$) lower than the rate recorded for fish induced with 0.3 mL kg⁻¹ Ovatide. The rates were not significantly ($p > 0.05$) different among each of the two hormonal treatments. Treatment VI, 0.3 mL kg⁻¹ Ovatide recorded the highest hatching percentage (64.31 ± 7.73). The mean survival rate of one-week old hatchlings. The highest ($38.33 \pm 14.81\%$) mean survival was recorded in treatment induced with 0.2 mL kg⁻¹ Ovatide but was not significantly ($p > 0.05$) different from treatment induced with Ovaprim at 0.5 mL kg⁻¹ and 0.75 mL kg⁻¹ respectively. The least survival rate (29.67 ± 10.49) was recorded in fish induced at 0.25 mL kg⁻¹ Ovaprim, and this was significantly lower ($p < 0.05$) than treatment induced with 0.2 mL kg⁻¹ Ovatide but was statistically similar ($p > 0.05$) to treatment III and IV.

The hatching rate recorded in this study indicates that 0.5 mL kg⁻¹ and 0.75 mL kg⁻¹ of Ovaprim dose and 0.2 mL kg⁻¹ and 0.3 mL kg⁻¹ of Ovatide were sufficient to achieve ovulation with similar results in *H. bidorsalis* due to their statistical insignificance. These results were different from the findings of Singh *et al.*⁴¹ who recorded significant ($p < 0.05$) increase in hatchability of *Anabas testudineus* as the dosage rate of Ovatide increased 48.7 ± 3.9 , 69.2 ± 4.9 and 92.3 ± 6.1 for 1, 2 and 3 mL kg⁻¹ respectively. The result is directly proportional to the findings of Ude *et al.*⁴² who obtained significantly higher hatching rate in *Clarias gariepinus* at higher dosage (30, 50, 70 µg kg⁻¹) of leutenizing hormone releasing hormone analogue (LHRHa) compared to the lower dose of 10 µg kg⁻¹. Marimuthu *et al.*⁴³ observed no significant difference in hatching rates between the medium and higher doses of Ovatide treated groups of *Channa punctatus* was induced at 0.2, 0.4, and 0.6 mL of Ovatide per kg of body weight. The hatching rate of *Clarias batriachus* sexual products also followed the same trend, and higher doses of Ovatide recorded better performance²⁷. However, lower dose of 0.4 mL kg⁻¹ body weight performed significantly better than higher doses 0.5 and 0.6 mL kg⁻¹ of body weight when Ovaprim was used to induce ovulation in *Clarias gariepinus*⁴⁴. The percent hatching rate recorded in treatment II to V in comparison with the result of Owodeinde and Ndimele¹⁹ who obtained similar results when Ovaprim and pituitary gland were used to induce ovulation in *Clarias gariepinus* cross with *H. bidorsalis*. However, the hatching rates recorded in this research were far less compared to Nwokoye *et al.*³⁰ who recorded 96.4 and 98.35% when *Heterobranchus bidorsalis* was induced with Ovaprim at 0.5 mL kg⁻¹ and homoplastic hormone respectively. The variations recorded so far could be as a result of high temperature, excessive handling, overcrowding and water

hardness. Temperature is an important environmental factor that affect sexual product development, hatch rates and disease susceptibility. Newly spawned eggs are very sensitive to temperature changes. Overcrowding causes poor water circulation and makes it easy for disease to transfer between egg masses. Embryos in the early development stages are sensitive to handling and should be handled as little as possible to prevent mechanical injury.

The percent survival rate after one week of nursing in a static renewal system was not significantly different among the treatments except for treatment V (38.60 ± 1.45) that was significantly different from treatment I. The survival rates were generally poor below 50%. This could be as a result of high temperature, other water quality parameters, insufficient utilization of the feed provided to the fry and the static renewal nature of water used for the trial. The low survival recorded is similar to the finding of Abubakar and Ipinjolu⁴⁵ who recorded poor survival of between 8.8-40% when feed concentrate was used in feeding *Clarias gariepinus* fry. There is need for further studies on how these factors affect larvae survival and how to improve on it.

Water quality parameters: The water quality parameters during the latency period up to final maturation and ovulation of eggs are presented in Table 2. The water quality parameters monitored during this study were within the acceptable limits for tropical freshwater fish culture. The water temperature which affect all the chemical and biological process is a very critical parameter when rearing larvae. The maximum and minimum values recorded during the two spawning trials were within the range of 25-32°C required for optimum growth for warm water fish survival and for catfish hatcheries^{46,47}. The pH which is a measure of the hydrogen ion (H^+) concentration in water, recorded in this study during the two spawning trials as presented (Tables 3 and 4) are also within the optimal pH range of between 6.5-9 required for catfish culture⁴⁶. However, the maximum values of 8.61 monitored during the ovulation and 1st day of incubation period were a bit higher than the optimal value recommended in De Graaf *et al.*⁴⁷. However, the pH values recorded in this study successfully induced ovulation, spawning and incubation, hatching of *H. bidorsalis*.

Conductivity is an index of the total ionic content of water, and therefore indicates freshness or otherwise of the water Ogbeibu and Victor,⁴⁸. The values monitored were within the required conductivity of freshwater which varies between 50-1500 hs/cm ⁴⁹. Dissolved oxygen is one of the most critical water quality variables in freshwater aquaculture and the oxygen levels depend on water temperatures, stocking rates. Low dissolved oxygen can be lethal to aquaculture species causing stress, increased susceptibility to disease, poor feed conversion efficiency, poor growth and even death. The result from this study (Tables 3 and 4) were observed to be within the desired range ($5-15 \text{ mg L}^{-1}$) for larvae survival^{46,47}. The ammonia concentrations recorded as monitored in this study showed that they fall within the range accepted for freshwater fish survival. According to Viveen *et al.*⁵⁰, fish are very sensitive to un-ionized ammonia and needs an optimum range of $0.02-0.05 \text{ mg L}^{-1}$.

Effect of induced spawning of *H. bidorsalis* using Ovaprim C (0.5 mL kg^{-1}) and ovatide (0.1 mL kg^{-1})

Second trial in year 2017: The results of spawning performance of *H. bidorsalis* using 0.5 mL kg^{-1} Ovaprim C and 0.1 mL kg^{-1} Ovatide are presented in Table 2. The results indicated no significant difference ($p > 0.05$) in all the reproductive indices of the two hormonal dosages. However, the absolute value of the weight of eggs, spawning fecundity, stripping percentage and relative fecundity and number of hatchlings were higher for the Ovatide treatment where the mean size of female brood fish is $901.67 \pm 33.21 \text{ kg}$. The mean size of the female brood fish used for the Ovaprim treatment was $843.33 \pm 29.63 \text{ kg}$. The water quality parameters during this experiment are presented in Table 4. This finding buttresses the effectiveness of these doses in

Table 3: Summary of water quality parameters during the spawning period in 2016 experiment

Parameters	Mean±SD	Minimum	Maximum
Water quality variables during ovulation			
Conductivity	512.77±1.74	510.00	516.00
pH	8.14±0.21	7.85	8.61
Water temperature (°C)	27.62±0.80	26.1	29.0
Hatchery temperature (°C)	28.17±1.49	26.6	32.5
Dissolved oxygen (mg L ⁻¹)	5.61±0.25	5.32	5.81
Water quality parameters during incubation (Day 1)			
Conductivity	510.08±5.21	500.00	520.00
pH	8.09±0.29	7.69	8.61
Water temperature (°C)	26.54±0.95	25.00	28.00
Hatchery temperature (°C)	26.81±1.54	25.00	29.00
Dissolved oxygen (mg L ⁻¹)	6.02±1.33	5.91	6.31
Water quality parameters during incubation (Day 2)			
Conductivity	513.88±3.98	508.00	520.00
pH	7.94±0.11	7.83	8.10
Water temperature (°C)	26.76±0.77	25.50	29.00
Hatchery temperature (°C)	26.81±1.54	25.00	29.00
Dissolved oxygen (mg L ⁻¹)	6.05±2.54	5.75	6.11
Water quality parameter during larval rearing (1- week)			
Conductivity	511±4.25	499.00	517.00
pH	7.67±1.66	7.69	8.22
Water temperature (°C)	27.82±0.32	23.10	28.00
Hatchery temperature (°C)	26.3±0.51	21.00	27.00
Dissolved oxygen (mg L ⁻¹)	5.31±1.22	5.01	5.53

Mean values in rows with same letter are not significantly different ($p > 0.05$).

Values in parentheses are minimum and maximum of data collected

Table 4: Mean water quality parameters during the spawning period of the second spawning exercise

Parameters	Morning	Afternoon	Evening
Water temp (°C)	29.18±1.08	29.42±0.88	29.35±0.98
Minimum	28.30	28.70	28.0
Maximum	30.0	31.70	30.55
Room temp (°C)	28.58±1.80	29.39±1.87	29.21±2.11
Minimum	27.11	27.033	27.14
Maximum	31.12	1.22	31.24
Dissolved oxygen (mg L⁻¹)	5.40±1.31	-	-
Minimum	4.89	-	-
Maximum	5.65	-	-
pH	7.72±0.12	7.81±0.04	7.81±0.17
Minimum	7.65	7.60	7.48
Maximum	7.88	8.01	8.20
Conductivity	439.85±5.62	443.10±4.92	446.20±0.72
Minimum	444.00	429.00	433.01
Maximum	444.22	450.03	448.10

inducing ovulation in *H. bidorsalis* in the study area because there were not many fluctuations in the results obtained in both treatments. However, the 0.1 mL kg⁻¹ Ovatide dose required for effective ovulation and spawning performance encourage cheap use of the product and reduce the cost of hatchery production of *H. bidorsalis*.

CONCLUSION

Latency period recorded for *H. bidorsalis* brooders in the study area was at average of 11-12 hrs, but increased marginally with reduction in dosage of the two hormones tested, and all the dosages induced ovulation in *H. bidorsalis*. The incubation duration of the fertilized eggs until hatching lasted for a period between 19-23 hrs. This finding buttresses the effectiveness of these two doses in inducing ovulation in *H. bidorsalis* in the study area because there were not many fluctuations in the results obtained in both treatments. However, the 0.1 mL kg⁻¹ Ovatide and 0.5 mL kg⁻¹ Ovaprim C dose required for effective ovulation and spawning performance encourage cheap use of the product and reduce the cost of hatchery production of *H. bidorsalis*.

SIGNIFICANCE STATEMENT

This study discovered that the latency period for attainment of ovulation and final maturation of *H. bidorsalis* is at the average of 11-12 hrs when Ovulin and Ovatide are used as inducing agents and duration of incubation last 19 h at average temperature of 27.62°C. Hatching of fertilized eggs is not uniform it lasts a period of 3 hrs before all eggs in the incubation trough hatch completely in the dry sub-humid region of study area. The hatching rate recorded in this study indicates that 0.5 mL kg⁻¹ of Ovaprim dose and 0.1 mL kg⁻¹ of Ovatide were sufficient to achieve ovulation with similar results in *H. bidorsalis* in the dry sub-humid region of Nigeria. These dosage rates are therefore recommended in the study area and it will help in reduction wastage of spent eggs and reduction in the cost of hormone due to the application of higher dose by farmers in the area.

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