



SPAWNING PERFORMANCE OF *Clarias gariepinus* ADMINISTERED SERIALLY DILUTED DOSES OF OVAPRIM

DOI: 10.7904/2068-4738-VI(11)-30

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Abstract. This study aim to study the spawning performance of African catfish induced with ovaprim serially diluted with 9% saline and coconut water at ratios of 1:1, 1:2, 1:3, 1:4, 1:5, 1:6, 1:7, 1:8 and 1:9 with 1:0 serving as the control, latency period was standardized using recommended time by the manufacturer. For saline water, egg hosed out with ease only for dilution of 1:1 and with harder press striping was possible till 1:7 beyond which striping was not possible, however using coconut water, eggs hosed out with ease up to 1:4 while harder press made striping possible till the last dilution (1:9). Fertilization decreased significantly ($P > 0.05$) as the dilution increases with both diluents. Hatching was presumed complete at about 28 hours after fertilization. Result obtained show that hatchability decreased with increasing dilution with saline while there was no statistical change in values obtain with coconut water up to 1:3 however value decreased thereafter, the study shows that hatchery operation for African catfish can use coconut water and saline water as diluents up to 1:4 and 1:1 dilution respectively at normal latency period without significant effect on spawning performance.

Keyword: Coconut water, Saline water, Fertilization, Hatchability, Synthetic hormone.

Introduction

The African catfish *Clarias gariepinus* is one of the most suitable species for aquaculture in Africa.

Since the 1970s, it has been considered to hold a great promise for fish farming because of its high growth rate, adaptability and resistant to handling and stress and ease of artificial propagation through induced spawning techniques for reliable mass supply of fingerlings [HECHT *et al.*, 1996, OLALEYE, 2005].

C. gariepinus are induced to spawn in captivity using different types of hormones.

Ovaprim, Suprefact mixed with motilium Methyltestrone; Deoxycotecosteron acetate (DOCA), Human Chorionic Gonadotropin (HCG). Ovaprim is probably the most popular synthetic hormone use among Nigerian farmers to induce ovulation in cultured species [SOLOMON *et al.*, 2011].

It is a mixture of the analogue of salmon gonadotropin-releasing hormone

(sGnRHa) and a dopamine antagonist domperidone [GOUDIE *et al.*, 1992].

Ovaprim is highly effective for many species, including both fresh water and marine species and has excellent storage characteristic and standardized dosage forms [ROWLAND, 1983].

Administration is recommended at 0.5 mL/kg of body weight of brood fish; however, Mahapatra reported a functional dosage of 2 mL/kg of body weight for females of the Asiatic catfish [MAHAPATRA, 2004], *Clarias batrachus* and Solomon and collab. reported ovulation of *Clarias gariepinus* treated with 0.5 mL/kg hormones diluted up till 900% [SOLOMON *et al.*, 2011].

Dilution of Ovaprim hormone is now often used in the breeding of African catfishes to reduce cost of production as the hormone which comes in 10 mL vial is relatively expensive and farmers do not recovered used hormone in un-successful breeding.

Hecht and collab. had reported that the main impediments in the spread of



catfish fingerlings are the cost of synthetic hormones although wild fry and fingerlings may be collected [HECHT *et al.*, 1996; BUTNARIU *et al.*, 2012], but are not sufficient for a viable stocking programme.

Hence to reduce cost of hatchery operation and increase availability of fish seed this research was designed to evaluate viability of ovaprim hormone diluted with two different diluents yet administered at recommended manufacturers rate.

Material and methods

Sixty six broodstocks of *Clarias gariepinus* was used for this study comprising male and female with the ratio of 1:1. The brood stocks were weighed prior to injection with ovaprim hormone.

The ovaprim hormone was serially diluted using 0.9% saline solution and coconut water. 0.9% saline solution was prepared by diluting 9g of pure sodium chloride in 1 litre of distilled water.

The dilution level were 1:0, 1:1, 1:3, 1:4, 1:5, 1:6, 1:7, 1:8, and 1:9. The 1:0 dilution levels served as the control.

The dilution level represented treatments and each of the treatments was replicated three times.

The serially diluted hormones were injected into the different batches of fish (using 2 mL syringe and hypodermic needle) intramuscularly at the base of the pectoral fin.

Stripping was standardized at recommended time of 12 hr (according to manufacturer's label) Stripping was confirmed possible when there is gentle press on abdomen of the fish and eggs ooze out easily in the control treatment, stripping were then tried with gentle press and harder press as the cases may be for the other treatments.

Where ever stripping was possible, the male fish were sacrificed to collect milt to effect fertilization, incubation and hatching.

The fecundity of the fish was determined for each treatment.

The female fish was weighed prior to injection.

After stripping, the fish were reweighed to determine the approximate weight of the eggs.

Then samples (1g) of the eggs were counted and multiply with the approximate weight of the eggs. Fecundity and

Relative fecundity was estimated as follows:

$$\text{Fecundity} = \text{weight of eggs} \times \text{No. of eggs in 1g of egg mass}$$

$$\text{Relative fecundity} = \frac{\text{Total no. of egg}}{\text{Wt of Fish}} \times \frac{100}{1}$$

The percentage hatchability rate was determined from each treatment.

During fertilization of the eggs, 1 g of the eggs were counted, fertilized and incubated in a 70 L plastic bowls.

Upon hatching, the hatchlings were counted.

$$\% \text{ Hatching} = \frac{\text{No. of hatched eggs}}{\text{Total No. of eggs in Batch}} \times \frac{100}{1}$$

Results and discussion

Observation on stripping reveal that using saline water as diluents hosed eggs out with ease only at 1:1 dilution however with harder press stripping was possible till 1:7, beyond this point stripping was not possible.

Using coconut water, eggs hosed out with ease up to 1:4 while harder press made stripping possible till the last dilution (1:9).

Table 1 reveals the brood stock characteristics and spawning performance of African Catfish administered serially diluted ovaprim with saline solution.

Result reveals that serial dilution lead to significant decrease in % weight of striped egg, no of egg per gram and fecundity as dilution increases with saline water, higher value of this parameters are recorded in the control and in 1:1 dilution and thereafter progressively decreased till the last dilution.

Spawning performance followed the same trend however with higher values recorded in the control with values of 67.5 % and 75 % recorded for fertilization and hatchability respectively however



fertilization and hatchability were observed impossible beyond 1:8 and 1:8

dilutions respectively.

Table 1.

Broodstock characteristics and spawning performance of *Clarias gariepinus* induced with ovaprim serially diluted with saline water

Treatment	Wt of females	% Wt stripped	No of Egg per gram	% Fecundity	% Fertilization	% Hatchability
1:0	600±0.00	19.67±0.00 ^a	700±0.5 ^a	93235.3±989 ^a	67.5±0.5 ^a	79±1.0 ^a
1:1	610±150	18.5±4.17 ^a	704±36.5 ^a	87345.9±2675 ^a	55±2.0 ^b	59±1.0 ^b
1:2	600±100	15.5±4.50 ^b	615.5±14.5 ^b	73051.5±3300 ^b	55±2.0 ^b	58.5±1.5 ^b
1:3	611±25.0	15.08±6.74 ^b	615±55 ^b	68643.4±939 ^b	54±4.0 ^b	56.0±2.0 ^c
1:4	602±60.0	9.38±1.04 ^{cd}	615±94 ^b	40374.3±365 ^c	49±2.0 ^c	53.0±2.0 ^d
1:5	601±150.0	10.13±6.88 ^c	600±40 ^b	42616.1±185 ^c	46±2.5 ^c	47.5±0.5 ^e
1:6	615±25.0	8.09±2.24 ^d	561.5±11.5 ^c	27936.5±1225 ^d	43±1.0 ^d	44.5±1.5 ^e
1:7	600±0.00	4.0±3.00 ^e	515±85 ^d	12360±1444 ^e	20±20.5 ^e	0.0±0.0 ^f
1:8	599±50.0	4.76±2.94 ^e	516±24 ^e	14712.16±230 ^e	0.0±0.00 ^f	0.0±0.0 ^f
1:9	612±25.0	3.85±3.53 ^e	506±53 ^f	11922.4±326 ^f	0.0±0.5 ^f	0.0±0.5 ^f
P-Value	0.760	0.003	0.006	0.001	0.001	0.001

Mean in the same row with different superscript differ significantly (P<0.05)

Using coconut water as diluents for ovaprim appears to give better performance as result shows no significant reduction in broodstock

characteristics such as % wt of fish striped, No of egg per gram, and fecundity till 1:6 dilution with coconut water (Table 2).

Table 2.

Broodstock characteristics and spawning performance of *Clarias gariepinus* induced with ovaprim serially diluted with Coconut water

Treatment	Wt of females	% Wt stripped	No of Egg per gram	% Fecundity	% Fertilization	% Hatchability
1:0	600±0.00	19.60±2.50 ^a	710±0.5 ^a	83496±989 ^a	67.5±0.5 ^a	86.0±1.0 ^a
1:1	610±250	19.83±4.17 ^a	709±20.5 ^a	85762.7±664 ^a	75.0±1.0 ^a	85.5±1.0 ^a
1:2	615±125	19.36±4.86 ^a	711±12.5 ^a	84654.5±634 ^a	74.0±0.0 ^a	86.5±1.0 ^a
1:3	611±175	19.67±0.00 ^a	690±40.0 ^a	82926.75±430 ^a	74.0±0.0 ^a	82.5±2.5 ^{ab}
1:4	605±195	17.67±5.67 ^a	700±16.0 ^a	74832.45±350 ^{ab}	71.0±0.0 ^{ab}	72.0±1.0 ^b
1:5	612±150	16.67±8.33 ^a	683±10.0 ^{ab}	69679.93±142 ^{ab}	68.0±0.0 ^b	71.0±0.0 ^b
1:6	620±125	16.68±7.44 ^a	680±51.0 ^{ab}	70322.88±484 ^{ab}	64.5±0.5 ^c	67.0±1.0 ^c
1:7	600±00.0	12.94±7.06 ^{bc}	675±15.0 ^b	53493.9±267 ^c	59.5±0.5 ^d	65.0±0.0 ^{cd}
1:8	614±0.00	12.67±0.67 ^{bc}	670±51.5 ^b	52121.84±227 ^c	57±1.0 ^d	63.5±1.5 ^d
1:9	612±50.0	10.21±2.52 ^c	627.5±82.5 ^c	39209.46±437 ^c	55.0±1.0 ^d	59.5±0.5 ^e
P-Value	0.999	0.605	0.05	0.001	0.001	0.001

Mean in the same row with different superscript differ significantly (P<0.05)

Spawning performance on the other hand shows that fertilization and hatchability of African Catfish were statistically same with increases dilution till 1:3 (74 % and 86 % respectively) and thereafter decrease progressively but not below 59.5% as recorded in 1:9.

Comparing both diluents (Table 3 and 4) result shows that both broodstock characteristics and spawning performance were better using coconut water as a diluents compared to saline water, however at 1:1 value for fecundity

and no of egg per gram were comparable to each other.

Early researchers have reported that mature *Clarias gariepinus* cannot ovulate in captivity without the use of synthetic hormone [BATTAGLENE *et al.*, 1996], currently, ovaprim is the most commonly used hormone to induce ovulation in fish because it is highly effective for many species, and has excellent storage characteristic and standardize dosage forms [ROWLAND 1983, SOLOMON *et al.*, 2011].

The present study observed that at a latency period of 12 hour eggs hosed



out with ease only at 1:1 dilution for saline water and up to 1:4 dilution with coconut water, harder press made striping possible till 1: 7 for saline water and to the last dilution (1:9) for saline water, Ataguba and collab. reveals that the aim of synthetic hormone administration for induced breeding is to ripen eggs in the

ovaries of the fish and it is evident in ease of stripping when gently pressed [ATAGUBA *et al.*, 2012], hence reduction in the potency of the hormone with dilution which was more for saline water compared to coconut water must be the reason for this observation.

Table 3.

Comparing Broodstock characteristics of African catfish administered serially diluted Ovaprim with saline water and coconut water

Treat.	% Wt stripped			No of Egg per gram			% Fecundity		
	Saline H ₂ O	Coconut H ₂ O	P-value	Saline H ₂ O	Coconut H ₂ O	P-value	Saline H ₂ O	Coconut H ₂ O	P-value
1:1	18.5±4.17 ^b	19.83±4.17 ^a	0.001	704±36.5	709±20.5	0.504	87345.9±2675	85762.7±664	0.543
1:2	15.5±4.50 ^b	19.36±4.86 ^a	0.001	615.5±14.5 ^b	711±12.5 ^a	0.001	73051.5±3300	84654.5±634	0.120
1:3	15.08±6.74 ^b	19.67±0.00 ^a	0.001	615±55 ^b	690±40.0 ^a	0.001	68643.4±939	82926.75±430	0.236
1:4	9.38±1.04 ^b	17.67±5.67 ^a	0.001	615±94 ^b	700±16.0 ^a	0.001	40374.3±365 ^b	74832.45±350 ^a	0.001
1:5	10.13±6.88 ^b	16.67±8.33 ^a	0.001	600±40 ^b	683±10.0 ^a	0.001	42616.1±185 ^b	69679.93±142 ^a	0.001
1:6	8.09±2.24 ^b	16.68±7.44 ^a	0.001	561.5±11.5 ^b	680±51.0 ^a	0.001	27936.5±1225 ^b	70322.88±484 ^a	0.001
1:7	4.0±3.00 ^b	12.94±7.06 ^a	0.001	515±85 ^b	675±15.0 ^a	0.001	12360±1444 ^b	53493.9±267 ^a	0.001
1:8	4.76±2.94 ^b	12.67±0.67 ^a	0.001	516±24 ^b	670±51.5 ^a	0.001	14712.16±230 ^b	52121.84±227 ^a	0.001
1:9	3.85±3.53 ^b	10.21±2.52 ^a	0.001	506±53 ^b	627.5±82.5 ^a	0.001	11922.4±326 ^b	39209.46±437 ^a	0.001

Mean in the same row with different superscript differ significantly (P<0.05)

Achionye–nzeh and Obaroh had reported earlier that Increase in the dosage of ovaprim reduced the latency period in this fish and vice versa [ACHIONYE–NZEH and OBAROH 2012].

However since latency period was standardized using manufacturers prescription, differences of this study with those of Solomon and collab. are strongly linked with varying latency period for which striping was attempted [SOLOMON *et al.*,

2011], although Ree and Itanelt, reported that delay of few hours in stripping of ovulated eggs of most warm water fishes can greatly reduce the success of fertilization [REE and ITANELT, 1990], it is however envisaged that if latency period was extended for this study, more time would have been given for ripening of the egg as the potency of the ovaprim was reduced.

Table 4.

Comparing Spawning performance of African catfish administered serially diluted Ovaprim with saline water and coconut water

Treatment	% Fertilization			% Hatchability		
	Saline H ₂ O	Coconut H ₂ O	P-value	Saline H ₂ O	Coconut H ₂ O	P-value
1:1	55±2.0 ^b	75.0±1.0 ^a	0.001	59±1.0 ^b	85.5±1.0 ^a	0.504
1:2	55±2.0 ^b	74.0±0.0 ^a	0.001	58.5±1.5 ^b	86.5±1.0 ^a	0.001
1:3	54±4.0 ^b	74.0±0.0 ^a	0.001	56.0±2.0 ^b	82.5±2.5 ^a	0.001
1:4	49±2.0 ^b	71.0±0.0 ^a	0.001	53.0±2.0 ^b	72.0±1.0 ^a	0.001
1:5	46±2.5 ^b	68.0±0.0 ^a	0.001	47.5±0.5 ^b	71.0±0.0 ^a	0.001
1:6	43±1.0 ^b	64.5±0.5 ^a	0.001	44.5±1.5 ^b	67.0±1.0 ^a	0.001
1:7	20±20.5 ^b	59.5±0.5 ^a	0.001	0.0±0.0 ^b	65.0±0.0 ^a	0.001
1:8	0.0±0.00 ^b	57±1.0 ^a	0.001	0.0±0.0 ^b	63.5±1.5 ^a	0.001
1:9	0.0±0.5 ^b	55.0±1.0 ^a	0.001	0.0±0.5 ^b	59.5±0.5 ^a	0.001

Mean in the same row with different superscript differ significantly (P < 0.05).

As a result fecundity, percentage of wt striped and no of egg per gram were significantly affected for fish induced ovaprim serially diluted with saline water

while trend of decrease was observe from 1:4 dilution with coconut water.

Although the scientific reason for the observed better performance of



coconut water compared to saline water is not well understood, however it is envisaged that coconut water likely contain active ingredient that lead to induce ovulation in gravid fish, this hypothesis is subject to further research.

Bagenal had describe fecundity as the ripe spawnable eggs (>1.0 mm vitellogenic oocytes in mature female) in the ovary of the fish [BAGENAL, 1978, FERENCZ *et al.*, 2012], hence, relative fecundity was observed to be significantly lower ($P < 0.05$) as the diluents level increase for saline water and beyond 1:3 for coconut water in this study, as stated earlier reduction in the potency of the hormone may have lead to reduction in the number of viable egg compared to values of control.

The study shows that fertilization was not possible beyond 1:7 and hatchability beyond 1:6 dilutions with saline water. Ataguba and collab. has established the fact that higher fecundity leads to higher fertilization and by extension higher hatchability, hence since dilution of the hormone administered significantly reduced fecundity, less ripe egg will be available for fertilization and consequently hatchability were reduced [ATAGUBA *et al.*, 2012, CRISTINA *et al.*, 2014].

This result is different from the study reported by Solomon and collab. for *Clarias gariepinus* and by Fada for *Cyprinus carpio* as it was observed that these fishes still responded to 1:9 and 1:8 Ovaprim dilution levels respectively with 9% saline water [SOLOMON *et al.*, 2011, FADA, 2013], however these authors reported that latency period extension were in some cases beyond twenty-four hours and similar lower hatching success as reported in this study were observed.

Hence it is envisaged that coconut water diluents would have given a better result than that reported in this study if latency period were increased, it is however important to note that latency period is a compromise between allowing the broodstock more time to complete ovulation for spawning and egg quality deterioration as stated by Shelton, hence,

extending latency period should be with outmost care [SHELTON, 1989].

Previous study targeted at reducing cost of hatchery operation has evaluated various material for induced breeding with varied success level, Nwadukwe reported the efficacy of Frog pituitary gland in Oocyte maturation [NWADUKWE, 1993], ovulation and spawning success of *Heterobranchus longifilis*, and reported hatchability of 63.08 % and deformity of 8.08 %, Fagbenro and collab. reported insignificant spawning performance of *Clarias isheriensis* administered Acetone-dried pituitary extracts obtained from (non-piscine sources) common toad [FAGBENRO *et al.*, 1993], *Bufo regularis*, African bullfrog, *Rana adspersa*, and chicken, *Gallus domesticus*, as replacement for piscine-sourced pituitary extracts.

However result of this study gave better spawning performance while achieving same goal of reduced breeding cost.

Conclusions

In conclusion, for higher fecundity and hatchability at a very low cost, hatchery operation for African catfish can use coconut water and saline water as diluents up to 1:4 and 1:1 dilution respectively at normal latency period without significant effect on spawning performance.

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Received: November 06, 2014

Article in Press: March 17, 2015

Accepted: Last modified on May 18, 2015

