

Physico-Chemical Parameters of Water in Holding Tanks of *Clarias gariepinus* Induced with Ovaprim and Ovulin Hormones

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Abstract: *The physical and chemical parameters of water in holding tanks of Clarias gariepinus induced with ovaprim and Ovulin hormones were evaluated. The study was done in the hatchery unit of Department of Fisheries, University of Port Harcourt, Choba, Nigeria, and lasted for a period of four weeks. The result obtained showed that dissolved oxygen (DO) ranged between 3.82±0.88mg/l and 6.36 ±1.85mg/l for ovulin and 3.82±0.88mg/l and 5.83±1.67mg/l for ovaprim. Temperature values ranged from 25.89±0.31°C to 26.00±0.00°C. Water hardness, chloride, pH, alkalinity and ammonia values in all the treatments were constants and were respectively: 17.10±0.00mg/l, 30.00±0.00mg/l, 6.50±0.00, 17.100±0.00 and 0.001±0.00mg/l. Application of the hormones did not impact negatively on the physico-chemical parameters as they were within the required levels recommended for a successful fish reproduction.*

1. INTRODUCTION

The physico-chemical parameters of a body of water is very important to the productivity, growth and survival of the aquatic organisms that are living in the water and thus play an important role in the biology and physiology of the fishes which are part of the organisms living in water (Adebisi, 1981; Owhonda *et al.*, 2007). Bichi *et al.* (2014) observed temperature values between 25.9 and 30 °C which was said to be in line with (Ayinla 1988), who reported that the time of interval between the start of embryonic development (fertilization) and hatching, (incubation period) changes with the increase in temperature. Adeniji and Ovie (1982) and Madu (1989) reported that the best temperature range for optimum production of *Clarias species* is 25-31°C. Afzal *et al.* (2007) recommended a temperature range of between 25°C to 32°C for good performance of fishes. The pH of the water ranged from 6.1 to 7.6 and this is in agreement with the world health organization International standard for the fresh water (pH 7.08). It also corresponds with works of Huet (1972), USDA (1996) and Robert (2007) who indicate that the best water for cultivation is that which is neutral or slightly alkaline with a pH range of 7 to 8. The value for dissolved Oxygen content of water observed by Bichi *et al.*, (2014) ranged from 5.0mg/l and these agreed with those of Ufodike and Garba, (1992) who reported that a minimum constant value of 4.0mg/l DO is adequate for most species and stages of aquatic life. Brain (2006) and Ita *et al.*, (1995) reported that increased DO level is needed to support an increase in metabolic rates and reproduction.

According to Valeta (2013) increase in temperature is known to speed up metabolism through biochemical activity stimulated by heat energy (Beveridge and McAndrew, 2000) which results in enhanced development of the fish eggs. There is potential for increased temperature between 25 and 29 °C to significantly reduce hatching period and increase hatchability and fry survival. For example, temperature is known to be the main environmental factor governing fish egg development (Blaxter 1992). Temperature affects certain morphological features, hatching rate and larval behavior. In a research carried out by Bhujel *et al.*, (2000), temperature influenced egg development and hatching in *O. niloticus*, *Tilapia zillii* (Omotosho 1988) common carp, *Cyprinus carpio*, (El-Gamal, 2009), and Cod, *Gadus morhua* (Page and Frank, 1989; Geffen *et al.*, 2006).

Physicochemical parameter such as pH has been considered as one of the major factors affecting the hatchability and fertility of fish egg. Yang *et al.*, (2011) in an experiment reported that hatching was first observed at pH 10, beginning at 27 h after fertilization and ending at the 31 h. A clear difference was observed between hatching times, ranging from 31 to 64 h and increasing in order with decreasing pH. Yang *et al.*, (2011) in similar experiment with the Catfish, *Silurus asotus* reported

hatching rates in acid solutions to be higher than those in alkaline solutions which was considered to be a wide pH range for hatching compared to other fish species. Yang *et al.*, (2011) observed that eggs could be fertilized at pH 3-12 while in a hatching experiment, mortality was first observed at pH 13, when all fertilized eggs died within 8 min, followed by pH 2 (30 min), pH 12 (60 min), pH 3 (4 h), and pH 11 (5 h). This was attributed to high level of acidity which must have destroyed the viability of the eggs before, during and after hatching.

Many reports of hatching failure under such acidic conditions have been published. For example, Trojnar (1977) reported that deformations and death of white sucker *C. commersoni* embryos occurred at pH 5.0. Mount (1973) found that deformations of fathead minnow *Pimephales promelas* eggs took place at pH 5.9. Johansson and Milbrink (1976) reported that the embryonic development of roach and perch stopped at a pH lower than 4.6.

Since good water quality is essential to hatching success of fish in aquaculture, this study therefore investigate the water quality parameters in holding tanks of *C.gariepinus* induced with Ovaprim and Ovulin .

2. MATERIALS AND METHODS

Determination of Water Physico-Chemical Parameters

The physico -chemical parameters measured were dissolved oxygen (DO), temperature, pH, alkalinity, chloride and ammonia. The pH was measured with pH meter and controlled by the addition of alkaline if necessary.

Dissolved Oxygen

A glass-stoppered DO bottle was filled with water and allowed to overflow for three minutes. No air bubbles were allowed in the bottle by inclining the bottle slightly and inserting the stopper with a quick thrust. The stopper was carefully removed from the bottle. 0.5ml each of Winkler solution 1 and solution 2 reagents were added to the contents of the bottle and was thoroughly shaken to mix properly. A flocculent precipitate was formed and allowed to settle. Also, 0.5ml of concentrated H₂SO₄ was added to liberate the iodine equivalent of dissolved oxygen in the titration with the thiosulphate (APHA, 1998). The 5ml content was carefully put in a mixing bottle, five droplets of sodium thiosulphate (Standard Solution) were added drop by drop to the contents of the mixing bottle.

Temperature

A mercury thermometer calibrated in degree centigrade (0 -100°C) was used in the determination of water temperature. The thermometer scale was read off after dipping into water. When immersed in the water column it was allowed to stand for 5 minutes and reading was taken immediately it was removed. An average record was taken after taking three measurements.

Hydrogen ion Concentration (pH)

The pH value of the water was determined in the laboratory using a pH meter EIL model 720 made by Hach of Japan. The pH was determined by simply dipping the electrode into a 200mls of water that had been stirred and the reading was subsequently read off the meter. The mean of two of such readings was recorded as the pH of the water.

Alkalinity

The total alkalinity value was determined from the titration of 0.02M tetraoxosulphate (vi) acid (H₂SO₄) until methyl orange indicator colour changed from yellow to pink. 100ml of the water sample was transferred into a 250ml conical flask followed by 2 drops of methyl orange indicator and titrated with 0.02N H₂SO₄ until there was a colour change from yellow to pink at the end point of the titration. This was repeated twice and the average concordant titre values were used to calculate the total alkalinity expressed in mg/l.

Total Hardness of Water

Fifty (50ml) of the sample solution was poured into the beaker and 1ml of ammonia buffer solution (0.5N) and two to three drops of Eriochromo Black with 0.1M EDTA solution were added and titrated to the end point of blue colour. The Total Hardness were similarly obtained for all the samples collected from the eighteen stations/tanks.

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$$\text{Total Hardness (mg/l)} = \frac{0.01 \times \text{titre} \times 40.1 \times 1000}{\text{Vol. of sample}}$$

Chloride

Fifty milli liter (50ml) of samples and blank were measured into a conical flask and 3 drops of mixed indicator above were added into the sample and the blank. Then 1ml of Nitric acid was added into each of the flasks (representing 18 tanks) using dropping pipette until the solution turned pale yellow. Mercuric II Nitrate was titrated against each solution until a colour change occurred. The endpoint of titration was the appearance of a blue colour.

Data Analysis

Data obtained from each treatment were compared by two way ANOVA test to determine the significant differences ($p < 0.05$) in reproductive performance parameters and physico-chemical parameters such as dissolved oxygen, temperature, Ammonia, chloride, total hardness and pH using Turkey Honest significant difference at 95% probability.

3. RESULTS

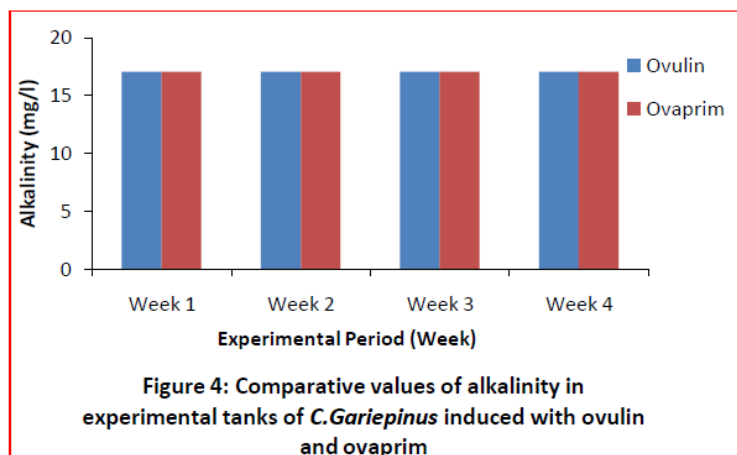
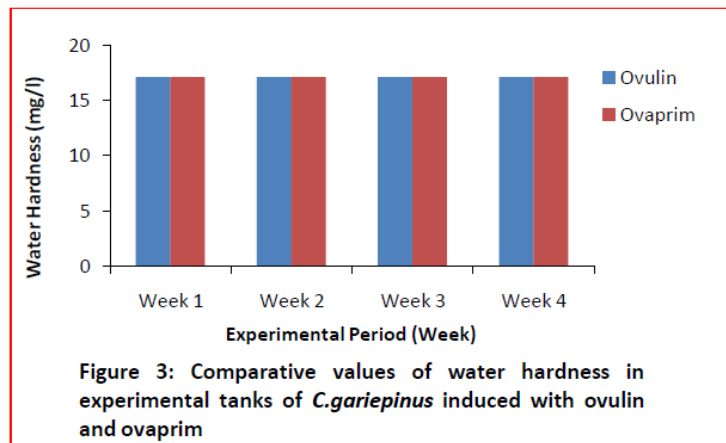
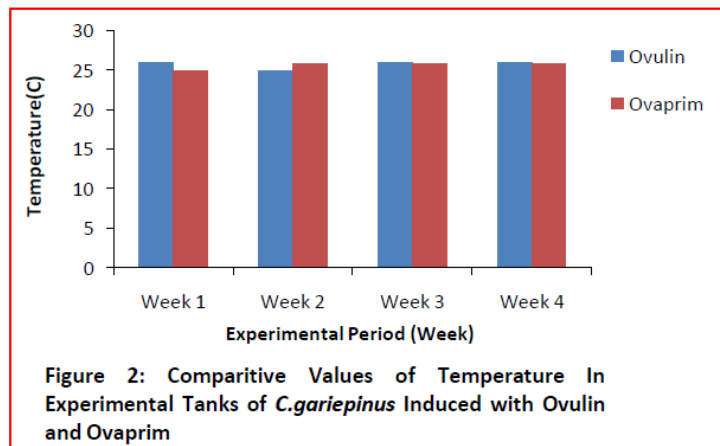
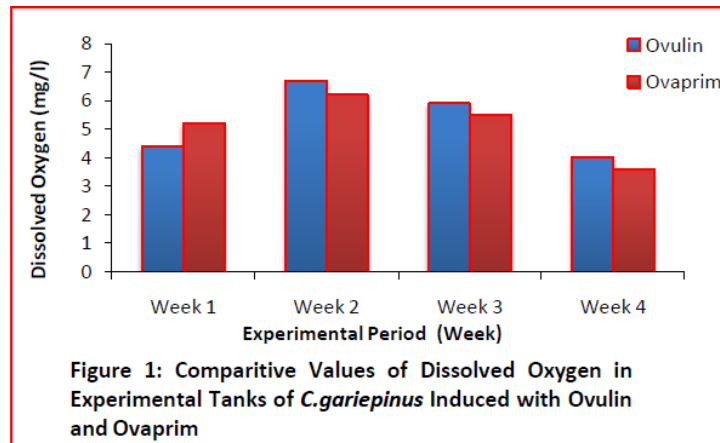
Table 4.1 shows the physico-chemical parameters of the water in the treatment tanks during the experimental period. Dissolved oxygen (DO) in the respective treatments (50, 75 and 100%) varied from $3.82 \pm 0.88\text{mg/l}$ to $6.36 \pm 1.85\text{mg/l}$ with some levels of slight significant difference for ovulin ($p < 0.05$) while that of ovaprim varied from $3.82 \pm 0.0\text{mg/l}$ to $5.83 \pm 1.67\text{mg/l}$ (Table 1). Dissolved oxygen decreases with increase in concentration of hormone in both ovulin and ovaprim treated water. Dissolved oxygen fluctuated in value between week 1 to 4 with the highest ($6.76 \pm 1.84\text{mg/l}$) and the lowest value ($4.00 \pm 0.59\text{mg/l}$) observed in weeks 2 and 4 for ovulin treated tank. Dissolved oxygen also fluctuated with the highest value (6.20 ± 1.24) and the lowest value ($3.63 \pm 0.53\text{mg/l}$) observed in weeks 2 and 4 for ovaprim (Figure 1). Dissolved oxygen showed significant difference between weeks ($p < 0.05$). Temperature values vary slightly from $25.89 \pm 0.31^\circ\text{C}$ to $26.00 \pm 0.00^\circ\text{C}$ with the highest value observed in treatment 3 (100%) for ovulin treated tank but fairly constant in ovaprim treated tank with temperature range of 25.80 ± 0.39 to 25.89°C (Table .1).

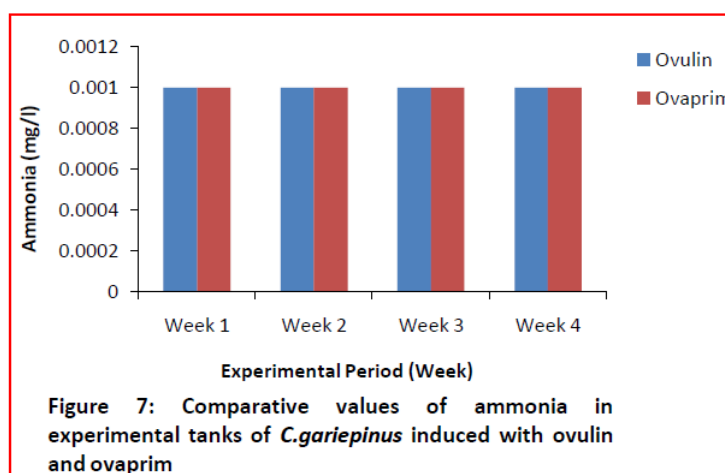
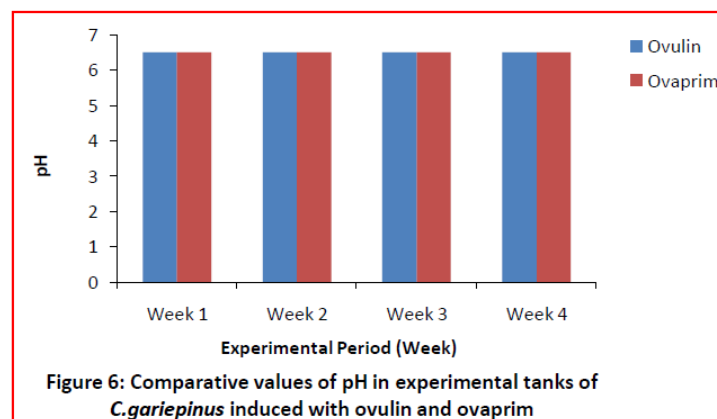
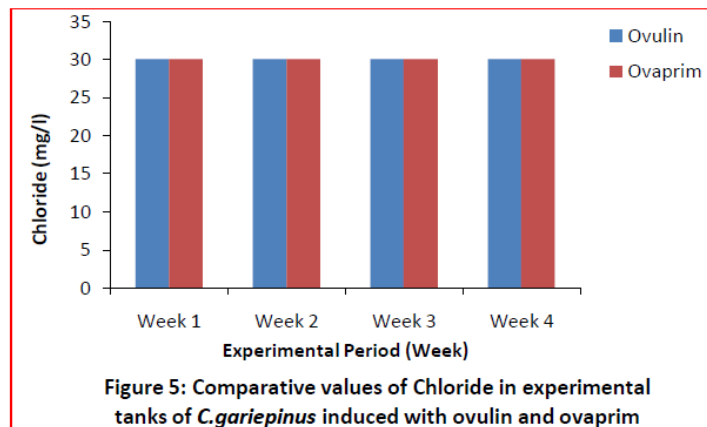
The weekly temperature value also show some degrees of consistency (Figure 2). Temperature value showed significant difference ($p < 0.05$) between weeks. Water hardness in all the treatments are constants/uniform (17.10 ± 0.00). Figure 3 also shows that the weekly water hardness were also uniform (17.100 ± 0.00). Chloride values were uniform, $30.00 \pm 0.00\text{mg/l}$ without significant difference ($p < 0.05$). pH values were also the same 6.50 ± 0.00 which is slightly acidic in the ovulin and ovaprim treated tanks without significant difference (Tables 1; Figures 4, and 5). Alkalinity values were uniform at 17.100 ± 0.00 in the ovulin and ovaprim treated tanks without significant difference (Table 1 and Figure 6). Ammonia values were constant throughout the study period in the ovulin and ovaprim treated tanks without significant difference (Tables 1 and Figure 7).

Table 4.1. Effect of Ovulin (A) and Ovaprim (B) on the physico- chemical properties of water. (means \pm SD)

Variables	Concentration (%)		
	50	75	100
Ovulin			
Dissolved oxygen (mg/l)	6.36 ± 1.85^a	5.71 ± 1.24^b	3.82 ± 0.88^c
Temperature ($^\circ\text{C}$)	25.89 ± 0.31^a	25.89 ± 0.31^a	26.00 ± 0.00^b
Hardness (mg/l)	17.10 ± 0.00^a	17.10 ± 0.00^a	17.10 ± 0.00^a
Alkalinity (mg/l)	17.10 ± 0.00^a	17.10 ± 0.00^a	17.10 ± 0.00^a
Chloride (mg/l)	30.00 ± 0.00^a	30.00 ± 0.00^a	30.00 ± 0.00^a
pH	6.50 ± 0.00^a	6.50 ± 0.00^a	6.50 ± 0.00^a
Ammonia (mg/l)	0.00 ± 0.00^a	0.00 ± 0.00^a	0.00 ± 0.00^a
Ovaprim			
Dissolved oxygen (mg/l)	5.83 ± 1.67^a	4.95 ± 1.29^b	3.82 ± 0.63^c
Temperature ($^\circ\text{C}$)	25.80 ± 0.39^a	25.87 ± 0.33^a	25.89 ± 0.31^b
Hardness (mg/l)	17.10 ± 0.00^a	17.10 ± 0.00^a	17.10 ± 0.00^a
Alkalinity (mg/l)	17.10 ± 0.00^a	17.10 ± 0.00^a	17.10 ± 0.00^a
Chloride (mg/l)	30.00 ± 0.00^a	30.00 ± 0.00^a	30.00 ± 0.00^a
pH	6.50 ± 0.00^a	6.50 ± 0.00^a	6.50 ± 0.00^a
Ammonia (mg/l)	0.00 ± 0.00^a	0.00 ± 0.00^a	0.00 ± 0.00^a

Different superscript in the same column shows significant difference ($P < 0.05$)





4. DISCUSSION

The dissolved oxygen (DO) obtained in this study were within the required levels recommended for a successful fish reproduction and in agreement with FAO (1996) except the fall in dissolved oxygen 3.82 ± 0.0 mg/l compared to 6 mg L^{-1} recommended by FAO but this can be attributed to the securely locked indoor hatchery used for the study. The value for dissolved Oxygen content also agreed with those of Ufodike and Garba (1992) that reported a minimum constant value of 4.0 mg/l DO is adequate for most species and stages of aquatic life. Brain (2006) and Ita *et al.*, (1995) reported that increased DO level is needed to support an increase in metabolic rates and reproduction.

Temperature ($^{\circ}\text{C}$) values recorded in this experiment and this is in line with Ayinla (1988), who reported that the time interval between the start of embryonic development (fertilization) and hatching, (incubation period) changes with the increase in temperature, Adeniji and Ovie (1982) and Madu (1989), reported that the best temperature range for optimum production of *Clarias species* is $25\text{-}31^{\circ}\text{C}$. Afzal *et al.* (2007) recommended a temperature range of between 25 to 32°C for good performance of fishes. The metabolism of cold blooded organisms (Poikilotherms) is dependent on

temperature as well as the solubility of dissolved oxygen and the density and viscosity of the water. Therefore, the survival, growth and behaviour of aquatic organisms are dependent on temperature. Sikoki and Veen (2004) noted that fish and many other aquatic organisms grow best at temperatures between 25 – 32°C especially in the tropic. Guy (1992) also reported that temperatures between 30 – 35 °C are tolerated by fish and many other aquatic organisms but above this range, aquatic life is threatened. According to Abdu raheem *et al.*(2012) the temperature range of 25.70 to 27.00 °C (mean 26.40 °C) recorded throughout their experiment was higher than 22 °C which Viveen *et al.*(1986) observed for *C. gariepinus* that exhibited latency period in excess of 15 hours, while Zonneld *et al.* (1988) obtained their best results at 25 °C.

pH values obtained in this study were uniform at 6.50 ± 0.00 which is slightly acidic in the ovulin and ovaprim treated tanks. This is in line with the pH ranged from 6.1 to 7.6 reported by Bichi *et al.* (2014) which was attributed to environmental factors and also in agreement with the world health organization. International standard for the fresh water is pH 7.08. It also corresponds with works of Huet (1972), USDA (1996) and Robert (2007), which indicate that the best water for cultivation is that which is neutral or slightly alkaline with a pH range of 7 to 8.

The pH of 7.00 to 8.00 was within normal range for culture fishes (Viveen *et al.*, 1986). Woynovorich and Horvath (1980) stated that a number of environmental factors such as temperature, pH, dissolved oxygen and calmness play decisive role in ovulation and that temperature is of vital importance. Yang *et al.*, (2011) in an experiment reported that hatching was first observed at pH 10, beginning at 27 h after fertilization and ending at the 31 h. A clear difference was observed between hatching times, ranging from 31 to 64 h and increasing in order with decreasing pH. Yang *et al.*, (2011) in similar experiment with the Catfish, *Silurus asotus* reported hatching rates in acid solutions to be higher than those in alkaline solutions which was considered to be a wide pH range for hatching compared to other fish species. The 0.001 ± 0.00 observed for ammonia in this study is in disagreement with the 0.50mg/l reported by Abdu raheem *et al.* (2013). This could be attributed to difference in environmental and physiological factors.

5. CONCLUSION

Application of both ovulin and ovaprim in spawning of *C. gariepinus* did not impact negatively on the water quality parameters except in dissolved oxygen, which can be augmented with the use of aerators. The results of this research will therefore be important to aqua culturists in both semi intensive and intensive systems in improving water quality for production of adequate quality fingerlings required for commercial farming of *Clarias gariepinus* in Nigeria and other developing countries. It will also help farmers improve the hatchability of *Clarias gariepinus* eggs and survival rate of *Clarias gariepinus* fry by routinely monitoring temperature and other physico-chemical parameters to avoid mortality during hatchery operations.

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