

# JIMMA UNIVERSITY COLLEGE OF NATURAL SCIENCES SCHOOL OF GRADUATE STUDIES DEPARTMENT OF BIOLOGY ECOLOGICAL AND SYSTEMATIC ZOOLOGY STREAM

The spawning response of African catfish, *Clarias gariepinus* (Claridae: Teleost) exposed to different piscine pituitary and synthetic hormone

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A Thesis submitted to the Department of Biology, College of Natural Sciences, School of Graduate Studies, Jimma University in Partial fulfillment of the requirement for the Degree of Master of Science in Biology (Ecological and Systematic Zoology).

Jimma, Ethiopia October, 2015

#### Abstract

African catfish (*Clarias gariepinus*) is generally considered to be one of the most important tropical fish species for aquaculture. However, its production has faced limiting factors like shortage of induced breeding technique, seasonality in availability and lack of good quality seed (fingerling) supply to farmers and producers. The present study was conducted to evaluate the spawning response of C. gariepinus exposed to different piscine (C. gariepinus and Cyprinus carpio) pituitary extracts and synthetic hormone (Luteinizing hormone releasing hormone + Domperidone). Nine C. gariepinus gravid females were divided in to three treatment groups and injected with hormones intramuscularly. Data on spawning fecundity, fertility rate (%), hatchability rate (%) and induction hour (hrs) were calculated. The highest mean in spawning fecundity (9731.6 eggs/g body weight/female) was observed in groups injected with pituitary extract of C. gariepinus followed by the group injected with pituitary extract of C. carpio (5813.8 eggs/g body weight/female). The lowest mean spawning fecundity (5666.6 eggs/ g body weight/female) was observed in group injected with synthetic hormone. However, the spawning fecundity/g body weight/female did not show significant difference (p = 0.073) among the groups. The highest mean fertility rate (84.3 %) was recorded in groups injected with pituitary extract of C. gariepinus followed by the groups injected with pituitary extract of Cyprinus carpio (C. carpio) (80.6 %) and synthetic hormone (74.9 %). The mean fertility rate recorded in this study showed no significant variation (P = 0.069) among the groups. The mean hatchability rate recorded in this study was high (73.3 %) in groups injected with pituitary extract of C. gariepinus and followed by the group injected with pituitary extract of C. carpio (63.5 %). The lowest (51.5 %) mean hatchability rate was observed in groups injected with synthetic hormone. The mean hatchability rate showed significant difference (P = 0.04) among the groups. In general, the present experiment indicated that the use of pituitary extract from C. gariepinus was more effective for induction of spawning in C. gariepinus during artificial reproduction as compared to C. carpio pituitary extract and synthetic hormone.

Keywords: African catfish, artificial propagation, pituitary hormone, synthetic hormone

#### Acknowledgements

Foremost my thanks go to my research advisers Mr. Mulugeta Wakjira, Mr. Tokuma Negisho and Mr. Megerssa Endebu for their advice, encouragement, assistance and unreserved support both during the preparation of the proposal and the write up of the thesis. No words are good enough to extend my heartfelt gratitude to them. Their unreserved sharing of their knowledge and experiences is greatly acknowledged.

Next, I would like to thank Jimma University for allowing me to join this MSc program and ZFRRC for letting me to carry out the experiment on the premises of the research center. My due thanks also go to Mr. Derribew Hailu who is worker at ZFRRC for his kind assistance during laboratory activities. I would like to express my deepest gratitude to my instructors and all staff members of Biology Department for their kind cooperation and assistance throughout my study.

My everlasting thanks goes to my beloved wife Wesene Niguse and my daughter Yanet Gadisa for their unconditional love, support and encouragement, your words were a source of inspiration to me that has enabled me to withstand the "stress" that comes with this kind of work . Afterward, my thanks go to my father Natea Nadha and to my mother Sorsani Bulti for your vision and decision to devote in intellectual property for my study. Lastly, my gratitude goes to all those who contributed in one way or another to the successful realization of this study and who deserve deepest appreciation to me.

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# List of Acronyms

ANOVA	Analysis of Variance
CAFAN	Catfish Farmers Association in Nigeria
DOCA	Deoxycorticosteroid Acetate
FAO	Food and Agricultural Organization of the United Nations
FR	Fertility Rate
HCG	Human Chorionic Gonadotropin
HR	Hatchability Rate
LHRH	Luteinizing Hormone Releasing Hormone
NaCl	Sodium chloride
NASS	National Agricultural Statistics Service
PAST	Paleontological Statistics
SPSS	Statistical Package for Social Sciences
WRC	Water Research Commission
ZFRRC	Ziway Fisheries Resource Research Center

#### **1. Introduction**

#### 1.1. Background

Aquaculture is the farming of aquatic organisms including fish, mollusks, crustaceans and aquatic plants. Farming implies some sort of intervention in the rearing process to enhance production, such as through regular stocking, feeding or protection from predators (WRC, 2010). Aquaculture is developing, expanding and intensifying in almost all regions of the world (FAO, 2012). It is the fastest growing food producing sector in the world today (FAO, 2014). Global aquaculture production has grown at 10 to 11 percent a year over the past decade and is projected to continue increasing (FAO, 2010). Aquaculture can serve as one of the best alternatives for the over-exploited natural aquatic resources. Therefore aquaculture can help to reduce pressure and maintain the ecological balance of the natural ecosystems. It is probably one of the most promising answers to a world population growth in providing additional food and employment. FAO (2006) reported that out of the total tonnes of fish consumed in the world every year, almost half is produced in a controlled environment. The current supply trends combined with ever increasing population, the per capita consumption of fish in Africa is stagnating. To change this condition and boost production of fish, aquaculture remains the most feasible option that can sustain adequate fish supply in Africa (Asaminew, 2012).

Aquaculture in Ethiopia is remains more potential than in actual practice. This is the contribution of aquaculture is almost negligible despite the fact that the country's environmental and socio-economic conditions support its development (FAO, 2005). The consumption and demand of fish as a cheap source of protein is increasing in Ethiopia, but the fish supplies mostly were come from the major lakes and rivers in the country. Consequently, capture fisheries from those natural environments presently causes over exploitation and seem to have reached their natural limits. As the report of Asaminew (2012) indicates the fisheries production of the major lakes of Ethiopia is declining in an alarming rate. As a result of high population growth in the country, there is high

competition to be engaged in fisheries activities around the lake region. Thus, as the experience of different countries in the world shows, if aquaculture can be developed in a sustainable way, it can be the best alternative to tackle this kind of problems (Yang *et al.*, 2006; Watanabe, 2002).

Aquaculture development in Ethiopia is faced with a number of challenges which include lack of human power, lack of cheap and efficient locally available fish feeds, lack of locally selected and certified fish seeds, the problem of land ownership policies in the country, over-reliance on capture fisheries, lack of licensed fish seed multiplication centers and lack of institutional capacity in the area of training, research and technology transfer (Asaminew, 2012).This shows that aquaculture development for poverty mitigation and food self-sufficiency in the country cannot be over emphasized. Yet, aquaculture can be an important mechanism for local food security, food availability and improves access to nutritious and healthy food for the rural poor. It also plays an important role in poverty alleviation as it provides employment to millions of people, both in the sector itself as well as in support services (Asaminew, 2012; Faruque *et al.*, 2010).

African catfish is widely distributed throughout Africa and it is one of the most important commercial freshwater fish species in many parts of the continent. It is known for its adaptability and hardiness to adverse environmental conditions (Macharia *et al.*, 2002). Goose and Richter (1996) identified *C. gariepinus* as the most important species for aquaculture in Africa due to its omnivorous eating habits, resistance to handling stress, high meat content, ability to handle low water levels by breathing atmospheric air and its ability to withstand high stocking densities. This species has potential for highly intensive culture without prerequisite pond aeration or high water exchange rates. The meat quality is first-rate containing high nutritive values and is particularly favored as there are no intramuscularly spines and more flesh than cultured tilapia species.

Additionally, *C. gariepinus* is considered to be one of the most important tropical fish species for aquaculture and a fish of great promise for fish farming in a wide number of

African consumers; because of its fast-growing, wide range of temperatures  $(8-35 \ ^{0}c)$  and water quality (Faruque *et al.*, 2010; Munro, 1967). However, farming this species has many limitations; collection of fry from wild, requires specialized breeding techniques, seasonality in availability, uncertainty of species of fish seed, diseases and limited quantity of harvestable fish seed. Though, Catfish fingerling production is often perceived as a bottleneck in tropical countries for commercial fish farming (Shepherd and Bromage, 1992). To change this condition and boost production of African catfish, artificial propagation and appropriate larvae treatment remains the most feasible option that can sustain adequate fish supply, generally in Africa and specifically in Ethiopia (Asaminew, 2012).

According to Dadebo *et al.* (2014) Ethiopian lakes and river systems have great potential for the production of *C. gariepinus* and it is one of the commercially important fish species in the country. It is a fast growing fish and tolerates harsh environmental conditions. It has great economic importance next to Nile tilapia in the country. However, there is a research gap in using hormone (natural or synthetic hormone) for acceleration of this fish farming through induced spawning (Gadissa and Devi, 2013). Therefore, in order to address the aforementioned gap the present study evaluated the spawning response of African catfish exposed to different piscine (*C. gariepinus* and *C. carpio*) pituitary as well as synthetic hormone.

#### **1.2. Statement of the problem**

Ethiopia has high potential for production of *C. gariepinus*. But, continuous supply of fingerlings and harvesting high production is not well established and generally at an immature stage (Asaminew, 2012). There are many factors that constrained aquaculture growth in this country. Some of these are; lack of adequate fish feed, difficulty with induced breeding method, lack of sufficient fingerlings for stocking and shortage of man power in aquaculture sector (Atieno, 2011).

In Ethiopia there is no available domestic fingerling producing hatchery yet, and fingerlings are always obtained from the wild. Though, the stock quantity is limited, wild caught fingerlings are of poor quality and slow in terms of growth (Zerihun, 2015). As a result of high population growths in Ethiopia, competitions in fishing activities around the natural lakes remain high. Consequentially, most of the fish producing lakes are already overexploited, triggering an alarmingly decline rate (Dadebo *et al.*, 2014). Therefore, this study aimed at evaluating the spawning response of *C. gariepinus* exposed to different piscine pituitary and synthetic hormone.

# **1.3. Objectives**

#### **1.3.1.** General objective

To evaluate the spawning response of African catfish (*C. gariepinus*) exposed to different piscine pituitary and Synthetic hormone.

# **1.3.2.** Specific objectives

- To examine the spawning fecundity of African catfish (*C. gariepinus*) exposed to different natural piscine pituitary (*C. gariepinus* and *Cyprinus carpio*) and Synthetic hormone.
- To examine the fertility rate of African catfish (*C. gariepinus*) exposed to different natural piscine pituitary (*C. gariepinus* and *C. carpio*) and Synthetic hormone.
- To examine the hatchability rate of African catfish (*C. gariepinus*) exposed to different natural piscine pituitary (*C. gariepinus* and *C. carpio*) and Synthetic hormone.
- ↓ To compare the induction response (reaction rate) of *C. gariepinus* exposed to natural piscine pituitary (*C. gariepinus* and *C. carpio*) and Synthetic hormone.
- **4** To identify the best hormone for spawning of African catfish (*C. gariepinus*).

### **1.4. Significance of the study**

Importance of aquaculture development for nutritional security, source of income and creating job opportunity in Ethiopia is well stated (Mitike, 2014). However, using artificial propagation and producing fingerlings are the major limiting factors in aquaculture development in the country. The seasonality and scarcity of *C. gariepinus* larvae from wild were made the need of artificial spawning an important concern in Ethiopia (Zerihun, 2015). The study has a great role in identifying the best pituitary hormones that would produce better spawning response and hatchlings in *C. gariepinus* culture. Therefore, results of the present study will help to improve the availability and continuous supply of *C. gariepinus* fingerlings to farmers. The study also provides baseline information for further research.

#### 2. Review of related literature

# **2.1.** Taxonomic position, morphological features and habitat of African catfish(*C. gariepinus*)

The African catfish belongs to the kingdom Animalia, phylum Chordata, sub-phylum Vertebrata, class Actinopterygii (ray-finned fishes), order Siluriformes (catfish) and family *Clariidae* (Nelson, 2006). The family *Clariidae* consists of two notable genera; namely *Clarias* and *Heterobranchus*. They are highly diverse. More than 100 different species have been studied in Africa based on their morphology and anatomical features (Teugels, 1984). *Clarias gariepinus*, commonly named as African sharp tooth catfish, is scaleless and "air breathing". The genus *Clarias* was reviewed in the 1980s and some species were synonymized with *C. gariepinus*. These were *Clarias lazera* of West and North Africa, *Clarias capensis* of Southern Africa, *Clarias mossambicus* of Central Africa, in the name *C. gariepinus* (Teugels, 1986).

*Clarias* can be defined as displaying an eel shape, having an elongated cylindrical body with dorsal and anal fins being extremely long (nearly reaching or reaching the caudal fin). Both fins contain only soft fin rays. The outer pectoral ray is in the form of a spine and the pelvic fin normally has six soft coats. The head is flattened, highly ossified with the skull bones (above and on the sides) forming a calque. The body is covered with a smooth scaleless skin (De Graaf and Janssen, 1996). The skin is generally darkly pigmented on the dorsal and lateral parts of the body. The color is uniform marbled and changes from grayish olive to blackish according to the substrate. Usually, varies from dark to light brown in color and often spotted with shades of olive and grey. On exposure to light the skin the color generally becomes lighter (Skelton, 2001). They have four pairs of unbranched barbels, one nasal, one maxillar (longest and most mobile) on the vomer and two mandibulars (inner and outer) on the jaw. Tooth plates are present on the jaws as well as on the vomer. The major function of the barbels is prey detection. De Graaf and Janssen (1996) found that *C. gariepinus* with barbels are 22.6 % more efficient at catching prey than those without. This could indicate that tactile behavior is important in the prey

catching processes. Slow, methodical searching and grasping their prey by suction is the normal predatory tactic of *C. gariepinus*. A negative pressure (suction) is created by a sudden increase of the bucco-pharyngeal chamber. An important aspect of predation by *C.gariepinus* is their ability to switch feeding from one type of prey to another. For instance, in Lake Sibaya (South Africa), catfish ignore (cannot catch) fish prey during daylight and feed mainly on invertebrates, which are abundant and relatively easy to catch. In contrast, at night when fish prey becomes more vulnerable, they switch their feeding habits to fish prey (Bruton, 1979b; 1979a).

A supra-branchial or accessory respiratory organ, composed of a paired pear-shaped airchamber containing two arborescent structures is generally present. These arborescent structures located on the second hand of the forth branchial arcs, are supported by cartilage and covered by highly vascularised tissue which can absorb oxygen from atmospheric air (Moussa, 1956). The airchamber communicates with the pharynx and with the gill chamber. The accessory air breathing organ allows the fish to survive for many hours out of the water or for many weeks in muddy marshes. The male and females of *C. gariepinus* can be easily recognized as the male has a distinct sexual papilla, located just behind the anus. This sexual papilla is absent in females (De Graaf and Janssen, 1996).

African catfish (*C. gariepinus*) lives in a variety of freshwater environments including lentic waters like lakes, ponds and pools. It is also very prominent in flowing rivers and around dams. It is very adaptive to extreme environmental conditions and can live in pH range of 6.5-8.0. They are able to live in very turbid waters and can tolerate temperatures of 8-35  $^{\circ}$ c. Their optimal temperature for growth is 27-30  $^{\circ}$ c (Verreth, 1993). African catfish spend most of their time on the bottom of lakes and rivers and do most of their feeding there (NASS, 2010). However, they are able to move across land to another water source during damp conditions (Skelton, 2001) and thus can breathe atmospheric air by mean of a supra-branchial or accessory respiratory organ. They simply extend their strong pectoral fins and spines and begin crawling through shallow pathways. This species can live in very poorly oxygenated waters and is one of the most recent species of fish to live

in such uninhabitable place by other fish species (Eyo and Mgbenka, 1992). They are also able to secrete mucus to prevent drying and are able to burrow in the muddy substrate of a drying body of water (Skelton, 2001).



Fig. 1. African catfish (Clarias gariepinus)

#### 2.2. Geographical distribution and aquaculture potential of African catfish

African catfish (*C. gariepinus*) is considered as an indigenous and most important species for aquaculture in the African continent (Akankali *et al.*, 2011). It has wide distribution in Africa, ranging from the Nile to West Africa and from Algeria to Southern Africa (De Graaf and Janssen, 1996). African catfish is the most sought species by fish farmers and consumers and as commands it has very good commercial value in Nigerian markets (Gaffar, 1996). In 2009 investment in catfish farming was documented by catfish farmers association of Nigeria (CAFAN) and reported over 24 billion naira profits annually (Akankali *et al.*, 2011). African catfish is an endemic species having a ubiquitous distribution in rivers, streams, dams and lakes in Kenya. All the *Clarias* species reported in Kenya inhabit wetlands or wetland open interface. These groups of fish (Siluriformes) are widely consumed. Successful culture/captive breeding of this species and thus fingerling rising has been done in the country. It can be raised in high densities resulting in high yields and fetches a higher price than tilapia as it can be sold live at the market (Macharia

*et al.*, 2002). In Ethiopia, the species is found almost in all lotic and lentic water bodies (Dadebo *et al.*, 2014). *Clarias gariepinus* is both an ecologically and economically important fish in Ethiopia. Hence, knowledge of its biology could have significant importance for the development of catfish aquaculture in the country (Dadebo *et al.*, 2014).

African catfish is now widely farmed in many regions of Europe and Asia specially; it occurs in Israel, Syria and South of Turkey insignificantly (De Graaf and Janssen, 1996). In India it is recognized as exotic species and now widely farmed. *Clarias gariepinus* was brought to India from neighboring Bangladesh and cultured initially in the two northeastern states of West Bengal and Assam and the southern state of Andhra Pradesh, together with the Indian major Carp (De Graaf and Janssen, 1996).

*Clarias gariepinus* has been considered as an ideal species for the development of aquaculture in Africa (Gadissa and Devi, 2013). It is very important to the sustainability of the Aquaculture industry in the continent. In recent past, fish fingerlings were captured from the wild however, due to problems associated with wild fish seed, seasonality in availability, uncertainty of species of fish seed, diseases and limited quantity of harvestable fish seed; it has become an unreliable source for commercial fish farming (Shepherd and Bromage, 1992). *Clarias gariepinus* is relatively insensitive to disease and does not have high water quality requirements. It tolerates high concentrations in the water of ammonia (NH<sub>3</sub>) and nitrite (NO<sub>2</sub>) in water. Low oxygen concentrations are tolerated because the fish utilizes atmospheric air as well as dissolved oxygen since it has well developed air breathing organs (De Graaf and Janssen 1996). It can be raised at high densities, resulting in high yields and fetches a higher price than other fish species as it can be sold live at the market. Moreover, *C. gariepinus* can be fed with large variety of agricultural by products and make this species an excellent candidate fish for aquaculture (Macharia *et al.*, 2002).

One of the many challenges faced in catfish (*C. gariepinus*) is fingerling production. The optimal environmental conditions and feeding behavior during the early life stages are poorly understood and the tiny size of the organisms and the concomitant need for small

food particle sizes poses specific strains on the feed technology (Awaïss and Kestemont, 1998). In spite of huge efforts to use artificial feeds, the culture of fish larvae during the primary nursing phase still depends heavily on natural food. Live feeds include rotifers, artemia and other tiny organisms that are often used as the first foods in the aquaculture food chain. Feeding live prey for fish larvae are most essential because during first few days of their life stage they lack a completely developed digestive system. Moreover, feeding live prey increases feed intake by predatory larvae, resulting in reduced cannibalism in some species like *C. gariepinus* and enhance larval production (Faruque *et al.*, 2010).

#### 2.3. Reproduction of African catfish

#### 2.3.1. General mechanisms of hormonal action in fish reproduction

The internal mechanism that controls the process of reproduction in fish is the brainhypothalamus-pituitary- gonad chain. Hypothalamus produces the gonadotropin-releasing factor (GnRH) which acts on the pituitary gland. This gland controls synthesis and release of the gonadotropin hormones (GtHs), whose role is to lead the gonads (ovaries and testes) to produce the gametes. Gonadotropin hormones (GtH) which consists of follicle stimulating hormone (FSH) and Luteinizing hormone (LH) act on the ovaries and testes (gonads). Steroids and prostaglandins appear to be the local ovarian mediators of GtH action causing release of the eggs (Rottmann et al., 1991). High GtH in blood levels trigger two distinct ovarian processes. These are final maturation of the egg, which appears to be stimulated by steroids (e.g. progesterone) that are produced by the follicle and rupture of the follicle (ovulation), which evidently is stimulated by prostaglandins. Steroids also appear to induce spermiation in the male. The pituitary gland also produces dopamine, which on the contrary, has an inhibiting effect on the process. The primary substances used for hormone-induced spawning was Lutinizing hormone releasing hormone analogs (LHRHa) alone or in combination with dopamine blockers which enhance the potency of LHRHa to stimulate the pituitary (Yousefian and Mousavi, 2011).

Injections of Luteinizing Hormone-Releasing Hormone (LHRH) have been used experimentally to mimic the fish's GnRH. Recently, synthetic LHRH analogs, referred to as LHRHa have been applied on fish for artificial propagation. LHRHa stimulates the fish's own pituitary to produce and release the GtH necessary for spawning. Dopamine inhibits the release of hormones from the pituitary, effectively blocking the pituitary's positive response to injected LHRHa. Dopamine blockers act either by preventing the release or by inhibiting the binding of dopamine. Experimental results indicate that the use of dopamine blockers prevents this negative feedback, enhancing the effectiveness of LHRHa for fish species (Zohar *et al.*, 2010).

#### 2.3.2. Natural reproduction of African catfish

The natural reproduction of *C. gariepinus* shows a seasonal gonadal maturation which is usually associated with the rainy season. The maturation processes of the species are influenced by annual changes in water temperature and photoperiodicity and the final triggering of spawning is caused by a rise in water level due to rainfall (de Graaf et al., 1995). Naturally, African catfish participate in mass spawning and lay their eggs in vegetation. The larvae are able to swim and feed within 2 to 3 days and growth is faster in males than females (NASS, 2010). Spawning usually takes place at night in the shallow inundated areas of the rivers, lakes and streams. Courtship is preceded by highly aggressive encounters between males. Courtship and mating takes place in shallow waters between isolated pairs of males and females. The mating posture, a form of amplexus (the male lies in a U shape curved around the head of the female) is held for several seconds. A batch of milt and eggs are released followed by a vigorous swish of the female's tail to distribute the eggs over a wide area. The pair usually rest after mating from seconds up to several minutes and then resume mating (De Graaf and Janssen, 1996). There is no parental care to ensure the survival of the offspring except the careful choice of a suitable site. Development of eggs and larvae is rapid and the larvae are capable of swimming within 48-72 hours after fertilization at 23-28 °C (De Graaf and Janssen, 1996).

#### 2.3.3. Artificial propagation of African catfish

Artificial propagation is adopted in order to have mass production of the fish all year round. It is the most promising and reliable way of ensuring availability of good quality fish seed continuousily and also vital for sustainability of the aquaculture industry. It involves the use of natural (hypophysation) or synthetic hormones to induce ovulation and spawning in farmed fishes (Olumuji and Mustapha, 2012; Ononuju *et al.*, 2007). In captivity the African catfish does not spawn spontaneously since the environmental factors such as the rise in water level and flood of shallow areas do not occur on the fish farms. Since the early 1970's several techniques have been developed with or without hormone treatment for the artificial reproduction of the African catfish (De Graaf and Janssen, 1996).

Semi natural or hormone induced reproduction within ponds or concrete tanks can be used on small farms to produce their own larvae and fingerlings. However, the method has not proved to be a reliable method for mass production needed for larger fish farms or distribution centers of catfish fingerlings (de Graaf et al., 1995). Therefore, artificial propagation under more controlled conditions including; stripping of eggs, collection of the sperm, followed by fertilization of eggs has been developed. Eventhough, artificial propagation of African catfish by more or less similar to that which occurs during the route of natural reproduction (from fertilized eggs to fingerling size) has several advantages. These include better rates of fertilization and hatching, protection against enemies and unfavorable environmental conditions, better conditions for growth and survival, guaranteed for supply of fish seed all the year round, fish seed obtained outside the natural environment of the fish, increased survival rate of the fry and improved quality through breeding (i.e. hybridization). Eventhough artificial propagation of catfish seems, the promising method for better production of this species, the major constraints to fish breeders is the cost of procurement of these hormones (Khan, 2014; Olumuji and Mustapha, 2012; Woynarovich and Horvath, 1980).

According to Madu (2006), hormone input accounts for about 50-60% of the recurrent expenditure of a catfish fingerling production in Nigeria. For instance the cost of ovaprim, ovatide, human chorionic gonadotrophin (HCG), decorticosterone acetate (DOCA) and luteinizing hormone releasing are equally expensive (Olumuji and Mustapha, 2012). There are also other disadvantages of artificial propagation where, the donor fish has to be sacrificed in most cases and hence resulting in loss of fish. Furthermore, the whole process is laborious, highly technical and also very expensive in that it requires proper housing, constructions of tanks and installation of jars in a closed recirculatory system. It should be noted that artificial (i.e. naturally induced or through hypophysation) production of fish seed are carried out in enclosure system in hatcheries which are composed of both an indoor and outdoor facilities. Therefore, they require inputs such as brood stock, adequate water supply and suitable feed to obtain best result (Okechi, 2004).

#### 2.3.4. Artificial Propagation through hypophysation

The hypophysation technique uses the pituitary gland (the hypophysis) to induce spawning in fish and can be carried out at any time of the year and under any environmental conditions. The technique ensures fish seed availability all year round. In this technique, a single catfish (*C. gariepinus*) is induced to spawn many times within a year between successive spawning, even though catfish breeds naturally only once a year (De Graaf and Janssen, 1996).

# **3.** Materials and Methods

# **3.1. Description of the experiment site**

The experiment was conducted from March to May 2015 at Ziway Fisheries Resources Research Center (ZFRRC) located at 160 km south of Addis Ababa in Batu Town, Oromia Regional State, Ethiopia. The research center is situated near the shore of Lake Ziway within the mid Ethiopian Rift Valley system (7.919oN and 37.727°E) at an elevation of 1638 meters above sea level (Gadissa and Devi, 2013). The fish specimens and fish offal (trash fish) for the experiment were obtained from Lake Ziway.

# 3.2. Pituitary gland extraction

The pituitary glands which are pinkish-white globule-like organ located on the ventral side of brain (Fig. 2) were taken from fresh fish head offal at the source (Lake Ziway) from *C*. *gariepinus* and C. *carpio*. The head of the fish was dissected with knife and the pituitary glands were picked up by forceps. Then the pituitary glands were transferred to vials and preserved in absolute acetone (98.2 %) in a refrigerator at a temperature of  $4^{0}$ C (Brzuska, 2004).



(a)

(**b**)

Fig .2. Extraction of pituitary glands from fish skull (a) and the extracted glands (b).

#### 3.3. Synthetic hormone used for the experiment

For the present experiment, the maturation, ovulation and spawning of female *C. gariepinus* was induced by injecting a combination of luteinizing hormone-releasing hormone analogue (LHRH-A<sub>2</sub>) and Domperidone (DOM). In the synthetic hormone treatment, LHRH-A2 acts by stimulating the release of gonadotropin from the pituitary and the Domperidone by suppressing the action of a natural hypothalamic gonadotropin release-inhibiting factor (GRIF) which has been identified as dopamine (Ahmadnezhad *et al.*, 2013).



Fig. 3. Synthetic hormone, luteinizing hormone-releasing hormone analogue (LHRH-A<sub>2</sub>) and Domperidone (DOM). (NINGBO SECOND HORMONE FACTORY http://www.nbshusheng.com)

#### **3.4. Broodstock collection and maintenance**

The Broodstocks for the experiment were collected from Lake Ziway and transported to ZFRRC and kept in concrete ponds having size of 7x5x1m. Broodstocks for breeding purpose were selected by consideration of some external morphological characteristics according to (De Graaf and Janssen, 1996). Female brooders having a well distended abdomen and weigh more than 500g were selected. For male broodstock selection color of

genital papilla (reddish) and also weight greater than 200g were considered (Gadissa and Devi, 2013; De Graaf and Janssen, 1996). Prior to the experimental setup the broodstocks were acclimatized in separate concrete pond for one month. The stock was fed twice daily at 7-9 am and 4-6 pm with pellet feed prepared from Noug cake and wheat bran (2:1) at the rate of 5% of their body weight. A seine net was used to gently capture the brood fish. After collection of the fish from the conditioned pond, fish are treated with formalin (25 ml/ L for 30 minutes) in bath to prevent the transfer of pathogens from fish to hatching system, eggs and fry (Floyd, 1996).



Fig .4. Concrete pond used for broodstock maintenance

#### **3.5.** Hormone treatment

#### 3.5.1. Piscine pituitary treatment

The extracted pituitary glands were homogenized in a tissue grinder (mortar and pestle) with 2 ml of 0.9 % salt solution. The solution was administered to matured female African catfish by intra-muscular injection into the dorsal muscle above the lateral line just below the posterior part of the dorsal fin (Fig. 5. b) with a syringe (5 ml volume) at a dose of 2 mg/kg body weight (De Graaf and Janssen, 1996). The injected area of each fish was massaged gently with a finger in order to distribute the hormone evenly into the muscle while slowly retracting the needle.



Fig. 5. Preparation of pituitary gland (a) and injecting the fish with pituitary gland extracted (b).

**(b)** 

#### **3.5.2.** Synthetic hormone treatment

(a)

The preparation of the Synthetic hormone for injection was based on the direction of a manufacturer. A vial of LHRH-A<sub>2</sub> containing 125  $\mu$ g powder was dissolved in 12.5 ml of saline solution which gives a concentration of 10  $\mu$ g/ml. Then a vial of Domperidone containing 100 mg was mixed with a 10ml saline solution to make a concentration of 10 mg/ml. Finally, equal volume of each solution was taken and mixed together to inject the fish. The prepared mixture of synthetic hormones was injected in to the experimental fish at a dose of 1 ml/kg body weight of the fish (NINGBO SECOND HORMONE FACTORY http://www.nbshusheng.com).

#### **3.6. Experimental design**

The spawning response trials were carried out in three treatments in a completely randomized design. Each treatment group consisted of three individuals of female African catfish which were selected randomly from the pond. The three treatments included: fish treated with *C. gariepinus* pituitary extraction, *C. carpio* pituitary extraction and synthetic hormone designated as CG, CC and SH, respectively.

# 3.7. Stripping of the eggs

After observing the 11hrs of ovulation period, each female broodstock were removed from the tank carefully. The tank was previously adjusted with the re-circulating water, aerator and boiler at 27 <sup>o</sup>C. The fish were held firmly with a wet towel at both ends and the abdomens were gently pressed with thumb from the pectoral fin region to the genital papilla into a dry tray as in (Fig. 6a) (De Graaf and Janssen, 1996). After stripping, the spent female was carefully returned into the concrete pond.



Fig. 6. Hand stripping of female catfish (a) and striped eggs (b).

## 3.8. Milt collection and fertilization

Milt was collected from male *C. gariepinus* by sacrificing the fish. Then, the fish was dissected dorsally from the anal to the head by sharp knife and tests were collected. The testes were dissected by using a sharp blade (De Graaf and Janssen, 1996). The milt collected from one male fish was used to fertilize eggs of two female fish. It was squeezed over the eggs and then wet fertilization was performed with saline solution (0.9 %) and water. It took only 1minute to accomplish fertilization by mixing the striped eggs with the collected milt solution using a bird feather (Fig. 8b).



(a)

(b)

Fig .7. Milt extraction from male catfish (a) and extracted milt for the experiment (b).



(a) (b) Fig. 8. Artificial fertilization, milt on eggs (a) and mixing process (b).

# **3.9. Incubation**

The egg incubation system was filled with clean tape water. The water in the system was allowed to re-circulate having its temperature adjusted at 27  $^{O}C$  in the lab before the start of the incubation. Incubation was done promptly after fertilization i.e. about 60 seconds after mixing egg mass and water. A Nylon mesh (1 mm) mosquito net was suspended as substrate for spreading of the fertilized eggs. The eggs were spread in single layers on the

suspended nylon mesh net for incubation. The fertilized egg masses were incubated in the spawning trays for a period of 24 to 48 hrs at 27<sup>0</sup>C (Brzuska, 2002; Verreth, 1993).

#### 3.10. Hatching

The process of development from fertilized egg to hatching, like all other biological processes, is dependent upon water temperature. The higher the water temperature the faster the eggs hatch (de Graaf *et al.*, 1995). In the present experiment the fertilized eggs remained in the plastic tank at a temperature of 27  $^{0}$ C.



Fig. 9. Catfish fry produced from the experiment.

## 3.11. Separation of larvae from spoiled eggs

The healthy larvae were swimming into the shadowed part under the cover and clustered at the edges of the plastic tank. Only viable larvae were looking for shelter and pass through the perforated bottom of the tray by actively swimming while leaving behind the dead and unfertilized whitish eggs. The trays were removed as soon as hatching was completed and normal larvae had gathered under the incubation trays (de Graaf *et al.*, 1995). The dead and unfertilized eggs were removed by siphoning (Woynarovich and Horvath, 1980).

#### 3.12. Water quality parameters

Maintenance of good water quality is essential for both survival and optimum growth of fish at all stages of their life in general and specifically for spawning response of gravid females, egg incubation and rearing fingerlings (Okere *et al.*, 2015). In present study temperature, pH and dissolved oxygen of the water of hatching system were monitored daily using a thermometer, pH meter and oxygen meter respectively.

#### **3.13. Data analysis**

Data on spawning fecundity (SF), fertility rate (FR %) and hatchability rate (HR %) were calculated. Spawning refers to the process of releasing the eggs by the female. Spawning fecundity refers to the amount of eggs obtained per each female fish after hormonal treatments (Ayoola *et al.*, 2012). The absolute spawning fecundity was calculated as the total number of eggs carried by a female fish, was then standardized as number of eggs/g body weight/female fish.

Fertility rate was calculated as the number of eggs fertilized as a percentage of the total number of ova collected (Shourbela *et al.*, 2014; Francis *et al.*, 2013). The opaque eggs at the center and white translucent at periphery were considered as fertilized and those which were whitish at all part considered as unfertilized eggs. The percentage of fertility rate was calculated as described by (Francis *et al.*, 2013).

Fertility Rate (FR %) = 
$$\frac{Numbre of fertilized eggs}{Total number of eggs collected} x100$$

Hatchability rate was calculated as the number of hatched eggs as a percentage of the total number of eggs fertilized (Shourbela *et al.*, 2014; Francis *et al.*, 2013). The mean number of hatchlings in each treatment was obtained by direct counting of unhatched eggs as well as the number of hatchlings in the incubation tray. Percentage hatchability was also calculated as described by (Francis *et al.*, 2013).

Hatchability Rate (HR %) =  $\frac{Number of hatchlings}{Total number of eggs fertilized} x100$ 

One-way analysis of variance (ANOVA) was used to analyze the mean differences in spawning fecundity (SF), fertility rate (FR %) and hatchability rate (HR %) among the three treatment groups. When significant differences were observed Post-Hoc Tukey test was employed to determine specific groups with significant differences. Test of homogeneity of variance among the groups was done using Leven's statistic. When Leven's test returned significant, a more robust Welch statistic was used for the test. A 5% level of significance was used for all the analyses. The statistical analyses were conducted using software package of SPSS (version 16.00) and PAST (Version 2.17).

# 4. Results

# 4.1. Spawning fecundity

The standardized spawning fecundity for the three treatment groups is given in (Table 1 and Fig .10).

Table 1. Summary of the mean and range of the standardized spawning fecundity (eggs/g body weight) among the three treatment groups (Mean  $\pm$  SE)

Treatments	Ν	Mean	SE	Range
CG	3	9731.6	1.860	7098.31-13325.89
CC	3	5813.8	5.028	4896.99-6630.17
SH	3	5666.6	3.264	5131.25-6257.80



Fig.9. Spawning fecundity (mean  $\pm$  SE) among the three treatment groups

The mean difference in spawning fecundity of the three treatment fish groups was statistically not significant (Welch F statistic = 0.27, p = 0.073). However, the effect of size measured as Partial Eta squared was 58.2 % and the power of ANOVA test was 50.3 %.

# 4.2. Fertility Rate (FR %)

Results for the fertility rate are given by Table 2 and Fig.11.

Table 2. Comparison of the fertility rate (FR %) among the three treatment groups (Mean  $\pm$  SE)

Treatments	Ν	Mean	SE	Range
CG	3	84.3	2.112	80.50-87.80
CC	3	80.6	1.778	78.30-84.10
SH	3	74.9	2.818	70.80-80.30

The mean value of the Fertility rate (%) was showed by graphical presentation.



Fig.10. Fertility rate (%) (mean  $\pm$  SE) for the three treatment groups

The mean value of fertility rate among the three groups was higher in CG than CC and less in SH recipient female *C. gariepinus*, respectively. The One-way ANOVA result for fertility rate among the three hormone treatment groups were statistically not significant (p = 0.069). However, the effect of size measured as partial Eta squared was 59 % and the power of ANOVA test was 51.7 %.

# 4.3. Hatchability rate (HR %)

Results for the hatchability rate is given by Table 3 and Fig.12.

Table 3. Comparison of the hatchability rate (HR %) among the three treatments groups (Mean  $\pm$  SE)

Treatments	Ν	Mean	SE	Range
CG	3	73.3	6.24	60.90-80.80
CC	3	63.5	4.38	58.20-72.20
SH	3	51.5	1.79	48.60-54.80



Fig. 11. Hatchability rate (%) (mean  $\pm$  SE) for the three treatment groups

The One-way ANOVA result for hatchability rate among the three hormone treatment groups was significant (p = 0.04). The effect of size measured as partial Eta squared was 65.9 % and the power of ANOVA test was 64.5 %. The post-Hoc Tukey test for hatchability rate (HR %) showed that the difference existed between fish treated with pituitary extracted from *C. gariepinus* (CG) and fish treated with synthetic hormone (SH) with p = 0.033).

#### **4.4. Induction Hours**

Induction hour (latency period) is the period from injection till the onset of ovulation (hrs) or time taken from injection of female brood fish to time of stripping (Shourbela *et al.*, 2014). In the present experiment the induction responses for each treatment recipients of *C. gariepinus* females was almost similar (Fig. 12). The females *C. gariepinus* injected with synthetic hormone responded faster than the other two treatments.



Fig.12. Induction responses (hrs) for the three treatment groups

# 4.5. Water quality parameters

The mean value of water quality recorded during experimental period was present in Table 4.

Table 4.	The mean	value of	water	quality	parameters	recorded	in the	hatching	system
					1			0	2

Parameter	Result		
Appearance	Clear with no visible particles		
рН	6.7		
Dissolved oxygen (DO) mg/ l	5.6 mg/L		
Temperature	27.0°C		

Temperature, pH and dissolved oxygen of the water of hatching system were checked daily using thermometer, pH meter and dissolve oxygen meter respectively.

#### 5. Discussion

The best spawning fecundity result was recorded from treatment group whose ovulation was stimulated with injection of *C. gariepinus* pituitary extract with a mean of 9731.6 eggs/g body weight/female. The mean values for spawning fecundity of females *C. gariepinus* injected with pituitary extracted from *C. carpio* and synthetic hormone were 5813.8 and 5666.6 eggs/g body weight/female, respectively. One-way ANOVA for spawning fecundity among the three hormone treatment groups was statistically not significant (Welch F statistic = 0.27; p = 0.073). However, the effect size or magnitude of mean difference measured as partial Eta squared (58.2 %) was found to be very large (Pallant, 2007). Moreover, the power of ANOVA test (50.3 %) was poor, i.e. < 80 % (Pallant, 2007). The lack of statistical significance among the treatment groups could be due to the poor power of ANOVA test, which in turn is because of the small sample size (n = 3) per each treatment group. Therefore, based on the justification for the poor power of ANOVA test and the very large effect size, the spawning fecundity of the group treated with the *C. gariepinus* pituitary can be considered as a better reaction than that of the other two treatment groups (*C. carpio* pituitary and synthetic hormone).

The absolute mean spawning fecundity results of the natural hormone treatments (*C. gariepinus* and *C. carpio*) in the present study (63,653.66 and 49,150.66 eggs per female, respectively) were less than the findings of Gadissa and Devi (2013) (69,939.00 and 54,633.00 eggs per female, respectively. The lower values in the present study could be attributed to the hormone dosage differences where higher dosage (3 ml/kg body weight) was used than the present study (2 ml/kg body weight). Eventhough, this report showed large figures than the present study it does not state the body weight of the brooder fish. The body weight of the brooder fish is the most important factor which affects the spawning fecundity of the brooder fish. For instance, De Graaf and Janssen (1996) reported that brooder fish can spawn 5 % of their body weight. Similarly, spawning fecundity of the present experiment with synthetic hormone recipients was less than the report of (Fame *et al.*, 2005). The spawning fecundity reported by this study was 77,000.00

whereas the result of the present experiment was 64,189 .00 mean spawning fecundity. The difference in the spawning fecundity of the present experiment and the report of Fame *et al.* (2005) could be attributed due to the types of synthetic hormone and dose applied. The present experiment used the combination of LHRH-A<sub>2</sub> + Domperidone at dose of 1 ml/ kg, but this report used only LHRHa at dose of 2 ml/ kg.

The fertility rate recorded in the present experiment was high with a mean of 84.3 % in C. gariepinus females injected with pituitary extracted from C. gariepinus and followed by C. *carpio* pituitary recipients with a mean fertility rate of 80.6 %. The lower mean fertility rate 74.9 % was recorded in C. gariepinus female's injected with synthetic hormone. The One way ANOVA for fertility rate among the three hormone treatment groups was statistically not significant (p = 0.069). Leven's statistic was applied to check test of homogeneity of variance among the groups and shows greater than 5 % level of significance (p = 0.063). The effect size (partial Eta squared) shows 59 %, which was considerably large (Pallant, 2007). Additionally, the power of test was also poor i.e. 51.7 % (Pallant, 2007). The lack of statistical significance among the treatment groups could be due to the poor power of ANOVA test. This means that the fertility rate among the groups was practically large, but it could not be detected by ANOVA, which could be because of the small sample size (n = 3) per each treatment group. Therefore, based on the justification for the poor power of ANOVA test and the very large effect size, the fertility rate of the group treated with the C. gariepinus pituitary can be considered as a better reaction than that of the other two treatment groups (C. carpio pituitary and synthetic hormone).

The fertility rate of fishes that were injected with *C. gariepinus and C. carpio* (84.3 % and 80.6 %, respectively) in the present study were somewhat higher than the report of Gadissa and Devi (2013), (76.9 % and 80.5 % fertility rate, respectively). The difference in fertility rate between this report and the present study could be due to differences in the condition at which eggs incubated and hatchery facilities (pH, temperature and dissolved oxygen). Similarly, the fertility rate of the present experiment was higher than the work reported by

Ayoola et al. (2012) in Nigeria. This report showed, pituitary extracted from C. gariepinus and synthetic hormone (Ovulin) was applied for the inducing of female C. gariepinus. From both treatments fertility rate yielded was 60.7 % and 67 %, respectively. On the other hand, the fertility rate of the present experiment with the synthetic hormone recipients of the female C. gariepinus was similar with the reports of Adobayo (2006); Fame et al. (2005) which showed 75 % fertility rate. These reports used synthetic hormone (LHRHa) and (Ovaprim) at a dose of 2 ml/kg and 0.5 ml/kg body weight of the female C. gariepinus respectively. In the contrary, the present experiment showed poor fertility rate when the synthetic hormone is applied on female C. gariepinus. This become reasonable when compared to other reports (Shourbela et al., 2014; Ndimele and Owodeinde, 2012; Olumuji and Mustapha, 2012). These reported as 83.17 %, 88.4 % and 88.7 %, fertility rate respectively from females C. gariepinus. The appeared variation between the present experiment and those reports on the fertility rate result could be due to the synthetic hormone type. This shows that GnRha + Dompridone for the former one and Ovaprim for the last two respectively. Additionally incubation of eggs, hatchery facilities (pH, temperature and dissolved oxygen) and fertilization time and situation may also play the role.

Table 3 illustrates that, in the present experiment the highest mean hatchability rate 73.3 % was recorded from eggs collected from *C. gariepinus* pituitary extract recipients of females *C. gariepinus*. Meanwhile, the mean hatchability rate for *C. carpio* pituitary recipient was 63.5 %. The hatchability rate was comparatively less (51.5 %) for synthetic hormone recipient female C. *gariepinus*. The One-way ANOVA result for hatchability rate among the three hormone treatment groups was significant (p = 0.04). The effect size measured as partial Eta squared (65.9 %) was high value (Pallant, 2007). The Power of ANOVA test was 64.5 % which appears to be less when compared to the standard (power > 80 %) to be good power of test (Pallant, 2007). In fact, the Post-Hoc Tukey test of the hatchability rate recorded in the present study showed significant variation (p = 0.033) between the *C. gariepinus*. In the present experiment, the mean hatchability rate recorded with the

natural pituitary extract i.e. pituitary from *C. gariepinus* and *C. carpio* was 73.3 % and 63.5 % respectively. This record was better when compared to similar report that used the same source of pituitary for inducing female *C. gariepinus*. For instance, the report of Gadissa and Devi (2013) showed that, the mean hatchability rate was 45.3 % and 42.9% from females *C. gariepinus* which were injected by *C. gariepinus* and *C. carpio* pituitary extract respectively. The enhanced result recorded by the present study could be due to the hatchability facilities. Even if, the other parameters does not reported by this study the temperature at which eggs incubated was  $23^{\circ}$ c, which is lessthan the optimum water parameters sited for the eggs and larvae incubation. But, in the present study eggs are incubated at temperature of  $27^{\circ}$ c, which is reported as the optimum temperature for egg and larvae incubation by (Potongkam and Miller, 2006). Similarly, the hatchability rate recorded by the present experiment (73.3 %) was good as compared to the result (60.7 %) of Ayoola *et al.* (2012), using *C. gariepinus* pituitary treatment.

Likewise, the hatchability rate of the present study was noteworthy when the synthetic hormone treatment used, because more than half 51.7 % was achieved. In contrary, it is less when compared to other works (Shourbela et al., 2014; Ndimele and Owodeinde, 2012; Olumuji and Mustapha, 2012; Adebayo, 2006). These reports showed 89.1 %, 71.7 %, 57.7 %, and 65 % hatchability rates respectively. All these studies used female C. gariepinus for treatment application purpose and the synthetic hormones used were Ovaprim for the first three and combination of GnRha + Dompridone for the last one respectively. The difference in the hatchability rate result between those reports and the present experiment were mainly due to the synthetic hormone variation in addition to the dose. In the same way the hatchability rate of the present experiment was not as much of the report of Fame et al. (2005) with the application of synthetic hormone. The present experiment recorded only 51.5 % for C. gariepinus induced with LHRH-A2 + Dompridone but, Fame et al. (2005) reported 67 % hatching success for C. gariepinus induced with LHRHa. The Hatching rate success difference in between this report and the present experiment could be due to hormonal dose. The hormone dose of this report was 2 ml/kg body weight while only 1 ml/kg body weight in the present experiment. Moreover,

condition at which eggs incubated and hatchery facilities (pH, temperature and dissolved oxygen) could play a great role on the hatchability rate of the incubated eggs.

Induction hour (latency period) can be described as the time interval between injection of the female fish and stripping of eggs. There are some other factors to be considered simultaneously with latency period. The water in which the female brood-stock is kept after being injected has to be of the right temperature, as this can affect the latency period. The higher the temperature the lower the latency period (Crandell *et al.*, 1995). The optimum temperature to keep the fish is 25 to 28°C and the fish can be ready in about 11-13 h (FAO, 1996).The ripening of the ovary after injection depends on the type of hormone used to introduce the female fish (Crandell *et al.*, 1995). The fish breeder must properly monitor the exact latency period of the fish to avoid over-ripeness and under-ripeness of the eggs in order to achieve maximum spawning, fertilization, hatchability and survival of the hatched ones (Zonneveld *et al.*, 1988).

The induction hour of the present study was varied from 11.5hrs to 12.5hrs which shows only one hour difference. The group which react fastlly at 11.5hrs was those *C. gariepinus* females received synthetic hormone. The others which injected by *C. gariepinus* and *C. carpio* pituitary extracts were responded at 12hrs and 12.5hrs respectively. For all the three treatments the induction hours were not statistically shows significant variation while all incubated at temperature of  $27^{\circ}$ c. Though, the induction time result of the present study was nearly similar with that of Brzuska (2003) who reported that combination of GnRha with Domperidone shows latency time of 13hrs. There was a difference of 1.5hrs between this report and the present experiment with the synthetic hormone applied on *C. gariepinus* females. Additionally, from the same work Brzuska (2003) reported that, the latency time of 12hrs when *C. carpio* pituitary extract was applied on *C. gariepinus* females for induction purpose. This was analogous with the present experiment when *C. gariepinus* females were injected with the *C. carpio* pituitary extract. In the same way, Shourbela *et al.* (2014) reported that the latency time was 11.5hrs in female *C. gariepinus* injected with Domperidone +GnRha. Similarly, the report of Olumuji and Mustapha (2012) showed that

female *C. gariepinus* induction time was 11.5 hrs when injected with Ovaprim. Even though, the present experiment used both the natural and synthetic hormone for inducing reproduction of *C. gariepinus* females the induction time was note shows this much variation. Although, other reports used different sources for the inducing purpose, but the induction time shows more or less similar with the present experiment.

Monitoring water quality parameters in culture systems is very important as the variables influence fish physiological processes. However, in African catfish negative impacts of low water quality are quite rare, as adults are relatively tolerant to a range of water parameters, but opposed to its juveniles. This is obvious that water temperature, oxygen and pH affect the physiology of gravid brooder, the incubated eggs and larvae. Subsequently, the growth and survival of fish that results from stressed eggs are also affected (Okere et al., 2015; Musiba et al., 2014). The water parameter monitored during the present study were Temperature, pH and dissolved oxygen at hatching system and the average result was shows as 27<sup>o</sup>c, 6.7 and 5.6 mg/L respectively. The recorded water parameters of the present study were more or less similar with the average water quality suggested for egg and fingerling incubation by Potongkam and Miller (2006), who reported as the best medium for egg and fingerling incubation at Temperature (25-30°C), pH (6.5-8.0) and Oxygen (5-8 mg/L). Additionally, the values of water quality parameters of the present study were somehow similar with the reports of Ndimele and Owodeinde (2012) which used as water temperature, pH and dissolved oxygen values of  $28^{\circ}$ C, 6.7 and 4.3 mg/L respectively. Therefore, the present study was conducted at the optimum water parameters for the broodstock, eggs and larvae. This implies that there was no influence of these parameters on the result of the present study.

# 6. Conclusion and Recommendation

#### 6.1. Conclusion

The present study showed the possibility of artificial propagation of African catfish using various hormone treatments, which gives hope for the continued supply of fingerlings in the aquaculture of African catfish.

The use of piscine pituitary hormone treatment from fish offal, particularly an extract from *C. gariepinus*, is more preferable in the artificial propagation of African catfish. This is because in the present study better performance in SF, FR and HR was obtained from a group treated with natural pituitary extracts, especially from *C. gariepinus*.

Furthermore, the use of pituitary extract from fish offal is more feasible as it is locally available at relatively cheaper price than synthetic hormones which are only hardily available and more expensive.

# **6.2. Recommendations**

Further study on feed preference and survival rate of larvae is recommended to augment the present study and thus for better insight about artificial propagation of African catfish.

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