Sudan University of Science and Technology

College of Animal Production Science and Technology

Department Of Fisheries and Wildlife Science

Induced ovulation and spawning of African catfish (*Clarias gariepinus)* using different dose of ovaprim

أثر مستويات مختلفة من هرمون الاوفابريم علي معدل الفقس و البقاء في أسماك القرموط

A dissertation Submitted in Partial Fulfillment of the Requirement of the B.Sc. Degree in Fisheries and Wildlife Science (Honor).

Prepared By:

1. Ahmed Adam armed Mohamed
2. Babeker Ibrahim Osman
3. Tajaldeen Mohamed Abdalmwla

Supervisor: Dr.Fouzi Ali Mohamed

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الآيـــة

قال تعالى :

أعوذ بالله من الشيطان الرجيم

بسم الله الرحمن الرحيم

((وهو الذي سخر البحر لتأكلوا منه لحماطريا وتستخرجوا منه حلية تلبسونها وترى الفلك مواخر فيه ولتبتقوا من فضله ولعلكم تشكرون 14)) صدق الله العظيم

سورة النحل ص 268

DEDICATION

To my Parent

And all my family

To My Friends And all my Teachers

I hope all your dreams come true

To all we are grateful

With all our doaa

Our sincere thanks also extends to all members of our department and faculty. Beloved family's especially for the mom's Rogya Hessen, Hawa Abubker, Fatima Mohammed.

Acknowledgement

All gratitude is goes to Allah who guided us to bring forth to light this study. We feel indebted to our supervisor Dr.Fouzi Ali Mohamed for his skilfulGuidance and invaluable suggestion at various stages of this work, we simply cannot find the right words to express our gratitude to him, patience, and advice and unlimited support were our light to find out our way throughout the study period.

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ABSTRACT

The main objectives of this study w induced to spawning using different dose of synthetic hormone Ovaprim. As the spawning performance of *Clarias gariepinus* induced to spawningusing different dose of synthetic hormone Ovaprim. This study was conduct at the Fish Hatchery Unit, at Department of Fisheries Science and wildlife, College of Animal Production Science and Technology, Sudan University of Science and Technology, during the period from July to October 2018.The brood stock used for the study were collected and conditioned for a week before the inject and feeding by control feed. Fish were fed at 2,5% of body weight twice daily. The selected brood stock were kept separately in different ponds without feeding them, after they were injected with 5 ml Ovaprim per kg live weight and then left for 12 hours latency period as a post adulatory maturation period and to ensure high hatching rates and low proportion of deformed larvae. Data collected was analyzed using simple destructive statistics to determine the mean and percentage .The results showed that high larval production (50%) in the treatments II out of which 10% survived rate, indicates an overall good egg quality and effectiveness of ovaprim, including ovulation and spawning in the African catfish. There was difference among number of fertilized eggs, number of hatched eggs, % larval production and survival in all three treatments.

Keywords: *Spawning, Ova prim, hatching, C.gariepinus.*

الخلاصة:

الهدف من هذه الدراسة معرفة كفاءة مستويات مختلفة من هرمون الاوفابريم علي معدل الفقس في اسماك القرموط الافريقي (القرموط) اجريت الدراسة الحالية بمفرخ الاسماك بقسم علوم الاسماك والحياة البرية – كلية علوم و تكنولوجيا الانتاج الحيواني بجامعة السودان للعلوم والتكنولوجيا خلال الفترة من شهري يوليو الى اكتوبر2018. الامهات التي استخدمت في هذه الدراسة تم عزلها واقلمتها قبل اسبوع من اجراء الحقن وتم التحكم في الغذاء بتغذيتها بنسبة 2.5% من وزن جسمها مرتين في اليوم . وتم وضع الامهات المنتخبة في احواض منفصلة عن بعضها البعض الذكور والاناث كل على حدى وتم ايقاف التغذية قبل 24ساعة من الحقن . وبعدها تم حقن كل انثى ب(5مل) من الهرمون وبعد 12ساعة من الحقن تم تحفيز الاناث وتم الحصول على معدل عالي من البيض المخصب مع وجود كمية قليلة من من البيض غير المخصب.البيانات المتحصل عليها من الدراسة تم تحليلها احصائيا بواسطة تحليل احصائي بسيط لتحديد المتوسط والنسبة المئوية لمعدلات الفقس. اظهرت النتائج معدل تفقيس عالي من اليرقات بمعدل 50% في التجربة الثانية وبمعدل بقاء 10% وهذا مؤشر جيد لتاثير هرمون الاوفابريم في التحفيز لانتاج بيض ذو جودة عالية في سمكة القرموط الافريقي. واظهرت النتائج فروق في البيض المخصب ومعدل التفريخ واليرقات المنتجة ومعدل البقاءفي الثلاث تجارب.

الكلمات المفتاحية. *التفريخ ،أوفابريم، الفقس، اسماك القرموط*

CHAPTER ONE
INTRODUCTION

1.1 Background

The African catfish, *Clarias gariepinus* Burchell 1822, is the favorite fish for aquaculture in West Africa (**AdewumiandOlaleye, 2011**). This species of fish dwells in a variety of freshwater environments including still water(lakes, ponds and pools); and flowing water (rivers, rapids and dams). *C. gariepinus*has been vastly cultured and subjected to intensive research in aquaculture due to its ease of culturing, fast growth rate, high resistance to disease, tolerance of a wide range of temperature, low dissolved oxygen as well as high salinity levels and most importantly high commercial value (**Teugels, 1986a).**

1.2 Study problem

The major limitation in the aquaculture of *C. gariepinus*is that the species does not breed freely in captivity (**Adebayo and Fagbenro, 2004**). This is as a result of stress induced ovarian atresia (**Lubzens et al., 2010**).

1.3 Justification

Hormonal induction of reproduction is necessary for the catfish to overcome atresia. This is done either by injection of pituitary gland from the donor fish of equivalent weight to the female spawned (**Fagbenro*et al.*, 1998; Salami *et al.*, 2006**) or by injection of synthetic gonadotrophin releasing hormone analogues (GNRH-a). The synthetic equivalents are quite expensive for many local farmers and hatcheries in Africa while the use of raw pituitary gland could be cumbersome. Pituitary gland is the main source of the major hormones responsible for reproduction in animals. The pituitary

cells have been reported to undergo continuous mitotic process (**Yeung *et al*., 2006**). The pituitary gland can be cultured and proliferated *in vitro* in order to use their secretions for induction of spawning in catfish (**Chen et al., 2010**). Cells growth, division and multiplication can be achieved by addition of fetal bovine serum and culture medium to the pituitary cells in a culture plate at 30°C and 5% CO2 (**Lubzens*et al*., 2010**).

4.1 **objectives**

1. To investigate the spawning performance of *Clarias gariepinus* induced to spawning using different dose of synthetic hormone Ovaprim.

CHAPTER TOW
LITERATURE REVIEW

2.1 Taxonomy

Species:*Clarias gariepinus* (**Burchell, 1822**)

Family: Clariidae

Order: Siluriformes

Class: Actinopterygii

African sharptooth catfish *Clarias gariepinus* is a typical air-breathing catfish with a scaleless, bony elongated body with long dorsal and anal fins, and a helmet like head. Colorvaries dorsally from dark to light brown and is often mottled with shades of olive and grey while the underside is a pale cream to white (Skelton 2001). It can grow very large with a maximum reported length of 170 cm (IGFA 2001) and weight of 60 kg (Robbins et al. 1991).The genus Clarias was reviewed in the 1980s, which resulted in several widespread species being synonymized (*Clarias capensis* of southern Africa, C. mossambicus of central Africa and C. lazera of west and north Africa) under the name *Clarias gariepinus* (**Teugels 1986**).

Although more than 100 different species of the Genus Clarias have been described in Africa, a recent systematic revision based on morphological, anatomical and biographical studies has been carried out by **Teugels (1982a, 1982b, 1984**), who recognized 32 valid species. The large African species which are of interest for aquaculture belong to the subgenus Clarias. In earlier systematic studies on the large African catfish species **Boulenger (1911)** as well as **David (1935**) recognized five species of within this subgenus. Both authors used morphological criteria such as form of

vomerine teeth, ratio of vomerine to premaxillary teeth band and the number of gill rakers. The five species were;

1. *Clariasanguilarus*
2. **Clariassenegalensis*
3. *Clariaslazera*
4. *Clariasmossambicus*
5. *Clariasgariepinus*

2.2 Natural distribution and habitat

The native range of C. gariepinus covers most of the African continent, with the exception of Maghreb, Upper and Lower Guinea, and the Cape provinces of South Africa (**Picker & Griffiths 2011**) According to **Skelton (2001**) it is probably the most widely distributed fish in Africa. **Jubb (1967)** describes its natural distribution as occurring as far south as the Orange River system in the west and the Umtamvuna River in the east of South Africa. *C. gariepinus* is widely tolerant of many different habitats, even the upper reaches of estuaries, but is considered to be a freshwater species. It favours floodplains, slow flowing rivers, lakes and dams (**Skelton 2001**). It can tolerate waters high in turbidity and low in dissolved oxygen, and is often the last or only fish species found in remnant pools of drying rivers (**Safriel&Bruton 1984, Van der Waal 1998**).

2.3. Biology

2.**3.1. Description of the genus and species**

The catfish genus 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 containing only soft fin rays (Figure3). The outer pectoral ray is in the form of a spine and the pelvic fin normally has six soft trays. The head is flattened, highly ossified, the

skull bones (above and on the sides) forming a casque and the body is covered with a smooth scaleless skin. The skin is generally darkly pigmented on the dorsal and lateral parts of the body. The colour is uniform marbled and changes from greyish olive to blackish according to the substrate. On exposure to light skin the colour generally becomes lighter.

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.

Asupra-branchial or accessory respiratory organ, composed of a paired pear-shaped air-chamber containing two arborescent structures is generally present. These arborescent or cauliflower-like structures located on the secondhand forth branchial arcs, are supported by cartilage and covered by highly vascularised tissue which can absorb oxygen from atmospheric air **(Moussa, 1956**). The air-chamber communicates with the pharynx and with the gill-chamber. The accessory air breathing organ allows the fish tosurvive for many hours out of the water or for many weeks in muddy marshes.

2.3.2. Diet and mode of feeding

Clariasgariepinus is considered to be omnivorous displaying both scavenging and predatory behaviour (**Bruton 1979a**). It is known to have an extremely varied diet consuming fruits and seeds, all types of aquatic invertebrates and small vertebrates, small mammals and even plankton **(Bruton 1979a, Skelton 2001**). Larger individuals show a specific dietary shift towards fish as they grow bigger (**Willoughby &Tweddle 1978**). However, inactive foods, which it detects with its sensory barbells before securing with its array of very fine teeth prior to gulping, are generally preferred (**Bruton 1979a, Skelton 2001**). Alternatively, it can be an

efficient predator and even hunt in ‘packs’ where it may herd shoals of small fish against submerged aquatic vegetation before devouring them **(Merron 1993)**. Solitary feeding, social hunting and coordinated pack-hunting foraging behaviours and even feeding migrations have all been observed **(Bruton 1979a, Merron 1993**).

2.3.3. Habitat

Clarias spp. inhabit calm waters from lakes, streams, rivers, swamps to floodplains, some of which are subject to seasonal drying. The most common habitats frequented are floodplain swamps and pools in which the catfish can survive during the dry seasons due to the presence of the accessory air breathing organs **(Bruton, 1979a; Clay, 1979).**

2.3.4 Reproduction

Shoals of the fish migrate upstream or to the shores of still water bodies prior to breeding (**de Moor &Bruton 1988**). Courtship, spawning and egg laying takes place at night often after rain (**Bruton 1979b**). Eggs usually adhere to submerged vegetation, either aquatic or terrestrial vegetation that has recently been submerged as a result of seasonal water level rise (**Bruton 1979b**). Hatching of the eggs occurs soon after spawning, usually after 24 to 36 hours (**Bruton 1979b).** There is no parental care of the young (Hecht et al. 1988). Average fecundity of Lake Sibaya C. gariepinus was found to be approximately 45 000 eggs for a 2 kg fish (**Bruton 1979b**).

2.3.5 Breeding

In order to boost fish production on a level that will be able to serve people as reliable source of protein, there is a need to be able to reproduce fish on a large basis in and out of season to ensure regular supply. Artificial reproduction and selective breeding has become very popular nowadays in the age of science and technology as a means of ensuring large scale

production of fish seeds throughout the year. Artificial propagation methods constitute the major practicable means of providing enough quality seed for rearing in confined fish enclosure such as fish ponds, reservoirs and lake (**Charo and Oirere, 2000).**

Once they reach a weight of approximately 200 g.Reproduction infish is controlled by several factors which include sex steroids in the regulation of reproductive processes (**Kime, 1993).** These eproductive processes are controlled through the brain-pituitary-gonadal axis. The brain is stimulated by environmental cues like water rise, temperature, feeding, rainfall, and photoperiod to release gonadotropin releasing hormones **(Zohar *et al*., 2010).** Ovulation and spermiation are effected as a result of the sex steroids that have been produced. Administration of gonadotropin releasing hormone analogue has been reported to increase levels of plasma sex steroids in fish (**Zhuo*et al*., 2011).**

The use of synthetic hormones in female fish is now popular as a means of artificially inducing the female fish in order to ovulate. However, Zhuo et al., 2011 has shown from his study that Gonadotropin releasing hormone analogue multiple injection potentially accelerated testicular maturation of male yellow catfish. The use of both synthetic and natural hormones brings about quick ovulation and higher percentage of hatched fish, though synthetic hormone gives higher yield than the natural hormones **(Krol*et al*., 2006).**Gonadotropin Releasing Hormone analogue (GnRHa) is now the best available biotechnological tool for the induced breeding of fish. Ovaprim and Ovatide are both synthetic hormone preparation containing salmon gonadotropin releasing hormone analogue and domperidone(SGnRHa + Domperidone) which are usually used for spawning induction in catfishes to get quality seed **(Sahoo*et al*., 2005)**.

As shown by researchers, synthetic hormones are best in inducing ovulation in female fish towards the yield of viable seeds on regular basis, there is a need to also cause the same effect of viable seed production by inducing spermiation in male fish. The use of hormones in female fish is gaining popularity each day while there is little work on inducing spermiation in male fish. This study was necessary to test and compare the effects of synthetic hormones on the milt quality of *C. gariepinus* and the resultant effects of the induced milt on seed yield and quality. African Catfish *(Clariasgariepinus*) is widely considered as the leading cultured fish in the country. Some of the credentials of African catfish are: high growth rate reaching market size of 1 kg in 5–6 months under intensive management conditions: highly adaptable and resistant to handling and stress; can be artificially propagated by induced spawning techniques for reliable mass supply of fingerlings; commands a very high commercial value where it is highly cherished as food in Nigerian homes and hotels (**Olaleye, 2005**). This fish shows a seasonal gonadal maturation which is usually associated with the rainy season. The maturation processes of *C. gariepinus* in nature are generally influenced by annual changes in water temperature and photoperiodicity and the final triggering of spawning is usually caused by a raise in water level due to rainfall **(De Graaf*et al*., 1995).** The female African catfish has a fully developed ovary which contains "ripe" eggs all year round, if kept in ponds and water temperature kept above 22 0C. The eggs of a "ripe" female make up 15-20% of the body weight. In captivity the African Catfish does not spawn spontaneously since the environmental factors such as the rise in water level and inundation of shallow areas do not occur on the fish farms. Under natural condition of spawning, lower hatching rates have been reported for *Clarias gariepinus* by various authors.

De Graaf*et al* **(1995)** reported an average rate of 59.1% in the rainy season for C. gariepinus in the Republic of Congo, **while Macharia*et al*** **(2005)** reported a rate as low as 4% for C. gariepinus eggs incubated on a nylon substrate. Fertilization, hatching and early survival of larvae are vital for successful aquaculture of the African catfishes **(Ataguba*et al.*, 2009).Odedeyi (2007**) noted that the largest mature *C. lazera (gariepinus)* would usually give the best spawn weight in induced breeding, but there is no literature available as to whether the fish with the best spawn would equally give the best fry survival and best growth performance. A major pre-requisite for successful fish farming enterprise is a reliable and consistent source of fish seeds (fingerlings) of the commercially important species **(NwubaandAguigwo, 2002**).Induced breeding by hypophysation was developed in India for catfishes such as *Heteropneus tesfossilis* and *Clariasbatrachus***(Dehadrai et al.,1985**), the doses of pituitary extract required for singhi was found to be very high (30 mg/kg). In recent years, a combination of dopamine antagonists and LHRH analogue (LHRAa) has been found successful in ovulation and induced breeding in some teleosts**(Fremin, 1991).** Carp pituitary extract (CPE) and luteinizing hormone-releasing hormone analogue (LHRHa) are two well known hormones for controlling ovulation in channel catfish (**Fobes 2013**). Since the 1990s, a drug known as ovaprim has been commonly used as a spawning hormone in fish breeding .Ovaprim, which is a combination of salmon GnRH analog combined with a dopamine agonist Domperidone, has proved to be extremely successful in breeding of carps with a spawning rate of about 100%. In comparison, the success rate of spawning using GnRH analogs in other fishes are 100% in Grouper (*Epinephelus salmoides*) and 99% in Milkfish (Channoschanos) **(Kelley and Lee, 1986).** Catfishes are a

favorite food in India and Southeast Asia. Of the three species that are chiefly cultivated viz. *Clarias batrachus, C.gariepinus* and *C. macrocephalus,* the first and third are extensively cultured in Asia. Although *C.batrachus* breeds naturally in ponds, the efficiency and rate of induced spawning of catfish with ovaprim has been found to be less than 50% (Ngamvongchon et al., 1988), which is very low compared to that of carps. Sex steroids in female fish perform major roles in oocyte maturation, ovulation and spawning.

Synthesis of vitellogenin and increase in ovarian size during final oocyte maturation is controlled by 17b-estradiol. 17bestradiol is directly related to gonadosomatic index **(*Coccia et al., 2010).***

In fish reproduction under controlled conditions attempts are made to obtain eggs of the highest weight possible and of the best quality, and hence to produce the highest possible numbers of good quality hatch. For this purpose various preparations stimulating ovulation are experimentally tested to find stimulators that would ensure such effects. It is obvious that appropriate maternal (and paternal) material should be used to obtain satisfactory results of stimulated fish breeding. With respect to African catfish (*Clariasgariepinus),* the species of a well-grounded position in European fish culture **(HuKuczyński*et al.*,1999).**

Up to 1995 in the Institute of Ichthyobiology and Fish Culture of the Polish Academy of Sciences at Gołysz ovulation stimulation was carried out in this fish species using carp pituitary homogenate (**Adamek, 1995).** In later years within a program of investigations on the effects of reproduction after ovulation stimu¬lation numerous experiments were carried out using various preparations (of both natural and synthetic origin) (**Brzuska, 2002).**

The satisfactory results of ovulation stimulation with Ovopel observed in various fish species **(Brzuska, 2001)** and numerous merits of thispreparation (i.e. the possibility of precise dosing without weighing the stimulator, the simple method of its preparation for the treatment of fish, the elimination of an additional injection of dopamine receptor blocker, and the possibility of repeated application are a short interval if no ovulation occurs)

distinctly show its high value. Ovopel contains a mammalian GnRH analogue, D-Ala6, Pro9NEtmGnRH- a and water-soluble dopamine receptor antagonist – metoclopramide. The concentrations of D-Ala6, Pro9NEt-mGnRH and metoclopramide are 18–20 μg/pellet and 8–10 mg per pellet, respectively (**Horváth et al., 1997).** Since the preparation was developed for Cyprinidae and the induced reproduction of this family is more effective when the ovulation stimulation is performed with two doses of carp pituitary, **Horváth*et al*. (1997)** recommended applying two doses of Ovopel.The results of studies conducted on European catfish (Silurusglanis L.) show that in the case of Ovopel the two doses (i.e. 1/5 pellet/kg body weightas the priming dose and 1 pellet/kg as the resolving dose) are not necessary if the stimulation is carried out during the season of natural spawning and the spawners are in a good reproductive condition. With the application of one dose of Ovopel (1 pellet/ kg body weight) the results of reproduction were satisfactory **(Brzuska, 2003).**The effects of ovulation stimulated in African catfish (*Clarias gariepinus)* with two doses (1/5 +1 pellet/kg) or one dose (1 pellet/kg) of Ovopel show that the differences in the weight of eggs and their quality (expressed by fertilization percentage and by the percentage of living embryos after 24-hour incubation) between the groups treated with the above doses were statistically insignificant **(Brzuska *et al*., 1998)**. The treatment either with one Ovopel dose or with

two doses did not affect the percentage of fish yielding eggs (in rela-tion to all the fish are hormonal stimulation) or the latent period.

In the investigation conducted by **Tan-Fermin*et al.* (1997)** on Asian catfish *Claria smacrocephalus* Gunther (a tropical freshwater fish of the order Siluriformes) only one dose (0.05 μg/g body weight) of D-Ala6,Pro9LHRH-ethylamide was used as an ovulation stimulator. However, Legendre et al. (2000) reported that both in experimental reproduction and in fry production in Indonesian fry production units of Asian catfish *Pangasius hypophthalmus* (the most common cultured pangasiid catfish throughout Southeast Asia) two injections of Ovaprim, a preparation containing sGnRH-a (D-Arg6,Trp7,Leu8,Pro9Net) and domperidone, were applied.In conducting controlled fish reproduction particular attention should be paid to the reduction of stress threatening the spawners during numerous manipulations accompanying it. Among other factors hormonal injections are responsible for the stress.The reduction of their number seems justified if it does not negatively affect the reproduction results. The African catfish *(Clarias gariepinus)* – a valuable species with a well-grounded position in European aquaculture **(Huisman and Richter, 1987)** is highly sensitive to stress. Therefore it is advisable to reduce the number of manipulations associated with ovulation stimulation in this species.Artificial propagation methods constitute the major practicable means of providing enough quality seed for rearing in enclosures such as fish ponds, reservoirs and lakes **(Charo and Oirere, 2000).** Fish seed production efficiency of many fish farms' hatcheries throughout sub-Saharan Africa or developing countries like Nigeria is mostly low due to poor handling of brood stock **(Aiyelari et al 2007).** Many hatcheries in Nigeria are facing the problem of poor spawning and low hatchability of *C. gariepinus* although it is widely

cultured in Nigeria. Poor egg quality and low hatching rates are amongst the difficulties most often reported by fish farmers **(Adewumi and Olaleye 2011).** Fertilization, hatching and early survival of larvae are vital for successful aquaculture of the African catfishes **(Haniffa *et al* 2000)** and this has been investigated earlier **(Ataguba *et al* 2009).** Proper handling and health status of female brood fish has been reported to be of great importance in the reproductive performance of fish (**Aiyelari*et al* 2007).** Generally research studies had focused on hatching success in relation to environmental variables such as temperature **(Oyelese, 2006),** and CaCO3 water hardness (**Silva *et al* 2003).** A small but sexually matured fish is an indication of either genetic inheritance or nutritional deficiency. Size is directly proportional to fecundity and egg size **(Bromage and Roberts 1995).**

2.3.6 Growth

Clarias gariepinus is considered to have a rapid growth rate (in length and weight), the rate of which strongly depends on ambient conditions and habitat **(Bruton&Allanson 1980, Hecht &Appelbaum 1987, Britz&Pienaar 1992)**. Growth has been found to be positively density dependent **(Hecht &Appelbaum 1987).** Individuals have been recorded to reach 200 mm SL within a year (**Bruton&Allanson 1980, Skelton 2001).** In females, the growth rate decreases after 3 years resulting in the males reaching larger sizes **(Skelton 2001).** Individuals of this species are known to live for eight or more years **(Bruton&Allanson 1980).**

2.3.7 Environmental tolerance ranges

Clariasgariepinus can endure extremely harsh conditions **(Skelton 2001).** It is able to tolerate very low oxygen concentrations and even survive for considerable periods out of water, via the use of a specialized

suprabranchial organ **(Safriel&Bruton 1984, Hecht et al. 1988).** This organ is a large paired chamber with branches above the gill arches specifically adapted for air breathing **(Maina&Maloiy 1986)** and allows it to move over land even when not forced to do so by **drought (Welman 1948, Johnels 1957)**. Water temperatures between 8 and 35°C, salinities of 0 to 10‰ and a wide pH range are all tolerated (**Safriel&Bruton 1984).***C. gariepinus* exhibits high growth rates between 25 and 33 °C, with optimum growth recorded at 30°C **(Britz& Hecht 1987).** The ability of the fish to be able to tolerate these extreme conditions allows it to survive even in moist sand or in borrows with an air-water interface **(Bruton 1979c, Van der Waal 1998).**

CHAPTER THREE
MATERYALS AND METHOD

3.1 Study area

The study was conduct at the Fish Hatchery Unit, Department of Fish Science and wildlife, College of Animal Production Science and Technology, Sudan University of Science and Technology, during the period from July to October 2018.

3.2 Catfish brood stock for the trial and hormone injection

The brood stock used for the study were collected and conditioned for a week before the injection and feeding by control feed. Fish were fed at 2,5% of body weight twice daily. The selected brood stock were kept separately in different ponds without feeding them, after they were injected with 0.2 ml Ovaprim per kg live weight and then left for 12 hours latency period as a post ovulatory maturation period and to ensure high hatching rates and low proportion of deformed larvae. The development process from fertilized eggs to hatching is dependent upon water temperature while hatching rate is, next to egg quality, dependent on the temperature, oxygen level. After stripping of the induced female brood stock, the eggs were weighed. The male was sacrificed to obtain the gonads which house the milt. The mixture of eggs and milt was stirred gently for about 1.0-2.0mins to allow contact and adequate fertilization. Within a few minutes after fertilization, the eggs absorbed water and could become sticky so the eggs were distributed in netting suspended in the hatching trough (70cm x 40cm x 30cm). The incubated eggs were monitored and temperature maintained between 26 0C -27 0C for incubation between 24– 30 h.The percentage (%)

fertility and hatchability were determined subjectively after 24-30 hours of fertilization by identifying the healthy developing eggs which were transparent green brownish in colour**(Coppens, 2007)** while the dead eggs were also estimated:

% Fertility = (No. of fertilized eggs / No. of Extruded eggs) X 100%

% Hatchability = (Total no. of fertilized eggs-Total no.of unfertilized eggs) X 100%÷Total no. of fertilized eggs

This was done by allowing the newly hatched larvae of all the treatments and that of the control to live on the remains of their yolk sacs for the first 3 days **(Heicht*et al*., 1996)** after hatching out of the eggs and thereafter carefully removed from the hatching troughs and were fed with nutrient.

Irregularities in the activities of the fry in terms of feeding, movement in water was observed at the same time taking note of the dead fry which were removed immediately to avoid contamination of water. Survivability evaluation which was observed for a period of about 3 – 4 weeks was done for each stage of the experiment together with fertility and hatchability for fresh (control experiment). The post-hatching survivability was evaluated as follows:

$$\% \text{ Survival} = \frac{(\text{Total no. of larvae} - \text{No. of dead larvae}) \times 100\ \%}{\text{Total no. of larvae}}$$

3.3 Statistical analysis

Data collected was analyzed using simple destructive statistics to determine the mean and percentage using SPSS (16).

CHAPTER FOUR

RESULTS

According to our results, fish of TI and TIII did not respond to injection and thus spawning did not occur. There were differences between experimental groups in terms of hatchability rate (table1).The highest values of spawning rate were observed in TIIand the lowest spawning rate among all experimental groups TI(table 2). The highest values of egg weight/g.bw obtained in fish administrated with ovaprim (5 ml/kg.bw: TII).

Table (1): latency period pseudo- gonad somatic index of C.grapeinus varying female: male brood stock ratios.

Parameters	**Treatment I**	**Treatment II**	**Treatment III**
No. males	2	1	2
No. female	3	3	3
Total weight	3469	2540.9	2272.8
Mean weight	693.8	635.2	454.6
Total dosage	10	15	11
Ovaprim dosage	3.3	5	3.7
Hatchability	(-)	(+)	(-)

Table (2): Mean values of percentage larval production and survival in ovaprim induced C.grapeinus under varying brood stock (female: male) ratios

Parameters	**Treatment I**	**Treatment II**	**Treatment III**
No of stripped eggs	2690	2860	6360
No of fertilized eggs	896	953	2120
No of hatched eggs	0	500	0
Percentage larvalproduction	0	50	0
Percentage larval survival	0	10	0

CHAPTER FIVE
DISCUSSION

The induction of ovulation and spawning in the African catfish *C. gariepinus* using ovaprim injection was effective on a single intramuscular injection of 5 ml for female brood fish 454.6 g each. The maximum latency period of 12 hrs, recorded in this study.

The pseudo-gonadosomatic index also used as index of sensitivity to ovaprim reached up to 50%, indicative of the fact that a high number of eggs could be collected when fish is induced with ovaprim dosage 5 this result was agreement with (**Brzuska*et al.*, 1998**) who stated that *Clarias gariepinus* with two doses (1/5 +1 pellet/kg) or one dose (1 pellet/kg) of Ovopel show that the differences in the weight of eggs and their quality (expressed by fertilization percentage and by the percentage of living embryos after 24-hour incubation) between the groups treated with the above doses were statistically insignificant. The temperature range of 26 to 27 and 30C˚ (mean 27.7C˚) obtained the best results this result with underline with **(Oyelese, 2006)** who reported generally research studies had focused on hatching success in relation to environmental variables such as temperature.In the treatments II out of which 10% survived rate, indicates an overall good egg quality and effectiveness of ovaprim, including ovulation and spawning in the African catfish this results was agree with (**Sharaf, 2012)** who reported that synthetic hormones and specifically, Ovaprim are known to significantly increase ovulation in African catfish.

There was difference among number of fertilized eggs, number of hatched eggs, % larval production and survival in all three treatments.

CHAPTER SIX

CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

This study was conducted that high larval production (50%) in the treatments II out of which 10% survived rate, indicates an overall good egg quality and effectiveness of ovaprim, including ovulation and spawning in the African catfish. There was difference among number of fertilized eggs, number of hatched eggs, % larval production and survival in all three treatments.It was confirmed from this study that the effective dosage of ovaprim indicated from this study was 5 ml.

6.2 Recommendations

1. Further studies were needed to determine effect of other synisitic hormone on African catfish ovulation and spawning.

2. Further studies are required to examine the development and growth Performances of larvae and fry produced by the induced breeding using ovaprim hormone.

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