

INDUCED BREEDING AND REARING OF PANGAS (*Pangasius pangasius*) USING SYNTHETIC HORMONE WOVA-FH AT SHREE SANJAY FISH AND FRY PRODUCTION CENTRE, SANTNAGAR VILLAGE (SIRAHA), NEPAL



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Submitted to:

Central Department of Zoology

Institute of Science and Technology

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Kirtipur, Kathmandu, Nepal

September, 2022

DECLARATION

I hereby declare that the work presented in this thesis entitled “**INDUCED BREEDING AND REARING OF PANGAS (*Pangasius pangasius*) USING SYNTHETIC HORMONE WOVA-FH AT SHREE SANJAY FISH AND FRY PRODUCTION CENTRE, SANTNAGAR VILLAGE (SIRAHA), NEPAL**” has been done by myself, and has not been submitted elsewhere for the award of any degree. All the sources of the information have been specifically acknowledged by reference to the author(s) or institution(s).



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RECOMMENDATION

This is to recommended that the thesis entitled “**INDUCED BREEDING AND REARING OF PANGAS (*Pangasius pangasius*) USING SYNTHETIC HORMONE WOVA-FH AT SHREE SANJAY FISH AND FRY PRODUCTION CENTRE, SANTNAGAR VILLAGE (SIRAHA), NEPAL**” for the partial fulfillment of Master’s degree of Science in Zoology with special paper Fish Biology and Aquaculture. This is her original work and has been carried out under my supervision. To the best of my knowledge, this thesis work has not been submitted for any other degree in any institutions.


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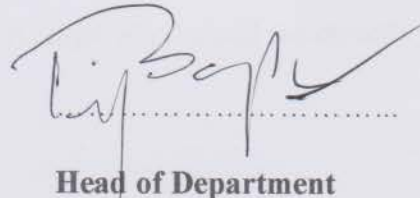
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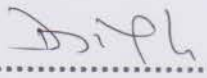
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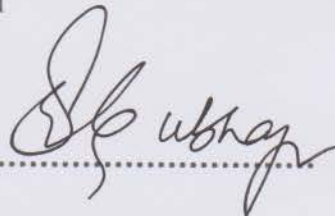
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LIST OF ABBREAVATIONS

Abbreviated form	Details of abbreviations
BW	Body weight
CPE	Carp Pituitary Extract
DOFD	Directorate of Fisheries Development
FAO	Food Agriculture Organization
SSFPC	Shree Sanjay Fish and Fry Production Centre
GDP	Gross Domestic Production
GnRH	Gonadotropic Release Hormone
MOAC	Ministry of agriculture and Co-operative

ABSTRACT

“Induced Breeding and Rearing of Pangas (*Pangasius pangasius*) using Synthetic Hormone WOVA-FH at Shree Sanjay Fish and Fry Production Centre, Sant Nagar Village (Siraha), Nepal” was the topic of the current study. The current field study, which lasted 4 months from May to August 2021, examined physical factors, Pangas biology, fecundity, Gonado Somatic Index (G.S.I), fertility rate, hatching rate, embryonic development, and growth (fingerlings). Single dosages of WOVA-FH hormone were successful in spawning pangas. At the same time, single dosages of WOVA-FH were administered to both the male and female broods. Dose for females is 0.5-2.5ml/kg. 0.5-1.5ml/kg of body weight for male. The latency period lasted 10 to 12 hours. Different pond’s temperatures ranged from 24 to 34°C over the study period. The total number of eggs spawned was found ranged from 279,000-374,000, the fertility rate (70-90%), the G.S.I. (7.80-13.19%), and the hatchling rate (45-56%) were all discovered in Pangas. After 4hrs, the embryo’s development was sustained, and between 26 and 28hrs, it would hatch. After five days of transfer to nursery ponds for hatching, the fry were fed artificially made feed containing 35-40% feed on a daily basis. From the first week to the tenth week, both the length and weight of the fry were constantly rising.

1. INTRODUCTION

1.1 Background of the Study

Nearly half of the fish used for human consumption worldwide are produced by the aquaculture industry, one of the sectors that produces animal food most quickly. One of the most frequently traded and produced fish in the world is pangas. Many nations with hot climates currently raise this fish economically. The unique quality of this fish is its ability to produce vast quantities of seeds even from a small reservoir area, and to eat them with great gusto. The predatory character of this fish. Pellet grains are the primary feeding source for this fish in commercial production. The quantity of grains we feed the fish in the pond determines how much fish is produced. This fish produces 400 million tons per hectare per year in Vietnam. Bangladesh is the home to the endangered species *Pangasius*. Two decades ago, there were plenty of fish in Bangladesh's rivers and estuaries. As a result of habitat destruction, overfishing, water pollution, improper management, and other anthropogenic factors, the number of fish gradually decreased (IUCN Bangladesh, 2015). However, Bangladesh, Indonesia, India and Vietnam are presently experiencing daily growth in the *Pangasius* culture. *Pangasius* catfish are mostly produced and exported by Vietnam. In India, West Bengal received it in 1997 via Bangladesh. Similar to this, a small number of farmers in Nepal's Terai region introduced the *Pangasius* catfish from India on their own initiative and have been successfully cultivating it for a few years. However, compared to catfish, Indian large carps make a bigger contribution to our nation's aquaculture.

By providing opportunities for improved food security, employment, and income among other things, fishing and aquaculture can address the issues of enduring poverty. With the commencement of traditional fisheries activities in 1952, fisheries development history began. For the past many decades, the Nepali Government has given fisheries a high priority. For the purpose of reducing hunger and malnutrition. Setting up Fisheries Development Centers in Nepal has been a top objective for the Nepali government. "Agriculture Development Project" financed by the Asian Development Bank, systematic aquaculture, in particular pond fish culture, was started in 1981/82. In 1981-1982, the aquaculture production program was anticipated to be around 750 million tones. Furthermore, in 1986-1987, it rose to 4939 tones. With the

creation of numerous private fish hatcheries to increase the activities related to fishing during this time, the production technology was successfully transferred to the private sector. The country's fish production increased to 11,925 mtons in 1996/1997 from 4939 tons in 1986/87 with the help of international organizations as the UNDP, FAO, ADB, JICA and IFAD. A large rise in aquaculture production occurred at the project's conclusion, reaching 20,000 tons by 2003/2004. (MOAC, 2004). Pond fish culture was transformed into a major production system in 2003/2004, accounting for more than 95% of production, according to the overall development of aquaculture production trends.

With an annual growth rate of 9% aquaculture is one of Nepal's fastest-growing industries (FAO, 2016). The Terai plain, in the southern region of the nation, is home to 94% of the nation's fish ponds, which accounts for the majority of the production of pond fish. The agricultural sector accounts for more than 35% of Nepalese GDP and employs more than two thirds of the population (GDP). The potential of aquaculture for enhancing citizen nutrition and economic prospects has recently come to the attention of the Nepalese government. Yet despite recent initiatives to support and encourage local aquaculture, annual fish yield from aquaculture is still low (3.89 t ha⁻¹), and citizens continue to lack access to fish. Access to fish is sufficient, keeping consumption rates moderate (2.1 kg/person/year: DoFD,2013).Low-quality feeds frequently results in fish yields that are far below meals, which causes serious worry about under yielding (Iiuma et al., 1999).

A significant rise in polyculture production could result from better feeding techniques, which would then boost local residents availability to animal-source protein diet (Dashila et al., 2006). Developing low-cost alternatives is crucial because the majority of small-scale farmers lack the financial means necessary to support their own aquaculture system using just complete feed. Under -5-years-olds are disproportionately affected by chronic malnutrition and micronutrient deficiencies in Nepal and other southern Asian Nations (UNICEF, 2012) moderately stunted. An estimated 24% of all women are undernourished. As animal products are expensive to include in daily diets, protein deficiency is common among Nepalese people. In 201, a total of 105.96 tons of aquaculture products were produced worldwide, with a value estimated at US \$162.76 billion (FAO, 2017).

There are various fish development and research centers in Nepal that focus on the production of fingerlings, fry, and fish. There is only one Fisheries Center in Province 1, but there is no Fish Development Center. There are three fish development centers in province 2 but no fish research centers. Province 3 has two Fisheries Centers and one Research Center. There are two Fisheries Centers and one Research Center in province 4. There are two fish development centers and one fisheries research center in province 5. There is no Fish Development and research centre in province 6. Province 7 has a single fish research and development center, FDTC, 2018/19.

A few farmers from the Terai region of Nepal introduced pangas from India on their own initiative and have been cultivating them successfully in the private sector for a few years. The main barriers to expanding this company, however, seed and culture technologies because Nepal has not yet made significant progress in the breeding and culture of this species. Pangas have lately been artificially propagated in some government farms in Nepal, with varying degrees of success. The Regional Agriculture Research Station, Tarahara has researched the identification of the mating season and the influence of temperature and precipitation to promote spawning (NARC). In the aquaculture industry, *Pangasius* catfish are now regarded as the third-most significant freshwater fish group (FAO, 2016). Carps can be included in a polyculture or monoculture of *Pangasius*. It is now one of the main freshwater fish species raised for domestic consumption and will continue to make a significant contribution to the nation's expanding aquaculture industry (Shete et al., 2017). The Pangasipdon hypothalamus species is the most well-known for commercial farming of the several *Pangasius* fish species. This is referred to as Baikhi/Baikha in India and Nepal.

Some Terai districts of Nepal have already begun to practice commercial pangas fishing. In last few years, some leading farmers in Jhapa, Sunsari, Morang, Siraha, Dhanusha, Sarlahi, Chitwan, Rupandehi, Kapilvastu, Dang, Banke and other districts have been able to earn good income by raising *Pangasius* fish. Some farmers in Nepal produce this fish on 20-25May. Tons per hectare per year has been found. It is possible to get much more productivity by providing technical services and support. With more than 3 fish being imported annually in our country, the commercial expansion of Pangas fisheries in our country will make the Country self-sufficient in fish and farmers will be able to earn comparatively more income.

Induced breeding is a very reliable method for reviving the declining natural and for supplying the growing demand for seeds from farmers. The economically significant fish-those that do not reproduce in captivity-are bred using the process of induced breeding, which involves artificial stimulation. Induced breeding is a method whereby mature fish breeders are encouraged to reproduce in captivity by introduction of pituitary or other synthetic hormones. The prompt stimulation encourages the production of sperm and eggs. Spawn that has been gathered from natural water is not pure since they may have introduced undesired wild species into the culture pond. In those phases, sorting pure seed is completely difficult. Later stages make it possible, but they take time. Spawning occurs when sexually active substances (ova in the case of females and milt in the case of males) are discharged to the outside of the body (Basaran et al., 2008). Exogenous hormones are introduced into the bodies of fish that are mature parents in a process known as “induced breeding” (Heggbert, 1996). Only fish with advanced stages of gonadal maturation were used. For the fertilization processes, females were chosen based on external physical traits suggesting that they were prepared for the spawning induction procedure, such as red urogenital papilla and bulging coelomic cavity.

Nowadays Fisheries Development Centre use different synthetic hormone for induced breeding of Indian major carps and Chinese carp and Pangas catfishes.

Advantages of Induced breeding (Jain, 2003; Srivastava, 2005)

- The target species pure spawn is made available. The spawn collected from the river is not entirely pure. Because of this, it is impossible to separate pure seed from mixed spawn of the other species because they are mingled with the spawn of other species.
- The seed spawn is timely available, but it's difficult to predict when it will be available from natural sources.
- Pure spawn can be made available any number.
- Although the seed spawn is timely available, its supply from natural sources is somewhat unsure.
- Pure spawn can be provided in any quantity. The chosen species is made available as pure spawn. The spawn collected from the river is not pure. They mix unfeasibly with the spawn of the other species.

1.2 WOVA-FH (Salmon Gonadotropin Releasing Hormone Analogue Injection)

A fish breeding hormone called WOVA-FH is used to effectively encourage the breeding of several species of fishes such as Indian Major Carp, Exotic Carp, Cat Fish, Tilapia, Pacu and others. In a variety of teleosts, WOVA-FH is increasingly employed to induce ovulation and spawning.

Benefits:

- No stress is caused to the broodstock after hormone delivery.
- Broodstock can survive in harsh environmental circumstances.
- Simple injectability.
- Better conditions for growth survival and growth.
- Excellent rates of fertilization, spawning and hatching.

1.3 Embryonic and Larval Development of Pangas

The sperm entering the egg is the first step in the embryonic development. Impregnation is the name of the process. The larval and embryonic stages of their life cycles are the most delicate and important. Therefore, research on early larval and embryonic development is crucial for effective raising of larvae, industrial farming large-scale seed production. There are 5 different development stages of fish: embryonic, larval, fry, juvenile and adult. The fertilized egg, cleavage, morula, blastula, embryonic body development, production of the optic and auditory vesicles, closing of the blastopore, and tail development as well as hatchling phases are all included in this stage. Following the embryonic stage, the hatchling goes through organogenesis and develops into with the arrival of their parents, the larval stage comes to an end. These kinds of research on embryonic and larval development are crucial for fisheries because they provide information on spawning times and locations, temporal shifts in spawning seasons, mature stock size at ovulation, mortality rates at the end of spawning period, and the environmental relationships. In order to increase fisheries production and decrease fry mortality, research of embryonic and larval development are crucial. Although there are reports of embryonic and larval development in Nepal, this knowledge is very helpful.

1.4 Descriptions of *Pangasius pangasius*

The pangas catfish (*Pangasius pangasius*), a recently arrived foreign fish species in Nepal, is a member of the family Pangasiidae of the order Siluriformes. In Nepal, this fish is popularly referred to as “baikhi” or “Pangas”. Pangas catfish were introduced to various nations, including Malaysia, Indonesia, and China, from the Mekong River in Vietnam to the Chao Phraya River in Thailand (FAO, 2016). Recently, Pangas commercial production and culture have drastically increased in certain Asian nations, particularly in China, Thailand, Vietnam, Bangladesh, India and Nepal in southeast Asia. In other parts of southeast Asia, where it is native, *P. pangasius* is frequently consumed as food. More than 130 countries throughout the world buy this fish, mostly as white fillets. Pangas is currently ranked as the third-most significant group of freshwater fish in the aquaculture industry (FAO, 2016). Due to its omnivores feeding behavior, quick growth rate, high stocking capacity, simple culture method, high disease resistance, good market demand, and tolerance to a variety of environmental changes, this species has become increasingly popular (Sarkar et al., 2007; Ali et al., 2005; Rohul Amin et al., 2005). Another crucial point is that, because it is a voracious omnivore fish, this fish is easily acclimated to the artificial diet under controlled circumstances. As this fish contains more erythrocytes than any other fish, as well as an extra respiratory organ, and can breathe through bubbles and skin to assist it endure an environment lacking in dissolved oxygen, pangas can be raised in high stocking densities (Shrestha et al., 2015). The very tiny, adhesive eggs of this fish were fertilized eggs, which were pale creamy or brown in color. When they reach a particular size, larvae and tiny juveniles switch to eating phytoplankton instead of eggs and live food (Zooplankton, *Artemia naupli*).

Fish producers in Nepal have recently expressed interest in growing this species in ponds in an effort to boost profitability and meet the steadily rising demand for fish on the market. These Pangas can adapt to a variety of culture conditions and have a high correction factor and rapid development. These fish’s reproductive season was observed to last from June to August (David, 1963), and from July to October in Bangladesh (Rahman, 1992). *P. Pangasius* reaches sexual maturity at 3+ years of age in pond environments, and spawning typically occurs in the summer and monsoon months, from early June to September, when the water is quite warm due to the downpour or the flooded upper levels of streams and rivers. The temperature during spawning varied

from 26 to 31°C. Pangas has been an excellent choice for carp pond monoculture and polyculture (Rahman, 1992). Due to their reduced market price and lack of internal spines, pangas are in high demand in Nepalese marketplaces. Additionally, because of this fish's delicate flavor and high fat content, the vast majority of people eat it. It suggests that this fish has potential to significantly increase fish production, reduce poverty, and improve livelihoods in Nepal. After two years of being raised in captivity, the fish would achieve a size range of 2.5kg to 3.0kg and exhibit maturity traits if supplementary feed containing 25% crude protein was given to them at a rate of 4% of their body weight every day.

Pangas are typically raised in intense aquaculture systems entirely on supplemental feed. Pond water in monoculture obtains a significant amount of inorganic nutrients through the microbial breakdown of wasted fish meal and metabolic wastes due to the usage of a large amount of supplemental feed. These nutrients encourages excessive phytoplankton growth in pond water, which supports more planktivorous fish without additional feed or maintenance expenses (Sayeed et al., 2008). Thus, Pangas and planktivorous fish polyculture could benefit both fish production and water quality.

1.4.1. Morphology of *Pangasius pangasius*

Fish morphometric measurement are crucial for systemic, taxonomic and growth variability studies (Tandon et al., 1993) and they also provide valuable information on the precise identification. A strong method of characterizing stocks known as morphometric statistical analysis involves the identification of appropriate variations in shape (Sharma et al., 2015). Body is scale-free, elongated, and laterally compresses. The tail is slightly extended before the caudal peduncle and is constricted behind the adipose fin. The head and abdomen are flat. The occipital process is used to reach the dorsal fin's basal bone, and the snout is fairly prominent. The head is slightly granulated above. The eyes are located in the lower portion of the head and partially on the anterior half. The upper jaw is longer than the lower jaw, and the mouth gape is of average size. Cleft of mouth is used to reach opposite the centre of front edge of the eye. Four groups of teeth are present on the palate; palatine teeth are in a crescent row, vomarine patches are separate from or nearly confluent with those on palate. Barbels are two pairs; the maxillary pair reaches the base of pectoral fin and the mandibular pair is half as long as

the head in length. First dorsal fin is with a moderately strong spine which is strongly serrated on its inner edge but finely serrated on its outer edge. Adipose dorsal fin is short, posteriorly free, and originates almost opposite to the middle of the anal fin. Pectoral fin spine is serrated, strong and as long as dorsal spine. Anal fin is large and well developed. Caudal fin is deeply forked; upper lobe is slightly the longer. Body color is silvery, darkest along the back and glossed with purple on sides; cheeks and the under surface of head is golden; caudal fin is bright yellow.

1.5 Objectives

1.5.1 General Objectives

To evaluate the Induced breeding and Rearing of *Pangasius pangasius* by using WOVA-FH (Salmon Gonadotropin Releasing Hormone Analogue Injection) at Shree Sanjay fish and fry production centre, Santnagar (Siraha).

1.5.2 Specific Objectives

1. To investigate Fecundity rate, Gonado-Somatic Index, Fertility rate, Hatching rate of *Pangasius pangasius*.
2. To observe various embryonic stages of development of *Pangasius pangasius*.
3. To evaluate growth rate of *Pangasius pangasius* from fry to fingerlings stage.

1.6 Significance of Study

The study of fish like Pangas is partially success as a food sources but their detail study about breeding, embryonic and rearing details has not been studied. This study will help to fill the knowledge gap to some extent and research of these species.

2. LITERATURE REVIEW

2.1 History of Induced Breeding

After developing pituitary extract Hussay in 1930, when viviparous fish were injected with the hormone to cause preterm birth, the technique of induced breeding was developed in Argentina. Pituitary extract was successful in inducing breeding in Brazilians in year 1934.

On the *Cirrhinus mrigala*, Khan (1937) attempted induced breeding for the first time in India. This method was later used by Choudhuri in 1955 applied with minor carps (*Esotrus danricus*) and (*Pseudeotropius atherinoids*). By injecting hormones, Ramaswamy and Sunderaraj (1956, 1957) reported successfully breeding of the cat fishes, *Heteropneustes fossilis* and *Clarias batrachus* in the years 1955 and 1956, respectively. By injecting carp pituitary extracts into the *Labeo rohita*, *Cirrhinus mrigala*, *Cirrhinus reba*, *Labeo bata* and *Puntius sarana* in 1957, Chaudhari and Alikunhi were successful in getting the animals to reproduce.

After the Brazilians, the Russians are thought to be the next to use hormone treatment in fish farming. Gerbilskii (1938) didn't achieve success until 1937, but ever since then, this method has been successfully used in the Soviet Union to produce sturgeon eggs at the farms along the lower Volga, Ural, Kuban and others river fishes .

By injecting the hypophysis of Common carp into brood fish of Silver carp and Bighead carp cultivated in ponds, aquaculture experts in Gungdong province used the first hypophysial procedure in China in 1958. In 1960, the pituitary gland of common carp in China replaced artificial grass carp reproduction.

2.2 Artificial breeding of Catfishes

Rahman et al. (2020) reported that induced breeding of 6 pairs of *Pangasius Pangasius* by different doses of Pituitary gland (PG) per kg body weight. The highest Ovulation rate (100%), fertilization rate (30.0±0.3%), hatching rate (24.8±0.8%) and survival rate (47.0±12.3% after 72hr) respectively.

Gnanamuthu et al. (2018) reported breeding with hormones is a common practice among edible fishes. They also reported use of synthetic hormone WOVA-FH, in gold fish at the rate of 0.1ml per gold fish of 25gm yielded a potential fry stock of (150±158),

percentage of hatchings (60.99%) and percentage of survival (72.56%) is high when compare to the non-induced ovulated fishes.

Singh et al. (2018) experimented on *Pangasiandon hypophthalmus* in Semi-arid agro-climated using Pituitary gland (PG) and reported fertilization and hatching percentage was 76.50% and 58.20% respectively. They also reported that after fertilization. Eggs became water hardened within 20 min.

Dhara et al. (2018) experimented on *Clarias batrachus* using synthetic inducing agent, Alpa-FH. They reported by using hormone Alpa-FH at the dose of 0.8ml/kg, the rate fertilization (80.4%), hatching (84.1%) and some common morphological deformities (1.27-3.83%).

Nesa et al. (2017).They reported fertilization and hatching rates of 62.33 ± 4.51 , 46.67 ± 5.86 and 37.33 ± 2.52 , 41.33 ± 5.69 , respectively, using hand the stripping approach and non-stripping method. Similar results were observed for 2-cell, 4-cell, 8-cell, 16cell, 32- cell embryonic development mean times, as well as for morula, blastula, germinal ring, gastrula, yolk plug, twisting movement, immediately before hatchling.

Islam et al., (2016) reported fertilization rate 87%, hatching rate 82%, deformity of larvae 6% and survival rate 80% by using pituitary hormone. The breeding performances of *P. hypophthalmus* were found to be satisfactory for the commercial production of this fish in Bangladesh.

Moses et al. (2016) experimented on *Pangasiandon hypophthalmus* using GnRH based inducing agents (Synthetic hormones) viz., GONOPRO FH and WOVA FH. Of the two induced breeding agents used WOVA FH has comparatively given good results compared to GONOPRO FH in this study. Generally an incubation time of 12 to 15 hrs is provided for successful hatching of eggs.

Ferosekhan et al. (2015) reported the blastomeres looked overlapped during these multicell stages and the size got reduced during morula stages onwards. The transparent larvae were 3-4mm in length with compact and oval shape yolk sac of 1.4-1.6mm in length at hatching. The 11 dph larval possessed dorsal, pelvic and caudal fin possessed 6-7, 6-7, 5-6 and 19-20 fin rays respectively.

Singh et al. (2015). They experimented on *Pangasius hypothalamus* using ovaprim and reported the percentage fertilization rate 60-80%. They also reported for fertilization of one million eggs of *P. hypothalamus*, one ml milt was used.

Denson et al. (2007) compared Human Chorionic Gonadotropin (HCG) and the luteinizing hormone releasing hormone analogue LHRHa for ovulation induction in Back sea Bass *Centroproistis strita*. The monsoon season matured Sea Bass (200-800gm) received two intramuscular injection of HCG spaced 24 hours apart (n8) or one intramuscular injection of HCG at dose of 330IU/kg. Between HCG (75.6±11.4%) and LHRHa (55.6±27.6%), a substantial difference in infertility was discovered.

Adebayo et al. (2007) compared Synthetic and non-synthetic hormones for artificial breeding of *Clarias* catfish. They reported that the synthetic hormone (Ovaprim) is more effective than the Frog Pituitary Extract (FPE) and *Clarias* Pituitary Extract (CPE). The highest Percentage fertilization (84.5%) and Percentage hatchability was ranged from 51.5-73.0% and latency period was 12hrs.

Islam A. (2005) experimented on Thai Pangas and reported embryonic and larval development was during peak (May-July) and late spawning (August-October) periods. The incubation period ranges from 24-36 hrs at a temperature of 20-30 °C.

In Captive settings, Sial et al. (2005) conducted research on *Pangasius sutchi* (*sluroid* catfish) utilizing carp pituitary extract, ovaprim, and ovatide. A dose of 21.0 to 25.0 mg/kg body weight given to female brooders of carp pituitary extract led to effective spawning in *Pangasius sutchi* following a latency period of 10-11 hours. Similar results were obtained with ovaprim, which improved fertilization and hatching rates in fish by inducing ovulation and spawning, respectively.

Khan and Mollah (2004). They reported 100% ovulation in females, the dose of 10mg/kg body weight of PG demonstrated the best result in consideration to fertilization and hatching rates of eggs. Hatching of fertilized eggs occurred between 28 and 32 hrs of incubation at 25 to 28°C. Hatching rate was 65%.

2.4 Induced Breeding in Nepal

The first successful induced breeding of fish was accomplished in Nepal in 1972 using the Woynarovich (1969) hypophysation technique on Chinese carp. Based on the number of fresh individuals recruited as brood stock each year and the variance of the

reproductive performance, Wagle and Pradhan (2003) evaluated the effective population size and rate of inbreeding of common carp.

At the Mukhya fish farm Nanupati, Dhanusha, Sah (2012) reported using ovaprim to encourage breeding in Common carp (*Cyprinus carpio*). According to her, between 201,000-333,000 eggs were hatched in all. 83% and 53.6% respectively, of eggs were fertilized and deposited. Four hours after fertilization, she claimed, the eggs began to cleave.

Common carp (*Cyprinus carpio*) and silver carp (*Hypophthalmichthys molitrix*) were used to induce ovulation in a study by Banjade (2015). According to him, ovaprim therapy led to greater rates of fertilization in common carp (75-92.5%) and silver carp (72.6-93.3%). Similar numbers showed that Common carp was 44-58% and Silver carp was 44-59% respectively. 4 hrs after fertilization, the egg was seen to cleave. 48 hrs after fertilization, the embryo continued to develop and hatched. The fry were fed a 45% protein artificially prepared meal at a rate of 5 to 10% body weight, and growth checks were performed every week.

Ovulin hormone was used to encourage breeding in the Rohu (*Labeo rohita*) and Naini (*Cirrhinus mrigala*) according to Sah (2017). She stated that ovulin treatment revealed the percentage fertilization of Rohu (77.77% to 88.33%) and Naini (71.05 to 82.56%). Similar, to Rohu, Naini had a hatchling percentage of 71.29-82.56% and 60.86-79.28% respectively.

LHRH-A Hormone was used by Yadav (2018) to encourage breeding in Silver carp (*Hypophthalmichthys molitrix*). She provided data on the percentage fertilization (72.5-92.5%) and hatching rate (65.21-82.60%) of Silver carp treated with LHRH-A.

Sah (2018) experimented preliminary observations on breeding and fry rearing of pangas (*Pangasius hypophthalmus*) using Ovulin (LHRH-A). She reported Ovulin at dos 0.5ml/kg spawner was found effective to induce spawning after 8-14 hrs of latency period. Mean fertility rate and hatching rate was estimated $90.1 \pm 5.9\%$ and $73.2 \pm 11.6\%$ respectively.

3. MATERIALS AND METHODS

3.1 Study Site

The present work was carried in Shree Sanjay fish and fry production centre, Dhangadhimai municipality-4, Santnagar (Siraha). Santnagar is a village of Dhangadhimai municipality-4 which is in Siraha District in Madhesh Province in Sagarmatha Zone of South-eastern Nepal. Shree Sanjay fish and fry production centre, Dhangadhimai municipality-4, Santnagar (Siraha) was established in 2046 B.S. by Ramu Chaudhary. It lies approximately 3km south from Zeromile bariyarpati Marg. The total area occupied by this farm is 9 hectare and for Reservior tank 1.5 Kattha with 61 Pond of various size for the purpose of nursing, rearing, stocking, brood management. The main water source of the farm is deep boring. There are 23 Nursery ponds with area in the range of 0.05-0.1 hectares and 7 brood fish ponds with area in the range of 0.1-0.0.25 hectares in this farm. There is only one hatchery:- 6 incubation tanks each with capacity of 2.5 cubic metre (100l eggs holding capacity) with 1m height, 1m diameter. 1 holding tanks with capacity of 3 cubic metre and 3 spawning tank with capacity of 10 cubic metre and 1m height, 2.5m breadth, 7m length. Shree Sanjay fish and fry production centre was established with following main objectives-

To produce carp's hatchlings, fry and fingerlings to supply local fish farmers.

To produce table fish to consumers to reduce import from India.

To collect different types of warm water fishes for culture practice

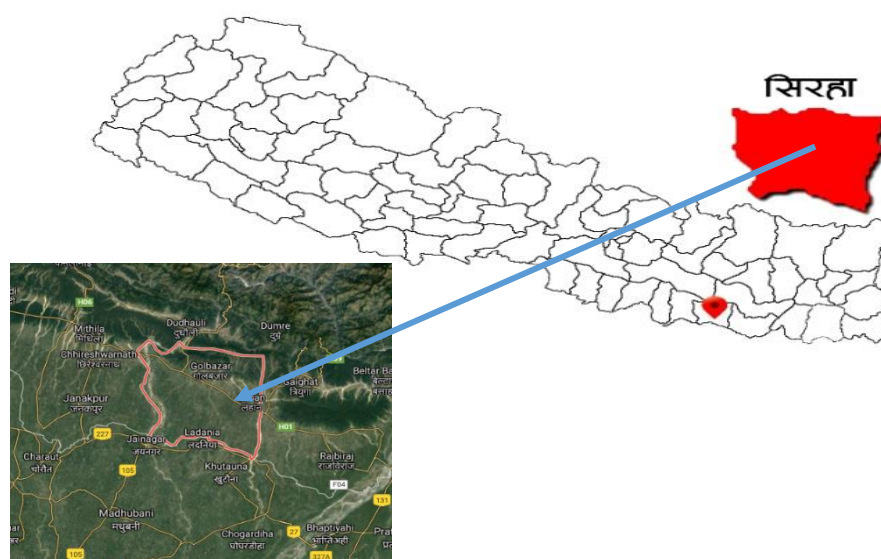


Figure 1: Map of the Study Area

Table 1: The Number of Pond and their Area in Study Site SSFPC

SN	Name of pond	Number	Area(ha)	Used pond	Unusedpond
1	Brood stock pond	7	1.5	All	-
2	Nursery pond	23	3.5	All	-
3	Rearing pond	11	2	9	2
4	Production pond	14	3	12	2
5	Other pond	6	1	3	3
	Total pond	61	11		7

There are 61 ponds having 7 Brood stock ponds, 23 Nursery pond, 11 Rearing pond, 14 Production pond and 6 Other ponds. Nearly 7 ponds are unused in this farm. (Table 1)

3.2 The Availability of Pangas Seed

Hatchling are produced between (June-August) of the month, fry between (June-September) and fingerlings between (August- November).

Study Period

From May 2021 until August 2021, a 4-month field research was conducted. Breeding took place between June and August of 2021 (Pangas). Fry growth monitoring and larval rearing began in June, 2021.

3.3 Physical Parameters

During the experimented period, the following physical parameter was investigated:

Nature of day: Through visual observation. It was noted while working in the field.

Color of the water: After placing pond water in a Petridish on white paper, the water color was assessed.

Temperature: A common digital meter was used to test the water temperature. Direct measurement was made by dipping the thermometer bulb into the water for two minutes, then recording the reading.

3.4 Fish Species

Pangas was selected for the study. The fishes were cultured in fresh waters along with other carp for polyculture.

3.5 Collection and Maintenance of Brood Fish

We choose healthy, disease-free, farm-raised brooders for our Pangas breeding efforts. The brooders ranged from 3 to 4+ years and weight from 3.5 to 6.5kg.

The mature male and female who are prepared to spawn are referred to as “Broods”. To prevent self-fertilization, male and female were placed in distinct pools. Each pond had been set up in accordance with accepted pond management techniques for stocking and raising brood stocks. The specimens chosen had no physical deformities and were in good health. They were maintained in stocking ponds throughout the first phase of brood care. Because the pond’s filamentous algae or macro-vegetation were insufficient, broods were fed on decomposed plant materials that contained bottom-dwelling creatures. The species-specific fecundity regime of brood stocks.

3.6 Identification of Male and Female Brooders

The secondary sexual characteristics of the male and female brood fishes were used to distinguish them at breeding time. The dorsal surface of the pectoral fin on the ripe male was rough, whereas it was extremely smooth on the female. By touching the surface of the fins close to the body, one can feel the roughness in the pectoral fins. The shape of the body, the health of the vent, and production of milt in males were other characteristics used to identify adult male and females. The table below explains the characteristics that can be used to identify mature male and female brooders.

Table 2: Characteristics Features of Sexually Mature Male and Female Fishes Used for Induced Breeding Trails

S.No.	Characters	Male	Female
1.	Brood body & Size	Good looking & healthy. Brood slender, more translucent & less pigmented. Relatively smaller than female.	Good looking & healthy. Body robust & pigmented. Large in body Size.
2.	Abdomen	Abdomen normal, not bulky like female.	Abdomen bulging, elastic & soft.
3.	Genital opening	With a pinkish color & protruding genital opening.	Genital opening with pink color during breeding season.
4.	Vent	Normal vent.	Prominent reddish vent.
5.	Putting pressure on abdomen	On slight pressure above the vent on the abdomen milky white fluid (milt) runs out through the vent.	Pressure slight yellowish discharge or a few ova may come out through the vent.

Source, Sharma 2008

To make breeding pathways easier to manage, medium-sized fish were chosen. Each brooder was captured in a hand net one by one at the time of selection. The breeding sets of fully grown brooders were housed in breeding tanks after the sex hormone injection. According to Malik et al., (2014), the fish used for breeding were a 2:1 mixture of male and female fish that were almost the same size and weight.

3.7 Hormonal Doses and Schedule of Administration

3.7.1 Doses of Administration

WOVA-FH (Salmon Gonadotropin Releasing Hormone Analogue Injection) was administered intramuscularly to Pangas in varying doses according on body weight. Only a single dose of WOVA-FH hormone was administered at once to both male and

female. Dose of female is 0.5-2.5ml/kg and dose of male is 0.5-1.5ml/ kg of the body weight.

3.8 Steps of Breeding

3.8.1 Collection of Brood Fish for Injection

With the aid of a hand net made to resemble a bag, a few fish brooders were selected and removed from the breeding pond. Every fish's length and weight were measured using a measuring tape and recorded. After that, they were each individually wrapped in a nice towel and placed on a weighted machine.

3.8.2 Method of Hormone Injection

At the time of injection, two people were needed; one held the fish's head gently against the cushion, while the other squeezed the fish's tail with one hand while injecting it intramuscularly above the lateral line on the caudal peduncle. A hypodermal syringe with a gradient of 0.1ml and a volume of 3ml was utilized for the hormone preparation. BDH needle number 24 was the one used in syringe. The brooders were immediately released into the breeding tank after injection. The entire process of administering the hormonal extract to the fish, including transporting the brooders, weighing them, injecting them, and releasing them back into the breeding tank, took only 1 to 5 minutes. The fish was held gently while receiving the hormone injection in order to cause the fish as much stress as possible.

3.9 Latency periods

In experimental fish, it is the lag time, or 10 to 12 hours, between the injection of the triggering hormone and the time of spawning. Estrus is a phenomena when fish start chasing one other eagerly if the hormone injection has a normal effect on the fish who have been stimulated. When ripples on the water's surface developed as a result of breeders being chased in the middle or bottom layer, this was the beginning of the estrus phenomena. They are chosen for stripping when brooders start chasing at that point.

3.10 Hand Stripping and Incubation

Fish were checked every 30 minutes after eggs were found in the aquarium (Drori et. al., 1994, Brzuska, 2004) and ovulated fish were stripped by applying pressure on the abdomen region. Female were hand stripped from the WOVA-FH treated female and

gently pushed in the abdomen region. Bird feathers were delicately incorporated into the sperm and egg combination to ensure fertilization. Water was added while swirling to firm the eggs. Fertilized eggs were properly rinsed with water after three to five minutes. Several times, the eggs were washed. Fertilized eggs are placed in an incubator when they expand and begin to float.

Before transferring the eggs, malachite green (2-5 gm in 10 liter water) was treated with water to guard against bacterial and fungal attack. After loading the eggs, potassium permanganate was used to treat the water (5-8 gm in 10 liter water). For an experiment to evaluate fertility rate and hatching rate, 50 fertilized eggs were housed in three floating sieves. A small amount of time before the yolk sacs were completely absorbed, the hatchlings were given the proper nourishment. Milk powder was fed to them by just sprinkling it slowly over the water. This aids in preventing the food from spreading out quickly through the outlet. The same way, a water and egg yolk mixture was given twice daily.

3.11 Determination of Fecundity, Gonado-Somatic index (G.S.I), Fertility and Hatching Rate

The fecundity, fertility and hatchling rate can be determined by following formula, (Kaur and Dhawan, 1997).

3.11.1 Determination of Fecundity Rate

By taking a sample of a gram egg and multiplying it by the total weight of the female fish in the brood, fecundity was calculated separately. The stripped out dry eggs weighed one gram on an electric scale. With the aid of brushes, three of these samples will be taken and individually emerged in salt solution to count the eggs.

Fecundity = No. of egg per gram \times Wt. of total egg (gm) spawned by female fish

3.11.2 Gondo Somatic Index (G.S.I)

According to GSI, the body's weight is equal to the weight of the gonads. The weight of gonads, which are connected to the female fish, indicates the stage of maturity. The peak phase of fish maturity is when their GSI is at its highest, and it quickly declines spawning. The following formula was used to calculate the GSI (%) of all female fish.

$$GSI = \frac{\text{Gonadal weight of brood}}{\text{Weight of brood}} \times 100$$

3.11.3 Determination of Fertility Rate

By taking a sample of eggs at the early morula stage, it was possible to assess the fertility rate for each female individually. By taking a sample of eggs in the early morula stage, the fertility rate was determined for each female separately. After the embryo had developed for four hours, it could be accurately calculated. The eggs in the sieve were removed and examined for fertility in a plastic trough. Unfertilized eggs were seen to be drab and opaque, but fertilized eggs were seen to be clear crystal balls. The average of the eggs in the entire sample was used to calculate its fertility.

$$\text{Fertility rate}(\%) = \frac{\text{Total no. of fertilized eggs}}{\text{Total no. of eggs}} \times 100$$

3.11.4 Determination of Hatchling Rate

Hatchings number was determined by taking out the net bowl in a plastic trough and number of hatched ones and unfertilized eggs were counted.

$$\text{Hatching rate}(\%) = \frac{\text{Total no. of hatchling}}{\text{Total no. of fertilized eggs}} \times 100$$

3.12 Embryonic Stage of Pangas

To research the various fish embryonic phases, a sample of fertilized eggs was gathered. The egg was taken and dipped into the distilled water multiple times. Eggs were kept at different stages in 1% formula solution, and the investigation was conducted using a binocular microscope with a Redmi Y3. In the Central Department of Zoology, Kirtipur, laboratory, methyl orange and eosine were employed to add color.

3.12.1 Fertilized and Unfertilized Eggs of Pangas

As soon as of the eggs occurs. The eggs were laid singly and remained firmly adherent throughout the duration of incubation. The more they developed, the more transparent they became. Pangas fertilized egg appears to be brand-new. Pangas unfertilized egg has a drab appearance in nature.

3.13 Rearing of Hatchling Till Fry Stage

One week old hatchlings were moved into the farm's prepared nursery ponds after being nursed for five days inside the hatchery in the incubation tank. Collections of hatchlings were made through the incubation tank's outlet by placing a piece of cotton fabric over the mouth of the outlet. After that, the hatchlings were measured in measuring cup (at the rate of 50,000 per cup). In plastic bags with enough water, the hatchlings were moved to the nursery ponds to be raised there. 35-40 % crude protein meals were given to fry twice a day. This experiment was continued for 70 days. Hatchling with the use of a scoop net and measurements of its length and weight after being moved to the nursery pond. After being transferred to the nursery pond using a scoop net, the hatchling's length and weight (up to 70 days old) were measured. The nursery pond is stocked with the hatchlings. The survival rate is directly impacted by stocking density. A 30-40% survival rate is expected. The survival rate in well managed pond can reach 90%.

3.14 The Routine Management of Nursery Pond

3.15 To determine the amount of food and fertilizer to use, as well as whether or not to replace the water, it is necessary to observe fry activity in the morning and the afternoon. We must take care to keep hazardous animals out, including frog eggs and tadpoles, birds, snakes, and aquatic weeds.

3.16 Feed Composition for Fry and Fingerlings

Wheat (30%), Maize (25%), Soyabean (15%), Mustard oil cake (15%), Ricebran (10%). Fishmeal (3%), Vitamin (1%), Minerals (1%). These are the elements that go into making feed for fingerlings and fry. A correctly mixed ball of feed is placed in the designated location at the designated time. Fed at the rate of 5-10% body weight and containing 35-40% protein. For their optimal growth and weight, they require a healthy, protein-rich diet.

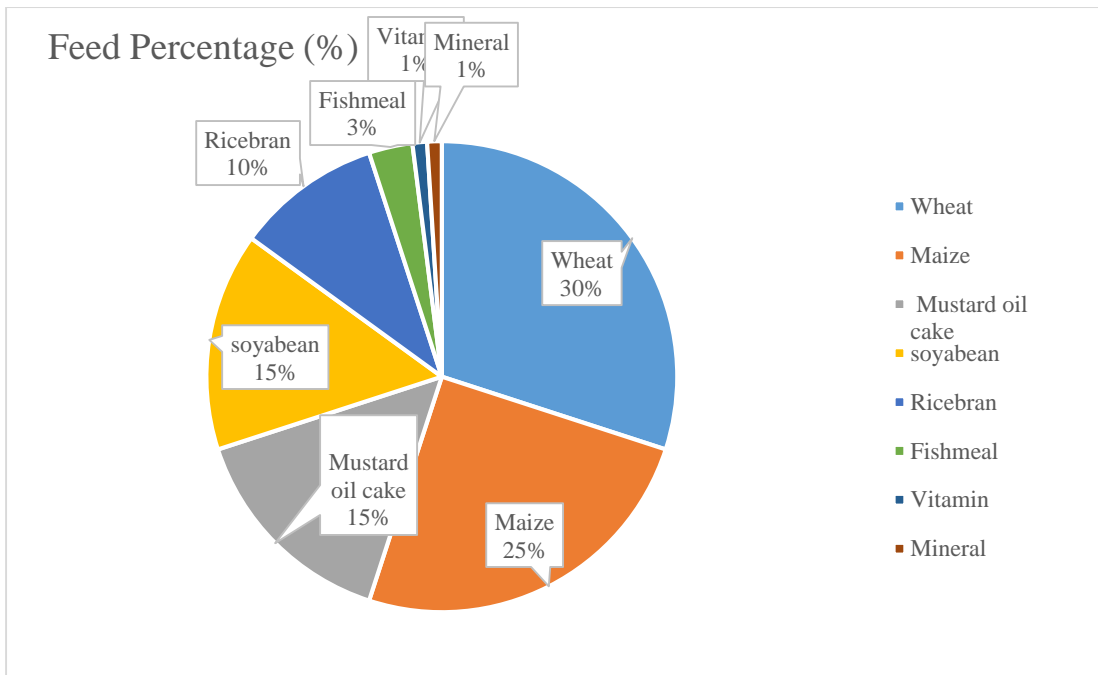


Figure 2: Composition of feed for fry rearing in nursery pond up to 70 days in percentage.

4. RESULTS

4.1 Biology of Pangas

The fecundity of Pangas ranged between 279,000-374,000 and G.S.I ranged from 7.80-13.19% of female brood fish (Table3).

Table 3: Fecundity and Gonado-somatic Index (G.S.I) of Pangas at Sanjay fish and fry production centre, Dhangadhimai municipality-4, Santnagar (Siraha).

No. of female	Weight of female (kg)	Weight of total eggs spawned (gm)	No. of eggs per/gm	Fecundity	G.S.I (100%)
1.	2.35	310	900	279,000	13.19
2.	3.5	310	990	306,900	8.86
3.	2.75	300	950	285,000	10.91
4.	4.1	320	1000	320,000	7.80
5.	3.7	340	1100	374,000	9.19

4.1.1 Fertility Rate and Hatching Rate (%)

The fertility rate of Pangas ranged between 70-90% and Hatching rate 45-56% (Table4).

Table 4: Fertility Rate and Hatching Rate of Pangas at Sanjay fish and fry production centre, Dhangadhimai municipality-4, Santnagar (Siraha).

Number of eggs	Number of viable fertilized egg	Fertility rate (100%)	Hatchling rate (100%)
50	40	80	53
50	45	90	56
50	36	72	46
50	35	70	45
50	41	82	52

4.2 Embryonic Development of Fertilized Eggs

4.2.1 Different Embryonic Stages

Unfertilized egg: Unfertilized egg of Pangas looks dull in nature eggs are opaque white and the nucleus disintegrate within one hour 10X. (PL I, Photo 1)

Fertilized egg: Round, clear, golden-brownish in color, and sticky in character, the fertilized egg was discovered. After the water was added, a slight swelling was seen. The pangas eggs was visibly transparent when the water had dried (PL I Photo 2).

1-cell stage (4 hrs): At the animals pole, an outgrowth known as a blastoderm grew over the vitelline sphere. The perivitelline space, a thin area that was evenly spaced, a thin area that was clear and free of yolk, completely separated the egg membrane (PL I Photo 3).

8 hrs embryo: The blastodisc is very elevated above the yolk (PL I Photo 4).

12 hrs embryo (Morula stage): Due to continual division, blastodermal cells were very tiny and gave animal poles a floral appearance (PL II Photo 5).

16hrs embryo (Blastula stage): The blastomeres are challenging to distinguish. Over the yolk sphere, the blastoderm was squeezed and covered more than half of the space (PL II photo 6).

20hrs embryo (Gastrula stage): Over the yolk sphere, a thick layer of blastoderm or the germinal ring took up $\frac{3}{4}$ of the space. A portion of the embryonic shield in the middle that extends into the germinal ring. Its broad end, which would later form the embryo's cephalon, was noted (PL II photo 7).

24hrs embryo (C-shaped embryo): The yolk sphere was covered by an embryo that seemed kidney-shaped with a distinct head and tail. Notochord and somite also showed up (PL II photo 8).

Hatching stage (26-28hr after fertilization): Chorion shell was shattered. Larvae that had just emerged from the egg were slim, straight, translucent, progressively tapering towards the tail, and capable of free swimming. The tail length accounted for 28% of the overall length, gives the larvae a silvery appearance. Larvae measured 3.5 ± 0.01 mm in size. The yolk sac was spherical, compact, light coloured and slightly transparent. Head is joined to the yolk sac as a result, seemed curved in the anterior part. The mouth

and mouth cleft are indistinguishable. There was no discernible back apparent eye (PL III photo 9).

1 day hatching: After hatching, the mouth was not immediately apparent. The tail gets bulkier. The mouth stayed open for the first day after hatching, and as the larvae grew older, the spaces between the jaws gradually shrunk. The eyes are tiny and resemble a black dot on the front of the head. Larvae measured 4.5-0.5 mm in length. Barbels were not clearly visible, although a slight elevation or bulging was visible below the lower jaw (PL III photo 10).

2 day hatching: Mouth of opening gets wider. There were saw structures like barbels. There were visible rays, and the tail fin was broad like a shark tail. With aging, a gradual shrinkage in the yolk sac was seen. Larvae were 5.5 mm in length and 0.5 mm in width at this stage (PL III photo 11).

3 day old hatching: Larvae are fully grown at this stage. The yolk sac lacks a spherical shape, has been completely absorbed, and has thinned out to form a conspicuous pectoral and pelvic fins fold. There was a noticeable mouth cleft. Operculum and pigmented dark eyes are both clearly apparent. Barbels on the upper jaw are also readily apparent. Compared to the top ones, the jaw barbell is longer. Pectoral, dorsal, and pelvic fins were present but not particularly noticeable. Larvae were 6.0 mm in this stage. (PL III and PL IV photo 12).

4 days old hatching: Color of larvae became shiny silvery and the eyes increase in size and pigmentation. No yolk material was seen. Pectoral fin fold well developed. Alimentary canal was visible. And also development of pigmentation observed on the body surface in this larvae, which was first appeared on the head surface followed over belly as well as just below the dorsal side and finally at the base of pectoral spine. At this stage length of larvae was 6.5 ± 0.5 mm in size. (PL IV photo 13).

5 days old hatching: The upper jaw is somewhat longer than lower jaw, and lips have fully formed. Mouth is still open, but the snout appears round. The barbels on the lower jaw are longer than those on the upper jaw. It is easy to see the air bladder. The eyes were prominent, which is 0.2 mm in diameter. The number of heads with pigmentation rose. The elementary canal coils up to the vent. The larvae was 7.5 ± 0.5 mm in size at this stage. (PL V photo 14)

PLATE I

Embryonic Development of Pangas at 10X

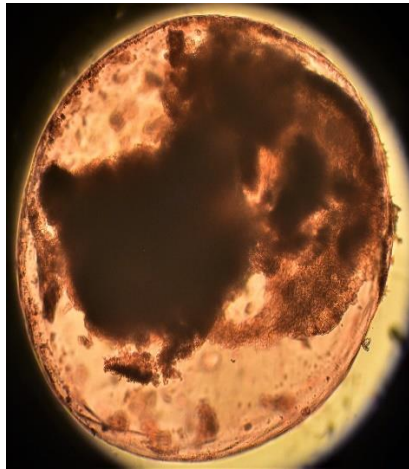


Photo1.Unfertilized egg10X



Photo 2: Fertilized egg 10X

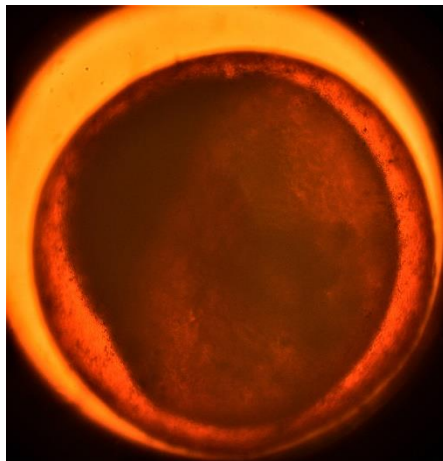


Photo 3: 4 hours embryo10X

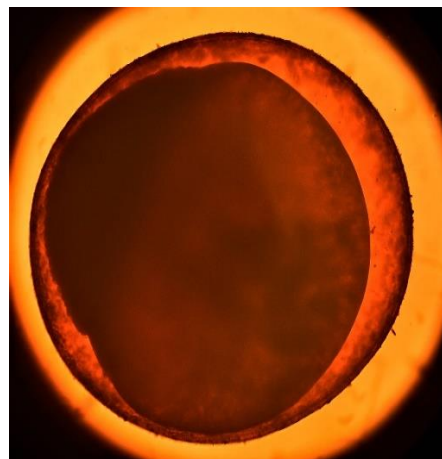


Photo 4: 8 hours embryo 10X

PLATE II

Embryonic Development of Pangas

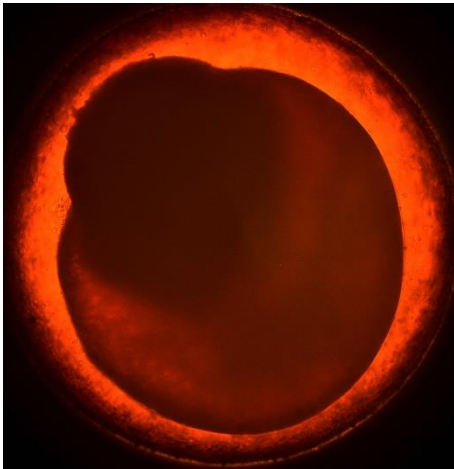


Photo 5: 12 hours embryo 10X

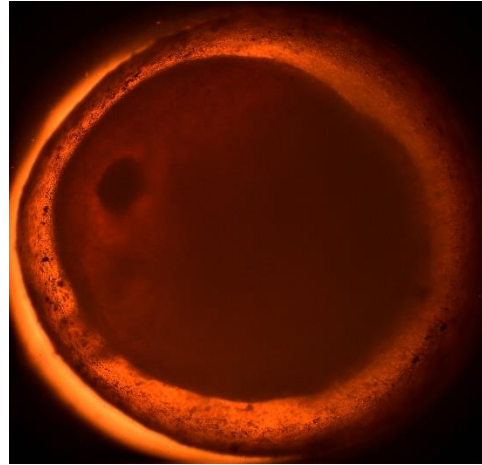


Photo 6: 16 hours embryo 10X

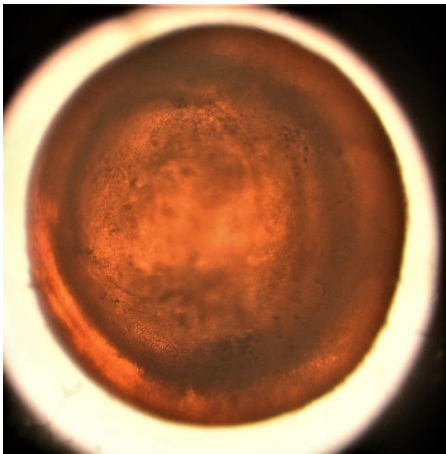


Photo 7: 20 hours embryo 10X

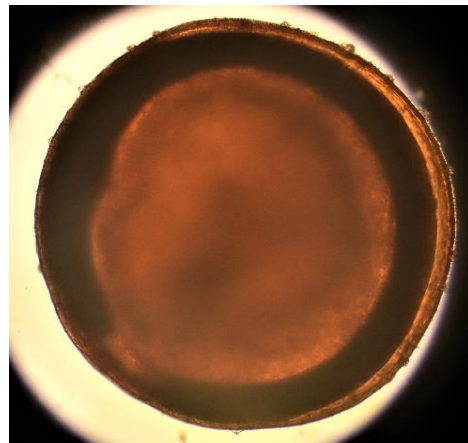


Photo 8: 24 hours embryo 10X

PLATE III

Hatching Stages of Pangas



Photo 9: Newly hatched at 28 hour 10X

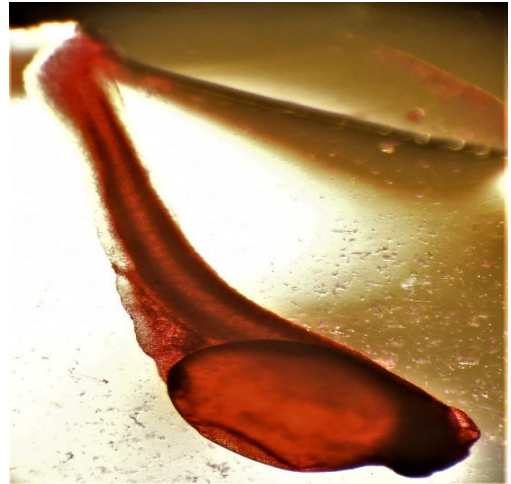


Photo 10: 1 day old hatchling 10X

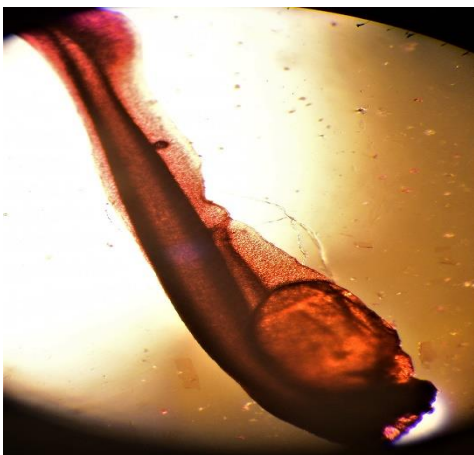


Photo 11: 2 day old hatchling 10X



**Photo 12: 3 day old hatchling 10X
(Head Portion)**

PLATE IV

Hatching Stages of Pangas

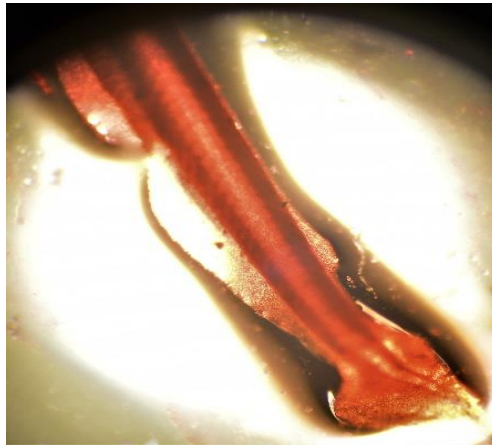
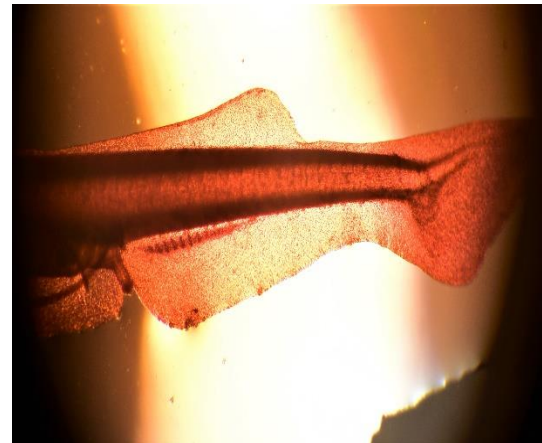


Photo 12: 3 day old hatchling10X (Tail Portion)



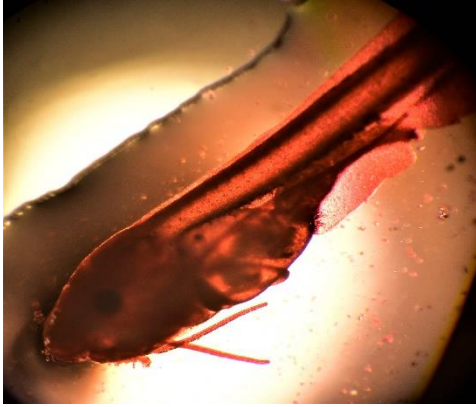
**Photo 13: 4 day old hatchling10X
(Head Portion)**



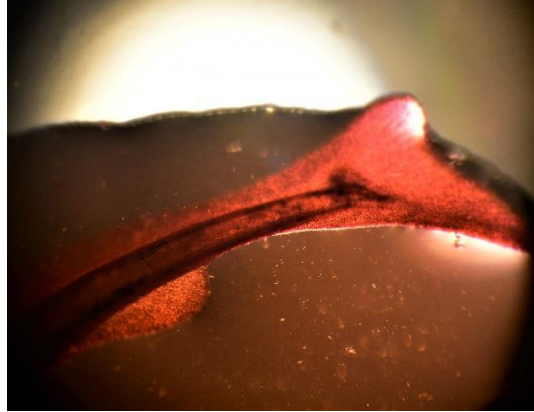
**Photo 13: 4 day old hatchling 10X
(Tail Portion)**

PLATE V

Hatching Stages of Pangas



**Photo 14: 5 day old hatchling 10X
(Head Portion)**



**Photo 14: 5 day old hatchling 10X
(Tail Portion)**

PLATE VI

HATCHING AND GROWTH RATE OF PANGAS



**Photo 15: 7 days hatching , Gas bladder
stage**

Photo 16: 7 days hatching



Photo 17: 20 days hatching



Photo 18: 30 days hatching

PLATE VII

Growth rate of Pangas



Photo 19: 40 days hatching

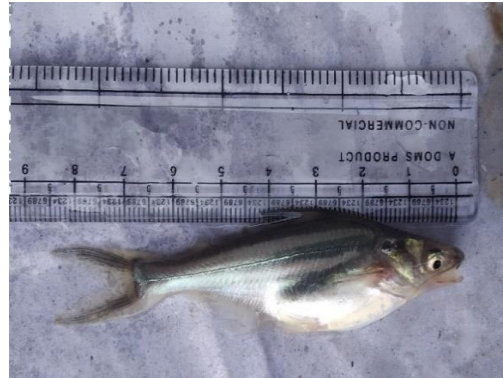


Photo 20: 50 days hatching



Photo 21: 60 days hatching



Photo 22: 70 days hatching



Photo 23: Growth check from right to left fry to fingerlings.

Growth of Length and Weight of Hatchlings

The length and weight of hatchlings were recorded gradually

4.2.2 Growth-check of Fry Up to 70 Days

After being moved to the nursery ponds for 5 days, the hatchlings were fed artificially-formulated feed containing 35-40% protein at the rate of 5-10% body weight. For their optimal growth and weight, they require a healthy and protein-rich diet. At intervals of 10 days, Pangas had its growth monitored. The fry's length and weight were seen to gradually increase.

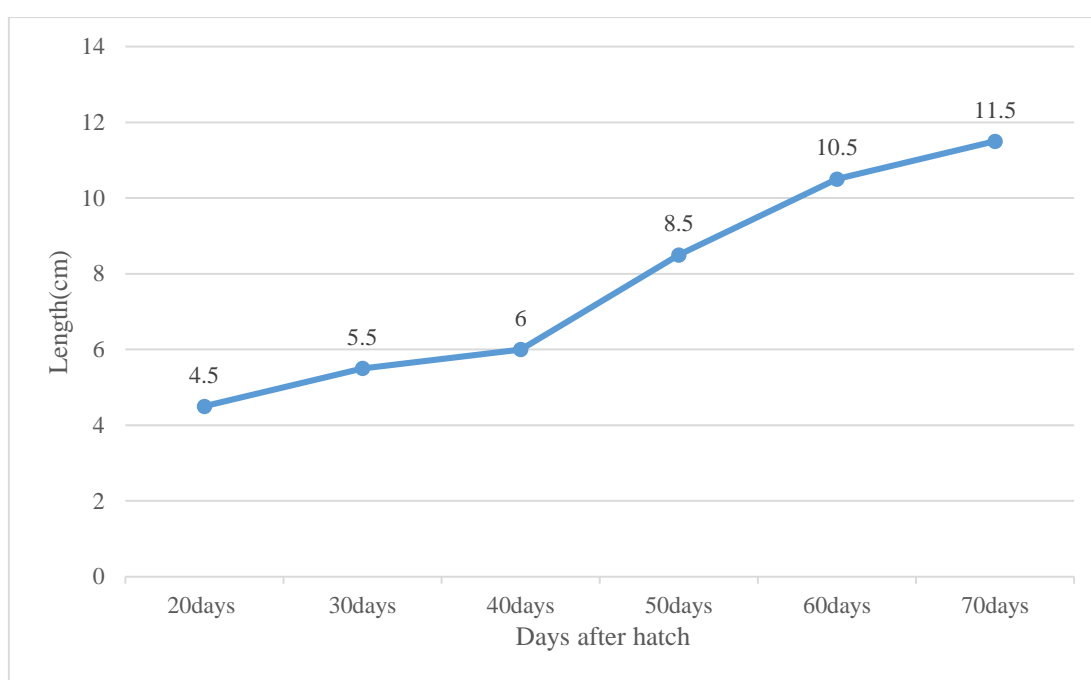


Figure 3: Growth Performance (Length) in the Fry under the Influence of Commercial Feed at Regular Interval (Photo 23).

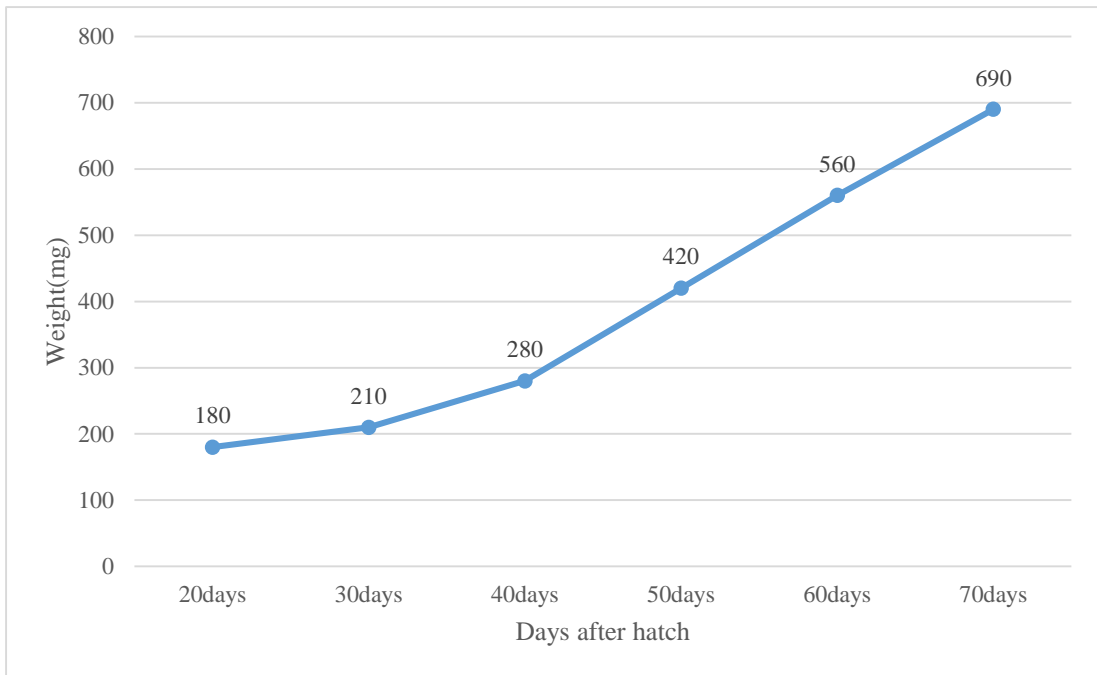


Figure 4: Growth Performance (Weight) of the Fry of Under the Influence of Commercial Feed at Regular Interval

4.3 Physical Parameter

The physical parameter had been examined from the water bodies sub-surface.

4.3.1 Nature of the Day:

Over the course of the study's nature was observed. There were 18 cloudy days, 15 partly cloudy days, 12 wet days, and 1 bright day recorded (clear sky).

4.3.2 Color of Water:

Throughout the whole study period SSFPC water was noted greenish in color.

4.3.3 Temperature:

The brood ponds temperature ranged from 25-31°C in the morning (6-10 am) and 26-36°C during the day. Incubation tank 24-26.5°C in the morning (6-10 am) during the day (25.5-26.5°C). In the morning (6-10 am), it was 30-34.4°C and throughout the day, it was 34-38°C.

5. DISCUSSION

5.1 Latency Time

Regarding the latency period, 10 to 12 hours after the WOVA-FH hormone injection, Pangas (*Pangasius pangasius*) started spawning. The current finding is in line with previous research on catfish induced spawning, including Pangas (Moses et al. 2016) and (Sah et al. 2018).

5.1.1 Induced Fecundity, G.S.I, Fertility Rate, Hatching Rate and Embryology of Fish

WOVA-FH, induced breeding was effectively accomplished in catfish. In the current study, Pangas (*Pangasius pangasius*) were induced to breed while receiving a dose of the hormone. Black sea Bass (Denson et al., 2007), *Carassius auratus*, ornamental fishes (Gnanamuthu et al., 2018), *Clarias* catfish (Daniel et al., 2020), *Pangasiandon hypophthalmus* (Rahman et al., 2020) have all been observed to spawn when given the same WOVA-FH hormone.

In the current study, each female was reported to have produced between 279,000 to 374,000 sticky eggs in a single spawning. The current outcome is compatible with finding Nesa (2017). The fertility of Pangas was estimated to be between 299,000-5,400,000 eggs per female by Singh (2018) and Rahman (2020). According to Sah (2018) research, female Pangas fish had a relative fecundity of 117,000 and 153,000. Compared to farm animals, most fish have higher genetic gains to be acquired through strong selection intensities. This implies that a very small number of people can significantly influence the genetic composition of next generations. Therefore, inbreeding rates may be substantial, which can lead to declines in fitness and other crucial traits (Rahman et al., 1988).

By using the WOVA-FH hormone, the G.S.I (Gonado somatic Index) ranged from 7.80-13.19% in the current study. These results were comparably similar to those reported by Sah (2018) Pangas, where the GSI ranged from 7.21-14.1%. The current outcome is comparable to Moses (2016) in Pangas GSI score of (8.52-12.52%) and Sai (2005) in Pangas GSI score of (9.23-13.67%).

According to Singh et al., (2018), PG had a 76.50% fertilization rate and 58.20% hatching rate. According to Sah et al., (2018), ovulin therapy resulted in the fertilization

and hatching percentage were 85.50% and 48.20% respectively. With PG tested, the percentages of fertilization and hatching percentages of 85.45% and 65%, respectively (Khan and Mollah., 2004). In the current study, WOVA-FH was more successful at promoting fecundity and increasing hatching rate.

At 32-36°C, embryonic development begins at 4hrs after fertilization. Sah (2018) said, in contrast to the current study, that development begins 3 hours after fertilization at 26-31°C. The most vulnerable stages of an animal's life cycle are the embryonic and larval development stages, which can result in mass mortality owing to inadequate husbandry, inadequate nutrition, and temperature changes. The diameter of fertilized eggs, according to Rahman et al., (2020), was 0.65mm. The eggs were very tiny (1.4-1.8mm in diameter), sticky in nature and had a pale, creamy, or brown in color when they were fertilized (Singh et al., 2018). However, Moses et al., (2016) discovered that the diameter of eggs was 1.09-1.28mm immediately before stripping and following fertilization was 1.2-1.45mm. The diameter of fertilized eggs was discovered 0.5mm by (Ferosekhan et al. 2015) and Singh et al., (2015)

In the current investigation, hatching occurred between 26 to 28 hrs after fertilization at a temperature 32-36°C. In Contrast to the current study, Singh et al., (2015) found that hatching occurred in the Pangas within 24-25 hrs at a temperature of 27-28°C. At a temperature of 20-30°C, the pangas hatched out in 24-36 hours, according to Islam et al. (2005). However, Khan and Mollah (2004) discovered that at 32°C, pangas hatched out in 28 hrs.

The usual Pangas developmental stages were divided into two subsequent periods in the current investigation under controlled conditions: the embryonic and larval periods. The blastodisc grew increasingly flat and gradually engulfed the yolk at the blastula stage. Dara et al., (2018) reported a similar outcome with *clarias* catfish. Additionally, Moses et al. (2016) describe a similar outcomes in Pangas.

5.2 Growth Rate of Fingerling

The surrounding aquatic environment, stocking density and food availability, food quality, the number of fish utilizing the same food source, as well as fish age, size and sexual maturity, all have a direct impact on the growth performance of fish.

With the aid of a scoop net, the hatchlings were removed from the pond and their weight and length were measured for up to 70 days. First measurements were taken after 20 days of raising, recording 4.5 cm in length and 180 mg in weight. At 30 days, a second measurement was taken, and hatchling had grown by 5.5cm in length and 210 mg in weight. In-depth, the third measurement in 40 days showed a length of 6 cm and weight of 280 mg. Fourth measurement at 50 days, with 8.5cm of length and 420 mg of weight. Weight was 560 mg and length was 10.5cm at the fifth measurement after 60 days. And the final measurement shows the peak length and weight after 70 days of rearing under the impact of artificial feed, reaching 11.5cm in length and 690 mg in weight under the appropriate physicochemical and environmental component. When artificial food or live food is applied, pangas grow quickly, reaching lengths 10 to 13 cm within the first year, according to reports from Sah (2018), Yadav (2018), Singh et al. (2017), and Khan (2015). Pangas also increase their length and weight quickly when artificial food or live food is applied. Moses (2016), in contrast to the current study, found that after 40 days of raising Pangas with Ovaprim hormone, they had grown to a length of 8 cm and weight of 450 gm.

6. CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

For the purpose of inducing Pangas to breed, the synthetic hormone WOVA-FH has been demonstrated to be more efficient. In the current study, WOVA-FH was more successful in causing ovulation and boosting fecundity and fertility rates. The most fragile stages of an animal's life cycle are the embryonic and larval stages, which can result in mass mortality owing to improper husbandry, artificial food and changes in water temperature. On artificial nutrition, fry and fingerlings grow and survive at their maximum potential. Therefore, the knowledge about the early development of Pangas gained from this would aid in the management and sustainable growth of aquaculture. Pangas has a six-month growth rate, which makes it good for bringing in money.

6.2 Recommendations

1. Fish that are mature and in good health should be chosen for breeding.
2. The water quality needs to be maintained for better spawn production, survival, and growth.
3. Using WOVA-FH hormone to induce Pangas breeding may be more advantageous.
4. More research should be done on the Pangas embryonic growth, survival rate, and hatchling rate.
5. Fish that will be used for breeding must be fully grown, ripe and gravid.
6. Numerous training sessions, workshop and seminar should be held both at Nationally and Internationally to help GN, NGO and INGO make this sector globally competitive.

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PHOTOGRAPHS



Identifying male and female



Packing fry and fingerlings



Injecting hormone



Stripping eggs and sperm



Observing embryonic stages



Feeding for hatchling