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Hypophysation in Fishes

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INTRODUCTION

Induced Breeding in Fishes

A complete scientific guide to hypophysation, hormonal induction, and controlled fish reproduction for sustainable aquaculture production.

What Is Induced Breeding (hypophysation)?

Induced breeding is the process in which fishes are stimulated to reproduce under controlled conditions when natural spawning does not occur in captivity. It is the technique where hormonal preparations are used to activate the maturation of gonads and to release the gametes. This is referred to as hypophysation because the pituitary gland (hypophysis) extract was the first hormone source used for this purpose.

Historical Background

Induced breeding began in the early 20th century to overcome natural spawning difficulties in captive fish.

- **1930** Houssay (Argentina) used pituitary extract to stimulate premature birth in viviparous fish first proof that hormones can control reproduction.
- **1934** Method repeated in Brazil; later adopted in America and Russia. Named hypophysation - crude pituitary glands from donor fish injected into brooders.
- **1937** Khan (India) First attempt of induced breeding on *Cirrhinus mrigala* using mammalian pituitary extract - first Asian application in aquaculture.

- **1955** Dr. Choudhuri applied the technique to minor carps (*Pseudeotropius atherinoides* and *Esomus danricus*).
- **1957** Dr. Choudhuri successfully induced Indian major carps: *C. mrigala*, *C. reba*, *Labeo rohita*, and *L. bata* a landmark achievement.
- **1963** Parameswaran & Alikunhi induced Chinese carps (*Tenopharyngodon idella*, *Hypophthalmichthys molitrix*) in India.
- **1984–90** Chondar described mass-scale breeding of IMC and silver carp in Bangla bundh using HCG combined with pituitary extract.

Principle of Induced Breeding (Hypophysation)

Induced breeding, or hypophysation, is founded on the concept of artificially stimulating the fish reproductive system by externally supplying hormones that the body would otherwise produce naturally. Instead of relying on environmental triggers such as temperature, rainfall, or water current, hormone injections are administered to initiate spawning under captive conditions. Normally, environmental signals activate the hypothalamus, prompting the pituitary gland to release gonadotropins FSH and LH which regulate gonad development. In hypophysation, this step is replicated by injecting pituitary extract or synthetic hormones into brood fish. These hormones travel via the bloodstream to the gonads, promoting the final ripening of eggs and sperm. Once maturation is complete, females release eggs through stripping or confined spawning, while males produce milt for fertilization. The fundamental principle, therefore, is

the deliberate replacement of the natural endocrine signaling process with an external hormonal input, enabling successful and controlled fish breeding independent of seasonal or environmental conditions.

Fish Pituitary gland

Pituitary gland is an endocrine (ductless) gland situated on the ventral side of the brain. It is a small, soft, whitish body whose size and shape vary with species. It is more or less round in carps; oval in catla, rohu and pear-shaped in mrigal. The pituitary is located in a concave cavity known as Sella turcica and enclosed by a thin membrane known as duramater. It may be attached to the brain by a short stalk called the Infundibular stalk.

Types of pituitary glands

Based on the presence or absence of the stalk, the pituitary is classified into

Leptobasic pituitary (with stalk)- e.g. Carps and catfishes

Platybasic pituitary (without stalk)- e.g. Murrels and glassfish (Ambasis sp.)

Steps For Induced Breeding

1. Collection of pituitary gland

Fish pituitary gland can be collected by dissecting and removing a portion of the scalp or through the Foramen magnum.

1.1 Dissecting and removing a portion of the scalp

In this method, the brain case (cranium) is obliquely cut using a butcher's knife/hand saw/bone cutter and the scalp removed. The brain is then exposed by removing grey matter and fatty substance with forceps and cotton. The anterior end (optic and olfactory nerves) of the brain is cut and the entire brain is lifted up and laid back, thus exposing the pituitary under a membrane. After removing the membrane and the fluid, the pituitary is lifted up by inserting the blunt end of the forceps and carefully transferred to a vial containing a preservative.

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1.2 Through the Foramen magnum

Foramen magnum is a large posterior aperture of the skull through which the spinal cord passes. The grey matter and fatty substance are first removed with the help of forceps and cotton (they are pulled out posteriorly). The brain is then exposed. After this, the anterior end (optic and olfactory nerves) of the brain is cut and the entire brain is lifted up and laid back, thus exposing the pituitary. After removing the fluid the membrane, the pituitary is lifted up by inserting the blunt end of the forceps and carefully transferred to a vial containing a preservative.

The first method is commonly practiced even though the second method is less time consuming and a large number of glands can be collected within a short time, with a good resale value of the fish.



2. Preservation of pituitary gland

2.1 Preservation in absolute alcohol

In this method, the gland, after collection, is immediately transferred to a vial/phial containing fresh absolute alcohol (ethanol). After 24 hours, the alcohol is removed and fresh alcohol is added and stored at room temperature or in a refrigerator.

2.2 Preservation in acetone

Immediately after collection, the pituitary gland is kept in ice-chilled acetone and stored in a refrigerator for 2-3 days. After this period, the acetone is changed and the gland stored in a refrigerator. Both absolute alcohol and acetone have de-fattening and dehydrating effect.

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2.3 Immediate freezing

In this method, the collected glands are frozen immediately and stored in a freezer.

3. Preparation of fish pituitary extract for injection

The extract preparation should be carried out just before injection. The required quantity of glands is taken out of vial and they are dried on a filter paper by allowing the alcohol to evaporate. The glands are then homogenized with distilled water or saline in a tissue homogenizer. If acetone-dried glands are used, they can directly be taken for maceration. One-third of the media is used for homogenization, while the remaining two-third is used for rinsing the homogenizer and the glass rod. Recommended dilution rate is 20-30 mg in 1 ml of the media. The extract is centrifuged at 5,000 rpm for 5 minutes. The clear supernatant solution containing gonadotropins is taken in syringe for injection.

Types of injection

Homoplastic injection: Injecting pituitary from one fish to another fish closely related to the donor fish. E.g. carp pituitary gland extract to carps.

Heteroplastic injection: Injecting pituitary from one fish to another fish distantly related to the donor fish. E.g. carp pituitary gland extract to catfish and vice versa.

Methods of injecting fish brooders

There are three methods of injecting brooders they are:

1. Intra-muscular injection:

It is administered into the muscle on the caudal peduncle or behind the dorsal fin, but above the lateral line. It is most effective, convenient, simple and less risky. It is widely practiced.

2. Intra-peritoneal injection:

It is given through the soft regions of the body, generally at the base of the pelvic fin or the pectoral fin. It is risky as it may damage the gonads or liver.

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3. Intra-cranial injection:

In this method, the injection is given through the cranium and is also risky as it may damage the brain. The pituitary extract is administered through a glass or disposable syringe, 2.0 ml capacity, having 0.1 ml graduation.

The size of the needle depends upon the weight of the brooder to be injected.

Needle number 22 is used for fish weighing 1-3 kg, No. 19 for larger fish and No. 24 for smaller fish. When 2 injections are given, one is given on the side that didn't receive the first injection.



4. Dosage of pituitary extract

Assessment of proper dosage is most important for successful spawning. In practice, the female receives two injections, while the male receives only one injection, i.e. at the time of second injection to the female. I Dose or Provocative or preliminary dosage and II Dose or effective or resolving dosage. The interval between the two doses is 6 hours.

Carp glands to major carps

	Female	Male
I Dose	2-3 mg/kg b.w.	Nil
II Dose	5-8 mg/kg b.w.	2-3 mg/kg b.w.

Carp glands to exotic carps

	Female	Male
I Dose	4-6 mg/kg b.w.	nil
II Dose	10-16 mg/kg b.w.	4-6 mg/kg b.w.

Breeding Hapa and Spawning

After injection, a set of one female and two males are released into the breeding hapa, which is a box-shaped container fixed with the help of four bamboo poles in ponds. The hapa is made of fine-meshed mosquito cloth. A thick cloth is not recommended as it does not permit proper circulation of water, causing suffocation. The dimensions of the hapa are usually 3.5 m × 1.5 m. The eggs and milt are passed out through the hapa. If the root which can be opened or closed is fixed on all sides except a part of its bottom, the hapa is fixed to bamboo poles in such a way that the lower part remains above the muddy pond surface and its lower surface remains submerged for circulation of water. Cemented cisterns are also used in place of cloth haps. After 2–3 hours of the second injection, the breeders start swimming actively and become excited and restless. Males begin chasing the female and pushing her with the snout. Spawning usually occurs within 6 hours of the second injection. Although injection may be given at any time in the day, it is better to select a cool and cloudy day, and injections are usually given in the evening when the temperature is low. Spawning occurs at midnight or a little later, but the hapa should not be disturbed till the next morning for observation of eggs. Fertilized eggs are crystalline, transparent and appear like pearls. They rise to the

surface on slight movement of water and are then transferred to hatching hapas for hatching.

Substitutes of fish pituitary gland
Synthetic hormone of fish spawning (Ovaprim, WOVA-FH, Ovotide)



CONCLUSION

Induced breeding has revolutionized freshwater aquaculture by ensuring a reliable and year-round supply of quality fish seeds. From Houssay's pioneering work in 1930 to modern hormonal techniques, this method has continuously evolved to meet the growing demands of fish farming. With the advancement from crude pituitary extracts to refined synthetic hormones, induced breeding has become more efficient, accessible, and widely practiced across the world. It remains a cornerstone of aquaculture, playing a vital role in food security and the sustainable development of the fisheries sector.

"The controlled stimulation of natural spawning processes - transforming seasonal breeding into a year-round, predictable science for a food-secure future."