

PREREQUISITE STEPS FOR DOMESTICATION OF HILSA (*TENUALOSA ILISHA*) IN FRESHWATER POND

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Hilsa (Tenuklosa ilisha) is an important commercial fish species in the eastern region of Indian subcontinent, particularly Bangladesh and West Bengal, India. It contains a number of essential bio-chemicals, including n-3 polyunsaturated fatty acids as health supportive to human beings. It takes a long journey from sea to freshwater for breeding. Until recent past, its population is declining in an alarming state. To conserve its population, ICAR-CIFA initiated its domestication in freshwater pond through following steps: breeding, rearing of seeds and grow-out culture of upto brooders.

Introduction

Fish is a unique creature in this planet, earth. The species is so diverse, spread over marine, estuarine, freshwater and coldwater habitats to cover a wide range of ecological spectra from Arctic to Antarctic horizontally and from hill stream to deep water sea vertically. Among a variety of more than 20000 known fish species until now, Hilsa (*Tenuklosa ilisha*) is a fascinating one (Fig.1). It is a eurihaline amphidromous marine fish belonging to the family Alosinae under order Clupiformes. Its silvery charming lustre, culinary flavour, delicious taste and medicinal importance in human health point of view adorn the species as a special status and so are widely accepted to a large number of populations, particularly eastern part of Indian sub-continent, West Bengal, India and Bangladesh¹. Hilsa is physiologically adapted growing and moving in the diverse ecological spectra while migrating across from sea via estuary and then to river – a wide range of environmental changes as

diverse as variable salinity from strong to medium to less and eventually to freshwater. In spite of its peculiar physiological behavior, ICAR-CIFA initiated its domestication in freshwater pond to save it from already depleting population.

Migration of Hilsa – from Sea to River Vice Versa River to Sea

Hilsa needs to produce offspring and so takes a long journey to migrate from deep sea to freshwater river. For the existence of survival, adults have to migrate in search of freshwater habitat to lay eggs and nurture progeny. In the Indian subcontinent, during breeding seasons both adult males and females of Hilsa migrate from Bay of Bengal to Hooghly River in India and to Padma River in Bangladesh twice a year. A number of males and females search a preferable as well as protected niche in the tidal freshwater river, take a sojourn, establish its breeding ground, and breed there. The fertilized eggs hatch and hatchlings grow to spawn which grow up to fry/fingerlings/ juveniles in freshwater river¹. With times, growing juveniles take journey towards sea - a new destination to live with accompanying parents, who would return home from river to sea via estuary – a journey accustomed in the Hilsa life cycle. The journey adopted by Hilsa from sea to river

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Fig. 1: A view of mature Hilsa

makes curiosity to fishery biologists because how the physiology of this species changes from saline to freshwater through continuously adjusting osmo-regulation is a matter of biological inquisitiveness.

Food and Feeding Habit to Support a Long Migration

Food is for energy. Hilsa requires substantial foods to support for a difficult journey and that to a long route of uncertainty indicating upwelling of water current, oceanic movement, salinity variation, attacks from predators, anthropogenic disturbance and more so. The species is planktophagus that means to feed on a variety of plankton comprising both phytoplankton and zooplankton. The method of feeding is same, but quality of foods changes along with changing niche from sea to river¹. Being a filter feeder it feeds by sieving the food particles through the gill rakers while swimming in the water, keeping the mouth open. It takes anything of plankton that enters into the mouth, and does not show any selectivity in feeding. This species is mainly a visual feeder and possesses well

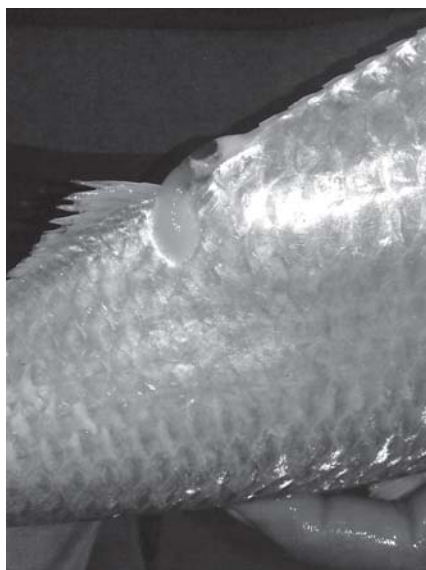


Fig. 2: A gravid female Hilsa ready to ooze out eggs

developed optic lobes and reduced olfactory lobes; both organs help it moving through planktons dominated water way. Juveniles of hilsa are voracious plankton feeders, although mainly feeds on zooplankton while staying in freshwater environment during their early stage of life¹. Quality of foods from sea to river seems a remarking variable: biochemical properties contained in deep sea

plankton is much richer than that of estuarine and of river planktons. When adults initiate distant long journey from sea they feed biochemically rich food of sea plankton that support their journey as compared to feeding planktons in other environments.

Hilsa as a Medicinal Food to Human Health

Not that it has only attractive taste, but that it contains a number of essential bio-chemicals which are health supportive to human beings. Research finds that eating Hilsa reduces the chances of sudden fatal ailment as heart attack. Hilsa is a good source of n-3 polyunsaturated fatty acids (PUFAs), particularly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). The n-3 PUFAs play a vital role in the development and function of the nervous system (brain), photoreception (vision) and the reproductive performances that prevents cardiovascular ailment, reduce the risk of coronary heart disease, inflammation, hypertension, hypertriglyceridemic effect, allergies, arthritis, autoimmune disorders and cancers^{2,3,4}. Besides, the rich content of nutrients such as protein (18-23%), fat, vitamins and minerals including phosphorous, calcium, magnesium, sodium, potassium, zinc, iron, copper and manganese are found in Hilsa that provides a health supportive food essential to human health⁵. The proteins contain the essential amino acids: arginine, histidine, isoleucine, leucine, lysine, methionine, phenyl alanine, threonine, tyrosine, valine and proline; umami taste amino acids: glutamic acid and aspartic acid and sweet taste amino acids: serine, glycine and alanine – all these appear to be human health supportive nutrients² as if Hilsa is made for protecting human health.

Domestication of Hilsa - a Daunting Task

Hilsa population dwindles rapidly in freshwater river systems as much as in alarming state due to indiscriminate catch, over exploitation, water pollution, and encroachment of its breeding and nursery grounds. Mature brooders

including males and females fail to recruit a substantial number of progenies through potential breeding, resulting in declines of off-springs in successive generations. To recover Hilsa population from its further declining, a strategic plan for their conservation is essential and in such case its domestication through breeding and rearing gets priority – a daunting task initiated. Breeding techniques for culturing different fish species is well established and now updated gradually. With such expertise, Scientists of RRC (Regional Research Centre), ICAR-CIFA (Indian Council of Agricultural Research - Central Institute of Freshwater Aquaculture), Kolkata has initiated domestication of Hilsa in freshwater pond as captive culture practice. They undertook several trials to overcome the hurdles of Hilsa breeding and rearing in control and eventually, succeeded through achieving the following steps: onboard breeding, transport of fertilized eggs, rearing fertilized eggs in hatching pool, rearing of larvae, fry and fingerlings through co-feeding, weaning the fish at early stage with supplementary feeds and pond management.



Fig. 3: A mature male ready to ooze out milt

Catching Hilsa in River to Necessitate a Coordination Between Scientists and Fishermen

Hilsa catch from river is a prerequisite task for breeding since Hilsa is not available in any closed water body. Hilsa migrates during February-March and then September-October in a calendar year and that happens around fool moon and new moon periods, therefore exclusive time/period matching is one of the critical factors for undertaking breeding⁶. For catching Hilsa, only skilled fishermen of traditionally long experience are able to do so by venturing of sailing boat against tidal current in the river. The fishermen keep dragging monofilament gill net below water surface against tidal current while sailing mechanized motor boat – the task as much risky as life threatening, but they make it as usual for livelihood. Before catching Hilsa, Scientists train the skilled fishermen

regarding handling brooders at the time of lifting them from net and then keep them inside boat water for a while till scientists reach to observe. Scientists who perform breeding have to be boarded in another motorized boat to carry all the essential equipments and chemicals and stay close to targeted fishing boat. Sooner the skilled fishermen catch the gravid fish, quicker they inform Scientists by raising hands. Hilsa being very sensitive die immediately after being lifted from water; however, experienced fishers could keep few broods alive (both males and females) with special care for breeding operation. Breeding needs to be operated as soon as possible on the boat itself.

Breeding of Hilsa – An Essential Biological Phenomenon

Breeding is an essential aspect among other biological activities for domestication of any fish species of high importance. That breeding is performed artificially in on-boat while boat floats on high tidal water current in river seems a difficult task. Some of the specific constraints which are usually encountered in Hilsa breeding include the following:

- Collection of sufficient number of both healthy females to have free oozing eggs (Fig. 2) and males with oozing milt (Fig. 3).
- Coincidence of time when both females with free oozing eggs and males with milt are available together – an important factor for Hilsa breeding. Sometimes, healthy females with free oozing eggs are caught, but not males with oozing milt available and vice versa in the condition.
- Females and males which are caught with monofilament gillnet are highly stressed and thus remain alive few minutes only.
- Sometimes, gravid females and males are available, but either of the sexes or both the sexes are not viable enough for breeding.

Scientists' Experience to Artificial Breeding of Hilsa

Extensive efforts were made in collecting viable broods of both males and females. Immediately after their catch from fishermen, their maturity status was to check. The criteria for selection of broods of both sexes were females having freely oozing yellow colour egg and males with freely oozing white colour milt. Having collection of both sexes of Hilsa brood, the necessary measures are required as:

- i) Dry stripping – a artificial technique to facilitate ‘On-board breeding’

The selected brooders were turned upside down by hand to remove surface water from buccal cavity, gills and body. Then both males and females were soaked with clean towel or tissue paper to absorb the least amount of surface water. Water to mix with eggs causes to lose fertility. A gentle finger or thumb pressure was put both sides of the upper region of female abdomen to move eggs towards vent and eventually to strip ovulated mass of eggs out through the vent. Stripped eggs were collected and spread in a clean steel/enamel tray. Then the male was gently pressed to strip milt out and put on the layer of eggs⁶. The entire activity which was performed while boarded in the boat is known as ‘On-board breeding’. Following the activities were necessary afterwards;

- With soft clean feather, eggs and milt were mixed thoroughly to achieve maximum fertility (Fig. 4).
- A small amount of river water (river water filtered through bolting silk hapa) was sprinkled gently as well as uniformly over the milt-eggs mixture to activate the process of fertilization.
- Sperms, stimulated with sprinkling water, started moving very fast, got adhered with egg membrane, remained active only 40-50 seconds, and then lose fertility.
- Much amount of water were added gently after 4-5minutes, and the mixture be kept undisturbed.
- Operation of post fertilization needed repeated washings with addition of freshwater to clean clogged milt, mucous, scales, faeces, blood, oil

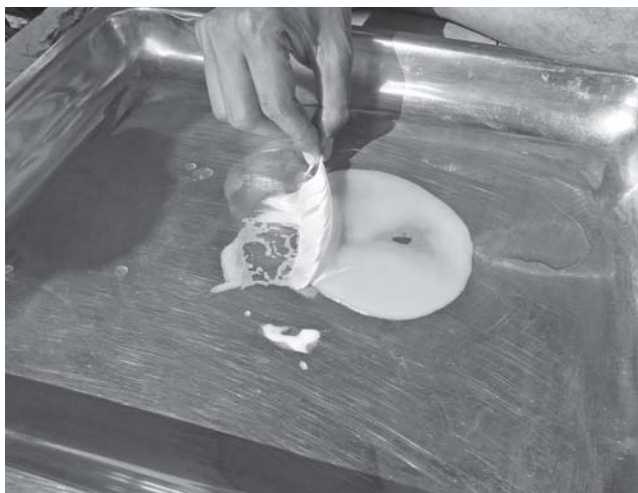


Fig. 4: Artificial fertilization performed through mixing of eggs and milt

droplets and other wastes so as to avoid bacterial or fungal infections.

- ii) A changing phenomena in the post fertilization

Fertilized eggs started swelling and got separated from each other. The condition when eggs get fully swelled in 45 minutes is known as ‘Water hardening’. The features of fully swelled fertilized egg as much as 2.0 mm diameter looked spherical, transparent, and yellowish yolk with numerous oil globules. By contrast, unfertilized eggs were measured around 1.8 mm in diameter with whitish yolk. Fertilized eggs which were separated from unfertilized ones were packed with oxygen filled polyethylene bags and transported by motor vehicle to Regional Research Centre, Rahara and its Kalyani Field Station, ICAR-CIFA, 120km away from the breeding spot. Fertilized eggs were put into hatching pool/aquaria with aeration facility to increase DO (Dissolved Oxygen) and fertilization rate varied between 95-98%^{6,7}. Within short time, eggs hatched to produce hatchings– an eternal biological phenomenon to begin life for its journey. All the hatchlings were reared in the aquaria till 4 days without giving any natural food or supplementary feed. The 4th day larvae as called as spawn measured as 5 mm/0.5 mg were able to swim in water. Then a total of 3.5 lakh larvae of 4-days old were stocked in the nursery pond for fry production.

Nurturing Hilsa to Grow from Spawn to Fingerlings

Larvae opened their mouth at 4th day of their age when they required exogenous natural food. The first feeding larvae (spawn) fed on *Chlorella vulgaris* as a starter food due to their small mouth gape. With increasing size, they fed on other phytoplankton, rotifers and large zooplanktons as Cyclops, Diaptomus, Moina, Daphnia through co-feeding – more than two types of foods used for feeding simultaneously. In co-feeding, larvae fed on both *Chlorella* and *Brachionus* sp. The supply of *Chlorella* served two purposes: (i) used as food to both Hilsa larvae and *Brachionus* which grew profusely within 2-3 days, and (ii) increase of DO into culture water through photosynthesis. When spawn grew to fry 40-60 days they started moving along the side of the ponds. They were provided with finely powdered supplementary feed following three ‘F’s –fixed time, fixed type of feed, and fixed place for weaning. Fry grew as fingerlings after 90-120 days of rearing. A huge amount of mixed zooplanktons was supplied daily along with powdered supplementary feed @ 5-10% of the individual body weight of larvae⁷.



Fig. 5: Scientists examine natural foods into pond conditioned for Hilsa culture in captivity

Grow-out Culture of Hilsa – a Way towards Domestication

Fingerlings grew as table sized fish as weighed around 100g after 5-6 months. Then they were stocked in the larger ponds, with supply of exogenous feed @ 5-10% of the individual body weight, along with sufficient amount of zooplanktons (Fig.5). When table sized fish grew adult, they were fed with exogenous feed @ 3% of their body weight. DO (Dissolved oxygen) was maintained above 6.0 ppm by operating with pond aerator. Usually, around 2 years of age, males became mature with freely oozing milt while females exhibited protruding eggs through the vent after nearly 3 year of age.

Conclusion

Hilsa population is decreasing in an alarming state. To rescue Hilsa from its state of endanger in near future, Scientists came forward to breed it artificially. The artificial propagation of Hilsa and its culture in captivity through extensive and continued research efforts offers the most appropriate strategy for its conservation. Until now, cycling of Hilsa life under captivity is yet to be accomplished biologically because captive brooders need to breed under pond condition. However, major of life cycle such as breeding, spawning, hatching, nurturing of juveniles including fry, fingerlings, growing of table sized fish, mature fish with gravid condition, their feeding with

both natural foods and supplementary feeding through weaning and culture practice is tested and established as a protocol. Scientific know-how from Lab- to - Land and Land - to - Lab is recognized through peer reviews and high power scientific body time to time. Scientists developed the technique to replenish the depleted Hilsa population through ranching of artificially bred fry and fingerlings. This success seems a

breakthrough in Hilsa breeding and culture that enables generations to see this natural creature sustained and protected in its habitats.

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References

1. V.R. Suresh, A.M. Sajina, S. Dasgupta, D. De, D.N. Chaattopadhyay, B.K. Behera, R. Ranjan, V. Mohindra and S. Bhattacharya, Current status of knowledge of Hilsa (ICAR-Central Inland Fisheries Research Institute, Barrackpore, W.B., Kolkata, 2017) p. 108.
2. K.S. Sidhu, *J. Reg. Toxicol. Pharmacol*, **38**, 336–344 (2003).
3. C. Von Sckacky, *Curr. Atherosclerosis Rep.*, **5**, 139–145 (2003).
4. A. Mnari, I. Boulel, I. Chraief, M. Hammami, M.S. Romdhane, M. El Cafsi and A. Chaouch, *Food Chem.*, **100**, 1393–1397 (2007).
5. M.A. Hossain, S.M. Almatrand and A.A. Al-hazza, *Indian J. Fish.*, **61** (2), 58-66 (2014).
6. D.N. Chaattopadhyay, A. Chakraborty, P.K. Ray, and P. Jayasankar, *Indian J. Fish.*, **66** (2), 125- 135 (2019).
7. D.N. Chaattopadhyay, A. Chakraborty, P.K. Ray, R.N. Mandal, S.K. Banik, V.R. Suresh and K. Ghosh, *Aquaculture Research*, **50** (3), 778-785 (2018).