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Silurid Butter Catfish *Ompok bimaculatus* (Bloch1794) of North region: A General Overview

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Abstract: ***Ompok bimaculatus*** **is the most demanding, economically important freshwater butter catfish. As a cost of development, water health is decreasing and so is aquatic life. Because of deteriorating water quality and uncontrolled fishing *O. bimaculatus* stock is under threat. To conserve this species and to fulfill consumer's demand of fish for food, captive breeding techniques are recommended as a solution. Like major carps, they are not responding to commercial gonadotropins. The reason may be with *O. bimaculatus* as it registered a wide genetic variation within the same species. Similarly, different habitat fish are different in their reproductive parameters too. To understand all these restrictions, one should have full detailed information on genetic, and physiological species differences to find out the best strain of *O. bimaculatus.* Reproductive parameters, availability, and habitat variation study of the Northern region will help in the breeding practice of *O. bimaculatus* in the region. This review will provide one-hand information on most of the important parameters to keep in mind while doing cultural practice.**

Index Term: **butter catfish, embryogenesis, gametogenesis, geographical variation, gonadosomatic index, oocyte diameter**

I. INTRODUCTION

Fish has huge economic significance among them catfish have special importance because of their easily digestible source of protein, full of other essential nutrients such as lipid, carbohydrate, minerals, and vitamins (Rajagopal & Davidar, 2008). In addition to direct economic benefits, fish are bio-indicator of water body biochemical profile in a life-supporting way (Bijukumar & Sushama, 2000). Due to increased water pollution as domestic, agricultural, and industrial effluent mix in natural water bodies, aquatic life is under threat. Among them, fish are more sensitive to changes in water quality. Deteriorating water quality influenced all physiology especially reproduction due to the compromised health conditions of fish. It is one of the reasons for the depletion of natural wild fish stock in addition to uncontrolled fishing. For successful fish culture and conservation program, an understanding of fish reproductive biology is essential. It will also include maintenance of a healthy population and optimization of appropriate broodstock (Shabanipour & Hossayni, 2010).

Ompok bimaculatus is an indigenous freshwater small fish commonly known as butter catfish confined in the South East Asian region. Due to its soft, nutritional, delicious value, it is highly priced in the Indian market and known as by the different vernacular names; butter catfish, glass catfish, one spot catfish, pabda, etc. The body of *O. bimaculatus* is elongated and the color is silvery grey. Among identifying features, is a black spot present on the shoulder, on the lateral line, behind the gill opening and above the middle of the pectoral fin. A small triangular black spot just above the lateral line on the caudal peduncle. The meristic traits of fish are used in order to discriminate them to their nearest species. The two-pair barbels are present in which one pair maxillary barbel extends beyond the proximal end of the anal fin while the other mandibular barbel is rudimentary as compared to maxillary barbels. The pelvic fin is not extending to the proximity of the anal fin. The

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large eyes of the species indicate its diurnal nature as compared to other silurids whose eyes are poorly developed which indicates their nocturnal nature (Raghunathan et al., 2003). The pectoral fin has 13-14 soft rays with a pectoral spine. The dorsal fin is not prominent however it has 4 rays and the caudal fin has 18 rays with forked in shape and separate from the anal fin (Srivastava, 2007). The anal fin becomes gradually narrower posteriorly with 60-75 rays (Jayram, 1981). The decline of the population of this species raises many questions regarding to their conservation in the *in-situ* as well as *ex-situ*. The biological causes of their declining population may include decreasing genetic diversity, sex reversal, immune-compromised health to their natural habitats etc. There is less knowledge of the species on the reproductive traits in order for their conservation and genetic stock assessment.

II. DISTRIBUTION

Systematic position

- Domain: Eukaryota
- Kingdom: Animalia
- Group: Pisces
- Phylum: Chordata
- Subphylum: Vertebrata
- Superclass: Gnathostomata
- Class: Osteichthyes
- Subclass: Actinopterygii
- Division: Teleostei
- subdivision: Euteleostei
- Superorder: Ostariophysi
- Order: Siluriformes
- Family: Siluridae
- Genus: *Ompok* (Lacepede, 1803) (Boeseman,1968)
- Species: *bimaculatus* (Bloch, 1794)

A more holistic understanding of *Ompok bimaculatus* in their native environment, including habitat preference, reproductive behavior, physiological behavior of stress, and diet, is necessary for both the refinement of standard protocols and the optimization of their use in a wide array of biomedical, behavioral, genetic as well as cognitive studies. In practice, little of the available ecological information has been used to develop husbandry protocols and it should be used for the different areas of research, such as reproductive profiling in a spatio-temporal way with relation to neuroendocrine mechanisms for the improvement of the care of butter catfish in research facilities. The maximum size of butter catfish, *O. bimaculatus***,** was reported at 45 cm (Talwar & Jhingran, 1991), while the size at first maturity for the male was 23.6 cm. However, for the females, it was unknown. As a potamodromous fish, the butter catfish was discovered in freshwater and brackishwater ecosystems with pH ranges of 6–8 (Riede, 2004). The butter catfish is a tropical fish and it tolerates 20–26° C. (Riehl et al., 1991). The food and agricultural organization (FAO) has confined the area of butter catfish to Asian-inland water bodies. It is found in a variety of ecosystems, including the Cauvery, Godawari, and Krishna rivers (Chandrashekhariah et al., 2000), the Indus (Ali et al., 2005), and the Chilka lake (Rao, 1995), as well as the oriental relam (Mirza et al., 1984), and the palearctic relam (Talwar & Jhingran, 1991). The butter catfish, *O.bimaculatus* is a native species of the Asian region and has been reported in Afghanistan (Talwar & Jhingran, 1991), Bangladesh (Rahman, 2001), India (Menon et al., 1999), Myanmar (Hla Win, 1987), Nepal (Shrestha, 2016), Pakistan (Mirza et al, 2002) and Sri Lanka (Pethiyagoda, 1994). The chromosome number of butter catfish was n = 40 (Arkhipchuk, 1999).

Butter catfish *(O.bimaculatus*) is indigenous to South East Asia and are broadly distributed across parts of India, Bangladesh, Borneo, Java, Laos, Malaya, Myanmar, Pakistan, Sumatra, Thailand, Vietnam, and Yumuna (Balasundram et al., 2001, Dua et al., 2009, Ghosh & Lipton, 1982, Hussain et al., 1998, Sarkar et al., 2010, Sarkar et al., 2011). This geographic region has a monsoon climate with pronounced rainy and dry seasons that have a profound effect on habitat parameters, including water chemistry and resource abundance. Butter catfish (*O.bimaculatus*) have been reported to occur in a wide variety of habitat types within this region, including man-made fish ponds, dams, and rivers. However, the results of field surveys conducted in India (Sarkar et al., 2012) provide the most comprehensive description of the habitat preference of this species to this date. Sarkar et al., (2011) sites within the Ganges River drainage in the Indian states including Uttar Pradesh, Jharkhand and West Bengal, They also reported from the tributaries of Ganga such as River Gomti, Ramganga, Ghaghara, Yamuna, Betwa, Dhawan etc. These butter catfishes are also found in the reservoirs of water bodies on river Subernlekha. *Ompok bimaculatus* inhibits in habitats with fast-flowing water, backwater shallow pools, and deep pools (Sarkar et al., 2010). The rivers are quite shallow ranging from 0.5-1.5 m in depth and are often muddy and murky. It occurs in streams and rivers of all sizes with currents ranging from sluggish to moderate. It also occurs in canals, beels and inundated (Pullin et al., 2007). The pH (6–8), hardness (4–28°dGH), and temperature (20–26°C) were preferred by *O. bimaculatus*. These data are useful in that they may be directly applied to the design of optimum water parameters for *O. bimaculatus* in the laboratory, as production in captivity may be highest when fish are housed in water that reflects that to which they are adapted in nature. Future field studies should fully investigate habitat conditions during the summer months, over a broader range of habitat types, to determine the full range of habitat preferences of butter catfish (*O.bimaculatus*). In nature, this makes it possible to better match water parameters for the laboratory stocks.

Habitat fragmentation, overexploitation, and anthropogenic causes are responsible for the decline of the butter catfish population. Recently, the International Union of Conservation and Nature (IUCN) has declared the species near threatened (Ng et al., 2010). It is now necessary to conserve the population of butter catfish, *O. bimaculatus* for its germplasm integrative as well as its bio-economical importance in aquatic ecosystems.

Aquaculture management and conservation require complete knowledge of the best wild strain to be propagated with full information on reproductive parameters. The reproductive success of fish species can be assessed based on size at maturity, the overall health status of the brooder, gonadosomatic index, gonadal maturation, fecundity, etc. (Fontoura, Braun & Milani, 2009). In the review, interspecies variation among O. bimaculatus in the Ganges and its tributaries, and its breeding practice in the North region were compiled in favor of the management of this near-threatened fish species' conservation strategies.

III. GENETIC AND ENVIRONMENTAL VARIATION

With the advancement of germ cell transplantation, well-organized approaches to germ cell conservation will be required. Initially, to investigate germ cell proliferation using various cognitive cues such as social status and learning motivations. Sensitization, habituation, etc. To determine cell proliferation, inject the thymidine analog 5-bromo-2-deoxyuridine (BrdU). BrdU is commonly used in vertebrates to examine gametogenesis (Spermatogenesis, Oogenesis) because it is incorporated into the S phase of mitosis and thus is an indicator of cell proliferation. Further research would be required in *O. bimaculatus* for determining the cell proliferation and its application in the area of germ cell transplantation for the better-yielding variety of butter catfish *O. bimacultus.*

The geographical distribution is known to influence reproductive parameters that include maturity, fecundity, gamete weight size, spawning, survival of young ones, etc. (Shinde et al., 2002). The fecundity and egg quality vary in different species and also in the same species because they are influenced by the local habitat and environment (Barmanh & Saikia, 1995; Kulshrestha et al., 1990; Senthilkumaran & Joy, 1995). In the Indian riverine system, the Ganga is the main freshwater river. It originates from Gaumukh in the Garhwal Himalayas and falls into the Bay of Bengal. The Ganga covers Uttarakhand (Western Himalaya), the North Plains (South and Eastern India), Nepal/Tibet, and Bangladesh (Gopal, 2000). It carries a rich biodiversity. Differences in species occur due to conserved gene variation and also as a result of different habitat adaptations. *O. bimaculatus* has relatively high genetic diversity within close streams, though these genetic variations are not free in genetic flow and they reserve the right to gene flow restriction among them (Lakra et al., 2010; Malakar et al., 2013). The genetic structure and phylogenetic relationships in *O. bimaculatus* from different habitats suggested distinct mtDNA lineages that had branched haplotypes. Because of its star-like branched lineage, *Ompok sps*. recorded high genetic variation and haplotypic diversity. (Kumar et al., 2016; Rashid et al., 2012).

The area of reproductive behavior would be fruitful in areas where the degree of genetic relatedness of individuals in shoals of adults is also unknown. This premise is supported by population genetics studies that have shown high levels of genetic variability and a weak genetic structure among fish collected from the four different sites in India, at least based on microsatellite markers (Gratton et al., 2004). Studying the reproductive traits of fishes on the basis of sex would be fruitful in order to compare their reproductive rhythmicity. The reproductive rhythmicity of fishes is controlled by the combinatorial effect of genetics in relation to environmental plasticity.

Genetic variation within species of O. bimaculatus was studied with an amplified Cytb gene (1,118 bp long). The analysis revealed that there are significant variations at the level of haplotypes and nucleotides within and between different habitats of O. bimaculatus. The analysis of molecular variance indicated 73.53% genetic variability between the populations and 26.47% genetic variation within the population. Different types of phenol may have different fitness, and selection leads to the emergence of proven phenotypes that provide ground for more genetic descendants (Malakar et al., 2013; Jebur, 2018) (Table 1).

Species	Population name	Examined individuals	Haplotype diversity	Nucleotide diversity	Haplotypes	Mean genetic distances	References
Ompok bimaculatus	Samarra Dam	20	0.11	0.4	2	0.01878	Jebur, 2019
	Shatt Al-Arab	20	0.12	0.6	3	0.01915	
	Hooghly	10	0.500	0.000	7, 3	0.005	Malakar, et al, 2013
	Girwa	13	0.298	0.001	6, 4, 3	0.063	
	Betwa	14	0.272	0.003	7,4, 3	0.003	
	Gomti	9	0.6000	0.000781	2	-	Kumar et al, 2016
	Ganga	6	0.7333	0.000781	3	-	
	Tapti	6	0.0000	0.000000	1	-	
	Satluj	6	0.0000	0.000000	1		

Table 1. Genetic variation among *Ompok bimaculatus* population based on mtDNA cytochrome b gene.

The environmental gradients such as temperature, pH, salinity, dissolved oxygen, water hardness, and the contents of minerals such as Ca, P, N, etc. also affect the physiological status of the organisms in relation to reproduction through neuroendocrine integration. The neuroendocrine integration provides signaling to reproductive hormones and neurotransmitters as a result of receiving specific environmental gradient stimuli. Few fishes exhibit such patterns as fractional spawning with group synchronous female reproductive tissue (Zhang et al. 2009). Seasonal reproductive cyclicity includes the annual regeneration of gonadal tissue, which is mediated by hormone regulatory pathway primarily involving gonadotropin-releasing hormone (GnRH), luteinizing hormone(LH), Follicle stimulating hormone (FSH), growth hormone (GH), melatonin, and other sex-steroid hormones (Lincoln et al., 1998; Trudeau et al., 1997; Thiery et al., 2002). Melatonin is secreted from the pineal gland and it is directly regulated by the photoperiod through the retinohypothalamic tract. Melatonin influences the neuroendocrine system through its receptors, which are widely distributed in the teleost brain (Ekstrom et al., 1992; Mazurais et al., 1999; Vernadakis et al., 1998**)**. Gonadotropin-releasing hormone (GnRH) is found in multiple forms in the brain. These multiple GnRH forms activate the anterior pituitary receptors to stimulate the expression and secretion of LH, FSH, and GH, which are all involved in controlling seasonal gonadal development and sex steroid synthesis. Moreover, the synchronization of fish reproductive behavior in the breeding season involves a well-defined sex pheromone response system, which indirectly affects the release of LH and induces ovulation in females and sperm release in males (Sorensen et al., 1998).

The closed structures of water bodies such as dams restrict the environmental gradient, which either directly or indirectly influences the reproductive potential of the species as compared to lotic ecosystems (Moussa et al., 2012). *O.bimaculatus* such as size at first maturity (L_{50}=23.6 cm) at Bhavanisagar reservoir in Tamil Nadu for the male. However, for the female, the size at first maturity was unknown. Thailand reported absolute fecundity (1700-68,000) of butter catfish, *O. bimaculatus* (Table 2). However, the gonadosomatic index was used as a marker in relation to body mass for determining the reproductive cycle. The gonadosomatic index is influenced by feeding intensity directly but not the gonad index (Cayre et al., 1996). Further research would be required in order to determine the comparative study of the reproductive potential of butter catfish *O. bimaculatus*, in relation to ecosystems. Reproductive strategies may deal with age and size at sexual maturity, fecundity, spawning conditions, parental behavior, sensitivity to environmental factors, egg, and larval morphologies, etc. Now it is necessary to study the reproductive traits of eggs, spawners (age at sexual maturity (years), relative fecundity, absolute fecundity, maximum GSI value (% of body weight), sexual dimorphism in males, and spawning.

Rivers	Condition factor	Oocyte diameter (mm)	Size at first maturity ($L_{50\%}$) Female	Size at first maturity ($L_{50\%}$) Male
Ganga	0.57 ± 0.14^{a}	0.5 ± 0.1^{a}	247	216
Gomti	0.63 ± 0.24^{a}	0.6 ± 0.1^{a}	232	227
Ramganga	0.55 ± 0.15^{b}	0.6 ± 0.1^{a}	262	242
Ghaghara	$0.60\pm0.0.17^{a}$	0.8 ± 0.0^{a}	228	198
Betwa	0.62 ± 0.11^{b}	0.5 ± 0.1^{a}	242	238
Sone	0.57 ± 0.07^{a}	0.6 ± 0.1^{a}	249	247

Table 2. Mean ± SD of condition factor and reproductive parameters of Ompok bimaculatus from six populations from different rivers of the Ganges basin (Praveen et al., 2017). Superscript *a* indicates no significant difference (p>0.05); *b* for significant difference (p<0.05) SD: Standard deviation.

The extensive comparative analysis of the reproductive traits of butter catfish is for the conservation and domestication of the species. In the pathway of steroidogenesis, cholesterol acts as a precursor for the formation of steroidal hormones which are responsible for reproductive events. Ultimately, 11-Ketotestosterone and 17β-Estradiol are responsible for the formation of spermatogenesis and oocyte growth in the male and females, respectively. During the breeding period, their titer reaches its maxima with a temporal expression in which the gonad somatic index is used as a marker for determining the reproductive season of the fish (Martin & Fabrice, 2009).

In butter catfish, no prior work has been reported about the reproductive hormones and their response system. The reproductive behavior of a few fishes was studied in patterns of the male attractiveness index. In spite of all these, No prior work has been reported on the behavioral plasticity of butter catfish *O.bimaculatus*.

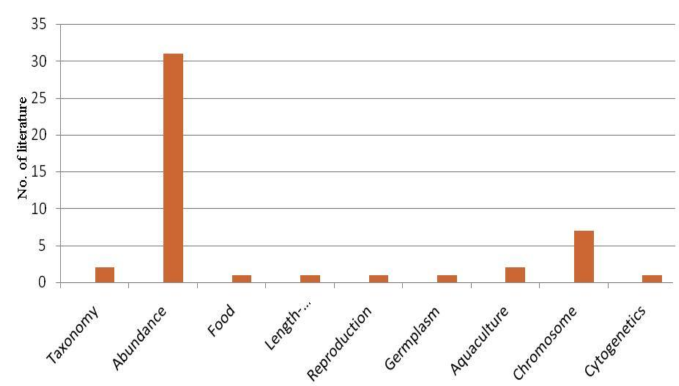

Fig 1**.** Availability of literature on different aspects of butter catfish *O.bimaculatus*

The analysis of fish biology of *O. bimaculatus* assists in elucidating that different aquatic habitats and environmental conditions influence the morphometric growth and reproductive performance of fish as they affect the maturity of fish. Due to the difference in geographical conditions of water bodies, these fish show a difference in their growth and breeding performance too (Sarkar et al., 2018). The reproductive parameters can be attributed to the differences in abiotic factors such as photoperiod, temperature, and rainfall as well as biotic factors like food availability and physiological characteristics which are generally considered as determining factors that trigger the reproductive cycle (Menezes and Vazzoler, 1992).

On the basis of variations in parameters of *O. bimaculatus* from 16 rivers (Betwa, Brahmaputra, Chambal, Ganga, Ghaghara, Gomati, Hooghly, Krishna, Mahanadi, Narmada, Ramganga, Sharda, Sone, Subarnarekha, and Tapti), fish can be divided into high absolute, moderate absolute, and low absolute categories. Though the result of different parameters from varied geographical rivers, they overlap with each other (Cavalcanti et al., 1999; Minos et al.,1995; Sabadin et al., 2010). Heterogeneity in these characteristics may be due to the difference in habitat. (Jenning et al., 2001) reported that variation occurs in morphometric characteristics among similar species of fish from different geographic locations and is influenced by environmental variables.

IV. MORPHOMETRIC VARIATION

Morphometry (size and shape) has been used in fisheries for taxonomy as well as best strain identification within species of different habitats as per local environmental variation (Chaklader et al., 2016; Sarower-E-Mahfuj et al., 2020). Several scientists worked on reproductive history and morphological comparisons of populations from different natural habitats to find out the difference within one species due to inherited body configuration and local changes (Petrakis & Stergiou, 1995). Based on wild catch from the Indian freshwater Ganga and its tributaries (viz., Betwa, Brahmaputra, Cauvery, Chambal, Ganga, Ghaghara, Gomati, Hooghly, Krishna, Mahanadi, Narmada, Ramganga, Sharda, Sone, Subernrekha, and Tapti River), fish were divided into morphology, reproductive parameters, and embryo survival categories (Mishra et al., 2016). A healthier and longer *O. bimaculatus* population was observed in the Narmada River at the Hosangabad site, and a small population of *O. bimaculatus* was found in the Hooghly River at the Naihati site. Heterogeneity in these characteristics was due to the difference in habitat and local environmental variables (Gupta et al., 2015; Mishra et al., 2016). Fish growth is called isometric growth when length increases in equal proportions to body weight. Fish from the rivers Betwa, Mahanadi, and Ganga showed isometric growth. Spencer (1864-67) described "cube law" for organism growth, i.e., W L3". The positive allometric growth for *O. bimaculatus* was exhibited for the Hooghly, Krishna, Ramganga, and Subernrekha Rivers. Negative allometric growth was represented by the Brahmaputra, Cauvery, Chambal, Ghaghara, Gomati, Narmada, Sharda, Sone, and Tapti Rivers. So, the length-weight relationship in fishes can be affected by habitat, availability of food, and degree of stomach fullness (Mishra et al., 2016). Research stated that there is a direct linear correlation between physical-morphometric dimensions and the gonad of *O. bimaculatus*. The highest correlation existed among fork length, standard length, and total length. The correlation level was high in body depth-fork length and body weight-total length. There was a high correlation between ovarian protein concentration-fecundity and gonadal weight-gonad somatic index (Mishra et al., 2018; Sarkar et al., 2017). This variation may be attributed to the differences in abiotic and biotic factors which are determining factors in maintaining the reproductive cycle (Vazzoler & Menezes, 1992).

V. REPRODUCTIVE PARAMETERS VARIATIONS

Fish breeding strategies not only considered the geographical best strain they also consider other important parameters such as gonadosomatic index (GSI), ovary weight-protein, fecundity, oocyte diameter-weight, and ovarian morphology. In the breeding season, somatic growth slows down and eventually stops but the gonads increase maximum in size reflected with the GSI pattern. In the breeding season proteins from somatic tissue are utilized in the reproductive organs (Chandrasekhara & Krishnan, 2011). The nutrient availability in fish viz protein, carbohydrate, and lipid content are important in the best gamete quality and embryo development followed by maximum survival after fertilization (Mishra & Devi, 2020). Due to differences in their local habitat i.e. river conditions, availability, and type of food variety, the wild population of *O. bimaculatus* had differences in their ovarian protein concentration and their fecundity as well in the different wild populations of different Indian rivers (Mishra et al., 2016, 2018). The ovarian protein concentration of *O. bimaculatus* from the wild population of Indian rivers varied with respect to rivers and with seasons as well. In the preparatory phase, Ganga river samples recorded high gonadal protein whereas, during the pre-spawning and spawning phase, Narmada river samples had high protein concentration. During the pre-spawning phase, fish remain hyperactive due to favorable environmental conditions and plenty of food supply. This further strengthens by an increase in the muscle protein content of fish that will later utilize in the spawning phase when there is nil feeding by gravid fish. Therefore increase in muscle protein will coincide with gonadal maturity. Hence, major macromolecular levels in muscle and gonads will be able to interpret gamete development and health along with fish morphometry (Bano, 1977; Mishra & Devi, 2020). The annual GSI of butter catfish was more or less the same in different wild habitats but the level of the GSI index varied with the water condition of their habitat. Geographical differences are clearly distinguished in *O. bimaculatus* as in the preparatory phase, Ganga river had high GSI but in Prespawning and spawning phase, Narmada River butter catfish had a high level. Similarly, in the preparatory phase, the lowest GSI was in the sample of Tapti River and Godavari River in the pre-spawning phase and Mahanadi River in the spawning phase (Mishra et al., 2016). Protein concentrations vary in Mahanadi and Godavari as of GSI. The fecundity was estimated highest in the Narmada river (Mishra et al., 2016). The gonadosomatic index showed a linear Pearson's correlation relationship with ovarian protein concentration in the different reproductive phases of *O. bimaculatus.* The ovarian protein level has a linear correlation with fecundity and GSI. Though it varied considerably in the different rivers in different reproductive phases. The histological sections of the ovary represented different stages of variation in different habitat fish (Mishra et al., 2018). Oocyte diameter does not show any type of correlation with ovarian protein but healthy oocytes reflect its internal and external environment support that is key to the successful breeding and survival of embryo (Mishra & Rawat, 2018; El-Sayed et al., 2003).

ANNUAL GONADAL CYCLE

The gonadosomatic index (GSI) is an important sign of reproductive activity for the gonadal maturation determination and helps in identifying the time of the season of breeding as the gonads swiftly increase in size and weight just before spawning (Mishra et al., 2016). The different aspects of the reproductive biology of *O. bimaculatus* are studied by scientists from time to time (Banik et al., 2012; Rao & Karamchandani, 1986).

A. Ovarian Cycle

The gonadal development stages were studied based on gonad and oocyte size variation, macro-and microscopic study of the whole oocyte, gonadal somatic index (GSI), and histology, which are different in different months as per change in photoperiod and temperature. The GSI becomes most reliable with other indicators such as gonadal macroscopic and microscopic observations and oocyte diameter to find out the reproductive cycle (Lowe-McConnell, 1994; Rimmer & Merrick, 1983). Oogenesis is a dynamic process in the ovaries in which the oocyte passes through three different developmental stages, which include primary multiplication, secondary growth, and the third maturation phase (Coward & Bromage, 1998; Cek et al., 2001). The primary phase is categorized by the previtellogenic oocyte, the secondary by the vitellogenic oocyte, and the maturation phase by the advanced, big vitellogenic oocyte undergoing germinal vesicle migration (Estay et al.,1998). Most Teleostean species are annual breeders. Teleost reproductive cycles are classified into four stages: preparatory, pre-spawning, spawning, and post-spawning (Sundararaj & Vasal, 1976). Adult female *Ompok bimaculatus* of the northern region exhibit a distinct annual ovarian cycle with five distinct reproductive phases. They are classified as resting (for the months of November, December, and January), preparatory (for the months of February, March, and April), pre-spawning (for the months of May, June, and July), spawning (for the month of August), and post-spawning (for the months of September and October) (Mishra & Rawat, 2018). These gonadal changes coincided with environmental clues. An increase in photoperiod and temperature promotes gonadal maturation and so the breeding response (Garg & Jain, 1984; Sundararaj & Vasal, 1976).

The resting-phase ovary of *O. bimaculatus* has the majority of oogonia stages. The ovary has increased the chromatin nucleolus developmental stage in order to transition from the early peri-nucleolus oocyte stage to the preparatory phase. In the preparatory phase, ovaries have perinucleolar oocytes with large nuclei and many different-sized nucleoli. The nucleoli have a significant role in vitellogenesis (Malhotra, 1963). The perinucleolar stage of the oocyte is further transferred to the yolk vesicle stage. The pre-spawning phase showed the growth of oocytes due to yolk deposition in vesicles. Yok vesicles or cortical alveoli are the characteristic features of vitellogenic oocytes (Guraya, 1993). Endogenously synthesized lipids, glycoproteins, and exogenously synthesized yolk protein were found in yolk vesicles (Guraya, 1986; Selman & Wallace, 1986). The spawning period was the month of August for *O. bimaculatus* In this period, the ovary was full of mature vitellogenic follicles with maximum weight. The GSI exhibited an increasing trend from February to August (spawning phase), which reflected a large quantity of yolk accumulation in mature ova (Hoque & Hossain, 1993). The oocytes' diameter was significantly increased. The germinal vesicle migration and breakdown were the important events. Yolk globules coalesce to form a translucent yolk mass. The cytoplasmic content was diluted due to hydration and thus appeared as the translucent and maximum size of oocytes (Foucher & Beamish, 1980). This fish shows a small spawning phase different from its sister fish, (Siddiqua et al., 2000; Chakrabarty et al., 2008). At the end of the spawning phase, the ovary decreases in weight due to ovulation and degeneration of left-out oocytes, which is referred to as atretic during the month of September. This post-spawning phase is supported by low GSI and oocyte diameter. Similar observations were reported in different fish species (Chakraborty et al., 2007; Lehri, 1968). Further, GSI drastically decreases in the post-spawning and resting phase from September onwards. The oocyte diameter also registered a sharp fall from September to October due to atretic follicles followed by the generation of new oogonia. The lowest value of GSI denoted intense spawning activity (Adamassu, 1996).

During the pre-spawning period of female *Ompok bimaculatus* (May to July), after the growth phase, follicles accumulate yolk to form post-vitellogenic follicles represent the best stage for conservation and propagation techniques with external gonadotropin induction (Mishra et al., 2016).

B. Testis Cycle

The annual testis cycle study helps in understanding reproductive physiology, standardizing captive breeding, artificial fertilization, and also sperm preservation. In teleosts, two types of spermatogenesis occur cystic and semi-cystic types. Spermatogenesis is completed within the cysts (cystic type) or spermatogenesis occurs partially outside the cysts (semi-cystic) (Mattei et al., 1993). In *Ompok bimaculatus,* spermatogenesis is of the cystic type. The tubular testis has different compartments where the complete cycle of gamete development takes place. The spermatogenic cells (SCs) form spermatogonia, spermatocytes after meiotic division. After mitotic division, spermatids and spermatozoa form. Different gamete stages can be grouped by size. Spermatogonia are large cells with a vesicular nucleus, whereas spermatocytes are smaller and further decreased as spermatids with little cytoplasm. The spermatozoa are the smallest and most mature male gametes. The interstitial vascular connective tissue of the testis contains Leydig cells, macrophages, and mast cells (Santos et al, 2001).

The testis annual cycle is divided into five phases based on the spermatogenic activity of testicular lobules and variations in GSI values: resting, preparatory, pre-spawning, spawning, and post-spawning, which coincides with the ovarian reproductive cycle in month distribution as well (Mishra & Rawat, 2017a). The preparatory phase is distinguished by maximum spermatogonial cells. The early growth phase is characterized by an increased transformation of spermatogonia into primary and secondary spermatocytes, a few spermatids, and spermatozoa. The number of spermatocytes increased to its maximum in the pre-spawning phase. With the progression of the pre-spawning phase, spermatocytes develop in spermatids and spermatozoa in testicular lobules (Dziewulska & Domagala, 2003). The spawning phase had a maximum number of spermatozoa and spermatids. The rainy season provides favorable environmental conditions to stimulate spermiation, which is completed in a very short time. The testes finally enter into post-spawning sexual quiescence with residual spermatozoa and a few cysts of spermatids along with a few dormant spermatogonial cells (Ahmed et al., 2013; Mishra & Rawat, 2017a).

Most of the fish spawn during July in wild conditions, but for *Ompok bimaculatus* of the Gomti river, it was August. The August highest testis GSI coincided with the ovarian GSI of *O. bimaculatus* This suggested natural adjustment to reduce interspecies competition for breeding grounds and, later on, for the survival of young ones. The monthly study regarding morphological, GSI, and anatomical changes of the gonads (testis and ovary) of *Ompok bimaculatus* from the Gomati River represents clear support for the annual reproductive cycle. Although microscopic studies record the presence of more than one stage gamete in the testis at one time. This reflects long-duration spermiation in male *O. bimaculatus* as a cyclic dynamic process with an annual reproductive cycle (Mishra et al., 2016, 2018; Mishra & Rawat, 2017a, 2018).

VI. BREEDING PRACTICE

Based on GSI, gonadal morphology, anatomy, gamete stage study (Fig. 4), etc., *O. bimaculatus* behaved as an annual breeder (Basavaraja et al., 2013; Mishra & Rawat, 2017a,b; Mishra & Rawat, 2018; Malla & Banik, 2015). In natural conditions, fish breed during their spawning phase only. To ensure a regular supply of seed, a healthy brooder may be induced by gonadotropin hormones for spawning before their breeding period (Surnar et al., 2015). Captive breeding is an essential requirement for mass seed production, culture, and management to conserve fish populations (Banik & Malla, 2014; Khan & Mollah, 1998; Sarkar et al., 2006). Catfish, like major carp fish, are incapable of reproducing in captivity, even after steroid hormone injection (Kiran et al., 2013). The earlier pituitary extract was utilized to induce fish for breeding in captivity (Zonneveld et al., 1988). The rising cost of donor pituitary, restrictions on the best donor of the same season, and the time-consuming process that leads to the use of other alternative synthetic hormones Different steroids, hormones, and their combinations were used to induce the breeding of fish in captivity. Nowadays, synthetic hormones and their combinations are available on the market to make fish breed in captivity, viz., human chorionic gonadotropin (hCG), luteinizing hormone-releasing hormone, ovatide, ovaprim (Goos et al., 1987; Haniffa etal., 1996; Manickam & Joy, 1989; Mishra & Rawat 2017b; Sarkar et al., 2005), etc. were used for induced breeding and have given good results in the breeding performance of fish.

The synthetic hormone combination includes domperidone and gonadotropin that facilitate oocyte maturation and smooth ovulation/spermiation. Some scientists compared the efficiency of the synthetic hormone in breeding performance and success. All these hormones were able to induce the fish to breed with the differences in latency period, spawning efficiency, fertilization rate, and hatchability along with a survival rate among offspring. These are critical parameters in determining the success of the synthetic. Induced breeding of freshwater catfish, *O. bimaculatus* was done and discussed thoroughly (Chaudhuri, 1962; Mishra & Rawat, 2017b; Raizada et al, 2013).

In induced breeding, male fish induction with synthetic hormones not only helps in synchronizing gamete maturation but also supports an increase in fertilization and the hatching rate. Scientists tried to breed catfish in captivity naturally after the application of spawning inducers like major carp, but major success was achieved by induced breeding with the stripping method (Mishra & Rawat, 2017b; Purkayastha et al., 2012; Rahman et al., 2013; Raizada et al., 2013; Sarkar et al., 2005). Hormone dose is species-specific and sometimes individuals may behave differently. Therefore, standardization of the dose for the individual fish set is always recommended (Shinkafi & Ilesanmi, 2014; Mishra et al., 2016; Sarkar et al., 2017). It supports the release of required steroids to promote oocyte maturation (MIS) and ovulation (progesterone and corticosterone) within the female body to help smooth and complete spawning. The higher hormone level in fully mature fish caused severe stress, plugging, and made them unable to spawn (Sahoo et al., 2008). Females who received a suboptimal dose of inducing hormone (insufficient lower hormone dose) were unable to ovulate (Fernandez-Palacios et al., 2014; Raizada et al., 2013). Later, after 24 h of fertilization, the hatching occurred. The hatching rate may also be varied as per the response of inducing hormones.

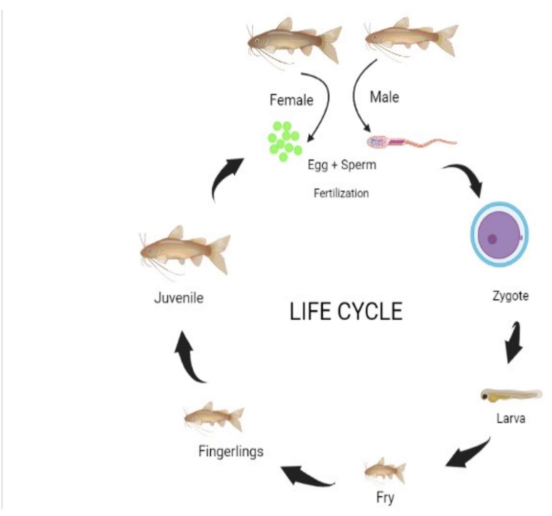

Fig 2. Stages of life cycle of *O. bimaculatus.*

VII. EMBRYONIC DEVELOPMENT

The embryonic development pattern of *O. bimaculatus* from fertilised ovum to fry was similar to all inducing agents. The mild difference in duration of organogenesis may be due to the difference in the local environment and variation in brooder stock (Chakrabarty et al., 2008; Chaudhuri, 1962; Mishra & Rawat, 2017b; Rahman et al., 2004; Raizada et al., 2013). The fertilized eggs were clear, light brown, whereas unfertilized eggs were creamy white and died shortly.

In the developmental stages, the blastodisc formation and cleavage are followed by the morula, blastula stage is completed within an early 2 hr after fertilization. After this, blastoderm cells increased rapidly in number and spread over the yolk. Later, the thick portion of the embryo was formed at one end, which is known as the cephalic region, and another thin portion was called the tail region. The hatching took place in general after 23 h of fertilisation due to increased internal pressure, tail movement, etc. The newly hatched larva was slender, transparent, with a large yolk liquefied on the anterior side. Hatch-out larvae have a distinct cephalic region with the brain, a trunk with a visible pumping heart and alimentary canal, and a long clear tail that helps in movement. Second-day larvae had a clear liquefied yolk with pigmentation in the cephalic and trunk regions. Larvae had big eyes and stumpy barbels. Larvae move with the tail only. Third-day larvae had a little yolk, an operculum with gills, and distinguished barbels. Pigmentation affected the whole body. Now, the mouth opening and fin were visible. After fertilization, the developing embryo converts into a hatch-out within 24 hrs, followed by a fry stage approximately three days later. Fry was a voracious eater and an active, fast swimmer (Basavaraja et al., 2013; Mishra & Rawat, 2017b). At this stage, their survival depends on the availability of suitable food and hygienic water parameters. (Fig. 3)

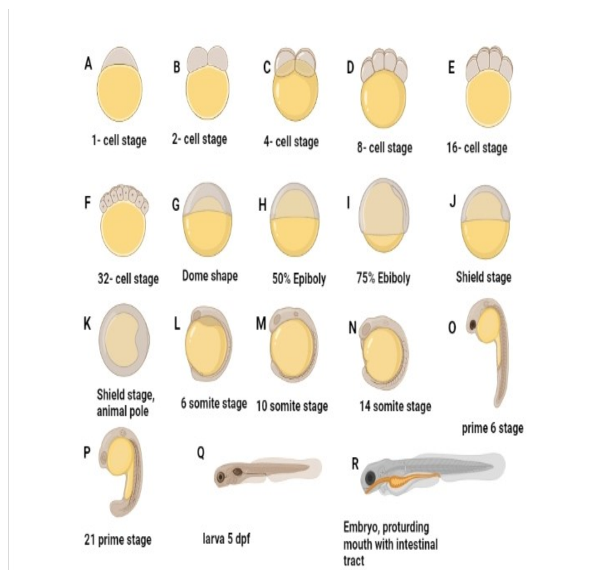

Fig 3. Embryonic development of *O. bimaculatus.*

To date, there have been no reported studies detailing the life span of butter catfish (*O. bimaculatus)* in the wild. However, it is speculated that butter catfish (*O. bimaculatus)* are primarily an annual species, based upon the size distribution of specimens collected in monthly samples over the course of a year and assessment of their reproductive strategy (spawning continuously upon maturity). However, these data are limited, and refer to only a single spawning season. Further collection of age-class data for wild populations will facilitate comparisons of the physiological ages of wild and caught animals, which will be useful for the design of biomedical studies, particularly those that involve aging.

VIII. DIET AND GROWTH

Information on the dietary preferences of wild butter catfish (*O. bimaculatus)* is of relevance to its conservation as well as fishery management programmes because it may aid in the design of diets and feeding protocols that better reflect species-specific digestive physiology and feeding behaviour than are currently used in captivity. There is some data available regarding the dietary habits of butter catfish (*O. bimaculatus)* in nature. The larvae of *O.bimaculatus* feed on zooplankton up to day 15. In the fry stages and onwards the fish can be fed with mixed food (rice polish, silkworm pupae and boiled**,** egg) at 100 % of the body weight of the population (Chakrabarty et al., 2008). They consume crustacean adults, crustacean larvae, and insects as their main food. The zooplankton, nektons, tadpoles, annelid worms, and mollusc are also observed as food items. The food substances observed in the gut, like vegetable matter, fish scales, crustaceans, mollusc, plant parts, and mud, are distributed throughout the different layers of the water bodies. Hence, it can be interpreted that both species of *O. bimaculatus* and *O. malabaricus* cannot be labeled as surface or bottom feeders. Both vegetable matter and fish scales as the observed food matters enable them to be identified as omnivorous. Butter catfish (*O. bimaculatus)* are generalists (euryphagous, carnivorous, omnivorous), consuming a wide variety of benthic and planktoni-vorus crustaceans in addition to worms and insect larvae (Saigal et al., 2006). Such gut content analysis was the microhabitat preferences of the species, as ponds and stream margins, where the butter catfish (*O. bimaculatus)* were most often found, were also areas where terrestrial insects were more likely to fall into the water. While these data are indeed suggestive, additional studies conducted during periods of high rainfall are necessary to gain a more complete understanding of the dietary habits of this species in nature.

Growth is a quantitative trait, and it may be measured at different times of the life cycle of butter catfish, *O. bimaculatus*. However, no literature is available about the life cycle of butter catfish. Many aspects are untouchable such as growth compensation or size-hierarchy effect (Brown, 1957; Ricker, 1958).

The growth rate of ectothermic teleost fishes is influenced by various environmental stimuli such as water current, the change in water temperature, photoperiod, trophic niche etc. which along with the internal state of the animal is processed, integrated, and responded through hormonally mediated pathways, to trigger developmental processes such as hatching, metamorphosis, (flatfishes, eels) or smoltification (salmonids), sexual maturation and spawning. These factors either directly or indirectly affect the growth of fish. The increasing rate of weight has a higher RNA:DNA ratio in fish such reports are known in intertidal fish *Scartichthys viridis*. Quantitative trait such as growth is normally under the control of polygenes, which each has a small effect on the phenotype, as well as environmental influence and a possible influence of major genes and or QTL (Falconer & Mackay, 1996). Further research would be required in butter catfish *O.bimaculatus* to compare their length-weight relationship spatially as well as to compare their RNA:DNA ratio for the justification of growth rate.

IX. LENGTH-WEIGHT-DENSITY

The length-weight of *Ompok bimaculatus* was studied by Shivakami in 1987. The length and weight range of specimens were 211-335mm, 38-196g respectively. She also reports the value slope (b) is 2.778. However in fish base, the maximum length of *O. bimaculatus* was reported 450mm (Fish base) while the maximum length (457mm) was reported by Ghosh & Lipton, (1982) from the northeast region of India, The available maximum length in the fish base show a conflicting situation over the fisheries sector on *Ompok bimaculatus*. The length-weight relationship would be used to compare stock discrimination, stock delineation as well as stock identification approaches. More study is also required for the length-weight relationship based on ecological habitats for determining the suitable habitat to increase the fishery management of the species.

The relative abundance of *O.bimaculatus* was highest in River Gomti (27.09%), followed by Ganga (24.30%), Ramganga (11.49%), Ghaghara (13.30%), Sharda (7.88%), Dhasan (2.13%), Betwa (6.90%), Subernlekha(2.96%), Yamuna(2.63%), Son (1.31%) Fig. 4 (Sarkar et al., 2012).

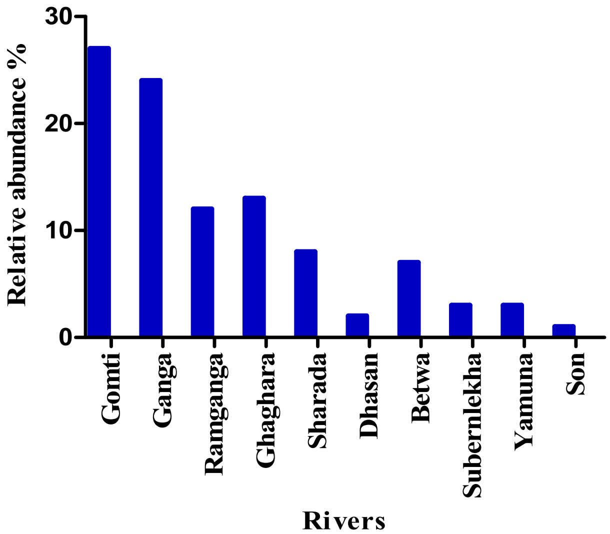

Fig 4. The relative abundance of *O. bimaculatus* from the different rivers.

The densities at which fish are held in culture exert a profound impact on their health, productivity, and welfare (Ellis et al., 2002). In general, exposing fish to crowded conditions results in decreases in growth rate (Procarione et al., 1999) and immune function (Suomalainen et al., 2005; Wedemeyer, 1996). The inverse relationship between these factors and rearing density has been linked to stress, indicated by the fact that the stress hormone cortisol is often elevated in the plasma of fish held in crowded conditions (Barton & Iwama, 1991; Vijayan & Leatherland, 1990). What ultimately evokes this stress response at elevated densities is unclear; the two most commonly proposed factors are an increase in aggressive behaviors or crowding itself (Ellis et al., 2002). Crowding also causes aggressive behavior in fish. Crowding conditions also lead to elevated cortisol levels in many species, including sea bream (*Sparus aurata*), common carp,(*Cyprinus carpio*), and rainbow trout (*Oncorhynchus mykiss*) (Pickering & Pottinger, 1987; Ruane et al., 2002; Tort et al., 1996).

butter catfish *O. bimaculatus* also appear sensitive to crowding, suggesting that some aspect of crowding itself elicited the negative effects. Reduced performance and impaired health of fish at elevated densities may also be caused by increased competition for resources and a decrease in water quality (Ellis et al., 2002). Despite all these, there has been no work about stress and its impact on survival, the endocrine response against reproduction, and other associated physiological processes.

The availability of butter catfish under biodiversity was categorized by the different workers to compare their abundance. However, in the other areas, there is a paucity of knowledge, such as taxonomy, food, length-weight, reproduction, germplasm, aquaculture, chromosomes, and cytogenetics. Now there is a need to fill up such a paucity in the areas of reproduction to compare their conservation across different parts of India for the welfare of the species.

X. CONCLUSION

O. bimaculatus registered species variation not only due to genetic variation but also due to geographical distribution. Therefore while selecting brooders for breeding or culture practice one should prefer the local best strain as per fecundity and gamete quality (Fig 5). But for the gene bank, the best strain of the different freshwater bodies should be conserved. The distant varied species may be used for breeding to get the best-fitted crop of *O. bimaculatus*.

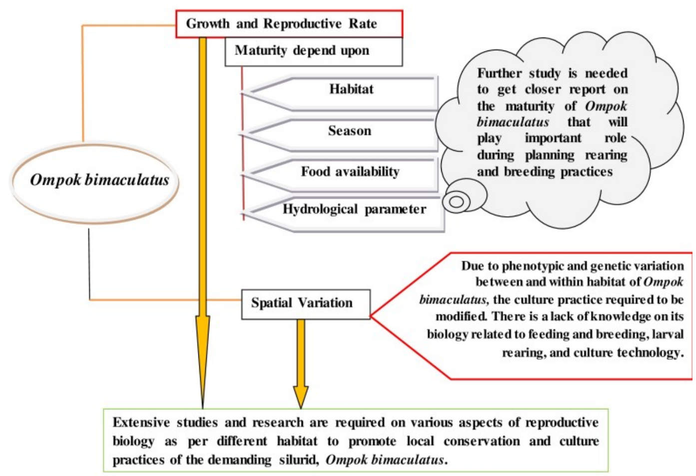

Fig 5: *O. bimaculatus* : Concern area to promote conservation and culture practices.

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