

# Karyomorphology of two major carps, *Catla catla* and *Labeo rohita*

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## Abstract

A detailed study of chromosomes of two species of Indian inland fish revealed the chromosomal number and type. It describes the relation between the two species. Their peculiar morphological characters have been recorded. These karyological observations provide strong evidence to conclude that the two species are closely related and have phylogenetic links.

## Introduction

Cytogenetics is the study of chromosome morphology and the behaviour of chromosomes during mitosis and meiosis. The chromosome number and its morphology is specific for a particular species. Fishes are of particular interest to Ichthyologists as well as cytogenetists as they occupy a very important position in the systematic differentiation of vertebrates. The benefits of this Karyological study among fishes are great values as fishes are economically important.

Karyological methods of ascertaining the taxonomic position of different species of fish have been in wide use in recent years among Russian and other ichthyologists (E. A. Salmenkova et al, 2005). The available data in fishes show that almost all forms of chromosomal rearrangements have played a role in the evolution of the fish karyotypes (E.D.Vasil'eva 2011).

## Materials and Methods

### Experimental species

Four different species of freshwater fish namely *Catla catla* Hamilton (*Catla*); *Labeo rohita* (Hamilton-Buchanan (*Rohu*);) were selected for the present study.

### Maintenance in the laboratory:

The live animals were collected from Poondi fresh water aquaculture station and they were transported to the laboratory in oxygenated polythene bags and maintained in fiber glass tanks containing enough amount of water. The water was changed once a day and the fishes were given food ad libitum. The fishes were acclimatized for about one week before the experiments were conducted. Feeding was stopped two days prior to the experiment. On the third day, the experimental fishes were injected with colchicine and introduced into the tank.

### Chemicals required

- Colchicine 0.01% (10mg colchicine was dissolved in 100ml of dis. water).
- Potassium chloride:0.4%, 400mg KCl was dissolved in 100ml of dis. water.
- Sodium citrate:0.9% 900mg of sodium citrate was dissolved in 100ml of dis. water.
- Carnoy's fixative: 3:1 ratio of methanol and Glacial acetic acid

### Giemsa Stain

Prepared by dissolving 2ml of stock Giemsa solution and 2ml of 10% disodium hydrogen phosphate to 4.6ml of distilled water (PH 6.8).

### Karyotyping

Procedure developed by Kligerman 1982 was followed with minor alterations.

Since blood samples could not be obtained in smaller fishes, chromosome preparation from the gill tissues were used for

chromosome preparation.

## Staining

The air dried slides were then stained with freshly prepared Giemsa staining solution (4%) for 15-18mts. The slides were then destained with distilled water and air dried. The slides were then screened for chromosomal spreads under the light microscope.

## Chromosomal complement in *Catla catla*:

The total diploid number was found to be 50(2n=50). This was confirmed by observing 157 metaphase plates which showed the diploid number 50.

Based on the idogram individual karyomorphology of the diploid set was analysed and the chromosome length was measured. The length ranges from  $8.5 \times 10^{-3} \mu$  to  $3.0 \times 10^{-3} \mu$  of the largest to the smallest chromosomes. In the diploid set four pairs are metacentric (4,9,15 and 20), Thirteen pairs are submetacentric (1,2,3,5,6,8,10,11,12,14,16,19 and 21) and remaining eight pairs are acrocentric (7,13,17,18,22,23,24 and 25). Relative length percent (RL%) ranges from 5.64 to 1.99. Nucleolar organiser region (NOR) and heterochromatin region (HCR) were also observed. (Fig 1, Table 1 and Plate 1 & 2).



Fig. 1: –  
An Idiogram of *Catla catla*.

Pair-wise						
Pair No	p(μ)	q(μ)	TL(μ)	RL%	Ic	TYPE OF CHROMOSOME
1	$2.2 \times 10^{-3}$	$6.0 \times 10^{-3}$	$8.3 \times 10^{-3}$	5.64	0.24	Submetacentric
2	$1.5 \times 10^{-3}$	$6.0 \times 10^{-3}$	$8.0 \times 10^{-3}$	5.31	0.19	Submetacentric
3	$2.0 \times 10^{-3}$	$4.5 \times 10^{-3}$	$7.0 \times 10^{-3}$	4.65	0.29	Submetacentric
4	$3.0 \times 10^{-3}$	$3.5 \times 10^{-3}$	$7.0 \times 10^{-3}$	4.65	0.43	Metacentric
5	$2.5 \times 10^{-3}$	$4.5 \times 10^{-3}$	$7.0 \times 10^{-3}$	4.65	0.36	Submetacentric
6	$1.5 \times 10^{-3}$	$4.75 \times 10^{-3}$	$6.75 \times 10^{-3}$	4.48	0.22	Submetacentric
7	$0.5 \times 10^{-3}$	$5.5 \times 10^{-3}$	$6.5 \times 10^{-3}$	4.31	0.08	Acrocentric
8	$2.0 \times 10^{-3}$	$4.0 \times 10^{-3}$	$6.5 \times 10^{-3}$	4.31	0.31	Submetacentric
9	$3.0 \times 10^{-3}$	$3.0 \times 10^{-3}$	$6.5 \times 10^{-3}$	4.31	0.46	Metacentric
10	$2.0 \times 10^{-3}$	$4.0 \times 10^{-3}$	$6.5 \times 10^{-3}$	4.31	0.31	Submetacentric
11	$1.75 \times 10^{-3}$	$4.3 \times 10^{-3}$	$6.3 \times 10^{-3}$	4.22	0.28	Submetacentric
12	$1.5 \times 10^{-3}$	$4.25 \times 10^{-3}$	$6.25 \times 10^{-3}$	4.15	0.24	Submetacentric
13	$0.75 \times 10^{-3}$	$5.0 \times 10^{-3}$	$6.25 \times 10^{-3}$	4.125	0.12	Acrocentric
14	$1.75 \times 10^{-3}$	$4.0 \times 10^{-3}$	$6.25 \times 10^{-3}$	4.15	0.28	Submetacentric
15	$2.75 \times 10^{-3}$	$2.75 \times 10^{-3}$	$6.0 \times 10^{-3}$	3.98	0.46	Metacentric
16	$2.0 \times 10^{-3}$	$3.5 \times 10^{-3}$	$6.0 \times 10^{-3}$	3.98	0.33	Submetacentric
17	$0.5 \times 10^{-3}$	$4.8 \times 10^{-3}$	$5.8 \times 10^{-3}$	3.85	0.09	Acrocentric
18	$1.0 \times 10^{-3}$	$4.25 \times 10^{-3}$	$5.75 \times 10^{-3}$	3.82	0.17	Acrocentric
19	$1.75 \times 10^{-3}$	$3.5 \times 10^{-3}$	$5.75 \times 10^{-3}$	3.82	0.30	Submetacentric
20	$2.5 \times 10^{-3}$	$2.5 \times 10^{-3}$	$5.5 \times 10^{-3}$	3.65	0.45	Metacentric
21	$2.0 \times 10^{-3}$	$3.0 \times 10^{-3}$	$5.5 \times 10^{-3}$	3.65	0.36	Submetacentric
22	$1.0 \times 10^{-3}$	$4.0 \times 10^{-3}$	$5.5 \times 10^{-3}$	3.65	0.38	Acrocentric
23	$0.5 \times 10^{-3}$	$2.5 \times 10^{-3}$	$3.5 \times 10^{-3}$	2.32	0.14	Acrocentric
24	$0.5 \times 10^{-3}$	$2.0 \times 10^{-3}$	$3.0 \times 10^{-3}$	1.99	0.17	Acrocentric
25	$0.5 \times 10^{-3}$	$2.0 \times 10^{-3}$	$3.0 \times 10^{-3}$	1.99	0.17	Acrocentric

p - Short arm  
q - Long arm  
TL - Total length  
RL% - Relative length percent  
Ic - Centromeric index

Table 1:  
Measurement of chromosomal complement of *Catla catla*.



Plate 1:

*Catla catla*.

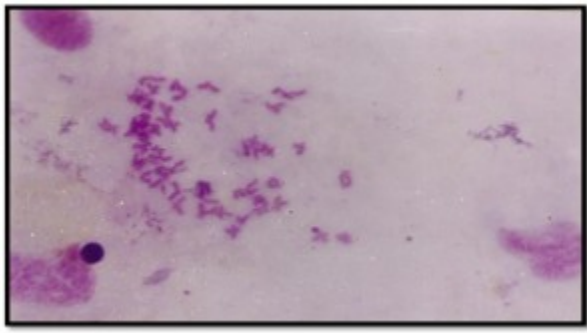


Plate 2:

Metaphase plate showing chromosomal complement  $2n = 50$  of *Catla catla*.

**Chromosomal complement in *Labeo rohita***

The diploid number was found to be  $50(2n=50)$ . This was confirmed by observing 133 metaphase plates which showed the diploid number 50.

The chromosomes are condensed in nature and darkly stained with distinct karyomorphology. Based on the idogram individual karyomorphology of the diploid set was analysed and the chromosome length was measured. The length ranges from  $6.5 \times 10^{-3} \mu$  to  $2.2 \times 10^{-3} \mu$  of the largest to the smallest chromosomes. In the diploid set eleven pairs are metacentric (2,3,4,5,10,12,14,15,20,23 and 24), Remaining fourteen pairs are submetacentric (1,6,7,8,9,11,13,16,17,18,19,21,22 and 25) and remaining eight pairs are acrocentric (7,13,17,18,22,23,24 and 25). Relative length percent (RL%) ranges from 6.43 to 2.18. Nucleolar organiser region (NOR) was found in pair one and heterochromatin region (HCR) were observed in all the other pairs. (Fig 2, Table 2 and Plate 3 & 4).

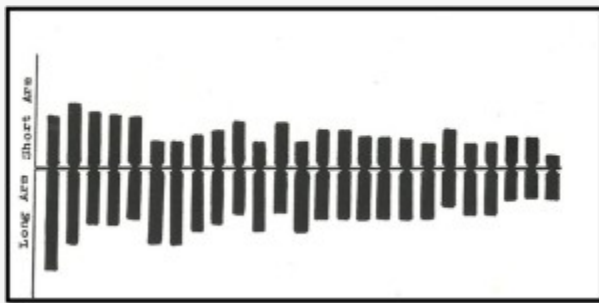


Fig. 2: –

An Idiogram of *Labeo rohita*.

Pair No	Pair mean		TL(μ)	RL%	lr	TYPE OF CHROMOSOME
	p(μ)	q(μ)				
1	$2.0 \times 10^{-3}$	$4.0 \times 10^{-3}$	$6.5 \times 10^{-3}$	6.43	0.31	Submetacentric
2	$2.5 \times 10^{-3}$	$3.0 \times 10^{-3}$	$6.0 \times 10^{-3}$	5.93	0.42	Metacentric
3	$2.2 \times 10^{-3}$	$2.2 \times 10^{-3}$	$4.9 \times 10^{-3}$	4.85	0.45	Metacentric
4	$2.0 \times 10^{-3}$	$2.25 \times 10^{-3}$	$4.75 \times 10^{-3}$	4.70	0.42	Metacentric
5	$2.0 \times 10^{-3}$	$2.0 \times 10^{-3}$	$4.5 \times 10^{-3}$	4.45	0.44	Metacentric
6	$1.0 \times 10^{-3}$	$3.0 \times 10^{-3}$	$4.5 \times 10^{-3}$	4.45	0.22	Submetacentric
7	$1.0 \times 10^{-3}$	$3.0 \times 10^{-3}$	$4.0 \times 10^{-3}$	4.45	0.22	Submetacentric
8	$1.25 \times 10^{-3}$	$2.5 \times 10^{-3}$	$4.25 \times 10^{-3}$	4.20	0.29	Submetacentric
9	$1.5 \times 10^{-3}$	$2.2 \times 10^{-3}$	$4.2 \times 10^{-3}$	4.15	0.36	Submetacentric
10	$1.8 \times 10^{-3}$	$1.8 \times 10^{-3}$	$4.1 \times 10^{-3}$	4.06	0.44	Metacentric
11	$1.0 \times 10^{-3}$	$2.5 \times 10^{-3}$	$4.0 \times 10^{-3}$	3.96	0.25	Submetacentric
12	$1.75 \times 10^{-3}$	$1.75 \times 10^{-3}$	$4.0 \times 10^{-3}$	3.96	0.44	Metacentric
13	$1.0 \times 10^{-3}$	$2.5 \times 10^{-3}$	$4.0 \times 10^{-3}$	3.96	0.25	Submetacentric
14	$1.5 \times 10^{-3}$	$2.0 \times 10^{-3}$	$4.0 \times 10^{-3}$	3.96	0.38	Metacentric
15	$1.5 \times 10^{-3}$	$2.0 \times 10^{-3}$	$4.0 \times 10^{-3}$	3.96	0.38	Metacentric
16	$1.25 \times 10^{-3}$	$2.0 \times 10^{-3}$	$3.75 \times 10^{-3}$	3.71	0.38	Submetacentric
17	$1.2 \times 10^{-3}$	$2.0 \times 10^{-3}$	$3.7 \times 10^{-3}$	3.66	0.23	Submetacentric
18	$1.2 \times 10^{-3}$	$2.0 \times 10^{-3}$	$3.7 \times 10^{-3}$	3.66	0.32	Submetacentric
19	$1.0 \times 10^{-3}$	$2.0 \times 10^{-3}$	$3.5 \times 10^{-3}$	3.40	0.29	Submetacentric
20	$1.0 \times 10^{-3}$	$2.0 \times 10^{-3}$	$3.5 \times 10^{-3}$	3.40	0.43	Metacentric
21	$1.0 \times 10^{-3}$	$1.85 \times 10^{-3}$	$3.35 \times 10^{-3}$	3.31	0.30	Submetacentric
22	$1.0 \times 10^{-3}$	$1.8 \times 10^{-3}$	$3.3 \times 10^{-3}$	3.26	0.30	Submetacentric

Table 2:

Measurement of chromosomal complement of *Labeo rohita*.

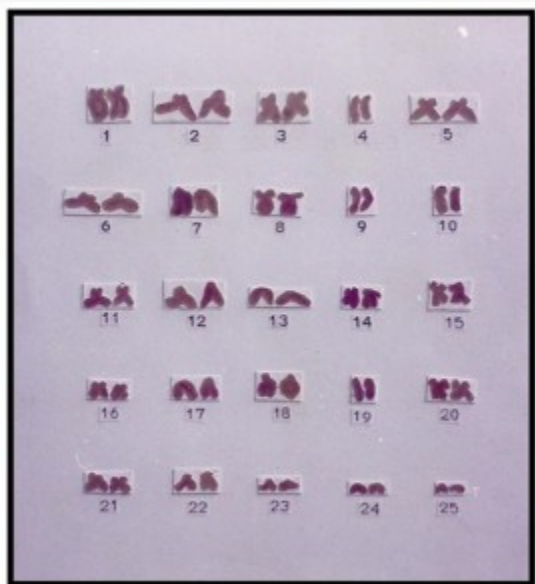


Plate 3:

Karyotype of *Catla catla*.

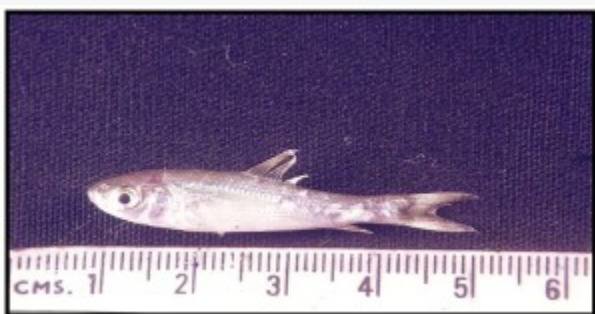


Plate 4:

*Labeo rohita*

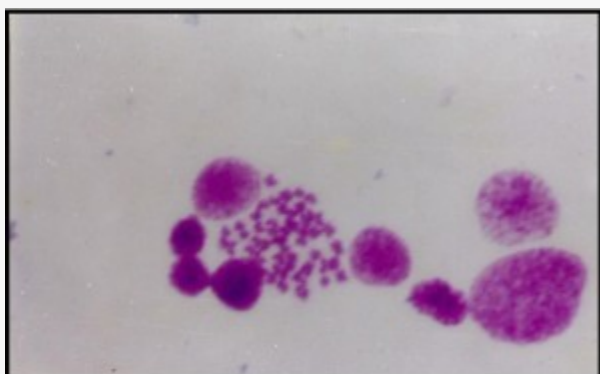


Plate 5:

Metaphase plate showing chromosomal complement  $2n = 50$  of *Labeo rohita*.

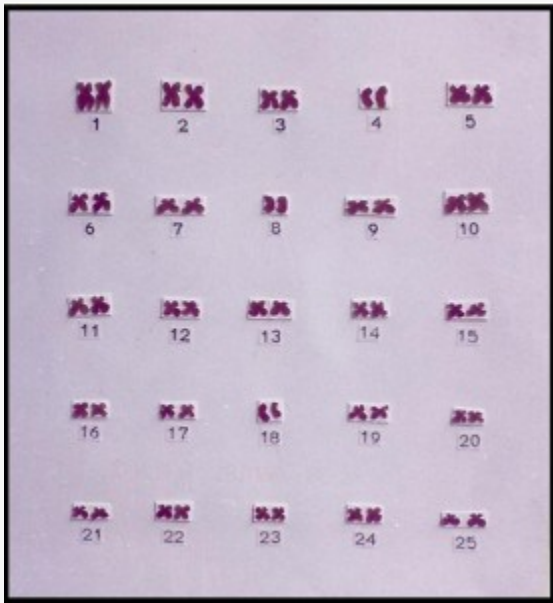


Plate 6:

Karyotype of *Labeo rohita*

## Discussion

Diversity is a land mark of evolution. Each species is characterized by a specific chromosome complement commonly referred to as karyotype. Karyodiversity is mainly due to the variation in the position of centromere. Centromere is the special region of chromosome which gets firmly attached to mitotic spindle at the time of metaphase during cell cycle. These are usually observed during the early stage of cell division as non-staining gaps. The chromatids, both attached to the kinetochore part of centromere and to the spindle during metaphase which can be observed by the influence of a drug, colchicines. Besides the centromere, non-stained gaps in the form of secondary constriction is also a common feature of chromosomes.

During the early and late metaphase, the chromosomes reach maximum degree of condensation and contraction, to take part in the on-going chromosomal division. Hence, metaphase holds the secret of cell cycle and expresses the full complement of chromosomes in the somatic cells of a species is referred to as somatic number and designated as  $2n$ . In the present study gill cells, conventional karyological technique of Kligerman and Bloom (1977) was adopted with due modification to suit the experimental fish. In the present study the diploid complement  $2n$  was 50 in both the species of carps.

The number of chromosomes per cell is rather a conservation characteristic and may be used as an indicator of closeness of species, within families. The number and position of the arms of the chromosomes is even more conservative than chromosome number and is often equally useful in taxonomic studies.

## Conclusion

In conclusion it is emphasized that the order Cypriniformes include fish groups which is not much variable from karyological point of view. But to analyse in detail the chromosome evolution processes and taxonomical relationship, it is necessary to collect more cytological banding methods.

## References

1. Abhay Singh Yadav, Anita Bhatnaga, Manjeet Kaur. 2013. Aberrations in the Chromosomes of *Cirrhinus mrigala* (Hamilton)

2. An L, Liu B S, Zhao Y H and Zhang CG. 2010. *Protolabeo protolabeo*, a new genus and a new species of labeonine fishes from the southwest China (Teleostei, Cyprinidae). *Acta Zootaxonomica Sinica* 35(3).
3. Anshumala Chaturvedi, Vindhya Mohindra, Rajeev K. Singh, Kuldeep K. Lal, Peyush Punia, Ranjana Bhaskar, Anup Mandal, Lalit Narain, Lakra W S. 2010. Population genetic structure and phylogeography of cyprinid fish, *Labeo dero* (Hamilton, 1822) inferred from allozyme and microsatellite DNA marker analysis *Mol Biol Rep* (2011) 38:3513–3529, Published online: 4 December 2010\_ Springer Science+Business Media B.V. 2010.
4. Bogutskaya N G, Kucuk F and Atalay M A. 2007. A description of three new species of the genus *Pseudophoxinus* from Turkey (Teleostei: Cyprinidae: Leuciscinae). *Zoosystematica Rossica* 15(2):335-341.
5. Dilkusha, Ponniah A G. 2001. Comparative cytogenetic studies in Indian major carps *Labeo rohita*, *Catla catla* and *Cirrhinus mrigala* (Cyprinidae, Pisces). *Chromosome Sci* Vol.5; No.3; Page.153-158.
6. Eschmeyer W N and Fricke R. (eds.), 2011. Catalog of fishes. Updated internet version of 05 May 2011. Catalog databases of CAS cited in FishBase (website).
7. FAO-FIES. 2010. Aquatic Sciences and Fisheries Information System (ASFIS) species list.  
REFERENCE LINK
8. Galetti P M Jr., Aguilar C T and Molina W F. 2000. An overview of marine fish cytogenetics *Hydrobiologia* 420: 55–62, 2000
9. Gopalakrishnan A and Basheer V S. 2000. Occurrence of *Labeo rohita* and *Cirrhinus mrigala* in Meenachil, Manimala and Pampa Rivers, Kerala. In: A.G. Ponniah and A. Gopalakrishnan (eds), *Endemic Fish Diversity of Western Ghats*, pp. 167-168. NBFGR-NATP Publication. National Bureau of Fish Genetic Resources, Lucknow, U.P., India.
10. Hänfling B, Bolton P, Harley M and Carvalho GR. 2005. A molecular approach to detect hybridisation between crucian carp (*Carassius carassius*) and non indigenous carp species (*Carassius* spp. and *Cyprinus carpio*). *Freshwat. Biol.* 50:403-417.
11. Huang Y F, Chen XY and Yang JX. 2007. A new labeonine fish species, *Parasinilabeo longiventralis*, from eastern Guangxi, China (Teleostei: Cyprinidae). *Zool. Res.* 28(5):531-538.
12. Kim T. Scribner, Kevin S. Page and Meredith L. Bartron. 2001. Hybridization in freshwater fishes: a review of case studies and cytonuclear methods of biological inference *Reviews in Fish Biology and Fisheries* 10: 293–323, 2001.
13. Kottelat M. 2007. *Rasbora dies*, a new species of cyprinid fish from eastern Borneo (Teleostei: Cyprinidae). *Ichthyol. Explor. Freshwat.* 18(4):301-305.
14. Kligerman A D and Bloom S E. 1977. Rapid chromosome preparations from solid tissues of fishes. *J. Fish. Res. Board Can.* 34:266-269.
15. Kligerman A D. 1982. The use of cytogenetics to study genotoxic agents in fishes. In: *Cytogenetic assays for environmental mutagens*, (Eds: T.C. Hsu, Allenheld). Osmum and Co., N.J., pp. 161-181.
16. Kushwaha B, Nagpure NS, Srivastava, Satish K and Ponniah A G. 2002. Cytogenetic studies in two geographical stocks of *Heteropneustes fossilis* (Bloch). *Indian Journal of Animal Sciences* 72(4): 348-350.
17. Liao TT and Tan HH. 2011. *Brevibora cheeya*, a new species of cyprinid fish from Malay peninsula and Sumatra *The Raffles Bulletin of Zoology* 59(1):77-82.
18. Mezhzherin SV and Pavlenko L I. 2007. A Case of Hybridization in Loaches (Osteichthies: Cobitidae: Cobitis) Determining Genetic Instability and Expansion Cytology and Genetics, 2007, Vol. 41, No. 4, pp. 218–225. © Allerton Press, Inc., 2007 Original Russian Text © S.V. Mezhzherin, L.I. Pavlenko, 2007, published in *Tsitologiya i Genetika*, 2007, Vol. 41, No. 4, pp. 26–35.
19. Santosh Sharma, Suman Kumaria, Pramod Tandon, Rao Rama Satyawada. 2012. Comparative karyomorphological study of some Indian *Cymbidium Swartz*, 1799 (Cymbidieae, Orchidaceae) *Comparative Cytogenetics* 6 (4)(2012):453-46.
20. Satish K. Srivastava, Kushwaha B, Nagpure NS, Sanjay Pandey, Shilpi Sharma, Ravindra Kumar and Verma M S. 2005. Genotoxic studies in *Cyprinus carpio* (Linneus, 1758) of river Yamuna *Indian Journal of Fisheries* 52(2): 235-239.
21. Tripathy S K, Gaur K K and Sarangi N. 2013 Morphotypes vis-a-vis genetic parameters of *Catla catla* (Ham.) and *Labeo rohita* (Ham.) backcrosses *African Journal of Biotechnology*, vol. 12(36), pp. 5503-5512.
22. Vasil'eva, Prazdnikov E D D V, and Vasil'ev VP. 2011. First Confirmed Occurrence of Syrman Goby *Neogobius syrman* (Gobiidae, Perciformes) in Sasyk Lake of the Black Sea Basin and Karyological Characteristic of Syrman Goby and Ginger Goby *N. eurycephalus* *Journal of Ichthyology*, 2011, Vol. 51, No. 7, pp. 513–520. © Pleiades Publishing, Ltd., 2011. Vol. 51, No.

4, pp. 472–479.

23. Zhu Y, Zhang E, Zhang M and Han Y Q. 2011. *Cophecheilus bamen*, a new genus and species of labeonine fishes (Teleostei: Cyprinidae) from South China. *Zootaxa* 2881:39-50.

24. Zhang S M. 1990. Inducing polyploidy and Karyological studies in Indian major carps *Catla catla* (Ham.), *Labeo rohita* (Ham.) and *Cirrhinus mrigala* (Ham.) and in eight other freshwater species of fish. A Report. The secondment of young Scientist Programme. (April 19, 1989–April 18, 1990). CIFA (ICAR).

25. Zhang S M and Reddy P V G K. 1991. On the comparative karyomorphology of three Indian major carps, *Catla catla* (Ham.), *Labeo rohita* (Ham.) and *Cirrhinus mrigala* (Ham.) *Aquaculture*. 97:7–12.