International Journal of Zoological Investigations

Contents available at Journals Home Page: www.ijzi.net

ISSN: 2454-3055

Comparative Study on DNA Quantification of Three Indian Major Carps of Middle Ganga Region

Rana Nisha^{1*} and Jain Seema ²

¹School of Life Science and Technology, IIMT University, Meerut, India

²Department of Zoology, RGPG College, CCSU, Meerut 250001, India

+

*Corresponding Author

Received: 13th October, 2020

Accepted: 24th November, 2020

Published online: 23rd December, 2020

<https://doi.org/10.33745/ijzi.2020.v06i02.014>

Abstract: The present study was conducted on DNA extraction and determination of DNA quantity of both male and female of Indian major carps *Catla catla*, *Cirrhinus mrigala* and *Labeo rohita* collected from three different sites of western Uttar Pradesh, India. DNA was isolated and gel electrophoresis was carried out and extracted DNA was analyzed using Nanophotometer to determine the quantity of DNA and its purity level. A total of 90 samples were analyzed. The DNA content of *Catla catla* female was 62-80 ng/μl and of male was 71-84 ng/μl; of *Cirrhinus mrigala* female was 62-74 ng/μl and of male was 64-78 ng/μl and of *Labeo rohita* female was 59-68 ng/μl and of male was 66-74 ng/μl. For evaluation of interspecific variations, the DNA content variation among sex, type (species), site were analyzed using generalized linear model by the analysis of the content variation which was done using ANOVA (Univariate Analysis of Variance UNIANOVA using SPSS 20.0, Followed by Posthoc Duncan's Multiple Range Test) and showed significance at 1% level in all parameters (site, type, sex).

Keywords: DNA extraction, Indian major Carps, Nanophotometer, Interspecific variation

Citation: Rana Nisha and Jain Seema: Comparative study on DNA quantification of three Indian major carps of middle Ganga region. Intern. J. Zool. Invest. 6 (2): 343-348, 2020.

<https://doi.org/10.33745/ijzi.2020.v06i02.014>

Introduction

Fish are the most diverse group of living vertebrates, with more than approximately 32,500 species currently known and order Cypriniformes contains planets largest monophyletic group of freshwater fishes, with

over 400 genera and 3000 species native to Asia, Europe, Africa and North America (Nelson, 2006). Fish has long been valued as a source of high quality animal protein and is relatively cheap for human nutrition.

Cyprinidae is one of the largest family of freshwater fishes represented by Indian carp species ranging from millimeters in length (minnows) to more than a meter (major carps) and a well-known component of East Asian freshwater fish fauna.

Aquaculture is the fastest growing food producing sector. Global fish production peaked at about 171 million tonnes in 2016, with aquaculture representing 47 per cent of the total and 53 per cent if non-food uses (including reduction to fish meal and fish oil) are excluded. The total first sale value of fisheries and aquaculture production in 2016 was estimated at USD 362 billion, of which USD 232 billion was from aquaculture production (FAO, 2018). The major carps of India fall under three genera-- *Catla*, *Labeo* and *Cirrhinus*. Due to their fast growing nature and good taste Indian major carps are commercially important group of fishes used in aquaculture and widely captured and form the mainstay of culture opportunities. The *Cirrhinus mrigala*, also known as the mrigal or Nain, *Catla catla* locally called Catla and Bhakur, and *Labeo rohita* is locally known as rohu.

These fishes though originally inhabitants of the Ganga river network in North India and the river of Pakistan, Bangladesh, Nepal and Burma, are also transplanted into other rivers in central as well as peninsular India. Besides this, these carps have been imported by many other countries such as Thailand, Vietnam, Laos (Welcomme, 1988).

The present study was conducted on the DNA isolation and determination of DNA concentration; its purity level of both male and female of major carps using

Nanophotometer and also to find out variation in DNA content of wild and cultured fish.

Materials and Methods

In the present study, experimental fishes viz. *Catla catla*, *Labeo rohita*, *Cirrhinus mrigala* were collected from different water bodies of western Uttar Pradesh, India (wild fishes from Ganga river and cultured fishes from Culture ponds/hatchery), morphologically identified (with the help of standard literature of Day's fauna 1875-78, 1989; Jayaram, 1981) and preserved for molecular studies (for DNA isolation and quantification). A total of 90 samples were analyzed including both male and female fish of wild and cultured major carps (30 samples of each). Voucher specimens were preserved in 10% formalin. DNA isolation was done following the method of Ruzzante *et al.* (1996) with minor modifications. Gel electrophoresis was carried out on 1.2% agarose gel. DNA quantification was done using nanophotometer (P330, Implens Germany) followed by manufacturer protocols.

Results

The present study was conducted on DNA extraction and quantification of both male and female of Indian Major carps *Catla catla*, *Cirrhinus mrigala* and *Labeo rohita* collected from three different sites of western Uttar Pradesh, India.

The DNA content of *Catla catla* female was 62-80 ng/ μ l and of male was 71-84 ng/ μ l, of *Cirrhinus mrigala* female was 62- 74 ng/ μ l and of male was 64-78 ng/ μ l and of *Labeo rohita* female was 59-68 ng/ μ l and of male was 66-74 ng/ μ l (Table 1). For evaluation of interspecific variations, the DNA content variation among sex, type (species), site

Table 1: Variation in DNA content of male and female major carps

S.N.		<i>Catla catla</i>		<i>Cirrhinus mrigala</i>		<i>Labeo rohita</i>	
		Female	Male	Female	Male	Female	Male
Site1	1.	62	71	62	66	60	66
	2.	62	72	66	64	60	67
	3.	63	72	63	66	61	70
	4.	68	74	62	68	60	72
	5.	67	78	64	78	64	70
Site 2	6.	62	77	70	78	66	70
	7.	62	77	65	70	68	70
	8.	64	78	60	74	62	73
	9.	65	76	67	72	59	70
	10.	62	80	67	74	61	74
Site 3	11.	77	78	70	67	62	73
	12.	77	79	70	70	60	67
	13.	78	82	74	74	61	70
	14.	76	84	70	72	60	72
	15.	80	82	68	67	60	67

Table 2: Mean and Standard error of all observations

Factor	No. of observations	Mean $\pm$ SE
Overall	90	69.089 $\pm$ 0.415
Site		
One	30	66.600 $\pm$ 0.718
Two	30	69.100 $\pm$ 0.718
Three	30	71.567 $\pm$ 0.718
Type		
<i>Catla catla</i>	30	72.833 $\pm$ 0.718
<i>Cirrhinus mrigala</i>	30	68.600 $\pm$ 0.718
<i>Labeo rohita</i>	30	65.833 $\pm$ 0.718
Sex		
Female	45	65.489 $\pm$ 0.586
Male	45	72.689 $\pm$ 0.586

measured were analyzed using generalized linear model by the analysis of the content variation which was done using ANOVA (Univariate Analysis of Variance UNIANOVA using SPSS 20.0, Followed by POST HOC: DUNCAN'S Multiple Range Test) (Table 2) and showed significance at 1% level in all parameters (site, type, sex).

The mean and standard error were calculated for all the fish species, for site of sample collection, sex, and species and subjected to ANOVA (UNIANOVA) to evaluate interspecific variations which are given in Table 2. The DNA content variation among sex, type (species), site measured were analyzed using generalized linear model by the analysis of the content variation was done using ANOVA (Univariate Analysis of Variance UNIANOVA using SPSS 20.0, Followed by Posthoc Duncan's Multiple Range Test), showed significance at 1% level in all Parameters (e.g., site, type, sex). DMRT was done and significant difference between the mean values was seen. The comparative study revealed that the highest content was seen in *Catla catla* female (Fig. 1) and also in *Catla catla* male (Fig. 2)

Discussion

The present study was conducted on DNA extraction and quantification of both male and female of Indian major carps *Catla catla*, *Cirrhinus mrigala* and *Labeo rohita* collected from three different sites of western Uttar Pradesh, India. The DNA content of *Catla catla* female was 62-80 ng/ μ l and of male was 71-84 ng/ μ l, of *Cirrhinus mrigala* female was 62-74 ng/ μ l and of male was 64-78 ng/ μ l and of *Labeo rohita* female was 59-68 ng/ μ l and of male was 66-74 ng/ μ l. Nanodrop is an extremely useful technology that allows

Quantification of DNA, RNA (A260) and protein (A280) concentrations and sample purity (260/280 ratio) over a large concentration range of 2-15,000 ng/L double standards DNA (Pratima *et al.*, 2014).

Figure 1: DNA content variation in female Indian major carps. The highest contents were seen in site 3 of female *Catla catla*.

Figure 2: DNA content variation in male Indian major carps. The highest contents were seen in site 3 of male *Catla catla*.

Prado *et al.* (2012) used nanodrop method for DNA quantification from different fishes based on nuclear target. Similar studies were also done by Adedunni (2014) on *Clarias gariepinus* by nanodrop spectrophotometer and nanodrop technique was also used by Shi *et al.* (2015) for DNA quantification in the process of molecular characterization of *Cynoglossus semilaevis*. Similar study on

diminution level of RNA/DNA ratio in tissue of *Labeo rohita* by exposure to some endocrine disrupting compounds was also done by Verma *et al.* (2016).

DNA quantification of male and female *Clarias batrachus*, *Clarias gariepinus* and *Clarias* hybrids was also done by Shobhna (2017). She reported that the value of DNA concentration in female of *C. batrachus* was in between 59 and 61 ng/μl and of male was in between 70- 76 ng/μl. The value of DNA concentration in female of *C. gariepinus* was 60 and 63 ng/μl and of male 75- 78 ng/μl. Highest DNA concentration was seen in hybrid individuals, in females the concentration was in between 74-76 ng/μl and in case of males value was between 88-91 ng/μl.

Similar studies on *Labeo angra* was also done by Rana and Jain (2018 a) for seasonal variation; Rana and Jain (2018 b) in female of wild *Labeo calbasu* in wild and cultured fishes; and Rana and Jain (2018 c) on *Cirrhinus mrigala*.

Rana and Jain (2019) worked on *Labeo bata* for the study of seasonal variation and the results revealed that the highest concentration was reported in cultured fishes in the month of July-August.

Acknowledgement

Authors are thankful to Department of Zoology, R. G. P. G. College Meerut for providing necessary laboratory facilities.

Adedunni A. (2014) Isolation and determination of DNA concentration of African catfish (*Clarias gariepinus*) using nanodrop spectrophotometer and agarose gel. A project report submitted to the department of aquaculture and fisheries management, University of Agriculture, Nigeria.

Day F. (1875-78) The Fishes of India: being a natural history of the fishes known to inhabit the seas and

fresh water of India, Burma and Ceylon. Williams and Norgate, London, XX, pp 816.

FAO. 2018. The State of World Fisheries and Aquaculture 2018 - Meeting the sustainable development goals. Rome. Licence: CC BY-NC-SA 3.0 IGO.

Jayaram KC. (1981) The freshwater fishes of India, Pakistan, Bangladesh, Burma and Srilanka - A Handbook. Zoological Survey India, Calcutta.

Nelson JS. (2006) Fishes of the world, Fourth edition. Wiley, New York.

Prado M, Boix A and Hols CV. (2012) Novel approach for the simultaneous detection of DNA from different fish species based on a nuclear target: quantification potential. Anal. Bioanal. Chem. 403:3041-3050.

Pratima N, Chaturvedi A, Manorama K, Sreedhar M, Ravicharan A and Bhagavatula K. (2014) Identification of fresh water fish species commonly consumed in Andhra Pradesh using PCR-RFLP profile of cytochrome B gene. Int. J. Food Nutri. Sci. 3: 33-37.

Rana N and Jain S. (2018 a) Seasonal Variation in DNA content of *Labeo angra* (Hamilton, 1822). Recent Trends Fish Molecular Biology Stress Physiol. ISBN 9788170196273, pp 83-91.

Rana N and Jain S. (2018 b) Analysis of DNA content of wild and cultured *Labeo calbasu* (Hamilton, 1822) using nanophotometer. Intern. J. Trends Scientific Res. Develop. 3: 906-909.

Rana N and Jain S. (2018 c) DNA quantification of wild and cultured *Cirrhinus mrigala* (Hamilton, 1822) collected from different sites of western Uttar Pradesh, India. Intern. J. Zool. Invest. 4: 181-185.

Rana N and Jain S. (2019) Variation in DNA content of wild and cultured *Labeo bata* (Hamilton, 1822) in different seasons. Bull. Env. Pharmacol. Life Sci., 8: 70-74.

Ruzzante DE, Taggart CT, Cook C and Goddard. (1996) Genetic differentiation between inshore and offshore Atlantic cod *Gadus morhua* off Newfoundland: microsatellite DNA variation and antifreeze level. Can. J. Fish. Aqu. Sci. 53: 634-645.

Shi B, Liu X, Xu Y and Wang S. (2015) Molecular characterization of three gonadotropin subunits and their expression patterns during ovarian maturation in *Cynoglossus semilaevis*. Int. J. Mol. Sci. 16: 2767-2793.

Shobhna (2017) Molecular characterization and DNA quantification of *Clarias batrachus*, *C. gariepinus* and their hybrids. Ph.D. Thesis CCS University, Meerut, India.

Verma R, Singh AK and Jaiswal K. (2016) Preliminary study on diminution level of RNA/DNA ratio in tissue of *Labeo rohita* by exposure to some endocrine disrupting compounds (EDCs). Aceh J. Animal Sci. 1: 16-20.

Welcomme RL. (1988) International introductions of inland aquatic species. FAO Fish. Tech. 294, 318. <http://www.fao.org/docrep/X5628E/X5628E00.htm>