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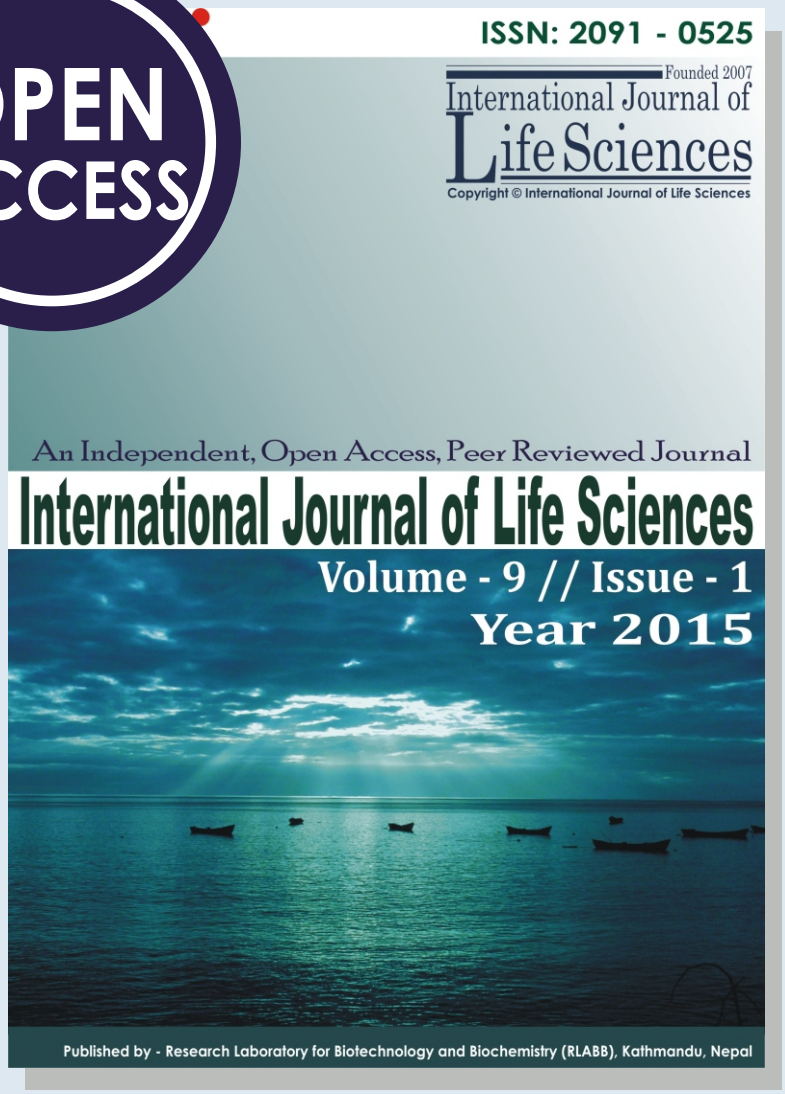

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Research Article

Genetic Variability Assay of Different Natural and Hatchery Populations of Rohu (*Labeo rohita*) in Bangladesh

Shikder Saiful Islam[1], Md. Saifuddin Shah[1], Foyez Ibn Shams[1], Md. Rayhan Ali[1] and Md. Lifat Rahi[1,*]

[1]*Fisheries and Marine Resource Technology Discipline, Khulna University, Khulna-9208, Bangladesh*

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ABSTRACT

The level of genetic variation determines the genetic status and provides the raw material for selective improvement of a stock. Randomly amplified polymorphic DNA (RAPD) technique was used to assess the genetic variability of 7 different natural (2) and hatchery (5) populations of Indian Major Carp, Labeo rohita (Rohu) in Bangladsh. In total, 140 fish samples were collected (20 from each of the populations). Genomic DNA was extracted from the muscle tissue, and 5 different oligonucleotide primers were used which revealed 80% polymorphic DNA bands. The polymorphic loci proportions were 0.71, 0.75, 0.75, 0.85, 0.84, 0.86 and 0.89 for Ma-Fatema hatchery, Chowdhuri hatchery, Niribili hatchery, Sonali hatchery, Kapotakha hatchery, the Halda river and the Baluhor Baor populations respectively. The pair-wise population differentiation (FST) values indicated a high level of genetic variation between different populations. The Unweighted Pair Group Method of Arithmetic Mean (UPGMA) dendogram based on Nei's genetic distances also revealed high level of inter-population genetic variation among the populations. The populations were segregated into two groups: the Halda River and Baluhar Baor hatchery in one group and Kapotakha, Ma-Fatema, Chowdhuri, Niribili and Sonali hatcheries in another group. Overall, RAPD results clearly indicate the reduced genetic quality of the hatchery seeds.

INTRODUCTION

The inland freshwater aquaculture of Bangladesh is mainly consisted of four Indian major carps (IMCs) species, *viz.*, *Labeo rohita* (rohu), *Cirrhinus cirrhosus* (mrigal), *Catla catla* (catla) and *Labeo calbasu* (kalibaush); these four species are the natural inhabitant of the Indus and Ganges river systems and widely available in different river systems of Bangladesh. IMCs contribute 30% of total inland aquaculture production in Bangladesh and Rohu alone contributes 12.65% (295,575 metric tons) (FSY, 2013). Good taste and glamorous look of this species attack the special attention of the consumers in entire Indian subcontinent (Chondar, 1999; Froese and Pauly, 2006; Phale *et al.*, 2009). Moreover, easy availability of seed and feed, rapid growth of this species warrant special interest among the farmers about the culture of this species. Before 1980s, culture of the species was based on the collection of natural seed but due to the decline in natural stocks and to ensure timely supply of seed, considerable numbers (total 922: 76 governmental and 846 private) of hatcheries have already been established in Bangladesh to meet the growing demand.

In an attempt to produce more seed, the hatchery owners and operators did not pay attention to the genetic attributes and thus seeds produced in hatcheries are of poor in quality (Shah and Biswas, 2004) which is evident by the wide spread claim of the farmers that hatchery produced seeds do not grow faster like the natural seeds. Slower growth performance of the hatchery seed is supposed to be due to inbreeding and/or reduced genetic variability of the seeds (Rahi and Shah, 2012). Avoiding pedigree record keeping, use of same broods repeatedly, negative selection as well as use of smaller broods, use of limited number of broods are the common phenomenon in most of the private hatcheries of Bangladesh. Another important factor is that hatchery operators are unaware about the genetic norms regarding brood stock

*** Correspondence to: Md. Lifat Rahi, Assistant Professor, Fisheries and Marine Resource Technology Discipline, Khulna University, Khulna-9208, Bangladesh.**
Email: lifatrahi@gmail.com; Phone no. - 0880 - 1726792607

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development, maintenance and breeding. Genetic quality losses are substantial and if somehow occur it's very tough to get rid of the problem unless corrective measures are taken. Increase of genetic variability in terms of heterozygosity is the best option to solve the problem within a short time (Hedrick and Kalinowski, 2000; Shah and Khan, 2008) and the most plausible way out could be the crossing between two genetically good stocks; prior to this, assay of genetic variability is an imperative (Frankham *et al*., 2002; Shah, 2010). Positive relation between growth and heterozygosity has been observed in a wide range of species around the world (Shah and Biswas, 2004; Simonsen *et al*., 2005; Rahman *et al*., 2009; Rahi and Shah, 2012). Unfortunately, little attention, however, has been paid to date in order to maintain the genetic quality of the Indian major carps in Bangladesh (Islam *et al*., 2005; Hansen *et al*., 2006). Genetic variability assay of different hatchery and natural populations would provide the genetic status of different populations. Hence, the better quality stocks could be identified and preserved for producing good quality seed in all the hatcheries that would help to combat the problem of slower growth performance.

Measuring genetic diversity in wild fish populations or aquaculture stocks is essential for interpretation, understanding and effective management of these populations or stocks. Genetic diversity could be measured indirectly and inferentially through controlled breeding and performances studies or by classical systematic analysis of phenotypic traits (Islam *et al*., 2005; Rahman *et al*., 2009; Rahi and Shah, 2012). Information on the genetic structure of cultivable fish and shellfish species is useful for optimizing identification of stocks, stock enhancement, breeding programs improvement, management for sustainable yield and preservation of genetic diversity (Dinesh *et al*., 1993; Garcia and Benzie, 1995; Tassanakajon *et al*., 1997). In the present study, efforts were made to assay the genomic DNA variability of different natural and hatchery populations of *Labeo rohita* (Rohu) in Bangladesh with the aid of RAPD technique using known primers.

MATERIALS & METHODS

Study area and sample collection

Fish samples (2-3 day old larvae) were collected from 1 natural (the Halda river) and 6 different hatchery (Ma-Fatema, Chowdhuri, Niribili, Sonali, Kapotakha and Baluhor Baor) populations. The Boluhor Baor population is a government hatchery where broods are collected from the 3 best natural populations (Halda, Padma and Jamuna rivers) of Bangladesh; there is an admixture of broods of these 3 rivers. In total 140 samples were collected (20 from each population) for DNA variability assay.

Genomic DNA Extraction of Larvae

Genomic DNA of the collected larval fish samples was extracted by using AccuPrep® Genomic DNA extraction kit (Bioneer, Korea) following the manufacturer's standard procedure. The extracted genomic DNA from all the 140 samples was preserved at -20ºC temperature until use.

Primer selection

Five different oligo-nucleotide decamer primers of random sequence (Operon Technologies, Inc., Alameda, CA, USA) were selected for the RAPD analysis of fish larvae collected from different populations. Selected primers were: OPA_1, OPA_{18}, OPB_2, OPB_{12} and OPC_3. Name of the selected primers, DNA sequence and G-C content have been presented in the table 1.

Table 1. Sequence of 5 different primers for RAPD analysis of *Labeo rohita*

Primer Name	Sequence (5´-3´)	Molecular Weight (g/mole)	G+C (%)	Temp. Profile	Number of bands scored	Number of Polymorphic Bands	Overall proportion of polymorphism (%)
OPA1	CAGGCCCTTC	2963.9	70	34	40	33	82.5
OPA18	AGGTGACCGT	3068.0	60	32	33	19	57.58
OPB2	TGATCCCTGG	3018.9	60	32	32	25	78.12
OPB12	CCTTGACGCA	2987.9	60	32	42	35	83.33
OPC3	GGGGGTCTTT	3090.0	60	32	28	28	100

Determination of DNA purity and adjustment of DNA concentration

The concentration and purity of extracted DNA samples were determined from the ratio of absorbance at A_{260} and A_{280} (absorbance at 260 nm and 280 nm) using a spectrophotometer against NaOH blank cuvette. DNA sample containing cuvette was washed properly before loading next sample. Thus, a list of data for the samples for two different absorbencies was found and saved. The protocol used in this experiment was designed for a double-beam spectrophotometer. The DNA concentration and purity was determined by the following formulas:

1. Double-stranded DNA concentration (C), µg/ml = Absorbance at $A_{260} \times 50 \times 500$
2. Purity = Absorbance at A_{260} /Absorbance at A_{280}

In case of all the extracted DNA samples, A_{260}/A_{280} values were <1 which indicates satisfying purity of the extracted DNA and there was no contamination. After determining the concentrations of extracted DNA, nuclease free de-ionized sterile distilled water was added in a required volume to adjust the concentrations of all the extracted DNA samples. The adjusted DNA concentration for PCR amplification was at 20-25 ng/µl.

PCR Amplification

The PCR reactions were performed in a 20µl reaction mixture containing 1µl DNA sample (template DNA), 2µl (10 pico-mole/µl) oligonucleotide primers, 2µl 10X reaction buffer (Bioneer, Korea), 2µl 10mM dNTPs mixture (Bioneer, Korea), 2µl Taq DNA polymerase (1 unit) and 11µl de-ionized sterile distilled water. The reaction mixtures were then placed in a DNA thermal cycler (C1000™, BIO-RAD, USA) for polymerase chain reaction. The PCR conditions were: initial extended step of de-naturation at 94 °C for 2 minutes followed by 35 cycles of de-naturation at 94 °C for 1 minute, primer annealing at 32-34 °C for 1 minute and elongation at 72 °C for 1 minute.

Evaluation of PCR products by agarose gel electrophoresis

After the completion of thermal cycling, 8 µl of each PCR products was analyzed electrophoretically by running through a 2% agarose gel and the amplified product size was determined by comparing with a 100 bp DNA size marker which is known as DNA Ladder (Bioneer, Korea). The DNA Ladder provided 13 different bands of 100 to 2000 base pairs (100, 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1200, 1600 and 2000 base pairs). The bands were observed on UV-transilluminator and photograph was taken by a Gel Cam Polaroid camera.

Data Analysis

DNA band patterns generated by RAPD were scored as 1 for bright bands (presence of bands) and 0 for their absence of bands. POPGENE (Version 1.31) software was used to determine gene diversity (Nei, 1973), gene flow (N_m), genetic distance (D), to construct an unweighted pair group method of arithmetic mean (UPGMA) dendrogram among the populations and to perform a test of homogeneity (at 95% confidence interval) in different locus between population pairs. Tools for population genetic analyses (TFPGA; Miller, 1997) software was used to estimate the population differentiation (F_{ST}) at 5% level of significance.

In this study, similarity coefficient was calculated across all possible pair wise comparison of individuals both within and among different populations using the method of Lynch (1990) with the formula:

$$SI = 2N_{AB} / (N_A + N_B)$$

Where,

N_{AB}= Number of fragments shared by individual A and B;

N_A and N_B=Number of fragments scored for each individuals,

nm was read with spectrophotometer.

RESULTS

The highest numbers of bands were generated by the primer OPB12, whereas the least number of bands were produced by the primer OPC3. A total of 175 distinct bands were produced by five primers of which 140 (80%) were polymorphic. Out of these five primers, OPB12 provided the maximum number of polymorphic bands and thus indicating a high level of polymorphism (Table 1). The RAPD profiles of different primers are shown in Fig. 1.

Within and between population similarity indices

Intra-population similarity indices (SI) was highest for the Halda river population (68.85%) followed by that of Kapotako (60.78%), Ma-fatema (56.67%), Sonali (48.55%), Niribili (52.98%), Baluhar Baor (56.88%) and Chowdhuri hatchery (39.04%) populations, respectively. Moreover, inter-population similarity indices (Sij) between the Halda River vs Kapotakha hatchery samples was higher (73.64%) than those for all other between-population indices for river and hatchery samples (Ma-Fatema-Halda: 63.52%; Halda-Sonali: 67.24%; Halda-Chowdhuri: 45.46%; Halda-Niribili: 60.66%; Halda-Baluhar Baor: 69.24%; Kapotakha-Ma-Fatema: 68.45%; Kapotakha-Sonali: 66.11%; Kapotakha-Chowdhuri: 64.59%; Kapotakha-Niribili: 69.26%; Kapotakha-Baluhar Baor: 52.78%; Ma-Fatema-Sonali: 62.1%; Ma-Fatema-Chowdhuri: 46.38%; Ma-Fatema-Niribili: 55.14%; Ma-Fatema-Baluhar Baor: 49.98%; Sonali-Chowdhuri: 56.34%; Sonali-Niribili: 52.58%; Sonali-Baluhar Baor: 51.08%; Chowdhuri-Baluhar Baor: 43.59%; Niribili-Baluhar Baor: 49.94% and Chowdhuri-Niribili: 58.335%).

Polymorphism in different populations

The highest (0.89) frequencies of polymorphic bands across all the five primers were observed in the Baluhor Baor population and the lowest (0.71) in the Ma-Fatema hatchery population (Table 2; Figure 1). Polymorphism in Chowdhuri, Niribili, Sonali, Kapotakha and Halda population were found to be 0.75, 0.75, 0.85, 0.84 and 0.86 respectively.

Table 2. Number and percentages of polymorphic bands, total number of bands and gene diversity obtained from different experimental populations

Parameters	MF	C	N	S	K	H	B
Total no. of bands	17	20	20	22	25	36	35
Total no. of polymorphic bands	12	15	15	17	21	31	31
Rate of Polymorphism (%)	70.6	75	75	77.3	84	86.1	88.6
Proportion of polymorphic bands	0.71	0.75	0.75	0.85	0.84	0.86	0.89
Gene diversity (H)	0.03	0.04	0.04	0.04	0.05	0.07	0.08

Overall no. of polymorphic bands across the populations: 49; Overall % of polymorphic loci across the populations: 90.7; Overall gene diversity for all loci: 0.08

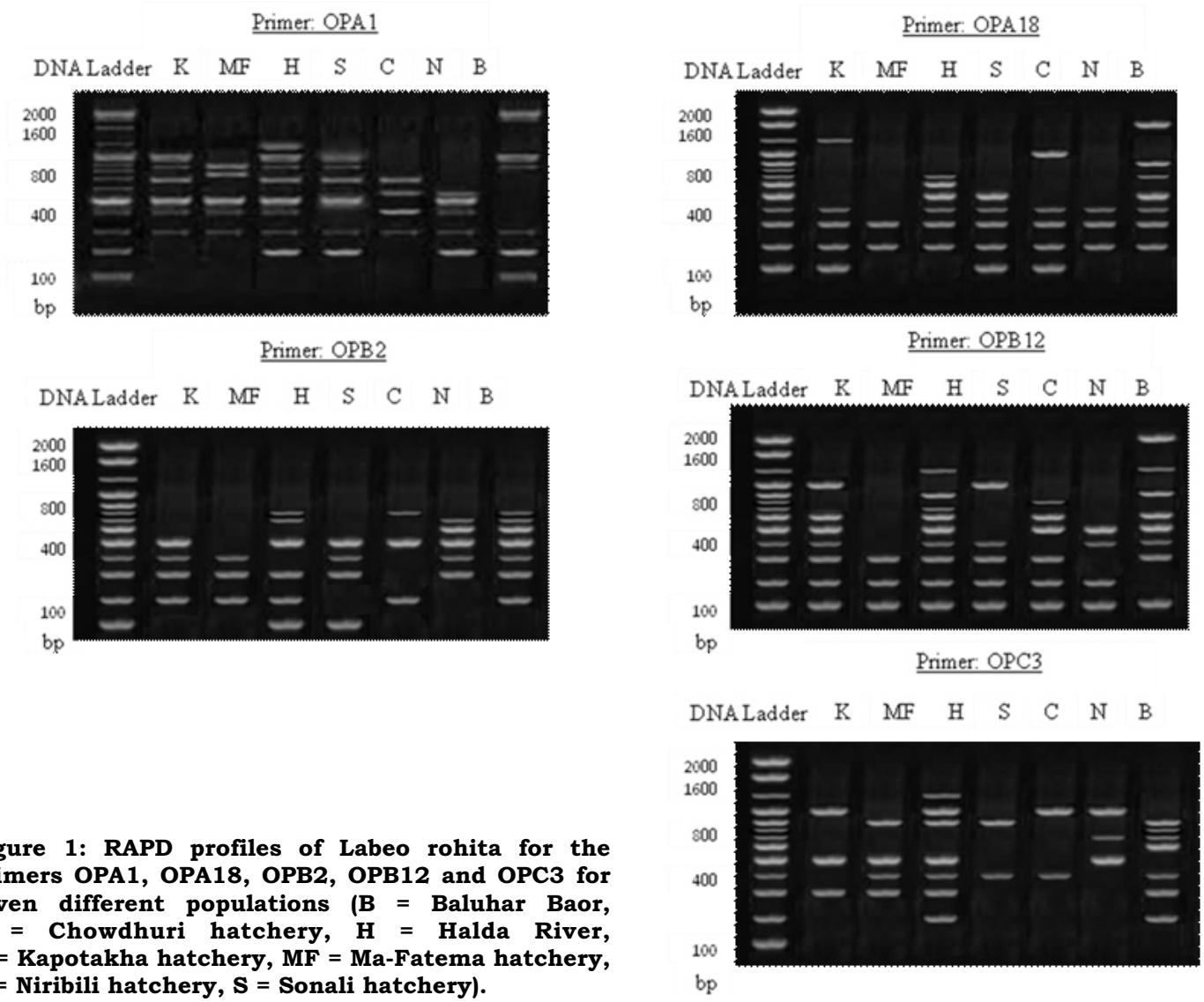

Figure 1: RAPD profiles of Labeo rohita for the primers OPA1, OPA18, OPB2, OPB12 and OPC3 for seven different populations (B = Baluhar Baor, C = Chowdhuri hatchery, H = Halda River, K = Kapotakha hatchery, MF = Ma-Fatema hatchery, N = Niribili hatchery, S = Sonali hatchery).

Nei's gene diversity

Overall gene diversity was found to be 0.08 across all populations for all loci. The study revealed that the Baluhor Baor population relatively higher gene diversity compared with the Halda river and other hatchery populations (Table 2). The gene diversity value for the Ma-Fatema hatchery population was the lowest. However, the 95% confidence intervals showed no significant differences in gene diversity between the populations.

Population differentiation (θ or F_{ST}) and gene flow (N_m)

The F_{ST} value was found to be highest between the Baluhor Baor and the Ma-Fatema hatchery populations but was lowest between Chowdhuri and Ma-Fatema hatchery populations. Moreover, the pair-wise F_{ST} values were found to be significant between Baluhor Baor vs. Kapotakha hatchery, Baluhor Baor vs. Sonali hatchery, Baluhor Baor vs. Niribili hatchery, Baluhor Baor vs. Chowdhuri hatchery, Baluhor Baor vs. Ma-Fatema hatchery, Halda vs. Kapotakha hatchery, Halda vs. Sonali hatchery, Halda vs. Niribili hatchery, Halda vs. Chowdhuri hatchery and Halda vs. Ma-Fatema hatchery populations (Table 3). The estimated gene flow (Nm) value across the populations was 15.42. The highest Nm value (32.08) was observed between the Chowdhuri hatchery and Ma-Fatema hatchery populations, whereas the lowest Nm (11.06) value was found between the Baluhor Baor and Ma-Fatema hatchery populations (Table 3).

Genetic distance

The genetic distance value for the Baluhor Baor vs. Chowdhuri, Baluhor Baor vs. Ma-Fatema and the Halda River Ma-Fatema hatchery population pairs were the highest (0.014) whereas the value for the Chowdhuri vs. Ma-Fatema hatchery population pair was the lowest (0.003) (Table 3). The UPGMA dendrogram indicates the segregation of the seven populations of rohu into two clusters: the Halda and Baluhar Baor populations made one cluster and other five populations (Sonali, Ma-Fatema, Chowdhuri, Niribili and Kapotakha) made another cluster. The second cluster was further separated into three subgroups (Figure 2).

Table 3. Population differentiation (θ or FST) at 5% level of significance, gene flow (Nm) and pair-wise genetic distance (D) values between the populations

Population pairs	F_{ST} ± SD	Gene flow (N_m)	Genetic Distance (D)
B vs. H	0.018 ± 0.011 (0.011-0.032)	27.43	0.006
B vs. K	0.049* ± 0.017 (0.024-0.078)	16.21	0.010
B vs. S	0.053* ± 0.028 (0.032-0.085)	14.85	0.011
B vs. N	0.054* ± 0.032 (0.032-0.109)	14.54	0.012
B vs. C	0.059* ± 0.036 (0.038-0.097)	12.77	0.014
B vs. MF	0.062* ± 0.037 (0.036-0.093)	11.06	0.014
H vs. K	0.047* ± 0.020 (0.023-0.081)	17.02	0.009
H vs. S	0.057* ± 0.029 (0.035-0.086)	13.93	0.012
H vs. N	0.049* ± 0.030 (0.029-0.092)	13.09	0.010
H vs. C	0.052* ± 0.031 (0.033-0.094)	13.04	0.011
H vs. MF	0.061* ± 0.034 (0.037-0.089)	9.89	0.014
K vs. S	0.022 ± 0.016 (0.013-0.045)	28.52	0.007
K vs. N	0.021 ± 0.017 (0.014-0.046)	28.74	0.007
K vs. C	0.023 ± 0.014 (0.012-0.042)	28.01	0.008
K vs. MF	0.025 ± 0.017 (0.015-0.043)	27.12	0.008
S vs. N	0.019 ± 0.012 (0.010-0.032)	27.31	0.006
S vs. C	0.017 ± 0.011 (0.009-0.031)	29.54	0.005
S vs. MF	0.022 ± 0.013 (0.010-0.033)	26.93	0.007
N vs. C	0.014 ± 0.009 (0.007-0.027)	31.76	0.004
N vs. MF	0.016 ± 0.009 (0.008-0.031)	30.67	0.005
C vs. MF	0.013 ± 0.007 (0.009-0.029)	32.08	0.003
Across all the experimental populations	15.42	-	-

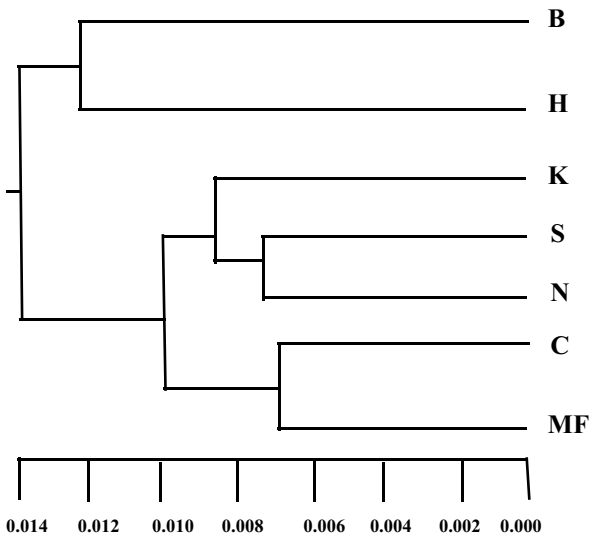

Figure 2. UPGMA dendrogram of *Labeo rohita* based on Nei's (1973) genetic distance.

DISCUSSION

The presence of genetic variation within populations and also between individuals is essential for their ability to survive and successfully respond to environmental changes (Ryman *et al*., 1995). The band patterns generated by RAPD assay in the present study were used to differentiate between 7 different populations of *Labeo rohita* and to deduce genetic relationship among them. In the present study, a higher rate of polymorphism was observed for all the populations. The percentage of polymorphic bands appeared to be more in the hatchery Ma-Fatema (70.6 %), Chowdhuri (75 %), Niribili (75 %), Sonali (77.3 %), Kapotakha (84 %) hatchery and the Halda (86.1 %), Baluhor Baor (88.6 %). Comparatively lower genetic variability was observed in almost all the hatchery populations (except Baluhor Baor) in the present study. This may be attributed to the maintenance of a limited number of individuals in the hatcheries that might caused inbreeding problem and reduced genetic variation in the hatchery population, as was also reported by Eknath and Doyle (1990) on the basis of effective population size.

In the present study, it was found that the Intra-population SI values for the hatchery populations were 60.78% for Kapotakha, 56.67% for Ma-Fatema, 48.55% for Sonali, 39.04% for Chowdhuri, 52.98 for Niribili and 56.88% for Baluhar Baor and the highest 68.85% for Halda river populations. All of the genetic quality indicators in terms of inter- and intra- population genetic similarity indices, population differentiation (θ or F_{ST}), gene flow (N_m), genetic distance (D), gene diversity (H) and proportions of polymorphi loci reveal the reduced genetic quality of hatchery populations. This genetic quality deterioration is the main reason of the farmer's interest loss to use hatchery seeds for aquaculture because the hatchery seeds exhibit slower growth performance and lower survival rates. This is because of the lack of genetic improvement programs in Bangladesh that might cause genetic drift, inbreeding, hybrid introgression and thereby serious genetic quality loss of Indian Major Carps which are studied by many authors (Shah and Biswas, 2004; Islam *et al*., 2005; Simenson *et al*., 2005; Shah and Khan, 2008; Rahman *et al*., 2009; Rahi and Shah, 2012). The band patterns of the hatchery populations provide the evidence that the source of the hatchery broods was from Halda river. It is very difficult to exactly determine the origin of hatchery broods in Bangladesh because most of the hatcheries are unaware about record keeping for pedigree history. In Bangladesh, 3 different natural populations of Indian Major Carps are considered for breeding program: the Padma river, Jamuna river and Halda river; to date, Halda river population is considered to be the best in terms of genetic quality and also for growth (Islam and Alam, 2004; Shah and Biswas, 2004;Hansen *et al*., 2006; Phale *et al*., 2009).

It has been found evident that if the hatchery populations are managed properly with better feed, good quality water and of course genetic with genetic aspects, it provides far more yield over the natural stocks (Barman *et al*., 2003; Shah, 2010; Rahi and Shah, 2012). The Baluhar Baor hatchery population could be a classical example in this regard that showed the highest genetic variation in this study. The reasons are: excellent brood management, pedigree record keeping, stocking huge number of broods, random breeding and collection of broods from different natural populations. Broods were collected from all 3 (Padma, Jamuna and Halda) major river populations of Bangladesh and later on, there had been an admixture of the broods. This is the reason of higher genetic variation of this hatchery population over the other hatcheries and even over the best natural population (Halda river) of Bangladesh. There is also a superb reputation of Boluhor hatchery seeds to the farmers who are highly interested to use this hatchery seed for aquaculture while opposite scenario is found for the most other hatcheries. The other 5 hatcheries sampled in this study, do not have a healthy brood stock and there is no clear information about source.

CONCLUSION

In the present study, RAPD based genetic variability assay of seven different natural and hatchery population of *Labeo rohita* (rohu) clearly showed the genetic quality deterioration of hatchery populations in Bangladesh. Consequently, the aquaculture of rohu has been suffering from growth inefficiency and lower survival but still there are promises to get rid of this problem. A massive program should be taken immediately to check the genetic status of different hatcheries as well as natural populations of Bangladesh in order to identify the good stocks which will act as a baseline to improve the seed quality of carps. After checking the genetic status of different populations, improved lines of fishes can be produced by crossing between highly genetically variable stock and less variable stock. This would create a positive impact in boosting aquaculture production of Bangladesh.

REFERENCES

Barman H, Barat A, Bharat M, Banerjee Y, Meher P, Reddy P, Jana R. 2003. Genetic variation between four species of Indian major carps as revealed by random amplifiedpolymorphic DNA assay. Aquaculture, 217: 115-123.

Chondar SL. 1999. Biology of Finfish and Shellfish (1st ed). SCSC Publishers, India, p. 514.

Dinesh KR, Lim TM, Chua KL, Chan WK, Phang VPE. 1993. RAPD analysis: an efficient method of DNA fingerprinting in fishes. Zoological Science, 10: 849-854.

Eknath AE, Doyle RW. 1990. Effective population size and rate of inbreeding in aquaculture of Indian major carps. Aquaculture, 85: 293305.

Frankham R, Ballou JD, Briscoe D. 2002. Introduction to Conservation Genetics. Cambridge University Press, Cambridge, UK.

Froese R, Pauly D. 2006. Fish Base. World Wide Web Electronic Publication. World Fish center (ICLARM). www.FishBase.Org.

FSY. 2013. Fisheries Statistical Yearbook of Bangladesh, Department of Fisheries, Ministry of Fisheries and Livestock, Government of the People's Republic of Bangladesh.

Garcia DK, Benzie JAH. 1995. RAPD markers of potential use in penaeid prawn (*Penaeus monodon*) breeding programs. Aquaculture, 130: 137144.

Hansen MM, Simonsen V, Mensberg KLD, Sarder MRI, Alam MS. 2006. Loss of genetic variation in hatchery reared Indian major carp, *Catla catla*. Aquaculture, 69(B): 229-241.
Hedrick PW, Kalinowski ST. 2000. Inbreeding depression in conservation biology. Annual Reviews in Ecology, Evolution and Systematics, 31: 139 - 162.
Islam MS, Ahmed ASI, Azam MS Alam MS. 2005. Genetic analysis of three river populations of *Catla catla* (Hamilton) using randomly amplified polymorphic DNA markers. Asian-Australasian Journal of Animal Sciences, 18: 453-457.
Islam MS, Alam MS. 2004. Random Amplified Polymorphic DNA analysis of four different populations of Indian major carp, *Labeo rohita* (Hamilton). Journal of Applied Ichthyology, 20: 407-412.
Lynch M. 1990. The similarity index and DNA fingerprinting. Molecular Biology and Evolution, 7: 478-484.
Miller MP. 1997. Tools for population genetic analysis, TFPGA, version 1.31. Computer software distributed by the author (mpm2@jan.ucc.nau.edu).
Nei M. 1973. Analysis of gene diversity in subdivided populations. Proceedings of the National Academy of Sciences of the United States of America, 70: 3321 -3323. http://dx.doi.org/10.1073/pnas.70.12.3321
Phale SR, Chauhan S, Bhute YV, Baile VV. 2009. Detection of Genetic Variation in the wild populations of Indian major carps using randomly amplified polymorphic DNA fingerprinting. Journal of Fisheries and Aquatic Sciences, 4(1): 63-70.
Rahi ML, Shah MS. 2012. Triploidization in rohu x mrigal hybrid and comparison of growth performance of triploid hybrid. Aquaculture Research, 43(12): 1867-1879. doi: 10.1111/j.1365-2109.2011.02996.x
Rahman SMZ, Khan MR, Islam S, Alam S. 2009. Genetic variation of wild and hatchery populations of the catla Indian major carp (*Catla catla* Hamilton 1822: Cypriniformes, Cyprinidae) revealed by RAPD markers. Genetics and Molecular Biology, 32(1): 197-201.
Ryman N, Utter F, Laikre L. 1995. Protection of intraspecific biodiversity of exploited fishes. Reviews in Fish Biology and Fisheries, 5: 417-446.
Shah MS. 2010. Sexuality, breeding biology and genetics of reproduction. In: Genetics of aquaculture and fisheries management (1st ed.). Sakib offset press, Khulna, Bangladesh, p.73.
Shah MS, Biswas BK. 2004. Growth and reproduction of wild and hatchery produced strains of Indian major carps. Bangladesh Journal of Fishheries, 27: 15-16.
Shah MS, Khan MMR. 2008. Randomly amplified polymorphic DNA analysis of three population of catla (*Catla catla*, Hamilton) in Bangladesh. International Journal of Bioresearch, 4(6): 76-81.
Simonsen V, Hansen MM, Mensberg KLD, Sarder RI, Alam S. 2005. Widespread hybridization among species of Indian major carps in hatcheries, but not in the wild. Journal of Fish Biology, 67:794-808.
Tassanakajon A, Pongsomboon S, Rimphanitchayakit V, Jarayabhand P, Boonsaeng V. 1997. Random amplified polymorphic DNA (RAPD) markers for determination of genetic variation in wild populations of black tiger prawns (*Penaeus monodon*) in Thailand. Molecular Marine Biology and Biotechnology, 6: 110115.

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