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Genetic variability in wild and hatchery populations of commercially important fish: The common carp (*Cyprinus carpio*)

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Abstract. *Ahmadi M, Kashiri H, Shabani A, Moghadam AA. 2018. Genetic variability in wild and hatchery populations of commercially important fish: The* common carp *(Cyprinus carpio). Biodiversitas 19: 1468-1474.* Common carp, *Cyprinus* carpio, is considered as one of the most important bony fish with high economic value in the Caspian sea. Since the population size of common carp has decreased during recent decades, restocking of the populations is done through releasing the hatchery-produced larvae into the Caspian Sea. In the present study, the genetic diversity of hatchery and wild populations of *C. carpio* was investigated using eight microsatellite loci (MFW7, MFW9, MFW13, MFW16, MFW17, MFW20, MFW26 and MFW28). A total of 145 different alleles were observed across all loci, with some of them being unique to each population. Although the allelic and gene diversity of hatchery populations tended to be lower compared to the wild populations, no significant differences ($P>0.05$) in genetic diversity parameters were observed among the wild and hatchery populations. In most cases, significant departure from Hardy-Weinberg equilibrium was observed, mainly because of the heterozygosity deficiency. Results from F_{ST}, R_{ST} and UPGMA analysis showed that the hatchery-reared population was the most differentiated and distant group. The results from this study are anticipated to provide important information for setting up more efficient strategies in conservation and restocking of *C. carpio*.

Keywords: Allelic diversity, *Cyprinus carpio*, hatchery, population, microsatellite

Abbreviations: A_E: Effective number of alleles, A_O: Number of alleles, A_U: Unique alleles, GRR: Gorganroud River, H_E: Expected heterozygosity, H_O: Observed heterozygosity, HWE: Hardy-Weinberg equilibrium, QR: Qaresou River, S: Allele size

INTRODUCTION

Common carp (*Cyprinus carpio*) belongs to the family Cyprinidae which is regarded as the largest family of freshwater teleosts (Nelson 1994). The common carp is native to Asia and Eastern Europe. This species has introduced throughout the world but the wild stocks are only present naturally in rivers draining to the Black, Aral and Caspian Sea (Kottelat and Freyhof 2007). It is believed that anthropogenic activities including damming on the rivers, overfishing as well as degradation of natural spawning grounds have caused considerable decline in natural populations of wild common carp through the world so that this fish is regarded as a vulnerable or endangered species in many places including the Caspian Sea basins (Kottelat and Freyhof 2007; Vazirzadeh and Yelghi 2015). To promote *C. carpio* population recovery, Iranian Fisheries Organization has proceeded to restock through releasing the hatchery-produced larvae in to the Caspian Sea basins since the late 1994. In this regard, at the Sijeval Bony Fishes Breeding Center in Golestan province, the hatchery-produced individuals from the breeders originated from Gorganroud River have been released annually in to the rivers of Gorganroud and Qaresou during the months of June and July. Although restocking by using hatchery-produced organisms is known as a common strategy for conservation of threatened wild populations, there is still uncertainty that whether this procedure is really efficient to improve the population size or not. However, despite the potential benefits, restocking through the supportive breeding may lead to possible problems including increase in genetic divergence between sites due to genetic drift and decrease in genetic variability (Roodt-Wilding 2007). Such adverse genetic effects on natural population restoration have been previously reported by Araki et al. 2007; Cheng et al. 2011 and Li et al. 2016. Nevertheless, there have also been reports on maintaining genetic variability in hatchery populations of other fish species (Pan and Yang 2010; An et al. 2013).

Since the genetic quality of hatchery-produced larvae may have a great impact on the efficiency of restocking programs (Li et al. 2016), conservation management strategies without the knowledge of genetic structure can have deleterious impacts on the gene pools of natural populations (Laikre et al. 2005). Food and Agricultural Organization of the nations also recommends genetic assessment of the populations used for restocking programs as well as the target ones to monitor any changes for sustainable management of populations. Therefore assessing the genetic diversity of hatchery-produced

compared to natural populations of common carp is urgently needed to produce high-quality individuals for successful management and sustainable use. More than 23 years have passed from the beginning of the massive release of common carp larvae into the Caspian Sea and natural spawning has decreased during this period. Unfortunately, despite that restocking programs have been widely implemented for common carp populations in southeast parts of the Caspian Sea, reports on the current genetic variation in hatchery populations are not available and the previous studies are limited to the wild populations in some regions of the Caspian Sea (Yousefian and Laloei 2011; Fallahbagheri et al. 2013; Laloei et al. 2013; Ghelichpour et al. 2013).

In the present study, we used eight microsatellite markers to evaluate the genetic diversity of wild and hatchery populations of *C. carpio*. The results from our study could help to provide important information for setting up more effective breeding strategies and appropriate management in conservation and restocking of *C. carpio*.

MATERIALS AND METHODS

Sample collection and DNA extraction

A total of 64 wild specimens of *C. carpio* were caught from southeast parts of the Caspian Sea, Iran, i.e., Qaresou River (QR) and Gorganroud River (GRR) (32 individuals for each group) (Figure 1) during April and May 2017. Similarly a further 64 hatchery-produced fish were obtained from two different groups in Sijeval Bony Fishes Breeding Center, Golestan province, Iran (32 individuals for each group). At this facility, two groups of wild and hatchery-reared breeders are used for restocking program: hatchery-reared population was established from the animals of Gorganroud River in the 2013s. The larvae produced from hatchery-reared breeders have been released annually into the Caspian Sea from 2015. The wild breeders are also caught annually from Gorganroud River during migration season and the larvae produced from these breeders have been released into the nature. Approximately 1×1 cm^2 of caudal fin was excised from each specimen and preserved in 96% ethanol at 4^0C for subsequent DNA extraction.

Total DNA was extracted from fin tissues using DNA extraction kit (GeneAll, Korea) according to the manufacturer's protocol. The quantity and quality of the extracted DNA were determined using a Biophotometer Spectrophotometer (Eppendorf, Germany) and 1% agarose gel electrophoresis, respectively. The genomic DNA was stored at -20°C until PCR reactions.

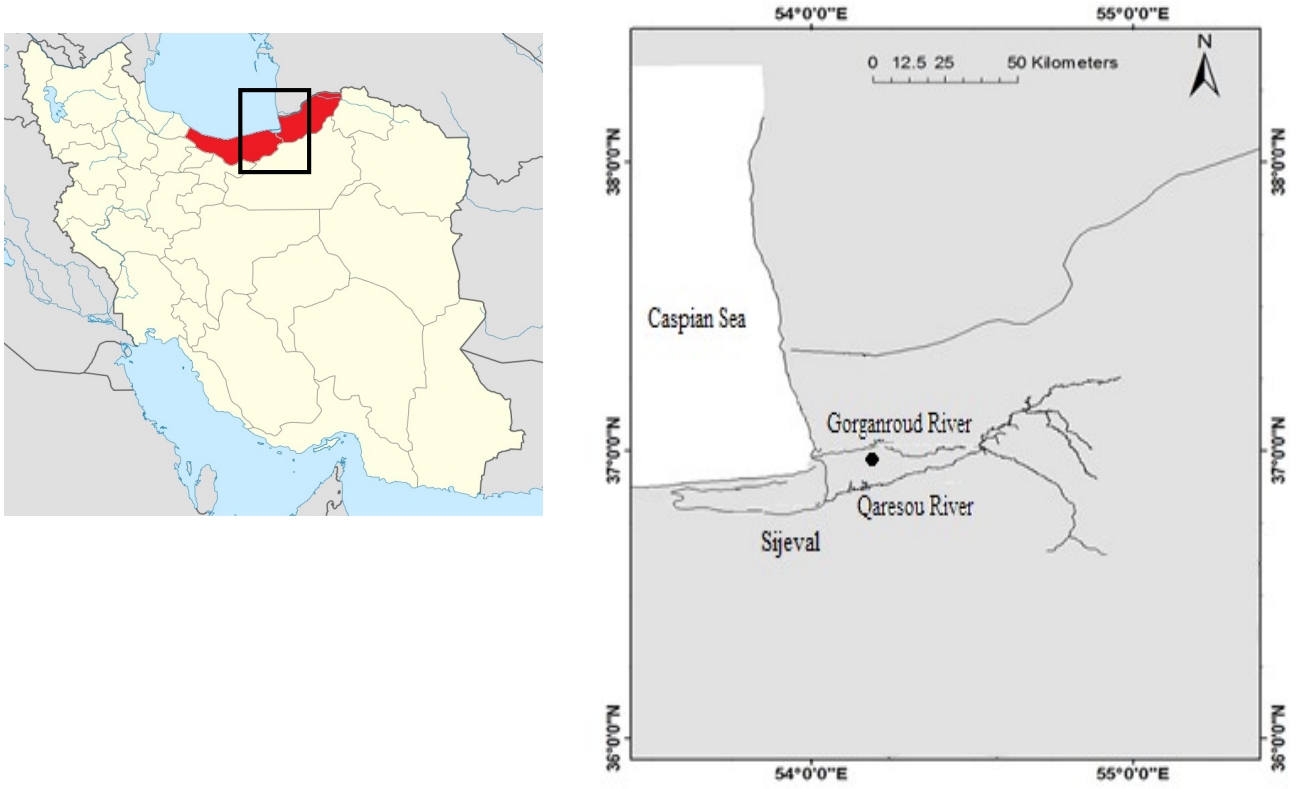

Figure 1. *Cyprinus carpio* sampling locations in Qaresou River (QR) and Gorganroud River (GRR), southeast parts of the Caspian Sea, Iran

Table 1. Characteristics of microsatellite loci used for screening of *Cyprinus carpio* genetic diversity.

Locus	Sequence		No. of alleles	Allele size	Anneal (°C)
MFW7	F: TACTTTGCTCAGGACGGATGC;	R: ATCACCTGCACATGGCCACTC	20	168-244	62
MFW9	F: GATCTGCAAGCATATCTGTCG;	R: ATCTGAACCTGCAGCTCCTC	14	144-196	58
MFW13	F: ATGATGAGAACATTGTTTACAG;	R: TGAGAGAACAATGTGGATGAC	16	176-236	56
MFW16	F: GTCCATTGTGTCAAGATAGAG;	R: TCTTCATTTCAGGCTGCAAAG	16	124-184	57
MFW17	F: CTCAACTACAGAGAAATTTCATC;	R: GAAATGGTACATGACCTCAAG	23	216-304	57
MFW20	F: CAGTGAGACGATTACCTTGG;	R: GTGAGCAGCCCACATTGAAC	20	208-284	60
MFW26	F: CCCTGAGATAGAAACCACTG;	R: CACCATGCTTGGATGCAAAAG	19	120-192	60
MFW28	F: GATCCCTTTTGAATTTTTCTAG;	R: ACAGTGAGGTCCAGAAGTCG	17	184-248	58

Polymerase chain reaction (PCR)

Eight nuclear microsatellite loci were used to measure genetic diversity among the common carp populations; MFW7, MFW9, MFW13, MFW16, MFW17, MFW20, MFW26 and MFW28 (Crooijmans et al. 1997). The characteristics of the applied loci are presented in Table 1. The PCR reaction was performed in 12.5 µL reaction mixture containing 2 µL DNA, 6.25 µL Red Load Taq Master (Jena Bioscience, Germany), 1 µL of each primer and 2.25 µL PCR grade water. PCR program was consisted of a pre-denaturation for 3 min at 95°C, followed by 35 cycles of denaturation for 30 sec at 95°C, annealing (Table 1) for 30 sec and extension for 30 sec at 72°C as well as a final extension for 3 min at 72^0C. The PCR products were separated on 10% polyacrylamide gel. A 100 bp ladder (Fermentas, USA) was applied as the indices for determining allelic size. The obtained gels were stained using silver nitrate method (Benbouza et al. 2006). After recording the gel images using a gel documentation system (Gel Doc XR, Bio-Rad, USA), the allele size was determined using Gel Pro Analyzer 3.9 software.

Data analysis

The possibility of the presence of null alleles, scoring errors and dropout of large alleles were checked using Microchecker program (Oosterhout et al. 2004). Cervus program 3.03 (Kalinowski et al. 2007) was used to determine the number of unique alleles (A_U). The genetic diversity parameters of each sample including the number of alleles (A_O), the effective number of alleles (A_E), allele size (S), the observed heterozygosity (H_O) and the expected heterozygosity (H_E) were calculated by Genealex 6.3 (Peakall and Smouse 2006). Differences in genetic diversity parameters were assessed using Wilcoxon-Mann-Witney test implemented in the SPSS software. Linkdis method implemented in Genetix was applied to investigate linkage disequilibrium (Belkhir et al. 1999). The probability of bottleneck in the populations was investigated using Bottleneck 1.2.02 (Cornuet and Luikart 1996) under TPM. This procedure is applied to analyze the deviation from mutation-drift equilibrium on the basis of excess or deficiency in heterozygosity. Deviation from Hardy-Weinberg equilibrium (HWE) was tested by Genepop 3.4 software (Raymond and Rousset 1995).

HWE tests were also carried out for each locus in each population to test if deviations from HWE were in the direction of deficit or excess of heterozygosity. The sequential Bonferoni correction was applied to adjust the significance levels for multiple tests (Rice 1989). The inbreeding indices (F_{IS}) and its significance were determined using FSTAT 2.9.3 program (Goudet 2001). Rstcalc (Goodman 1997) and FSTAT (Goudet 2001) were used to determine R_{ST} (Slatkin 1995) and F_{ST} (Weir and Cockerham 1984), respectively. Analysis of molecular variance (Amova) was run to determine the partitioning of genetic diversity within and among populations in Arlequin 3.1 (Excoffier et al. 2005). The significance estimates were obtained using 1000 permutations. The gene flow (N_M) among populations, unbiased genetic identity (GI) and genetic distance (GD) based on Nei (1978) were determined by Genealex 6.3 (Peakall and Smouse 2006). UPGMA dendrogram was constructed based on Nei's genetic distance using Population program 1.2.30 (Oliver Langella, CNRS UPR9034). The bootstraps levels were calculated by 10000 replicate across loci. The Treeview program 1.6.6 (Page 1996) was applied to visualize the constructed dendrogram.

RESULTS AND DISCUSSION

Population genetic diversity

Microchecker didn't show any evidence for large allele dropout and stutter-band scoring at the applied loci, but null alleles were observed at the loci MFW7 and MFW28. The estimated frequencies of null alleles in these loci were 0.208 and 0.173, respectively. All the applied loci showed polymorphism in all samples of common carp, and the polymorphism level varied depending on the locus. A total of 145 different alleles were observed across all loci, with some of them being unique to each population. Allelic diversity was 18.1, with the number of alleles per locus ranging from 14 (MFW9) to 23 (MFW17) (Table 1). The measures of genetic diversity parameters are presented in Table 2. The observed and expected heterozygosity values ranged from 0.573 to 1 (mean H_O: 0.836) and 0.0.865 to 0.956 (mean H_E: 0.92), respectively. No significant differences in actual and effective number of alleles as well as expected and observed heterosygosity were observed between the wild samples (Wilcoxon: $P>0.05$). In comparing the wild and hatchery populations, the mean

number of actual alleles was slightly higher in the wild samples which was not statistically significant (A_O: 14 for the wild samples versus 13.2 for the hatchery samples) (Wilcoxon: $P>0.05$). The average H_O and H_E of the wild samples also tended to be higher in comparison to the hatchery samples (H_O: 0.850 and H_E: 0.928 for the wild samples versus H_O: 0.821 and H_E: 0.912 for the hatchery samples) (Wilcoxon: $P>0.05$). 49 alleles were found to be unique to four groups: 15, 16, 10 and 8 in GRR, QR, Hatchery1 and Hatchery2, respectively (Table 2). To analyze the deviation from mutation-drift equilibrium in the studied populations, the bottleneck test was done under TPM. According to the results, no significant heterozygosity excess ($P>0.05$) was observed via the applied test, suggesting any of the populations have not experienced a recent bottleneck.

Significant departure from HWE was detected at most of the loci (Table 2). After adjusting the P values across 8 loci using the sequential Bonferroni method for multiple observations, 21 of the 32 tests (8 loci×4 populations) showed significant departure from HWE. A significant deficit of heterozygosity was noticed at some of the loci in the studied populations ($P<0.05$) (Table 2). The largest deficit was for Hatchery2 with the F_{IS} value of 0.389 at the locus MFW7. The average F_{IS} across all loci were 0.095 and 0.109 for the wild and hatchery samples, respectively. The heterozygosity excess detected at some loci was not significant in either population at any of the loci. No significant linkage disequilibrium was also noticed at any of eight loci after Bonferroni correction ($P>0.05$).

Genetic relationship among populations

The global F_{ST} over all samples was 0.017 ($P<0.01$). F_{ST} was in accordance with R_{ST} so that the lowest F_{ST} and R_{ST} were detected between GRR and Hatchery1 populations (Table 3). The Hatchery2 was the most divergent and differentiated group from all other populations ($P<0.01$). The Amova of all eight microsatellites revealed that most of the variations were found within individuals (96%; $P=0.010$). The levels of genetic variation between individuals within populations and between populations were also 1.84 ($P=0.024$) and 2.16 ($P=0.000$), respectively.

Table 2. Genetic diversity parameters for eight microsatellite loci in *Cyprinus carpio*

		MFW7	MFW9	MFW13	MFW16	MFW17	MFW20	MFW26	MFW28	Mean
GRR	A_O	11	9	12	15	19	17	15	14	14
	A_E	18.87	15.17	9.48	8.79	13.91	16.96	15.61	14.74	14.19
	H_O	0.628	1.00	1.00	0.712	0.866	0.801	1.00	0.753	0.845
	H_E	0.947	0.934	0.895	0.886	0.928	0.941	0.936	0.932	0.925
	A_U	0	1	2	2	5	2	1	2	1.9
	F_{IS}	**0.341**	-0.062	-0.105	0.216	0.081	0.165	-0.059	0.211	0.098
	HWE	***	ns	ns	*	*	**	ns	**	
QR	A_O	12	9	12	14	19	17	16	14	14.1
	A_E	22.68	16.14	11.90	9.38	19.25	14.71	16.92	13.71	15.59
	H_O	0.654	1.00	1.00	0.683	0.871	0.856	0.968	0.816	0.856
	H_E	0.956	0.938	0.916	0.893	0.948	0.932	0.941	0.927	0.931
	A_U	1	1	2	1	5	2	2	2	2
	F_{IS}	**0.326**	-0.051	-0.08	**0.241**	0.096	0.097	-0.027	0.136	0.092
	HWE	***	ns	ns	**	*	**	**	*	
Hatchery1	A_O	11	9	11	13	17	17	15	14	13.4
	A_E	15.18	12.53	10.77	9.35	7.97	15.36	11.36	20.34	12.86
	H_O	0.584	1.00	1.00	0.683	0.837	0.834	1.00	0.695	0.829
	H_E	0.934	0.92	0.907	0.893	0.874	0.935	0.912	0.951	0.916
	A_U	0	1	1	0	3	2	1	2	1.2
	F_{IS}	**0.381**	-0.062	-0.091	**0.243**	0.064	0.109	-0.078	**0.273**	0.105
	HWE	***	ns	ns	*	*	***	ns	**	
Hatchery2	A_O	11	9	10	13	17	16	15	14	13.1
	A_E	13.89	13.53	9.44	10.11	7.44	14.50	8.86	14.70	11.56
	H_O	0.573	1.00	1.00	0.632	0.812	0.852	1.00	0.647	0.814
	H_E	0.928	0.926	0.894	0.901	0.865	0.931	0.887	0.932	0.908
	A_U	0	1	0	0	3	1	1	2	1
	F_{IS}	**0.389**	-0.063	-0.112	**0.306**	0.090	0.102	-0.114	**0.308**	0.113
	HWE	***	ns	ns	***	*	**	ns	***	

Note: GRR: Gorganroud River; QR: Qaresou River; Significant amounts of F_{IS} are shown in bold; HWE = Hardy-Weinberg probability test after correction with sequential test of Bonferroni (Rice, 1989); ns = not significant,; * $P\leq0.05$; ** $P\leq0.001$; *** $P\leq0.0001$.

Table 3. Genetic relationships among *Cyprinus carpio* populations

Comparison	F_{ST}	R_{ST}	N_M	Nei's GD	Nei's GI
GRR vs. QR	0.012	0.025	20.59	0.203	0.814
GRR vs. Hatchery1	0.011	0.023	22.51	0.184	0.835
GRR vs. Hatchery2	0.023	0.043	10.63	0.296	0.717
QR vs. Hatchery1	0.013	0.027	18.97	0.221	0.793
QR vs. Hatchery2	0.025	0.044	9.78	0.327	0.685
Hatchery1 vs. Hatchery2	0.018	0.035	13.66	0.288	0.731

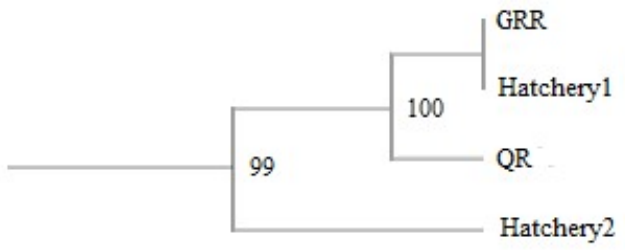

Figure 2. UPGMA dendrogram based on the Nei's (1978) genetic distance from 8 loci for the wild and hatchery populations of common carp

Cyprinus carpio populations presented high levels of gene flow (Table 3). The highest value of N_M (30.98) was observed between GRR and Hatchery1 samples. Nei (1978) unbiased genetic identity and distance among all samples are presented in Table 3. The highest GD and lowest GI were between Hatchery2 and QR while the lowest GD and highest GI were noticed among GRR and Hatchery1. According to the population relationships depicted in UPGMA cluster constructed from Nei's genetic distances, Hatchery2 was the most distant population while the nearest groups were GRR and Hatchery1 (Figure 2).

Discussion

No significant differences in genetic diversity parameters were observed among the wild samples of common carp. The genetic diversity observed in our study was higher than that previously reported by Laloei et al. (2013) for wild populations of common carp in south of the Caspian Sea (Guilan, Mazandaran and Golectan coasts) (H_O: 0.85 vs. 0.49 and H_E: 0.928 vs. 0.67) which could be related to the difference in applied loci, various sample size and sampling sites. In another studies by Yousefian and Laloei (2011) and Fallahbagheri et al. (2013) by RFLP, low genetic variation was reported for wild populations of common carp in southern Caspian Sea. In addition to the mentioned possible explanation, the fact that microsatellites are more efficient in detecting genetic diversity compared to RFLP (Shaw et al. 1999), may explain the lower concordance between our results and RFLP data. However, our results are in accordance with the study by Ghelichpour et al. (2013) who reported a considerable level of genetic diversity in the wild populations of common carp in southeast of the Caspian Sea. The observed genetic diversity in this study was higher than that reported by DeWoody and Avise (2000) for 13 other freshwater (A_O: 9.1, H_O: 0.46, H_E: 0.54) and 7 anadromous species (H_O and H_E: 0.68). We, therefore, suggest that the wild populations of *R. caspicus* in the investigated regions showed a proper level of genetic diversity.

In comparing the hatchery samples with the wild samples of *C. carpio*, despite non significant differences (Wilcoxon: $P>0.05$), the genetic diversity of hatchery samples was somewhat lower than that in the wild samples. A reduction in the number of unique alleles was also observed in the hatchery samples. Genetic diversity reduction in fish hatchery populations has been previously demonstrated (Li et al. 2016; Wenne et al. 2016). Selection, breeding with limited number of parents and consequently inbreeding depression appeared to be the main explanations for the decrease in genetic diversity of hatchery populations (Li et al. 2016). Tessier et al. (1997) also reported high genetic drift and 50% reduction of effective population size in hatchery populations of Atlantic salmon due to the very small number of parents used for establishing hatchery population. In our study, when two hatchery populations were compared with each other, the Hatchery2 population exhibited a very slightly lower level of genetic diversity which was not significant (Wilcoxon: $P>0.05$). However, the level of genetic variation in Hatchery2 was at an appropriate level. Also, according to the results from bottleneck test, the Hatchery2 population has not experienced a recent bottleneck. This was not surprising because less than 5 years have passed from the establishment of Hatchery2 population and the stocks have renewed annually from the wild during two recent years. However, the Hatchery2 showed the lowest level of genetic diversity compared to the other populations. Although the differences were not statistically significant, even the slightly lower level of genetic variation noticed in the hatchery-produced population should not be ignored because the risk of enhanced inbreeding, subsequent homozygosity and loss of alleles might be arisen over time (Wang et al. 2002). High rate of hatchery larvae release with decreased genetic diversity might produce negative effects on adopted gene pools and subsequently reducing the growth and survival fitness of wild populations (Thanh

et al. 2015). Therefore, to produce healthy young larvae of common carp for the effective hatchery release, it will be critical to maintain and promote the observed genetic variation in hatchery fish using well-organized broodstock management strategies.

Significant departures from HWE were observed in the both hatchery and wild populations after sequential Bonferoni correction. Additionally, significant heterozygosity deficit were detected at some of the loci. The biological reasons for such deficiency are not well detected (Raymond et al. 1997) and several factors are considered to explain it. In our study, the appearance of null alleles is suggested as an important reason for the observed deficiency at the loci MFW7 and MFW28. This has also been the reason for heterozygosity deficit suggested by Berdugo and Barandica (2014) and Biba et al. (2016). We also observed a high level of gene flow between the studied populations which can be a potential explanation for heterozygosity deficiency and deviation from HWE (Bhassu et al. 2004). In recent years, despite the massive release of hatchery-produced larvae in restocking programs, *C. carpio* populations have decreased mainly due to heavy fishing activity and increasing entrance of pollution. This would cause a corresponding decrease in population size which together with overfishing might increase the possibility of inbreeding in some populations (An et al. 2013). Therefore, inbreeding may be a potential explanation for heterozygosity deficiency observed in our study. In hatchery populations, founder effect is considered as a common reason for heterozygosity deficit (Lundrigan et al. 2005). However, only one factor cannot explain the deficit in heterozygocity as the interaction of various factors may contribute to it.

Genetic differentiation among the *C. carpio* populations was measured by F_{ST} and R_{ST} indices. R_{ST} is considered as more effective measure than F_{ST} due to using allelic size information in estimating population differentiation (Wachirachaikarn et al. 2009). For microsatellites, R_{ST} can be higher than F_{ST} under assumption of SMM as mutation model (Slatkin 1995). However, when differentiation is not dependent on mutation model under short time population separation and high rate of migration among populations, the F_{ST} values could be close to the R_{ST} ones (Slatkin 1995). In our study, there was no significant difference in F_{ST} and R_{ST} levels. However, although the R_{ST} values among the populations were slightly higher than those of F_{ST}, the R_{ST} levels were in accordance with those of F_{ST}. We found a low level of differentiation between the studied wild populations, GRR and QR. In addition to the life history, this low level of differentiation may be related to the stock enhancement strategies. The common carp larvae produced from the wild breeders in the hatchery are released yearly in to the rivers of Goraganroud and Qaresou while their breeders are caught only from the Gorganroud River. In this regard, high level of gene flow was observed between these two populations. So, beside the natural migration, releasing the hatchery produced-larvae into the nature without any attention to their parental catch location may be an important explanation for the observed high gene flow and subsequent low differentiation. In comparing the wild and hatchery populations, the lowest differentiation was observed among the Hatchery1 and GRR populations. This was not unexpected because the breeders were originated from the Gorganroud River. However, the highest levels of differentiation were noticed among the Hatchery2 and wild populations. The genetic distance and identity values were also in accordance with the differentiation measures so that the highest genetic distance and the lowest identity were among the Hatchery2 and QR while the lowest genetic distance and the highest identity were observed between the GRR and Hatchery1 samples. Similarly, closer genetic relationship among the GRR and Hatchery1 was revealed by UPGMA dendrogram in which these two samples were the nearest groups while the Hatchery2 was the most distant population which may be related to the hatchery operations such as artificial selection of contributing breeders.

In conclusion, more than 23 years have passed from the beginning of restocking programs for common carp populations. Unfortunately, there is not any information on genetic structure of *C. carpio* wild populations before starting the restocking programs to determine the effect of restocking on genetic diversity of the wild populations. However, according to the comparing our results with those reported for freshwater and anadromous species **(**Dewoody and Avise 2000), it can be said that despite some problems such as pollution, overfishing and restocking programs, the genetic diversity of *C. carpio* in the studied regions is at a proper level. According to our results, no significant differences have observed in genetic variation parameters among the wild and hatchery populations. But since long time has not passed from the establishment of hatchery2 population, even the slightly lower genetic diversity observed in this population should not be ignored. Therefore, we suggest an effort be undertaken to establish adequate broodstock strategies to promote and preserve the observed diversity.

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