African Journal of Agricultural Research Vol. 7(19), pp. 2946-2954, 19 May, 2012
Available online at http://www.academicjournals.org/AJAR
DOI: 10.5897/AJAR11.2170
ISSN 1991-637X ©2012 Academic Journals

Full Length Research Paper

Phylogenetic differentiation of wild and cultured Nile tilapia (*Oreochromis niloticus*) populations based on phenotype and genotype analysis

Samy Yehya El-Zaeem[1,4]*, Mohamed Morsi M. Ahmed[2,3], Mohamed El-Sayed Salama[4] and Walid N. Abd El-Kader[4]

[1]DNA Research Chair, Zoology Department, College of Sciences, P.O. Box 2455, King Saud University, Riyadh 11451, Saudi Arabia.
[2]Biological Sciences Department, Faculty of Sciences, P.O Box 80203, King Abdulaziz University, Jeddah 21589, Saudi Arabia.
[3]Nucleic Acids Research Department, Genetic Engineering and Biotechnology Research Institute (GEBRI), City for Scientific Research and Technology Applications, Alexandria, Egypt.
[4]Animal and Fish Production Department, Faculty of Agriculture, (Saba-Bacha), Alexandria University, Alexandria, Egypt.

Accepted 7 May, 2012

Variation in phenotype based on morphometric character indices and meristic counts and in genotype based on random amplified polymorphic DNA (RAPD) fingerprinting among different wild and cultured Nile tilapia (*Oreochromis niloticus*) populations were analyzed phylogenetically to study and compare the amount of differences in phenotype with the amount of differences in genotype and to help assess the degree of phenotypic plasticity shown by these populations. The results revealed that there were significant differences (P≤0.05) in most of morphometric character indices and meristic counts among different wild and cultured Nile tilapia populations tested. Data of genetic similarity coefficients among four populations of Nile tilapia based on RAPD data of all primers used, showed the highest interpopulation genetic similarity (64%) exhibited between Manzalah and Edku Lake populations, while the lowest genetic similarity (37%) was recorded between Manzalah Lake and cultured populations. The hierarchical cluster analysis based on each phenotype and genotype analysis grouped the four populations into two major category groups: Edku Lake, Manzalah Lake and Nile river populations group and cultured population group. Within these major grouping, wild Nile tilapia were grouped close together. Also, Edku Lake population appears to be more similar to that of Manzalah Lake population than that of Nile river population. Moreover, the great concordance between each phylogeny based on phenotype and genotype analysis revealed that the phenotypic plasticity may not be found in the different Nile tilapia populations tested and the relationship among them considered as intraspecific.

Key words: Phylogenetic, Nile tilapia, morphometric, meristic, random amplified polymorphic DNA (RAPD), phenotype, genotype.

INTRODUCTION

Identification of fish species plays a key role for the behavioral study. Different methods are used for identification but phenotype based on morphometric and meristic are considered as earliest and authentic methods for the identification of fish species in fish biology to measure discreteness and relationships among various taxonomic categories, and provide useful results

*Corresponding author. E-mail: selzaeem@yahoo.com, selzaeem@ksu.edu.sa, samy.elzaeem@alex-agrsaba.edu.eg. Tel: +201003552398, +966592299396.

Table 1. Minimum and maximum of weight and total length of Nile tilapia population samples collected from Manzalah Lake, Nile river, Edku Lake and cultured.

Trait	Manzalah Lake	Nile river	Edku Lake	Cultured
Average weight	124.69±46.98	184.54±41.62	179.82± 90.11	139.86±60.20
Minimum	46.60	111.90	65.00	30.00
Maximum	208.00	225.00	295.00	209.40
Average total length	18.92±2.02	21.30±1.79	20.43±3.14	19.26±3.31
Minimum	13.30	18.40	15.10	12.30
Maximum	21.70	25.00	24.50	23.20

used to differentiate fish populations in particular (Ihssen et al., 1981; Zafar et al., 2002; Costa et al., 2003; Barriga-Sosa et al., 2004; Doherty and McCarthy, 2004; Naesje et al., 2004). Morphometric is the external measurements of an organism, while meristic counts means serial counts of body elements (Talwar and Jhingran, 1992). Morphological characters including meristic counts and body proportions often vary clinically (that is, along a geographic gradient) (Lindsey, 1988).

The technique of random amplified polymorphic DNA (RAPD) marker (Welsh and McCelland, 1990; Williams et al., 1990) has been successfully exploited for stock identification and population analysis in fish. There are many well documented RAPD fingerprinting studies which provide evidence for stock discreteness (Partis and Wells, 1996; Dong and Zhou, 1998; Bartfai et al., 2003; Ruzainah et al., 2003; Wasko et al., 2004; Ahmed et al., 2004; Yan, et al., 2005; El-Zaeem et al., 2006; El-Zaeem and Ahmed, 2006).

Phenotypic plasticity is widespread in nature, and often involves ecologically relevant behavioral, physiological, morphological and life historical traits. Phenotypic plasticity can be inclusively defined as the production of multiple phenotypes from a single genotype, depending on environmental conditions (Hutchings, 2004; Miner et al., 2005).

Recent studies on the variation of molecular characters among fish populations have been made to compare the degree of variation in these molecular traits with that of morphological characters and, therefore, to help assess the degree of phenotypic plasticity shown by a species. Studies of morphological character variation are, therefore, vital in order to elucidate patterns observed in phenotypic and genetic character variation among fish populations (Beheregaray and Levy, 2000).

Therefore, the aim of the present work was to analyze the phylogenetic differentiation based on phenotype and genotype analysis and to compare the amount of differences in phenotype based on morphometric character indices and meristic counts with the amount of differences in genotype based on random amplified polymorphic DNA (RAPD) fingerprinting among different wild (Manzalah Lake, Nile river and Edku Lake) and cultured Nile tilapia (*Oreochromis niloticus*) populations, and to help assess the degree of phenotypic plasticity shown by these populations.

MATERIALS AND METHODS

The present study was carried out at Fish Breeding and Production Laboratory, Animal and Fish Production Department, Faculty of Agriculture (Saba-Bacha), Alexandria University and Nucleic Acids Research Department, Genetic Engineering and Biotechnology Research Institute (GEBRI), City for Scientific Research and Technology Applications, Alexandria, Egypt.

Specimen collection

From both sex, fifty mature individuals of each of wild and cultured Nile tilapia, (*O. niloticus*) populations were randomly collected from Manzalah Lake, Nile River, Edku Lake and cultured, by professional fishermen (Table 1).

Quantitative phenotype analysis

A total of 28 morphometric characters and 8 meristic counts were recorded within each population as described by Bagenal (1978), Ezzat et al. (1979), Taniguchi et al. (1996), Sahu et al. (2000), Costa et al. (2003) and Doherty and McCarty (2004). Condition factor of each species were measured using the following equation:

$K= (W/L^3)\ 100$ (Lagler, 1956)

Where W = body weight (g) and L= total length (cm). The morphometric characters and meristic counts are listed in Table 2. All morphometric characters were transformed by dividing the measurment by either total or standard length or head length of each fish to minimize the effect of fish size (Table 4). Data were statistically analyzed using the following model (CoStat, 1986):

$Y_{ij=}\ \mu_{+}\ T_{i+}\ e_{ij}$

Where, Y_{ij}: Observation of the ij^{th} parameter measured; μ: overall mean; T_i: effect of i^{th} population; e_{ij}: random error. Significant differences ($P \leq 0.05$) among means were tested by the method of Duncan (1955).

Phenotype differentiation among cultured and wild Nile tilapia populations based on morphometric character indices and meristic counts was analyzed by means of hierachical cluster analysis of the SPSS 12.0 (1999) software package. The cluster analysis using average linkage between-groups method (Sneath and Sokal, 1973) was performed on the matrix of Euclidian distances in order to depict hierarchically the shape differences. The results were presented as a dendrogram.

Genotype analysis

Genotype analysis was performed based on random amplified

Table 2. Quantitative phenotype traits based on morphometric characters and meristic counts used for differentiation analysis among cultured and wild Nile tilapia, *O. niloticus* populations.

Characters	Acronyms
Morphometric analysis	
Total length	TL
Standard length	SL
Head length	HL
Body depth	BD
Body width	BW
Head width	HW
Abdomen length	AL
Caudal peduncle length	CPL
Caudal peduncle depth	CPD
Caudal peduncle width	CPW
Orbit diameter	OD
Pre-orbital length	Pr-OL
Post-orbital length	Po-OL
Trunk length	TrL
Dorsal fin base length	DFBL
Pelvic fin base length	Pel FBL
Pelvic fin length	Pel FL
Anal fin base length	AFBL
Caudal fin length	CFL
Pectoral fin length	Pec FL
Length of longest dorsal fin spine	LLoDFS
Length of last dorsal fin spine	LLaDFS
Length of longest dorsal fin ray	LLoDFR
Length of last dorsal fin ray	LLaDFR
Length of longest anal fin spine	LLoAFS
Length of last anal fin spine	LLaAFS
Length of longest anal fin ray	LLoAFR
Length of last anal fin ray	LLaAFR
Meristic analysis	
Dorsal fin spines count	DFSC
Dorsal fin rays count	DFRC
Pelvic fin spines count	Pel FSC
Pelvic fin rays count	Pel FRC
Pectoral fin rays count	Pec FRC
Anal fin spines count	AFSC
Anal fin rays count	AFRC
Caudal fin rays count	CFRC

polymorphic DNA (RAPD) fingerprinting. DNA was extracted from liver tissues of each Nile tilapia population according the method described by Bardakci and Skibinski (1994).

In this work, ten base long oligonucleiotide primers were used to initiate PCR amplifications. Primers were randomly selected on the basis of GC content and annealing temperature for RAPD-PCR amplification (Table 3).

PCR amplifications were performed according the procedure of Williams et al. (1990, 1993). The reaction (25 µl) was carried out in a mixture consisting of 0.8 U of Taq DNA polymerase (Fanzyme), 25 pmol dNTPs, and 25 pmol of random primer, 2.5 µl 10× Taq DNA polymerase buffer and 40 ng of genomic DNA. The final reaction mixture was placed in a DNA thermal cycler (ependorf). The PCR programme included an initial denaturation step at 94°C for 2 min, followed by 45 cycles with 94°C for 30 s for DNA denaturation, annealing as mentioned with each primer (Table 3), extension at 72°C for 30 s and final extension at 72°C for 10 min were carried out. Samples were cooled at 4°C.

The amplified DNA fragments were separated on 2.5% agarose gel and stained with ethidium bromide. ΦX174 DNA Ladder marker (1353, 1078, 872, 310,……72 bp) was used in this study. The amplified patterns were visualized on an UV transilluminator and photographed by gel documentation system.

RAPD patterns were analyzed and scored from photographs. For the analysis and comparison of the patterns, a set of distinct, well-separated bands were selected. The genotypes were determined by recording the presence (1) or absence (0) in the RAPD profiles. Genetic similarity (GS) between individuals i and j was calculated according the formula given as (Nei and Li, 1979):

$$B_{ij} = 2\ N_{ij}/(N_i + N_j)$$

Where N_{ij} is the number of common bands observed in individuals i and j, and N_i and N_j are the total number of bands scored in individuals i and j respectively, with regard to all assay units. Thus, GS reflects the proportion of bands shared between two individuals and ranges from zero (no common bands) to one (all bands identical).

Genotype differentiation among cultured and wild Nile tilapia populations based on RAPD fingerprinting was analyzed by means of hierachical cluster analysis of the SPSS 12.0 (1999) software package. The dendrogram was constructed using the average linkage between groups, and the data matrix so generated was used for calculation of similarity matrix for all primers based on Jaccard's coefficients method (Jaccard, 1908).

RESULTS

Quantitative phenotype analysis

Mean values of morphometric character indices were compared among different Nile tilapia (*O. niloticus*) populations. The highest mean indices of BD/SL, Pel FBL/SL and LLoAFR/SL were obtained by Edku Lake population and differed significantly (P≤0.05) from those of the the other population tested. The highest mean value indices of BW/SL, HW/HL, Po-OL/HL, LLoAFS/SL and LLaAFS/SL were achieved by Manzalah Lake population, but did not differ significantly (P≤0.05) from those of Edku Lake population (Table 4).

The highest mean value of TrL/SL was recorded by cultured population and differed significantly (P≤0.05) from that of the other population tested. The highest mean value of CPL/SL was detected by Nile river population, but did not differ significantly (P≤0.05) from that of cultured population. The highest mean value of CPD/SL and LLaDFR/SL were found by Nile river population and differed significantly (P≤0.05) from those of the other populations. The highest records of CPW/SL, HL/SL and Pec FL/SL indices were observed by Nile river population, but did not differ significantly (P≤0.05) from

Table 3. The sequences, GC% and the annealing temperatures of the primers used.

Primers	Sequence 5`- 3`	GC%	Annealing temperature (°C/s)
1	AGG CCC CTG T	70	28/30
2	ATG CCC CTG T	60	28/30
3	AAA GCT GCG G	60	28/30
4	ACC GCC GAA G	70	28/30
5	AGG GGT CTT G	60	28/30
6	CCA GCC GAA C	70	28/30

Table 4. Means and standard deviation of quantitative phenotype traits based on morphometric character indices and meristic counts used for differentiation analysis among different Nile tilapia (*O. niloticus*) populations.

Characters	Populations			
	Manzalah	Nile	Edku	Cultured
Morphometric indices				
BD/SL	0.400±0.02^{b}	0.404±0.01^{b}	0.451±0.03^{a}	0.385±0.03^{c}
BW/SL	0.181±0.01^{a}	0.174±0.01^{b}	0.178±0.01^{ab}	0.163±0.01^{c}
TrL/SL	0.573±0.05^{b}	0.573±0.02^{b}	0.582±0.03^{b}	0.603±0.04^{a}
CPL/SL	0.115±0.01^{c}	0.128±0.01^{a}	0.123±0.01^{b}	0.126±0.01^{ab}
CPD/SL	0.153±0.01^{bc}	0.161±0.01^{a}	0.157±0.01^{b}	0.150±0.01^{c}
CPW/SL	0.055±0.01^{ab}	0.062±0.01^{a}	0.047±0.01^{b}	0.052±0.01^{b}
HL/SL	0.345±0.02^{a}	0.346±0.01^{a}	0.324±0.01^{b}	0.328±0.01^{b}
HW/HL	0.573±0.05^{a}	0.549±0.02^{b}	0.573±0.01^{a}	0.540±0.05^{b}
OD/HL	0.269±0.02^{b}	0.264±0.01^{b}	0.284±0.04^{a}	0.284±0.03^{a}
Pr-OL/HL	0.327±0.02^{b}	0.339±0.02^{a}	0.344±0.03^{a}	0.346±0.03^{a}
Po-OL/HL	0.495±0.03^{a}	0.480±0.02^{b}	0.485±0.03^{ab}	0.451±0.03^{c}
CFL/TL	0.215±0.01^{a}	0.207±0.01^{a}	0.206±0.01^{a}	0.209±0.02^{a}
LLoDFR/SL	0.250±0.03^{b}	0.268±0.03^{a}	0.258±0.04^{ab}	0.221±0.03^{c}
LLaDFR/SL	0.060±0.01^{c}	0.085±0.01^{a}	0.077±0.01^{b}	0.075±0.01^{b}
LLoDFS/SL	0.153±0.01^{a}	0.142±0.01^{b}	0.140±0.02^{b}	0.143±0.02^{b}
LLaDFS/SL	0.153±0.01^{a}	0.142±0.01^{b}	0.140±0.02^{b}	0.143±0.02^{b}
DFBL/SL	0.610±0.03^{b}	0.621±0.02^{a}	0.626±0.02^{a}	0.586±0.04^{c}
PelFL/SL	0.314±0.03^{a}	0.286±0.02^{b}	0.295±0.03^{b}	0.245±0.02^{c}
PelFBL/SL	0.052±0.01^{bc}	0.053±0.01^{b}	0.066±0.01^{a}	0.050±0.01^{c}
LLoAFR/SL	0.238±0.04^{b}	0.228±0.03^{b}	0.254±0.03^{a}	0.241±0.03^{b}
LLaAFR/SL	0.078±0.01^{a}	0.064±0.01^{a}	0.060±0.01^{a}	0.60±1.41^{a}
LLoAFS/SL	0.149±0.01^{a}	0.139±0.01^{b}	0.146±0.02^{a}	0.138±0.01^{b}
LLaAFS/SL	0.149±0.01^{a}	0.139±0.01^{b}	0.146±0.02^{a}	0.138±0.01^{b}
AFBL/SL	0.192±0.01^{b}	0.200±0.01^{a}	0.203±0.02^{a}	0.184±0.03^{b}
Pec.FL/SL	0.390±0.04^{a}	0.398±0.02^{a}	0.357±0.03^{b}	0.328±0.03^{c}
PecFBL/SL	0.067±0.01^{b}	0.075±0.01^{a}	0.075±0.01^{a}	0.069±0.01^{b}
Meristic character				
DFSC	16.46±0.50^{b}	17.00±0.00^{a}	16.54±0.50^{b}	17.00±0.00^{a}
DFRC	12.56±0.50^{c}	12.00±0.00^{d}	13.00±0.00^{a}	12.88±0.33^{b}
PelFSC	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00
PelFRC	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00
PecFRC	13.36±0.48^{b}	14.00±0.00^{a}	13.26±0.44^{b}	12.26±0.44^{c}
AFSC	3.00±0.00	3.00±0.00	3.00±0.00	3.00±0.00
AFRC	9.74±0.44^{b}	10.12±0.48^{a}	10.00±0.00^{a}	9.46±0.86^{c}
CFRC	16.00±0.00^{b}	16.00±0.00^{b}	15.82±0.56^{c}	16.28±0.45^{a}

Table 4. Count'd.

Length-weight relationship				
K	1.9±0.31	1.9±0.19	1.9±0.19	1.8±0.15

Mean values in the same row having the same letters do not differ significantly (P ≤0.05).

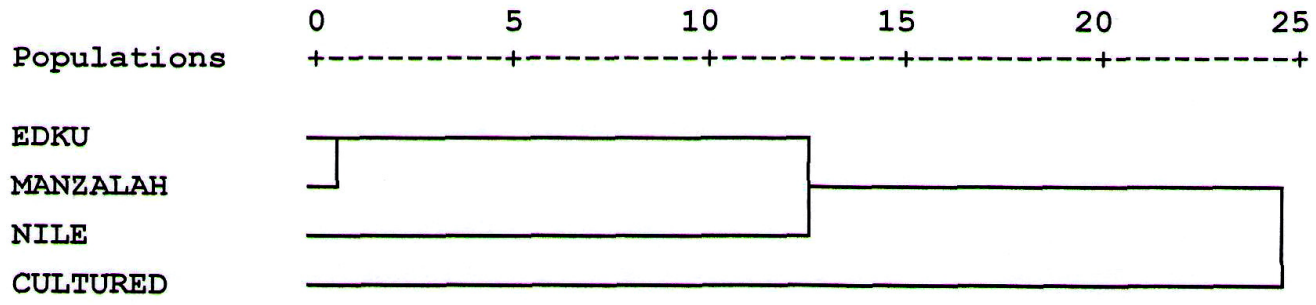

Figure 1. Dendrogram using average linkage (between groups) of different Nile tilapia populations based on morphometric character indices and meristic counts as shown by hierarchical cluster analysis.

those of Manzalah Lake population (Table 4).

The lowest mean value of OD/HL was obtained by Nile river population, but did not differ significantly (P≤0.05) from that of Manzalah Lake population. Moreover, the lowest mean value of Pr-OL was achieved by Manzalah Lake population and differed significantly (P≤0.05) from those of the other populations. Results of CFL/TL and LLaAFR/SL showed insignificant differences (P≤0.05) among different populations. The mean values indices of LLoDFR/SL, DFBL/SL and Pel FL/SL were significantly decreased (P≤0.05) by cultured population, showing lower mean compared with the other populations. The highest mean values of LLoDFS/SL and LLaDFS/SL were found by Manzalah Lake population and differ significantly (P≤0.05) from those of the other populations. The highest mean value indices of AFBL/SL was obtained by Edku Lake population, but did not differ significantly (P≤0.05) from that of Nile river population. The lowest mean value indices of Pec FBL/SL was recorded by Manzalah Lake population, but did not differ significantly (P≤0.05) from that of cultured population (Table 4).

The hierarchical cluster analysis based on quantitative phenotype (morphometric and meristic character indices), grouped the four populations into two major category groups; Edku Lake, Manzalah Lake and Nile river populations group and cultured population group (Figure 1). Within these major grouping, wild Nile tilapia were grouped close together. A dendrogram also showed that Edku Lake population appears to be more phenotypically similar to that of Manzalah Lake population than that of Nile river population (Figure 1).

Genotype analysis

All DNA samples from different Nile tilapia populations were examined using (RAPD) fingerprinting. Six random primers were used to determine DNA fingerprinting diversity in the different Nile tilapia populations. All the different primers used in this work, produced different RAPD band patterns (Figure 2). The number of amplified bands detected varied, depending on the primers and population. Moreover, to ensure that the amplified DNA bands originated from genomic DNA, and not from primer artifacts, negative control was done for each primer/population combination. No amplification was detected in the control reactions. All amplification products were found to be reproducible when reactions were repeated using the same reaction conditions (Figure 2).

Data of genetic similarity coefficients among four populations of Nile tilapia based on RAPD data of all primers used showed the highest interpopulation genetic similarity (64%) exhibited between Manzalah and Edku Lake populations. While, the lowest genetic similarity (37%) was recorded between Manzalah Lake and cultured populations (Table 5).

Moreover, RAPD analysis was used for constructing parsimony tree depicting relationships among the different Nile tilapia populations studied. The hierarchical cluster analysis based on RAPD fingerprinting, grouped the four populations into two major category groups. Within these major grouping, wild Nile tilapia (Manzalah Lake, Edku Lake and Nile river) populations were grouped close together. Also, a dendrogram showed that

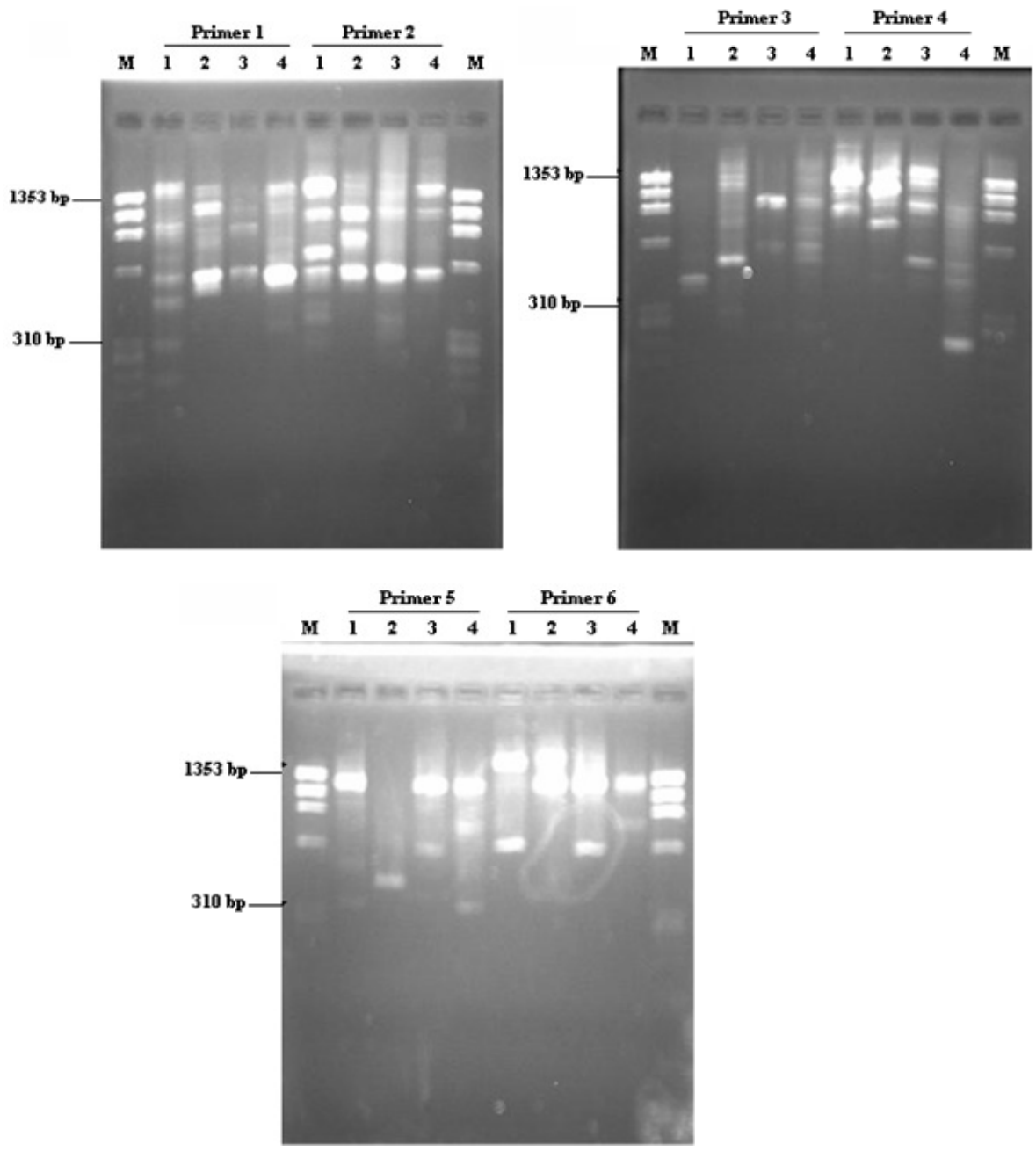

Figure 2. Patterns in different Nile tilapia populations obtained with different primers. Lane M: ΦX174 DNA markers, the lanes (1 to 4) of each primer are: Manzalah Lake, Nile River, Edku Lake and cultured populations, respectively.

Table 5. Genetic similarity coefficients among four populations of Nile tilapia based on RAPD data of all primers used.

Populations	Manzalah	Nile	Edku	Cultured
Manzalah	-	58.0	64.0	37.0
Nile	-	-	46.0	39.0
Edku	-	-	-	53.0
Cultured	-	-	-	-

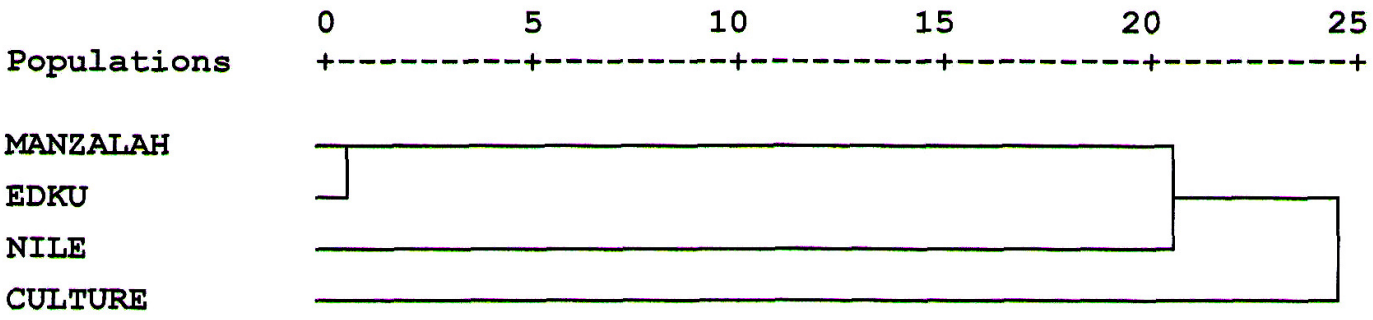

Figure 3. Dendrogram using average linkage (between groups) of different Nile tilapia populations based on RAPD fingerprinting as shown by hierarchical cluster analysis.

Manzalah Lake population appears to be more genetically similar to that of Edku Lake population than that of Nile river population (Figure 3).

DISCUSSION

Phenotypic based morphometric character and meristic counts have been commonly used in fisheries biology to measure discreteness and relationship among various taxonomic categories (Shepherd, 1991; Avsar, 1994; Haddon and Willis, 1995; Bembo et al., 1996; Turan, 1999; Anene, 1999; North et al., 2002; Costa et al., 2003; Barriga-Sosa et al., 2004). The main advantages of RAPD markers are the possibility of working with anonymous DNA and the relatively low expense; it is also a fast and simple to produce RAPD marker (Hadrys et al., 1992; Elo et al., 1997; Ali et al., 2004; El-Zaeem et al., 2011; El-Zaeem 2011a, b). Moreover, RAPD analysis might be useful for systematic investigation at the level of species and subspecies (Bardakci and Skibinski, 1994), and more sensitive and technically easier to perform and produce results with low statistical error, whereas, DNA fingerprinting detected greater genetic differentiation between Nile tilapia stains than other molecular techniques such as multilocus minisatellite marker (Naish et al., 1995). Sandoval-Castellanos et al. (2007) successfully used RAPD marker to evaluate and determine the genetic differentiation of different populations of jumbo squid (*Dosidicus gigas*). Chen et al. (2009) reported that RAPD marker can be used to distinguish gender in Yellow River carps (*Cyprinus carpio*). Klinbunga et al. (2010) showed that RAPD marker was successfully used for species identification of the blue swimming crab (*Portunus pelagicus*) in Thailand waters. The results of the present work are consistent with these findings.

Body shape in fish has been demonstrated to be influenced by environmental factors (Beacham, 1990; Robinson and Wilson, 1995). Plasticity in fish body shape has been hypothesized to be adaptive (Robinson and Parsons, 2002). Such phenotypic adaptations do not necessarily result in genetic changes in the population (Ihssen et al., 1981; Allendorf, 1988) and thus the detection of such phenotypic differences among populations cannot usually be taken as evidence of genetic differentiation.

Clayton (1981) reported that the major limitation of morphological characters at the intra-specific level is that phenotypic variation is not directly under genetic control but subjected to environmental modification. Hutchings (2004) demonstrated that phenotypic plasticity arises when the same genotype produces different phenotypes in different environments. The results of the present work are consistent with these findings.

The phylogeny of different Nile tilapia (*O. niloticus*) populations is considered problematic at the intraspecific level. Such a difficulty arises from highly homogeneous feature and morphology displayed by the different Nile tilapia populations and consequently, from the paucity of the key morphological characters suitable to address their phylogeny and evolution. In the present work, we have approached and compared the phylogenetic of different Nile tilapia populations based on each phenotype and genotype analysis. The results, presented by two dendrograms, proved that the amount of differences in genotype among wild and cultured Nile tilapia populations reflected the same amount of differences in phenotype among the same populations. Such phenotypic differences among these populations can be taken as evidence of genetic differentiation. Also, the results of genetic analysis confirmed the existing taxonomic system based on phenotype analysis. Moreover, the great concordance between each phylogeny based on phenotype and genotype analysis revealed that the phenotypic plasticity may not be found in the different Nile tilapia populations tested, and the relationship among them considered as intraspecific.

Therefore, it was observed in this study that either the phenotype analysis based on a large number of morphometric character indices and meristic counts, or the genotype analysis based on RAPD fingerprinting can

be used to discriminate fish populations with the same results up to the intraspecific level, or both the phenotype and genotype analyses can be used to assess the degree of phenotypic plasticity shown by populations.

ACKNOWLEDGEMENT

This project was supported by King Saud University, Deanship of Scientific Research, College of Science Research Center.

REFERENCES

Ahmed MMM, Ali BA, El-Zaeem SY (2004). Application of RAPD markers in fish: Part I- Some genera (*Tilapia, Sarotherodon* and *Oreochromis*) and species (*Oreochromis aureus* and *Oreochromis niloticus*) of Tilapia. Int. J. Biotechnol., 6: 86-93.

Ali BA, Huang T, Qin D, Wang X (2004). A review of random amplified polymorphic DNA (RAPD) markers in fish research. Rev. Fish Biol. Fish, 14: 443-453.

Allendorf FW (1988). Conservation biology of fishes. Conser Biol., 2: 145-148.

Anene A (1999). Morphometric and meristic description of *Tilapia mariae* (Boulenger, 1901) and *Tilapia zillii* (Gervais, 1848) from the Umuoseriche Lake in the freshwater reaches of the Niger delta floodplains. Acta Hydrobiol., 41: 211-218.

Avsar D (1994). Stock differentiation study of the sprat off the southern coast of the Black sea. Fish. Res., 19: 363-378.

Bagenal TB (1978). Methods for assessment of fish production in fresh waters (3rd ed). Oxford- London, Blackwell Sci, Publ., p. 365.

Bardakci F, Skibinski DOF (1994). Application of the RAPD technique in tilapia fish: species and subspecies identification. Hereditary, 73: 117-123.

Barriga-Sosa DLA, Jimenez-Badillo MDL, Ibanez AL, Arredondo-Figueroa JL (2004). Variability of Tilapias (*Oreochromis* Spp.) introduced in Mexico: morphometric, meristic and genetic characters. J. Appl. Ichthyol., 20: 7-14.

Bartfai R, Egedi S, Yue GH, Kovacs B, Urbanyi B, Tamas G, Horvath L, Orban L (2003). Genetic analysis of two common carp broodstocks by RAPD and microsatellite markers. Aquaculture, 219: 157-167.

Beacham TD (1990). A genetic analysis of meristic and morphometric variation in chum salmon (*Oncorhynchus keta*) at three different temperatures. Can. J. Zool., 68: 225-229.

Bembo DG, Carvalho GR, Cingolani N, Ameri E, Gianetti G, Pitcher TJ (1996). Allozymic and morphometric evidence for two stocks of the European anchovy Engraulis encrasicolus in Adriatic waters. Mar. Biol., 126: 529-538.

Beheregaray LB, Levy JA (2000). Population genetics of the silverside *Odontesthes argentinensis* (Teleostei, *Atherinopsidae*): evidence for speciation in an estuary of southern Brazil. Copeia, 2: 444-447.

Chen J, Wang Y, Yue Y, Xia X, Du Q, Chang Z (2009). A novel male-specific DNA sequence in the common carp, *Cyprinus carpio*. Mol. Cell. Prob., 23: 235-239.

Clayton JW (1981). The stock concept and the uncoupling of organismal and molecular evolution. Can. J. Fish. Aquat. Sci., 38: 1515-1522.

Costa JL, De Almeida PR, Cost MJ (2003). A morphometric and meristic investigation of Lusitanian toadfish *Halobatrachus didactylus* (Bloch and Schneider, 1801): evidence of population fragmentation on Portuguese coast. Sci. Mar., 67: 219-231.

CoStat (1986). CoStat 3.03, Copyright, Co Hort Software. P. O. Box 1149, Berkeley, CA 94701, USA.

Doherty D, McCarty TK (2004). Morphometric and meristic characteristics analysis of two western Irish populations of Arctic char, *Salvelinus alpinus* (L.). Biology and Environment: proceedings of the Royal Irish Academy, 104: 75-85.

Dong Z, Zhou E (1998). Application of the random amplified polymorphic DNA technique in a study of hetrosis in commen carp, *Cyprinus carpio* L. Aquacult. Res., 29: 595-600.

Duncan DB (1955). Multiple range and multiple F test. Biometrics., 11: 1-42.

Elo K, Ivanoff S, Vuorinen JA, Piironen J (1997). Inheritance of RAPD markers and detection of interspecific hybridization with brown trout and Atlantic salmon. Aquaculture, 152: 55-65.

El-Zaeem SY, Ahmed MMM (2006). Genetic differentiation between sex reversal and normal of full-sib Nile Tilapia (*Oreochromis niloticus*) based on DNA finger printing. Res. J. Fish. Hydrobiol., 1: 1-5.

El-Zaeem SY, Ali BA, Ahmed MMM (2006). Random amplified polymorphic DNA fingerprint and genetic similarity among four genera and between two phenotypes of cultured carps in Egypt. Int. J. Agri. Biol., 8: 111-115.

El-Zaeem SY, Ahmed MMM, Salama ME, El-Maremie HA (2011). Production of salinity tolerant Nile tilapia, *Oreochromis niloticus* through traditional and modern breeding methods: II. Application of genetically modified breeding by introducing foreign DNA into fish gonads. Afr. J. Biotechnol., 10(4): 684-695.

El-Zaeem SY (2011a). Phenotype and genotype differentiation between flathead grey mullet [*Mugil cephalus*] and thinlip grey mullet [*Liza ramada*] (Pisces: *Mugilidae*). Afr. J. Biotechnol., 10(46): 9485-9492.

El-Zaeem SY (2011b). Growth comparison of Nile tilapia (*Oreochromis niloticus*) and Blue tilapia, (*Oreochromis aureus*) as affected by classical and modern breeding methods. Afr. J. Biotechnol., 10(56): 12071-12078.

Ezzat AA, Hashem MJ, El Gharabawy MM (1979). Biometric variations in (*Solea vulgaris*) acclimatized in Lake Quarum, Upper Egypt. J. Fish Biol., 14: 39-46.

Haddon M, Willis TJ (1995). Morphometric and meristic comparison of Orange Roughy (*Hoplostethus attanticus*, *Trachichthyidae*) from the puysegur Bank and Lord-Howe-Rise, New-Zealand, and its implications for stock structure. Mar. Biol., 123: 19-27.

Hadrys H, Balick M, Schierwater B (1992). Applications of random amplified polymorphic DNA (RAPD) in molecular ecology. Mol. Ecol., 1: 55-63.

Hutchings JA (2004). Norms of reaction and phenotypic plasticity in salmonid life histories. In: Hendry AP; Stearns SC, eds. Evolution illuminated; salmon and their relatives. New York, NY: Oxford University Press, 154-174.

Ihssen PE, Booke HE, Casselman JM, McGlade JM, Payne NR, Utter FM (1981). Stock identification: Materials and methods. Can. J. Fish. Aquat. Sci., 38: 1838-1855.

Jaccard P (1908). Nouvelles recherches sur la distribution florale. Bulletin Soc. Vaudoisc Sci. Nat., 44: 223-270.

Klinbunga S, Thamniemdee N, Yuvanatemiya V, Khetpu K, Khamnamtong B, Menasveta P (2010). Species identification of the blue swimming crab *Portunus pelagicus* in Thailand waters using mtDNA and RAPD-derived SCAR markers. Aquaculture, 308: 539-546.

Lagler KF (1956). Fresh water fishery biology. W.M.C. Brown Company Publishers, Dubuque, Iowa, p. 421.

Lindsey CC (1988). Factors controlling meristic variation. In Fish physiology, Vol. XI-B (Hoar WS, Randall DJ, eds), pp. 197-274. San Diego, CA: Academic Press.

Miner BG, Sultan SE, Morgan SG, Padilla DK, Relyea RA (2005). Ecological consequences of phenotypic plasticity. Tren. Ecol. Evol., 20: 685-692.

Naesje TF, Vuorinen JA, Sandlund OT (2004). Genetic and morphometric differentiation among sympatric spawning stocks of white fish (*Coregonus lavaretus* L.) in Lake Femund, Norway. J. Limnol., 63: 233-243.

Naish KA, Warren M, Bardakci F, Skibinski DO, Carvalho GR, Mair GC (1995). Multilocus DNA fingerprinting and RAPD reveal similar genetic relationships between strains of *Oreochromis niloticus* (Pisces: *Cichlidae*). Mol. Ecol., 4: 271-274.

Nei M, Li WH (1979). Mathematical modelling for studying genetic variation in terms of restriction endonucleases. Proc. Natl. Acad. Sci. USA., 76: 5269-5273.

North JA, Farr RA, Vescei P (2002). Acomparison of meristic and morphometric characters of green sturgeon, *Acipencer medirostris*. J. Appl. Ichthyol., 18: 234-239.

Partis L, Wells RJ (1996). Identification of fish species using random amplified polymorphic DNA (RAPD). Mol. Cell. Prob., 10: 435-441.
Robinson BW, Parsons KJ (2002). Changing times, spaces, and faces: tests and implications of adaptive morphological plasticity in the fishes of northern postglacial lakes. Can. J. Fish. Aquat. Sci., 59: 1819-1833.
Robinson BW, Wilson DS (1995). Experimentally induced morphological diversity in Trinidadian guppies (*Poecilia reticulata*). Copeia, 2: 294-305.
Ruzainah A, Siti Azizah MN, Patimah I, Amirrudin A (2003). RAPD fingerprinting of the eel-loaches (*Pangio filinaris*) and (*Pangio piperata*) preliminary evolution. Aquacult. Res., 44: 959-965.
Sahu BB, Meher PK, Mohanty S, Reddy PVGK, Ayyappan S (2000). Evaluation of the carcass and commercial characteristics of Carps. Naga, The ICLARM Quarterly, 23(2): 10-14.
Sandoval-Castellanos E, Uribe-Alcocer M, Diaz-Jaimes P (2007). Population genetic structure of Jumbo squid (*Dosidicus gigas*) evaluated by RAPD analysis. Fish. Res., 83: 113-118.
Shepherd G (1991). Meristic and morphometric variation in Black sea Bass North of Cape Hatteras, North Carolina. Am. J. Fish. Manag., 11: 139-149.
Sneath PHA, Sokal RR (1973). Numerical taxonomy: the principles and practice of numerical classification. San Francisco: WH Freeman, p. 573.
SPSS (1999). SPSS 12.0. Copyright, IBM Corporation. Inc., 233 S. Wacker, 11th Floor, Chicago, IL. 60606-6307, USA.
Talwar PK, Jhingran AG (1992). Inland fishes of India. Rec. Ind. J., 3: 19-24.
Taniguchi N, Yamasaki M, Takagi M, Tsujimura A (1996). Genetic and environmental variances of body size and morphological traits in communally reared clonal lines from gynogenetic diploid ayu, *Plecoglossus altivelis*. Aquaculture, 140: 333-341.
Turan C (1999). A note on the examination of morphometric differentiation among fish population: The truss system. Tr. J. Zool., 23: 259-263.
Wasko AP, Martins C, Oliveira C, Senhorini JA, Foresti F (2004). Genetic monitoring of the Amazonian fish matrincha (*Brycon cephalus*) using RAPD markers: insights into supportive breeding and conservation programmes. J. Appl. Ichthyol., 20: 48–52.
Welsh J, McCelland M (1990). Fingerprinting genomes using PCR with arbitrary primers. Nucl. Acid Res., 18: 7213-7218.
Williams JGK, Kubelik AR, Livak KJ, Rafalski JA, Tingey SV (1990). DNA polymorphisms amplified by arbitrary primers are useful as genetic markers. Nucl. Acid Res., 18: 6531-6535.
Williams JGK, Hanafey MK, Rafalaski JA, Tingey SV (1993). Genetic analysis using RAPD markers. Meth. Enzymol., 218: 704-740.
Yan J, Liu S, Sun Y, Zhang C, Luo K, Liu Y (2005). RAPD and microsatellite analysis of diploid gynogens from allotetraploid hybrids of red Crucian carp (*Carassius auratus*) × common carp (*Cyprinus carpio*). Aquaculture, 243: 49-60.
Zafar M, Nazir A, Akhtar N, Naqvi SMHM, Zia-ur-Rehman M (2002). Studies on meristic counts and morphometric measurments of Mahseer (*Tor putitora*) from a spawning ground of himalayan Foot-hill River korang Islamabad, Pakistan. Pakistan J. Biol. Sci., 5(6): 733-735.