5. SUMMARY AND CONCLUSION

The present study was carried out to determine the morphometric and genetic variation within three populations of mullet (*Mugil cephalus*, *Liza ramada* and *Valamugil seheli)* collected from three different geographical areas in Egypt during the period from early April 2012 to the end of March 2013.

The laboratory work was carried out at Animal Production Department, Faculty of Agriculture, Fayoum University, Fayoum governorate, Egypt and amplified polymorphic DNA (RAPD) analysis was carried out at Genetic Engineering Laboratory, Faculty of Agriculture, Cairo University.

Seven hundred and fifty specimens of mullet from three geographic areas in Egypt were studied morphometrically. Nineteen morphometric traits were taken on each specimen. Data obtained were subjected to factor analysis using a principal component analysis (PCA) and discriminant function analysis (DFA). Morphometric measurements were: total length (TL), fork length (FL), head length (HL), distance of first dorsal fin (Dl), distance of second dorsal fin (D2), distance of ventral fin (DV), distance of the central of anus (DCA), distance of anal fin (DA), maximum body height (MBH), eye diameter (ED), caudal peduncle depth (CPD), dorsal fin length (DFL), snout length (NL), head depth (HD), width (W), pectoral fin length (PecFL), tail length (TailL), body weight (BW) and condition factor (CF).

5.1. Morphometric analysis

1- The results from principal component analysis (PCA) showed that, *Mugil cephalus* populations from Qarun, Burullus and Manzala lakes had higher degree of morphometric polymorphism. Which were reflected in variation primarily along three axes PC1, PC2 and PC3. Together these components accounted for more than 87.96% of variation. Univariate analysis exhibited significant differences among populations for most characters which were enabled to separate along PCI and PC2. BW, TL, FL, ED, and MBH ratios were selected by step-wise discriminant function analysis which indicated significantly three different morphotypes. Two canonical functions accounted for 100% of the variance, with only first function accounted 70.05% of variance. First function separated Manzala population which scored positively (> zero), while, the second function successfully separated Burullus population scoring negatively (< zero).

2- PC1, PC2 and PC3, accounted for more than 76.24% of observed morphometric variation in *Liza ramada* collected from Lake Qarun, Lake Manzala and Lake Burullus in Egypt. The PC1 accounted for 34.28% of variance. PC2 explained 22.51% of the variance. Morphological data of all populations of *Liza ramada* discerned the three major groups from each other. Univariate analysis exhibited significant differences among populations for all characters except body weight and eye diameter (ED) ratio, and enabled to separate along PCI and PC2. BW, CPD, PecFL, MBH and CF ratios were selected by step-wise discriminant function analysis indicated significantly three different morphotypes. Two canonical functions accounted for 100% of variance, First function accounted 80.11% of

variance. First function separated Qarun population which scored positively (> zero) while the second function successfully separated Burullus population which scored negatively (< zero).

3- PC1, PC2 and PC3, accounted for more than 77.87% of observed morphometric variation in *Valamugil seheli* collected from Lake Qarun, Lake Manzala and Lake Burullus in Egypt. The PC1 accounted for 43.2% of variance. PC2 explained 21.23% of the variance. Morphological data of all populations of Valamugil seheli discerned the three major groups from each other. Univariate analysis exhibited significant differences among populations for all characters which were separated along PCI and PC2. BW, CPD, DFL, PecFL, MBH and CF ratios were selected by step-wise discriminant function analysis indicated significantly three different morphotypes. Two canonical functions accounted for 100% of variance the First function accounted 80.72% of the variance. First function separated Manzala population scoring negatively (< zero) while the second function successfully separated Burullus population which scored positively (> zero).

4- PC1, PC2 and PC3, accounted for more than 64.76% of observed morphometric variation in *Mugil cephalus* collected from farms around Qarun, Manzala and Burullus in Egypt. PC1 accounted for 27.15% of variance. PC2 explained 22.47% of the variance. Morphological data of all populations of *Mugil cephalus* farms discerned the three major groups from each other. Univariate analysis exhibited significant differences among populations from all characters which were separated along PCI and PC2. BW, PecFL, DA, HD and CF ratios were selected by step-wise discriminant

function analysis from three *Mugil cephalus* populations, These characters indicated significantly three different morphotypes. Two canonical functions in the step-wise discriminant function analysis accounted for 100% of the variance (First function 68.58% and second function 31.42%). First function separated Burullus population which scored positively (> zero) while the second function successfully separated Qarun population which scored negatively (< zero).

5- PC1, PC2 and PC3, accounted for more than 77.76% of observed morphometric variation in *Liza ramada* collected from farms around lakes Qarun, Manzala and Burullus in Egypt. The PC1 accounted for 47.5% of variance. PC2 explained 17.39% of the variance. Morphological data of all populations of *Liza ramada* farms discerned the three major groups from each other. Univariate analysis exhibited significant differences among populations from all characters except body weight which were separated along PCI and PC2. CPD, HL, DCA and CF ratios were selected by step-wise discriminant function analysis indicated significantly three different morphotypes. Two canonical functions accounted for 100% of variance. The first function accounted 74.54% of variance. First function separated Manzala population scoring negatively (< zero) while the second function successfully separated Burullus population which scored positively (> zero).

5.2. RAPD-PCR analysis

For RAPD analysis, the 14 primers, which produced polymorphic patterns, were used generating a total of 239 reproducible bands. These bands ranged in molecular weight from approximately 200 to 1000 bp.

1- The primers that generated polymorphic amplification, produced high percentage of polymorphism in *Mugil cephalus* populations from Qarun, Burullus and Manzala lakes, (80%, % 75 and 70%, respectively) and a total of 1.8, 1.75 and 1.7 observed number of alleles of which 16, 15 and 14 loci were polymorphic. The total number of RAPD loci produced in populations were 60 loci, of which 45 loci were polymorphic (percentage of the number of polymorphic loci=75%). RAPD results suggested close relationship between *Mugil cephalus* Qarun farms and *Mugil cephalus* Qarun wild populations and separated *Mugil cephalus* Qarun wild population as an out-group of *Mugil cephalus* Manzala and Burullus wild populations.

2- Primers which generated polymorphic amplification, produced high percentage of polymorphism in *Liza ramada* Qarun, Manzala and Burullus lakes, (85%, 85% and 80% respectively) and a total of 1.85, 1.85 and 1.8 observed number of alleles of which 17, 17 and 16 loci were polymorphic. The total number of RAPD loci produced in populations were 60 loci, of which 50 loci were polymorphic (percentage of the number of polymorphic loci=83.33%). There were close relationship between *Liza ramada* Qarun wild and *Liza ramada* Qarun farms populations which separated *Liza ramada* Qarun wild population as an out-group of *Liza ramada* Manzala and Burullus wild populations.

3- The primers that generated polymorphic amplification, produced high percentage of polymorphism in *Valamugil seheli* populations from Manzala, Burullus and Qarun lakes, (80%, % 80and 75%, respectively) and a total of 1.8, 1.8 and 1.75 observed number of alleles of which 16, 16 and 15 loci were polymorphic. The total number of

RAPD loci produced in populations was 60 loci, of which 47 loci were polymorphic (percentage of the number of polymorphic loci=78.33%). There were close relationship between *Valamugil seheli* Burullus wild and *Valamugil seheli* Qarun wild populations which separated *Valamugil seheli* Manzala wild population as an out-group of *Valamugil seheli* Burullus and Qarun wild populations.

4- The primers that generated polymorphic amplification, produced the same percentage of polymorphism in *Mugil cephalus* Qarun, Manzala and Burullus and farms 80% and a total of 1.8 observed number of alleles of which 16 loci were polymorphic 80%, The total number of RAPD loci produced in populations were 60 loci, of which 48 loci were polymorphic percentage 80% of the number of polymorphic loci.

5- The primers that generated polymorphic amplification, produced high percentage of polymorphism in *Liza ramada* Qarun, Manzala and Burullus farms, (85%, 85% and 75%, respectively) and a total of 1.85, 1.85 and 1.75 observed number of alleles of which 17, 17 and 15 loci were polymorphic. The total number of RAPD loci produced in populations was 60 loci, of which 49 loci were polymorphic (percentage of the number of polymorphic loci=81.67%).

5.3. Genetic variation

Genetic variation within population indicated highest genetic variation within *Mugil cephalus* Qarun population 32.99% and lowest genetic variation within *Mugil cephalus* Manzalla population 24.97% and *Mugil cephalus* Burullus population had intermediate value 29.93%, respectively.

Liza ramada Manzala population had indicated highest genetic variation 37.08%, lowest genetic variation within *Liza ramada* Burullus population 30.09% and *Liza ramada* Qarun population had intermediate value 34.25%, respectively.

Valamugil seheli Burullus population had highest genetic variation 36.18%, lowest genetic variation was observed within *Valamugil seheli* Qarun population 31.56%, while, *Valamugil seheli* Manzala population had intermediate value 35.33%, respectively.

Mugil cephalus Qarun farms population had highest genetic variation 32.66%, lowest genetic variation was observed within *Mugil cephalus* Manzalla farms population 30.83%, while, *Mugil cephalus* Burullus farms population had intermediate value 31.21%, respectively.

Liza ramada Qarun farms population had highest genetic variation 34.47%, lowest genetic variation was observed within *Liza ramada* Manzala farms population 32.81%, while, *Liza ramada* Burullus farms population had intermediate value 33.86%, respectively.

5.4. Dendrogram analysis and genetic distance

Dendrogram linked the three species of mullet family Mugilidae in three selected sites farms and wild populations, the important points in the present study:

1- *Mugil cephalus* Manzala wild and *Mugil cephalus* Burullus wild populations (genetic similarity and genetic distance were 97.18% and 0.0286, respectively) were separated from *Mugil cephalus* Qarun wild

population as an out-group. RAPD results suggested close relationship between Manzalla and Burullus populations.

2- RAPD results suggested close relationship between *Mugil cephalus* Qarun farms and *Mugil cephalus* Qarun wild populations.

3- *Liza ramada* Manzala wild and *Liza ramada* Burullus wild populations (genetic similarity and genetic distance were 95.19% and 0.0493, respectively) were separated from *Liza ramada* Qarun wild population as an out-group. RAPD results suggested close relationship between *Liza ramada* populations from Manzalla and Burullus lakes.

4- RAPD results suggested close relationship between *Liza ramada* Qarun wild and *Liza ramada* Qarun farms populations.

5- *Valamugil seheli* Burullus wild and *Valamugil seheli* Qarun wild populations (genetic similarity and genetic distance were 95.52% and 0.0458, respectively) were separated from *Valamugil seheli* Manzala wild population as an out-group. RAPD results suggested close relationship between Burullus and Qarun populations.

Conclusions

1- Analysis of morphometric data obtained with PCA and DFA procedures showed a significant differentiation between mullet population's morphotypes.

2- Analysis of RAPD data revealed genetic differentiation within populations of *Mugil cephalus*, *Liza ramada* and *Valamugil seheli* populations in Egypt. The morphometric data showed a similar pattern, thus, the multivariate analysis of morphometric data can be used to support genetic documentation.

3- RAPD results suggested close relationship between *Mugil cephalus* wild populations from Manzalla and Burullus with genetic similarity and genetic distance 95.19% and 0.0493, respectively. Close relationship between *Mugil cephalus* Qarun farms and *Mugil cephalus* Qarun wild populations was observed.

4- Close relationship between *Liza ramada* populations from Manzalla and Burullus lakes. The genetic similarity and genetic distance were 95.19% and 0.0493, respectively.

5- Close relationship was observed between Burullus and Qarun of *Valamugil seheli* wild populations. The genetic similarity and genetic distance were 95.52% and 0.0458, respectively.

6- There was a higher genetic variation within *Mugil cephalus, Liza ramada and Valamugil seheli* wild populations. This may have been caused by the forces of genetic drift and natural selection pressure.

7- Further genetic studies on mullet populations in Egypt are needed and more samples of fish must be collected from many other localities which represent farm and natural habitats. The results of these studies will give more complete picture of the genetic resources of mullet in Egypt.

8- Development of new techniques for genetic studies and analysis of fish populations are needed. RAPD analysis provides more insights into total genetic diversity since the DNA surveyed represents the whole genome.