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Multivariate Analysis of Morphometric Parameters in Wild and Cultured Nile Tilapia *Oreochromis niloticus*

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ABSTRACT

In order to evaluate the phenotypic variation between natural population and cultured stocks of Nile tilapia (*Oreochromis niloticus Linnaeus*, 1758), the intra- and inter-population differentiations were analyzed using biometric approach. Thus, eight morphological measurements were taken from 300 specimens collected from Ismailia Canal (AB) as natural population and two cultured stocks from Kafr El-Shaikh (KE) and Fayoum (FA) Governorates. All specimens collected at juvenile stage in first generation which reared in the under environment al conditions. The univariate (ANOVA) and multivariate analyses (principal component analysis, PCA, and discriminant function analysis, DFA) showed a low variability among populations. The morphometric pattern observed in 3 stocks of Nile tilapia reflects variation primarily along three axes. Together these components accounted 81.20% of observed variation. Three characters were selected by step-wise discriminant function analysis on morphometric data. This study shows the existence of morphological differentiations between subpopulations derived from a single gene pool that have been isolated in separated sites for several decades although bred in relatively similar environments.

Keywords: multivariate, morphometric, Nile tilapia

INTRODUCTION

The rapid growth of Tilapia, their resistance to poor water quality, ability to grow under sub-optimal nutritional conditions, and high fecundity all make them well suited aquaculture. Tilapia is the second most cultivated fish in the world, only surpassed by carp, with almost 100 countries as producers (FAO, 2002). The worldwide use of Nile tilapia (*Oreochromis niloticus Linnaeus*, 1758) in aquaculture represents a somewhat unique scenario. According to recent statistics of the Egyptian General Authority for Fish Resources Development (GAFRD, 2009), tilapia consist 55.6% (477.458 tones) of the Egyptian production from fish culture sector (693.815 tones) in 2008. Also, Egypt produces 12% of the world farmed tilapia (2.121.009 tones) (FAO, 2007). Moreover, Egypt is by far the main tilapia producer in the Middle East and North Africa (MENA) region, where produced 477.458 tones in 2008 of tilapia, or 92.2% of all tilapia production in this region (Feidi, 2010). Yet, Nile tilapia is cultured in 23 African countries out of 32 countries that practiced tilapia culture in Africa. In Egypt, most of the aquaculture production of tilapia is derived from semi-intensive fish farms in earthen ponds, intensive systems, integrated intensive fish farms and cages (GAFRD, 2006).

Management of aquatic genetic resources should ideally involve a continuum of activities: documentation of genetic resources and the variety of ecosystems in which they are functional components, including the status of potential threats to these resources; characterization to determine the genetic structure or distinctness and conservation value of the resource; evaluation to estimate either direct or indirect economic potential; and utilization in sustainable genetic improvement schemes, with due regard to the emerging codes of practices of access to and benefit sharing of the genetic resources.

Tilapia hatcheries use only a few individuals as broodstock for natural or artificial propagation, that has been taken from other commercial farms or natural resources. Consequently, this may lead to inbreeding problems over several generations. Decreased genetic variability may have detrimental effects on commercial traits such as growth rate, survival, and disease resistance. Therefore, it is vital and critical to assessment of the genetic diversity among and within Nile tilapia populations in successive generations.

Morphometry is an important ecological character in fishes because it can affect reproductive success through the abilities to forage, defend territories, avoid predators, and attract mates. Reliable estimates of genetic and phenotypic parameters are needed for all traits of economic importance, to predict response to selection, to choose various breeding plans, to estimates economic returns, and to predict breeding values of candidates for selection (Gjerde and Gjedrem., 1984 and Taylor and McPhail, 1985). Therefore, external body shape as well as the shape of parts of the carcass might be traits of economic importance from a marketing point of view. The extent to which morphometric variation is determined by genetic and environmental factors is poorly understood for most fishes, including salamonids (Gjerde and Schaeffer, 1989). Schwanck and Rana (1996) these authors reported that the understanding the modes of inheritance of morphological characters in tilapia is of prime importance when suspected hybridization in farmed or wild stock has to be confirmed, as well as when hybrids are intentionally bred for better characters. So, the objective of the present study was to qualify and compare the morphometric variation within and between wild and cultured Nile tilapia populations. This study will provide additional information on the stocks of Nile tilapia used in aquaculture. This form was part of program on the characterization of natural populations and cultured strains of Nile tilapia in order to increase their production on the basis of rational use of genetic resources.

MATERIALS AND METHODS

This study was conducted at the Central Laboratory for Aquaculture Research (CLAR), Abbassa, Sharkia Governorate, Egypt.

Sample collection and treatment

Three Nile tilapia *Oreochromis niloticus* populations were used in this experiment. One natural population *O. niloticus,* Abbassa population (AB), collected from Ismailia Canal East of the Nile Delta near Abbassa, Egypt and two cultured populations from Kafr El-Shaikh (KE) and Fayoum (FA) were collected from commercial farms. Kafr El-Shaikh population of *O. niloticus* was derived from about 300 adults. Fish reproduced using equal numbers of females and males. Kafr El-Shaikh stock was undertaking selection program for growth performance improvement for several generations while Fayoum stock was undertaking selection program for pure line of Nile tilapia morphology strain.

Brood fish from each population were stocked in three concrete ponds ($15m^2$ each) during the last week of June 2009. Stocking density was 1 individual/m^2 at a sex ratio of 1:1 to maximize the effective population size (Falconer, 1989; Cameron, 1997).

Fish were allowed to reproduce and fry were reared in the same pond with their parents. Ponds were drained 35 days after stocking and fry were collected, counted, and held in 2 m^3 hapas to recover. In holding, they were fed ad libitum with isocaloric 40%-crude- protein powdered fish feed. After 1 month, fry from each group were counted, weighed, and stocked into three randomly assigned 15 m^3 concrete ponds at a density of 50 fish/m^3 after remove parents. Fish were fed daily with 25% protein. Feed was delivered about mid day. Water depth in the ponds was about 1 m. Dissolved oxygen, pH, temperature and Secchi disk visibility were measured daily, but remained at acceptable levels without intervention throughout the study. The average initial and final individual body weights (g) of AB, KE, and FA stocks for 90 days ranged from (6.08 – 45.18), (6.35 – 50.81), and (5.61 – 42.27) respectively. One hundred individuals of *O. niloticus* were randomly collected from each of three stocks for morphometric measurements.

Morphometric data

Eight morphometric measurements were studied according to Trewavas (1983). The morphometric characters were measured to nearest 0.01-mm using a digital caliper. The morphometric under study were body length (BL), standard length (SL), head length (HL), tail length (TL), trunk length (RL), body depth (BD), body thickness (BT), and head thickness (HT) (Figure 1). All morphometric measurements were transformed by dividing the measurement by the standard length of each fish to minimize the effect of fish size (Allendorf *et al*., 1987).

Analysis of morphometric data

All data from the sampled fish (300 individuals) were subjected to the multivariate analysis. Principal component analysis (PCA) and discriminant function analysis function (DFA) were then used to analyze the data using SPSS version 10. PCA was conducted on a covariance matrix. One-way analysis of variance (ANOVA) was performed to test the variation for each trait among fish populations.

RESULTS AND DISCUSSION

A major advantage of multivariate approach is its ability to incorporate covariation among morphometric parameters directly into the analysis of variation. Much of this morphometric variation may be hidden if analysis rely exclusively on univariate variation. If possible, a

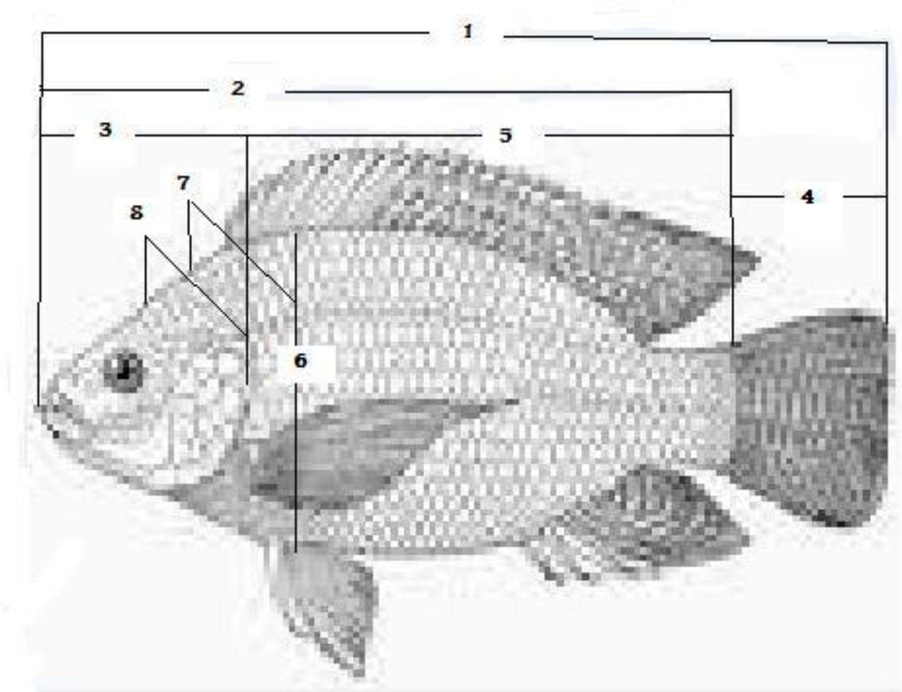

Figure1. Illustration of parts and measuring of Nile tilapia parts.

1- Body length (BL)	2- Standard length (SL)	3- Head length (HL)
4- Tail length (TL)	5-Trunk length (RL)	6- Body depth (BD)
7- Body thickness (BT)	8- Head thickness (HT)	

comprehensive analysis of multivariate morphometry should also take into account developmental aspects of morphometric variation that are likely to have consequences for fitness in the wild (Hard *et al.*, 1999). Three hundred specimens of *Oreochromis niloticus* from wild and cultured conditions were studied morphometrically. Data obtained were subjected to factor analysis using principal component analysis (PCA) and discriminant function analysis (DFA). In order to estimate the importance of the morphometric measurements for species identification, a univariate analysis of 7 measurements expressed as percentages of the standard length of fish was run (Table 1). All parameters exhibited significant differences among stocks ($P < 0.05$).

Table 1. ***Means[*] ± S.D of morphometric traits for three Nile tilapia stocks.***

	Populations		
Traits	**KE**	**FA**	**AB**
BL	**1.204^{a}±0.002**	**1.193^{b}±0.002**	**1.197^{b}±0.002**
HL	**0.337^{b}±0.001**	**0.351^{a}±0.001**	**0.339^{b}±0.001**
TL	**0.204^{a}±0.002**	**0.193^{b}±0.002**	**0.197^{b}±0.002**
RL	**0.663^{a}±0.001**	**0.648^{b}±0.072**	**0.660^{a}±0.041**
BD	**0.369^{b}±0.034**	**0.377^{a}±0.008**	**0.379^{a}±0.051**
BT	**0.159^{b}±0.010**	**0.169^{a}±0.011**	**0.172^{a}±0.009**
HT	**0.353^{b}±0.015**	**0.365^{a}±0.017**	**0.365^{a}±0.032**

[*] ***Means in the same row followed by different letters are significantly different (P< 0.05).***

(third axis). Together these components accounted for more than 81.20% of observed variation in morphometry (Table 2). Loading on PC1 were large and positive for body length and tail length and this component accounted for 39.99% of variance (λ = 2.79). PC2 was positive correlated with trunk length and negative correlated with head length,

Table2. ***Component matrix and Eigen values of morphometric traits for three Nile tilapia stocks.***

	Component				
Traits	1	2	3	4	5
BL	0.793	0.263	-0.549	-0.007	0.002
HL	0.711	-0.667	0.215	-0.021	0.021
TL	0.793	0.263	-0.549	-0.007	0.002
RL	-0.711	0.667	-0.215	0.021	-0.021
BD	0.491	0.443	0.501	-0.061	-0.553
BT	0.366	0.467	0.446	-0.548	0.383
HT	0.389	0.368	0.395	0.701	0.252
Eigen values λ	2.79	1.58	1.30	0.798	0.518
% of variance	39.99	22.59	18.62	11.40	7.39
Cumulative %	39.99	62.58	81.20	92.60	100.00

Factor loadings

The morphometric pattern observed in three stocks of Nile tilapia reflects variation primarily along the first three axes: 1) variation in body length, head length, tail length and trunk length (first axis), 2) head length and trunk length (second axis) and 3) body length, tail length and body depth explaining 22.59% of variance (λ = 1.58). PC3 was correlated negatively with body length and tail length, and positively with body depth. PC3 explained an additional 18.62 (λ =1.30).

Correlation among parameters

Correlation coefficients between all combinations of morphometric traits as ratios of standard length were calculated for all individuals in the three stocks (Table 3). The pairwise comparisons revealed significant correlations between:

A weak but significant positive correlation between body thickness and body depth (r=0.433, $P < 0.05$), a positive strong correlation between body length and tail length (r=1.00, $P < 0.05$) and a negative strong correlation between head length and trunk length (r=-1, $P < 0.05$).

Morphometric variation among stocks

The scatters of standardized scores of PC1 against PC2, PC1 against PC3 and PC2 against PC3 for all stocks examined are given in Figure 2. The plot of PCA scores for morphometric variables shows that all three stock of Nile tilapia located on the positive sector on the first and second component while all stocks overlap on the negative sector on the third component. This would indicate that populations of KE, FA and AB are similar in these morphometric traits. Hard *et al.*, (1999) indicated that

Table3. ***Correlation matrix of morphometric traits as ratios of standard length for three Nile tilapia stocks***

	BL	HL	TL	RL	BD	BT	HT
BL	**1.00**						
HL	**0.271***	**1.00**					
TL	**1.00***	**0.271***	**1.00**				
RL	**-0.271***	**-1.00***	**-0.271***	**1.00**			
BD	**0.230***	**0.152**	**0.230***	**-0.152**	**1.00**		
BT	**0.173***	**0.045**	**0.173***	**-0.065**	**0.433***	**1.00**	
HT	**0.184***	**0.106**	**0.184***	**-0.106**	**0.371***	**0.203***	**1.00**

****Significant at P < 0.05***

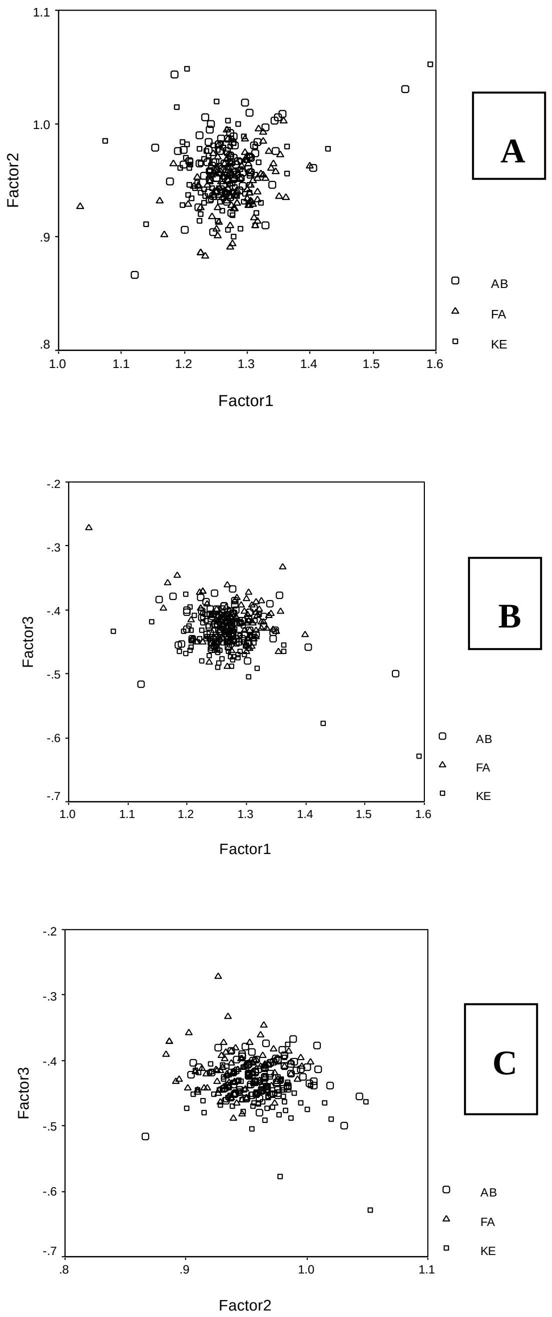

Figure 2. ***Scatter plot of standardized scores for three Nile tilapia stocks on (A) PC1 and PC2, (B) PC1 and PC3 and (C) PC2 and PC3.***

phenotypic patterns of morphometric variation in salmonid population identify opportunities for selection and show some similarities to morphological differences that have been observed between juvenile wild and hatchery salmonids. This variation may represent evolutionary opportunities for environmental pressures to act to differentiate the two groups morphologically (Swain *et al*., 1991). Selection on juvenile body size could indirectly alter fin size and positions as well as other aspects of body shape, for which differences are apparent within (Swain and Holtby, 1989; Swain *et al*., 1991; Taylor and McPhail, 1985) as well as among (Bisson *et al*., 1988) species. The correlated responses could affect performance in different habitat and consequently, patterns of habitat use in hatchery reared juveniles released to the wild.

Discriminant function analysis

Estimating patterns of morphological differentiation among populations is one important method of understanding how divergent selective regimes can generate and maintain phenotypic diversification (Langerhans and DeWitt, 2004; and Rogers and Bernatchez, 2007). Three characters were selected by step-wise discriminant function analysis (DFA) on morphometric data from three Nile tilapia stock. The characters selected on fish were total length, trunk length and body thickness ratios. The analysis of the morphometric variables produced two significant discriminant functions (Table 4), of which the first discriminant function separated groups. The first function accounted for 77.90% of the variance and second function 22.1% in the data. The first axis has a high negative loading for

Table 4. ***Standardized step-wise canonical discriminate function coefficients for morphometric traits for three Nile tilapia stocks.***

	Function	
Traits	**1**	**2**
TL	0.511	0.157
RL	0.584	0.803
BT	-0.747	0.651
Eigen value λ	0.416	0.118
% of variance	77.90	22.10
Cumulative %	77.90	100.00

body thickness ratio while the second axis has a high positive loading for trunk length. In bivariate plot of the two canonical functions, separated KE (positive sector) from FA and AB (negative sector) (Figure 3). Morphometric variation among the three stocks of Nile tilapia was used to test for the presence of stock structuring. Significant heterogeneity in morphology among the three Nile tilapia stock was revealed by univariate statistics and multivariate (PCA and DFA) (Table 5). Therefore, overlapping variation in morphometric characters lead to great difficulty in identifying the different stocks. Jerry and Cairns (1998) indicated that phenotype of an individual is a manifestation of its underlying genotype, as expressed in the local environment during development. Consequently, individuals that develop and mature in the same drainage area would be expected to share a similar phenotype, as they are likely to experience common environmental and genetic influences (Chambers, 1993). Morphometric data indicated that three Nile tilapia stocks are lowly structured morphologically. The step-wise canonical discriminant analysis generated standardized canonical coefficients for the 3 morphometric variables. These coefficients were subsequently used as an index to classify the specimens of the 3 Nile tilapia stocks.

Score = 0.511 (TL) + 0.584(RL) + -0.747(BT).

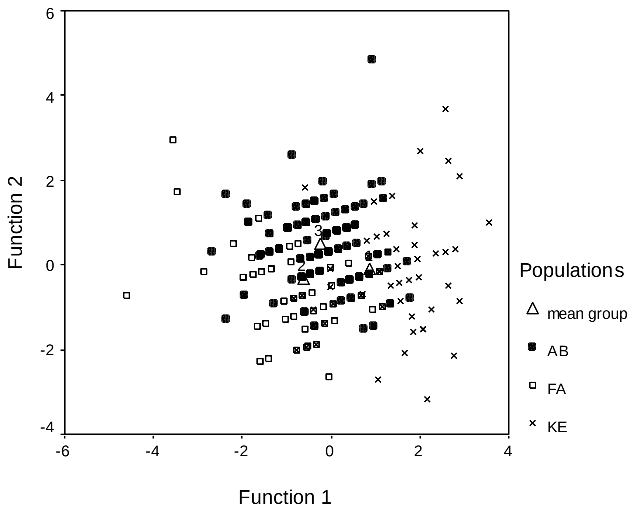

Figure3. Scatter plot of the two canonical discriminant functions from analysis of morphometric traits for Nile tilapia stocks.

Multivariate statistics was used as a tool to understand population differences and morphometric relationships. Canonical discriminant analysis has been useful in other studies of marine species where parallel use of genetic and morphometric studies led to the conclusion that DFA on morphometric data was equivalent to genetic analysis for group identification (Teugels, 1997 a and b); and Barriga, 2004; Nobah *et al.*, 2006). Moreover, the multivariate morphometry approach was able to identify covariation among the morphometric traits, which was not possible to identify by univariate analysis only (Murta, 2000).

REFERENCES

Allendorf, F; Ryman N; and Utter F; (1987): Genetics and Fishery Management: Past, Present, and Future. In: N. Ryman (ed.). Population Genetics and Fishery Management. The University of Washington, USA. p 1-20.

Barriga-Sosa, IDLA. (2004): Variability of tilapias (*Oreochromis spp*.) introduced in mexico: morphometric, meristic and genetic characters. J.Appl.Ichthyol. 20(1), 7-14.

Bisson, P.A., Sullivan, K. and Nielsen, J.L. (1988): Channel hydraulics, habitat use, and body form of juvenile coho salmon, steelhead, and cutthroat trout in streams. Trans Am Fish Soc, 117:262-273.

Cameron, N.D., (1997). Selection Indices and Prediction of Genetic Merit in Animal Breeding. CAB International, Wallingford, UK.

Chambers, R.C. (1993). Phenotypic variability in fish populations and its representation in individual based models. Transactions of the American Fisheries Society, 122: 404-414.

Falconer, D.S., (1989): Introduction to Quantitative Genetics. Longman Scientific & Technical, Longman Group, UK, 438 pp.

FAO, 2002. The state of world fisheries and aquaculture. FAO, Rome, Italy.

FAO, (2007): FAO yearbook, Fishery and Aquaculture Statistics, Aquaculture Production, Rome, Italy, pages:88.

Feidi, I.H. (2010): Tilapia markets in the Middle East and North Africa demand trends and outlook. Proceedings of the 3rd International Technical and Trade Conference and Exposition on Tilapia, October, 27-29, 2010 Kuala Lumpur, Malaysia.

GAFRD, (2006): 1990 – 2006 General Authority of Fish Resources Development yearbook. Statistics of fish production, Ministry of Agriculture and land Reclamation, Cairo, Egypt.

GAFRD, (2009): General Authority of Fish Resources Development yearbook. Statistics of fish production, Ministry of Agriculture and land Reclamation, Cairo, Egypt.

Gjerde, B.; and Gjedrem, T. (1984): Estimates of Phenotypic and genetic parameters for carcass traits in atlantic salmon and rainbow trout. Aquaculture 36, 97-110.

Gjerde, B.; and Schaeffer, L.R. (1989): Body traits in rainbow trout. II. Estimates of heritabilities and of phenotypic and genetic correlations. Aquaculture, 80: 25-44.

Hard, J.J., Winans, G.A. and Richardson J.C. (1999): Phenotypic and genetic architecture of juvenile morphometric in chinook salmon. The American Genetic Association, 90: 597-606.

Jerry, D. and Cairns, S. (1998): Morphological variation in the catadromous Australian bass, from seven geographically distinct riverine drainages. Journal of Fish Biology, 52:829-843.

Langerhans, R.B.; and DeWitt, T.J.(2004): Shared and unique features of evolutionary diversification. American Naturalist 164, 335- 349.

Murta, AG . (2000): Morphological variation of horse mackerel *(Trachurus trachurus)* in the Iberian and North African Atlantic: implications for stock identification. ICES J. Mar. Sci. 57, (4), 1240-1248

Nobah, Csk; Kouzamelan, EP; N, Douba,V; snoeks,J; Teugels,GG; Geore-Bi, G; Kone, T. and Falls, TM. (2006): The colour pattern of the caudal fin, a useful criterion for identification of two species of Tilapia and their hybrids. J.Fish.Biol. 69 (3), 698-707.

Rogers, SM; and Bernatchez,L. (2007): The genetic Architecture of ecological section and the association with signatures of selection in natural Lake whitefish (*Coregonus sp. Salmonidae*) species Pairs. Mol.Biol.Evol. 24 (6), 1423- 1438.

Schwanck, E; and Rana, K. (1996): Analysis of the morphometrics of three tilapias (*Tilapia zillii*, *Sarotherodon galilaeus* and *Oreochromis niloticus*) and their intergenericHybrids. The Third International Symposium on Tilapia in Aquaculture. CLARM Conf. Proc. no. 41. 550 p.

Swain, D.P. and Holtby, L.B. (1989): Differences in morphology and behaviour between juvenile coho salmon *(Oncorhynchus kisutch)* rearing in a lake and its tributary stream. Canadian Journal of fisheries and Aquatic Sciences, 46:1406-1414.

Swain, D.P. Riddell, B.E. and Murray, C.B. (1991): Morphological differences between hatchery and wild populations of coho salmon *(Oncorhynchus kisutch)* : environmental versus genetic origin. Canadian Journal of fisheries and Aquatic Sciences, 48:1783-1791.

Taylor, E.B.; and McPhail, J.D. (1985): Variation in body morphology among British Columbia populations of Cohosalmon, *Oncorhynchus kisuttch*. Can J Fish Aquat. Sci., 42: 2020- 2028.

Teugels, GG. (1997 a): Morphometric characterization of populations and strains of *Oreochromis niloticus, Sarotherodon melanotheron* (Cichlidae), Clariasanguillaris, *Clarias gariepinus* (Clariidae) and Chrysichthys nigrodigitatus (Claroteidae). Characterization of Ghanaian tilapia genetic resources for use in fisheries and aquaculture: extended abstracts and discussions. ICLARM Conf. Proc. no. 52. pp. 23-24.

Teugels, GG. (1997 b): Preliminary results on morphometric differentiation between natural populations of the Nile tilapia *Oreochromis niloticus* (Perciformes, Cichlidae). Proceedings of the Symposium Genetics and Aquaculture in Africa, Abidjan, 1-4.

Trewaves, E. (1983). Tilapine fishes of the genera *Serotherodon, Creochromis* and *Danakilia*, Trust. Brit. Mus-Henry Ling (Ltd), Dorset Press. England..

التحليل متعدد المتغيرات للصفات المورفولوجية في البلطي النيلي البرية والمستزرعة

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أجريت هذه الدراسة من أجل تقييم التباين المظهري للبلطي النيلي البري و المستزرع. تم تحليل التمايز داخل و فيما بين العشائر باستخدام معايير الصفات المورفومترية للأسماك. تم احضار الأمهات من ترعة الأسماعيلية (السلالة البرية) و سلالتين مستزرعتين من مزارع تجارية من محافظات كفر الشيخ و الفيوم و من ثم تفريخها و تربيتها فى بئية مماثلة حيث جمعت العينات من الجيل الأول و بالتالى حصرت ٨ معايير مورفولوجية من ٣٠٠ سمكة بلطي نيلي. أظهر جدول تحليل التباين (ANOVA) و تحليل المتغيرات المتعددة انخفاض فى مقدار التباين بين و داخل العشائر بوجة عام. لوحظ أيضا أن معظم التباين باستخدام PCA انعكس علي ٣ محاور الأولى بمقدار ٨١,٢٠ % من مقدار التباين الكلي. أوضح تحليل DFA أن هناك ٣معايير للتميز بين الثلاث عشائر السمكية. هذه الدراسة تبين وجود فوارق شكلية بين عشائر طبيعية مختلفة من نوع واحد و التى تم عزلها فى مواقع منفصلة لعدة عقود علي الرغم من أنها في بيئات مماثلة نسبيا.