MORPHOLOGIC CHARACTERIZATION OF TENCH (*TINCA TINCA* LINNAEUS, 1758) THROUGH THE CLASSICAL AND MODERN METHODOLOGY

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Abstract

The results of a profitable fish farming are based in the first place, on the genetic quality of fish (growth rate, prolificity, disease resistance, etc.) The genetic improvement of the fish is a continuous preoccupation at the global and national level, intending to create new, highly productive races and hybrids. The morphologic characterization used as an instrument of work in the evaluation of the morphometric changes of fish, is based on biometry. The study presented achieves a morphometric characterization of the tench through the classic biometric method and tests the possibilities of using 3D photographic method for fish measureing. The results show that the classic biometrics method with a high degree of aggression on fish, can be replaced with the 3D photometric method, used through an application on the mobile phone, that achieves better accuracy, saves time and completely avoids the stress and fish losses.

Key words: Morphometry, Tench, 3D Photometry

INTRODUCTION

One of the important factors which influence the profitability of fish farming besides the nutrition, health, and the growth conditions, is the genetic quality of broodstock and seed production. The efficiency of assimilation and feed conversion ratio which determine the specific growth rate, carnosity index, disease resistance, etc., depend on genetic quality of fish.

On a world scale and nationally, selection and breeding studies are being carried out to obtain hybrids that combine valuable hereditary characters and offer increased qualities to the offspring [2]. Romanian aquaculture requires programs for the renewal of genitors from fish species with high economic value, used for artificial breeding in hatcheries, to avoid inbreeding and the obtaining degenerate or unviable offspring caused by using the same broodstock over the recommended multi-annual period [5]. In order to diversify fish production but also to repopulate natural waters with fish species whose populations are declining, due the impact of anthropogenic factors and climate changes, we have chosen to study the tench [9].

Among the cyprinids, tench has the lowest growth rate, and the mass selection that has taken place in the last hundred years has made the size of tench to increase continuously to almost double body size today. Phenotypic evaluation of fish [7] requires their morphometry, which in the case of tench breeders can sometimes be traumatic or at least stressful, knowing that this species is sensitive to captive conditions and in the absence of proper acclimatization refuses to reproduce[1]. For these reasons, we have tested new methods of non-invasive 3D photographic biometry, which allows fish measuring and image rocessing, to avoid subjecting breeders to prolonged anesthesia and the mechanical stress of manipulation which affects mucus secretions or produces injuries that promotes fungal infestation of the skin. The photographic image can be measured directly in the fish farm or it can be stored in the application memory and the photographic biometrics can be performed later in the laboratory.

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MATERIAL AND METHOD

Morphometry offers the possibility of interspecies or intrapopulation comparison of fish with direct applications in the genetic improvement of valuable species of farmed fish. The purpose of classical, geometric, and photometric biometrics is to analyze and store the morphometric data of the genitors to comparatively evaluate the hybrid offspring. For the morphometric characterization, within a genetic improvement program, three methods were used: classical biometry, geometric morphometry, and 3D photometry.

For **classical biometry method** (*fig. 1*), 10 females and 20 males of tench broodstock provided from aquaculture were measured. The fish were previously anesthetized with clove oil at a concentration of 0.1 ml/l.

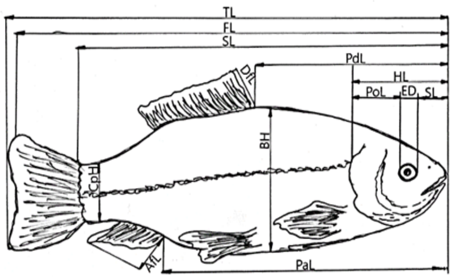


Fig. 1 Morphometric measurements on the specimen of *Tinca tinca* TL-total length, FL-fork length, sl-standard length, HL-head length, SL-snout length, ED-eye diameter, PoL-postorbital length, DfL-dorsal fin length, PaL-preanal length, BH-body height, CpH-caudal peduncle height, Fl-anal fin length.

The data were processed through statistical methods (arithmetic mean, standard deviation, variance, variation coefficient (VC=SD/meanx100) and the percentage ratio of morphometric characters to standard length (ML/SL%) were determined [11] and presented in *table 1*. The equation of the relationship between length and weight of fish and the coefficient of corelation [4]. The relation between the length (L) and weight (W) was determined by the formula, $W = a \times L^b$, and the coefficient of determination R^2 [11] and presented in the graph.

The characterization of fish by the **method of geometric morphometry** (*fig. 2*) was performed using the following software :tpsUtil32(used to create the TPS file), tpsDig232(used to add the landmarks on the TSP file), CoordGen8_Win(used to transform the file from TPS format to X1Y1), PCAGen8Win(used to create the chart according to the landmarks). Morphometric marks [10] and the distances between them [3] that underlie the characterization of tench genitors through the method of geometric morphometry [6], to create a database necessary for evaluations of the breeding processes results. The truss morphometric network of tench are shown in the figures below [8].

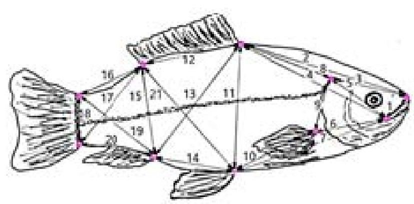


Fig. 2 The scheme of landmarks on tench

Fig. 3 Landmarks on tench TPS file in tpsDig232

Fig. 2,3 Morphological landmarks are pink coloured and morphometric distances between landmarks are numbered. The landmarks used in morphometric analysis of tench (in antero-posterior direction) are: tip of snout; posterior maxila; top of operculum; anterior origin of pectoral fin insertion; anterior origin of dorsal fin insertion; anterior origin of pelvic fin insertion; posterior end of dorsal fin insertion; anterior origin of anal fin; origin of upper insertion of caudal fin; origin of lower insertion of caudal fin; 1-mouth tip to premaxila; 2-mouth tip to dorsal fin; 3-mouth tip to operculum top; 4-pre maxilla to dorsal fin; 5-pre maxilla to operculum tip; 6-pre maxilla to pectoral fin; 7-pre maxilla to pelvic fin; 8-dorsal fin to operculum tip; 9-pectoral fin to operculum tip; 10-pectoral fin to pelvic fin; 11 dorsal fin to pelvic fin; 12-dorsal fin front to dorsal fin back; 13-dorsal fin to anal fin; 14-pelvic fin to anal fin; 15-dorsal back to anal fin; 16-dorsal fin back to caudal top; 17-dorsal back to caudal bottom; 18-anal fin to caudal top; 19-caudal top to caudal bottom; 20-anal fin to caudal bottom; 21-dorsal fin back to pelvic fin.

The results of the geometric morphometry data for genitors were stored in a database to be compared with the data of tench hybrids (fig. 4).

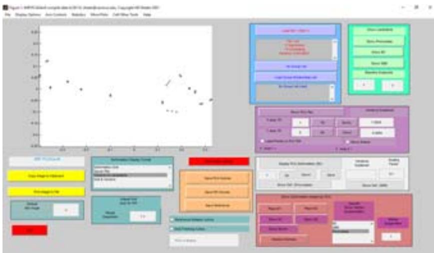


Fig. 4 The chart formed in PCA8GenWin

For the measurements made by the **3D photometry method**, we used an application downloaded on the mobile phone, presented in fig. (*fig.5, 6, 7, 8, 9, 10*). For photographic biometry, 10 fish specimens were used in 32 tests for different image resolutions. We used in triplicate 8 shootings at verticalities of 0.1, 0.2, 0.3 for each image to analyze their differences. The credit card type calibration variant was tested, A4 format and objects with given dimensions to compare the accuracy of the results, the testing of the A3 format was avoided because it is difficult to be used as calibration object in the field. Due to image calibration, the focal length was neglected. Only the total length of the fish was recorded in the field to verify the accuracy of the measurements compared to the measurements made with the roulette or ichthyometer. Figures 3-8 shows the application resolutions and options for the unit of measurement used, the type of geometric measurement, reference object models, how to perform the calibration, and how to perform the measurement.

Fig. 5 The option for selecting 32 different resolutions

Fig. 6 Option for selecting measurement units

Fig. 7 Option for selecting the desired geometric measurement

Fig. 8 Option for selecting the reference object

Fig. 9 Calibration of the reference object

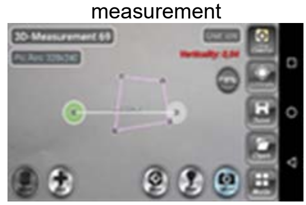


Fig. 10 Image-object measurement made after the calibration of the reference object

RESULTS AND DISCUSSIONS

The results of the characterization of tench brood stock by classical biometrics are given in *table 1*. Biometric data show that the distribution of values for the sample of fish studied are characteristic to a relatively homogeneous growth of females and males. The values obtained from the measurements highlight the sexual dimorphism in tench, which is characterized by a considerable increase in the values of morphometric characters in females compared to the values recorded in males.

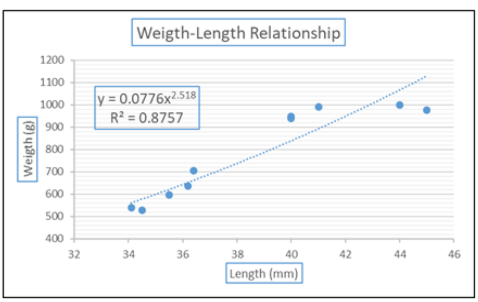
Table 1 Morphometric characters of *Tinca tinca*

Morphometric characters Females	Min.-max. (mm)	Mean (mm)	Standard deviation (mm)	Variance $(mm)^2$	Variation coefficient (%)	ML/TL (%)
Total length	400-440	411	±14.97	4.93	0.04	-
Fork length	390-425	401.4	±12.48	4.21	0.03	-
Standard length	340-370	347	±13.04	5.08	0.04	-
Predorsal distance	170-195	177.4	±9.95	7.59	0.06	43.16
Preanal distance	249-275	256	±9.82	5.19	0.04	62.29
Head length	80-90	85	±5.00	7.96	0.06	20.68
Snout length	30-35	33	±1.90	7.78	0.06	8.03
Eye diameter	7	7	±0.00	0.00	0.00	1.70
Postorbital length	43-48	45.6	±2.06	6.11	0.05	11.09
Interorbital distance	40-63	50.6	±7.31	19.55	0.14	12.31
Dorsal fin height	70-80	73	±3.79	7.03	0.05	17.76
Anal fin height	60-70	64	±4.90	10.36	0.08	15.57
Caudal peduncle height	41-45	42.4	±1.50	4.78	0.04	10.32
Body height	103-125	110.6	±7.71	9.43	0.07	26.91
Males						
Total length	341-361	352.4	±8.04	3.09	0.02	-
Fork length	329-357	343.2	±11.91	4.69	0.03	-
Standard length	287-300	291.4	±4.45	2.07	0.02	-
Predorsal distance	143-170	155.6	±11.37	9.89	0.07	44.15
Preanal distance	210-220	213.8	±3.71	2.35	0.02	60.67
Head length	67-84	73.2	±6.53	12.08	0.09	20.77
Snout length	23-34	27.6	±3.93	19.26	0.14	7.83
Eye diameter	7	7	±0.00	0.00	0.00	1.99
Postorbital length	37-43	38.6	±2.33	8.17	0.06	10.95
Interorbital distance	43-59	50	±6.03	16.32	0.12	14.19
Dorsal fin height	65-80	70.4	±5.16	9.92	0.07	19.98
Anal fin height	60-65	62.2	±1.72	3.74	0.03	17.65
Caudal peduncle height	38-42	40	±1.41	4.78	0.04	11.35
Body height	95-101	98.2	±2.64	3.63	0.03	27.87

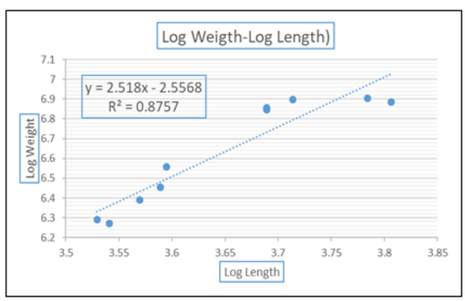
Legend: ML/TL(%)- The percentage ratio of morphometric characters to total length

The relationship between the length and weight of the fish are measurements that characterize the changes in robustness and health recorded in the evolution of fish populations The value of b=2.518 represents the slope and is lower than 3 which mean a negative allometry represented in graph 1 and 2.

The correlation coefficient is positive when its value is close to 1,in this case r=0.935 which indicates that both variables taken into account (fish weight and length) increase in a directly proportional way, according to the graphic below.



Graph 1



Graph 2

For the classical biometry, we compared the accuracy of the measurements made with the help of the roulette, with the photographic measurements.The comparative study of the classical measurements done at the fish farm with those of photometry at resolutions of 1280x720 (0.92MP), 1200x800 (0.96MP), 1280X768 (0.98 MP), with a variation of verticality values between 0-0.04 are presented in *fig. 11, 12*. The classical in comparison with photometric measurements of tench made in the laboratory are presented in *fig. 13,14*. The most accurate results are obtained by photometry under the conditions of a correct calibration. The size of the reference object is directly proportional to the desired mesurement accuracy.

Fig. 11 Measuring tench in the field with the roulette

Fig. 12 Measuring tench with the 3D photometry application, in the field

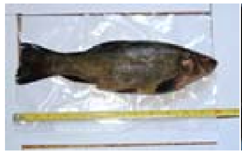


Fig. 13 Measuring tench with the roulette in the laboratory

Fig. 14 Measuring tench in the laboratory using the 3D photometry application

All values obtained in the left collum (classical method) are similar to the ones obtained in the right collum (3D photometry) with a ±2 mm error. For small fish specimens, the photometry method successfully replaces the classical measurement of fish by manipulation.

In the case of large fish, such as sturgeons, the only possibility of biometry is the option of measuring with rulette regardless of biometrics.

The geometric morphometry is a very accurate method but has two disadvantages, one is the difficulty of making photos on large fish and the other is related with the necessity for the fish to be well fixed (with needles on a substrate) which does not permit the recovery of the fish alive.

Method	Advantages	Disadvantages
Classical biometry method	-it is the most suitable method for measuring large size fish (e.g: sturgeons)	-requires the fish to be anesthetized -measuring done in the field with the roulette are not always precise -requires handling of the fish (it produces damage to the mucus secretion, scale losses, which allow fungal infestations)
Geometric morpho-metry method	-the better method for analyzing the differences in the characterization of fish species and hybrids. Being the most accurate method of morphometric evaluation used in genetic improvement of fish and species differentiation -it uses the landmarks for photograph measurements of fish and data are processed with performant PC-Softwares	-requires the fish to be dead (fixed with needles to position the fish fins in their natural stance, to allow easier placing of the landmarks in the right positions on the photo) -this method is more difficult to be used for measurements on very large fish
3D Photometry method	-does not require anesthesia of the fish -does not involve handling the fish -photographing the fish in the field can only take a few seconds -the images can be stored in the phone's gallery or the memory of the biometry application. The measurements on the photos can be continued in the laboratory or at any time when it is necessary.	-we have not tested the possibility of using this application for large fish

CONCLUSIONS

The method used for the morphological characterization of the fish must be chosen according to the accuracy, the size of the fish and the negative impact on it, due to the manipulation during the biometry.

3D photometric biometry can store the image of the fish in order to continue the measurements later or to reaccess the photos as many times as needed. To avoid the traumatic handling of fish in the field or repeated anesthesia, this method is recommended.

The use of 3D photographic biometrics provides accurate results compared to classical measurements performed on fish (depending on the correct calibration to the reference object, focal length, and small variations in verticality).

It is recommended to resume the tests and extend them in order to establish with certainty the degree of repeatability of the measurements and the possibility of using the 3D photometric method in large fish.

The geometric morphometry method is the most precise method but it needs special knowledge, a PC and software and for these reasons it is not accessible to the fish farmers.

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