



## First Report on the Abnormality Among Body Component Ratios in the Caught Caspian Sea Mahisefid (*Rutilus frissi kutum*, Kamensky, 1901)

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### Abstract

To study anomaly among body components in the Mahisefid population situated in one of the spawning rivers, the abnormal fishes were sampled (n=10). Twenty six morphological properties were measured to be statistically ( $P<0.05$ ) compared with normally caught fishes. The mentioned characters were standardized based on the total length. Eight ratios including standard length (SL), snout length (SNL), dorsal fin height (DFH), distance between pelvic fin to ventral fin (PV), distance between snout to anterior part of dorsal fin (PRB), distance between posterior part of dorsal fin to anterior part of caudal fin (POB), upper part of caudal fin length (UCFL) and mid part of caudal fin Length (CCFL) of abnormal fishes differed significantly ( $P<0.05$ ) from those of healthy fishes. The morphological characters were classified at three components. Discriminant analysis showed the ratios of fork length and standard length were the significantly ( $P<0.05$ ) variables to screen samples. Draining diverse pollutants and heavy metals to the receiving rivers, inbreeding and technical errors during propagation were the most important reasons threatening the life cycle of Mahisefid populations.

**Keywords:** Mahisefid, Abnormality, Morphological Components.

### Introduction

Mahisefid accounted economically one the most important teleost fish species living in the Caspian Sea. The highest distribution was reported in the south and south west of Caspian Sea basin (Kiabi *et al.*, 1999) and included more than 60% of total catchments (amounted 10000 to 18000 metric tones per year) in the Iranian part of this important reservoir (Abdolhay *et al.*, 2010). With an increase in the population of cities and industrialization process in the recent years, the habitats of this species have been exposed to various pollutions. Besides, overfishing caused the Mahisefid populations not to able to naturally reproduce. In this regard, Iranian Fisheries Organization (IFO) has proceeded to artificially propagation and stocking this species in the natural waters. IFO has being released more than 200 million fingerlings every year to improve the stocks (Abdolhay and Tahori, 2006).

It seems that anthropogenic effects including diverse pollutions leached to the environment and human manipulations caused different abnormalities in the Mahisefid populations. Also, artificial

propagation as one of the most important reasons of abnormalities would chronically reduce the growth of fish species (Valipour and Khanipour, 2009). The gradually destruction of gene bank and genetic stocks are the main parameters during long-term effects. Decreasing in growth rate, mean length, fecundity and increasing in the deformed larvae during artificial propagation will show their effects through a long term process (Valipour and Khanipour, 2009).

Regarding to one of the most important reasons in the deformities is technical problems during artificially propagation in order to recruit the stocks, it is necessary to consider preparing rivers and naturally spawning conditions and if it is possible to get semi-propagation conditions in the ponds that broodstocks are ready to select naturally each other even in a small scale (Valipour and Khanipour, 2009; Kashefi *et al.*, 2012).

In this regard, the most prevalent deformities in the Mahisefid populations in the Caspian Sea is to change body ratios. The aim of the present study is to evaluate morphological differences between healthy and abnormal fishes.

## Materials and Methods

### Sample Preparation and Measuring Morphological Characters

In spring 2009, a total of ten randomly abnormal fishes were caught from Mahisefid populations in Cheshmeh Killeh (Tonekabon, Mazandaran Province) ( $50^{\circ}51'$ ,  $21^{\circ}52'$ ) with salic netting. Samples were fixed in the formaline solution (10% w/w) and transported to the ichthyology laboratory of Faculty of Natural Resources, University of Tehran. Twenty six morphological characters were measured in the healthy and abnormal samples (Figure 1) and then standardized ratios based on the total length were compared (Kashefi *et al.*, 2012).

### Statistical Analysis

After confirming homogeneity of variance and normality of the data using Leaven and Kolmogorov-Smirnov tests (Zar, 1999), respectively, t-test was used to compare significant differences ( $P < 0.05$ ) the characters between abnormal and healthy fishes. Principal component analysis (PCA) from MANOVA procedure was used to screen the variables ( $P < 0.05$ ) with SPSS version 19. Also, discriminant analysis (DA) was used to determine the most important variables to classify samples.

## Results

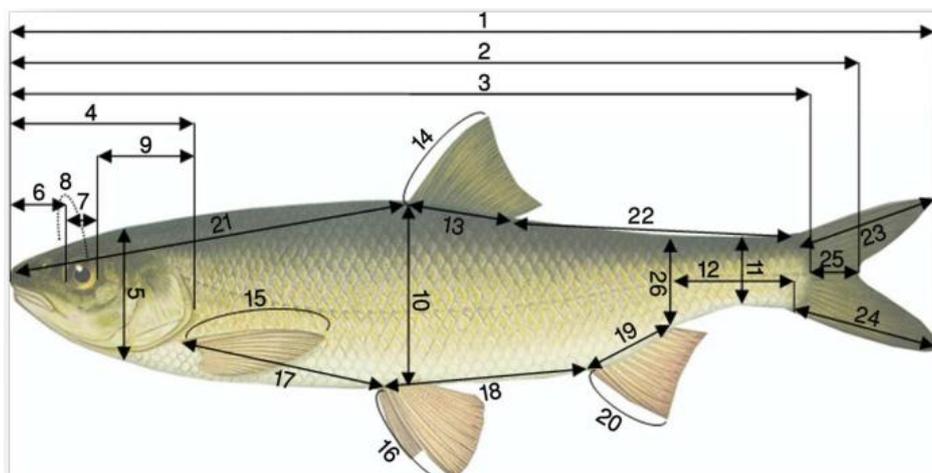
### Morphological Characters

To evaluate malformations and morphological differences, twenty six morphological characters were

compared between healthy and abnormal fishes (Table 1). The ratios of SL, SNL, DFH, PV, PRB, POB, UCFL and CCFL to TL (as SLr, SNLr, DFHr, PVr, PRBr, POBr, UCFLr and CCFLr, respectively) showed significantly ( $P < 0.05$ ) differences between healthy and abnormal fishes. The quantities of morphological characters (with exception HL, SNL, MAXH, PV, POB and UCFL) in abnormal fishes were higher than those of in healthy fishes (Table1).

### Multivariate Analysis (PCA and DA)

PCA showed that three components ( $F_1$ : 50.92%,  $F_2$ : 29.12% and  $F_3$ : 19.96%, respectively) described all 100% cumulative variance of the morphological characters. PCA reduced the twenty six variables into three principal components (PCs) which explained 100% of the variance cumulative. The PC1 and PC2 included approximately 80% of cumulative variance (not shown data). Loading coefficients obtained from the application of PCA are useful for showing the correlation between the original and the PCA transformed variables. The higher weighting, the more the variables have in common with the PC and the more it contributes to what the PC explains of the data structure. In this case, PC1 was high in UCFLr (0.991), POBr (0.962), HLr (0.809), PVr (0.803) and EDr (0.744), with positive values; and also high in PRBr (0.975), PFHr (0.973), SLr (0.935), CCFLr (0.922) and AFHr (0.899), but with negative values. PC2 was high in VAr (0.932), BEDr (0.928), MINHr (0.925) and HHr (0.888), with positive values and also high in DCFLr (0.534) and FLr (0.438), but with negative values. As shown in Figures 2 and 3, the morphological characters could be classified at three classes with similarity 40-50%.

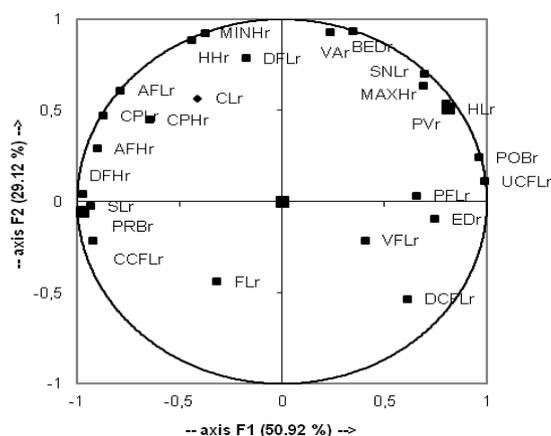


**Figure 1.** Morphological Characters used in abnormal and healthy fishes.

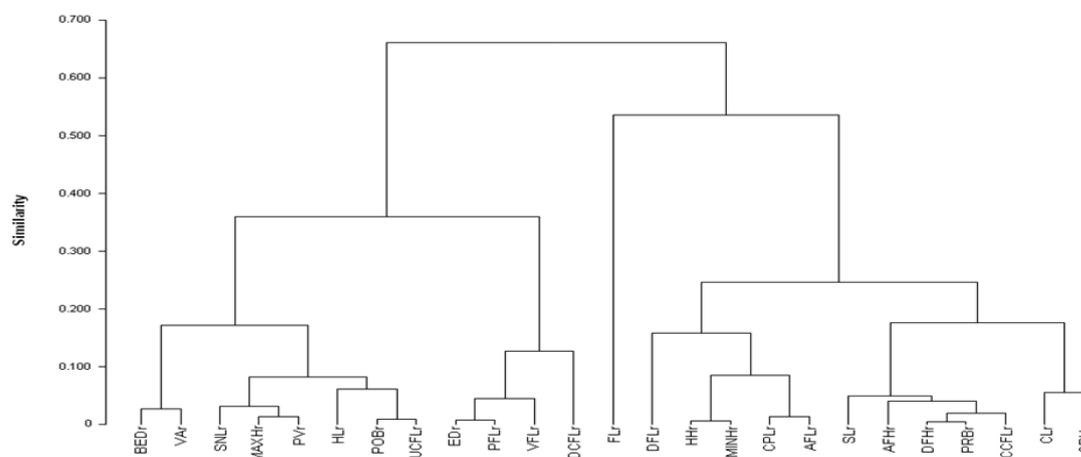
1: Total Length (TL); 2: Fork Length (FL); 3: Standard Length (SL); 4: Head Length (HL); 5: Head Height (HH); 6: Snout Length (SNL); 7: Eye diameter (ED); 8: Distance Between Eyes (BED); 9: Cheek Length (CL); 10: Maximum Height of Body (MAXH); 11: Minimum Height of Body (MINH); 12: Caudal Peduncle Length (CPL); 13: Dorsal Fin Length (DFL); 14: Dorsal Fin Height (DFH); 15: Pectoral Fin Length (PFL); 16: Ventral Fin Length (VFL); 17: distance between Pelvic fin to Ventral fin (PV); 18: distance between Pectoral fin to annus (VA); 19: Annus Fin Length (AFL); 20: Annus Fin Height (AFH); 21: distance between snout to anterior part of dorsal fin (PRB); 22: distance between posterior part of dorsal fin to anterior part of caudal fin (POB); 23: Upper part of Caudal Fin Length (UCFL); 24: Down part of Caudal Peduncle Length (DCFL); 25: Mid part of Caudal Fin Length (CCFL); 26: Caudal Peduncle Length (CPH)

**Table 1.** Morphological characters (mean, minimum and maximum; cm) of healthy and abnormal fishes and statistically comparison of body components ratios based on TL

Character	Healthy Fishes			Abnormal Fishes			P-value
	Mean	Minimum	Maximum	Mean	Minimum	Maximum	
TL	40.47	34	47.5	45.5	42	49.00	-
FL	36.58	31	43	41.4	38	44.80	0.627
SL	33.69	28.5	40	38.75	35.8	41.70	0.031
HL	6.67	5.45	8.45	6.30	6.2	6.40	0.055
HH	5.24	4.25	6.67	6.10	5.6	6.60	0.715
SNL	1.89	1.42	3.0	1.25	1.1	1.40	0.07
ED	1.08	0.9	1.22	1.10	1.1	1.10	0.269
BED	3.02	2.43	4.1	3.3	3.1	3.50	0.471
CL	3.74	2.99	4.65	4.35	4.2	4.50	0.557
MAXH	7.79	5.9	9.78	6.85	5.8	7.90	0.07
MINH	2.48	2.0	3.12	2.85	2.6	3.10	0.820
CPL	5.05	3.96	6.83	6.80	6.00	7.60	0.168
DFL	4.09	3.22	4.98	4.55	4.00	5.10	0.921
DFH	4.56	3.79	5.42	5.40	5.00	5.80	0.01
PFL	5.6	4.62	6.45	5.80	5.80	5.80	0.338
VFL	4.15	3.54	4.82	4.55	4.50	4.60	0.692
PV	9.91	7.82	11.68	9.70	8.80	10.60	0.031
VA	8.69	6.55	11.2	8.95	8.30	9.60	0.354
AFL	3.37	2.69	4.22	4.05	3.70	4.40	0.254
AFH	3.13	2.5	3.83	3.85	3.60	4.10	0.071
PRB	12.81	10.27	15.17	16.95	15.70	18.20	0.003
POB	17.07	14.3	20.13	13.75	12.70	14.80	0.0001
UCFL	6.91	5.87	8.04	6.80	6.40	7.20	0.003
DCFL	7.18	5.87	7.42	7.30	7.10	7.50	0.505
CCFL	1.76	1.5	2.12	2.60	2.50	2.70	0.004
CPH	3.28	2.53	3.9	3.80	3.60	4.00	0.286



**Figure 2.** Loading plot describing the relationship among the morphological characters derived from a PCA (Three clusters were shown with red lines).



**Figure 3.** Cluster diagram describing similarities among the ratios of morphological characters to TL

As observed in Figure 4, SNL was the first factor made significantly ( $P < 0.05$ ) different appearance between abnormal and healthy fishes. The DA showed that FLr and SLr had a canonical correlation 91.3%, meaning that FLr and SLr were the best variables to differentiate healthy and abnormal Mahisefid populations (not shown data).

## Discussion

Naturally, there are a number of abnormalities in the broodstock populations of live animals, but the more the interfering by human, the more abnormalities were observed. In wild, mutations (Line, 1997) and parasite infections (Brown and Nunez, 1998) had the highest effects on the formation of abnormality. In Mahisefid, the steps of artificial propagation and the stocking of larvae to obtain juvenile are associated with human intervention. As a result, this increases the probability of malformations. From genetic viewpoint, the most important objects in the recruitment programs of fish stocks is to maintain genetic variance. Thus, any selection programs are prevented and all caught fishes are used for artificial propagation. Selecting broodstocks with specific phenotype properties caused small genetic variance and finally producing fishes with pure loci that could not tolerate the environmental changes. As a result, general competence and ability to survive in its environment would reduce. Technical errors and lack of correct information would cause some abnormalities during the early stages of reproduction (Harris and Hulsman, 1991; Polo *et al.*, 1991).

Also, different factors including egg density, water pollution, decreasing dissolved oxygen, thermal, mechanical and salinity shocks, irradiation and light intensity could be effective on the incidence

of malformations through different incubation stages (Haya, 1989; Weis and Weis, 1989). The early life stages of fishes are the most sensitive stage. In the case of Mahisefid, this period was spent in the earthy ponds and under human considerations meaning that it is a rudimentary domestication as a transitional time during producing juveniles to stock into the natural water bodies (reservoir, river and ocean). Other factor influencing on the abnormality of Mahisefid population is hand feeding with the formulated diets. If the diet has deficiency on the vitamin C, tryptophan and phospholipids, it will increase the probability of abnormality (Weis and Weis, 1989). If environmental conditions including temperature and dissolved oxygen were not in the optimum ranges, they will increase the probability of abnormality in the same race or the following races. Different diseases can be one of the other reasons of abnormality (Dulin, 1979). In this regard, water habitats polluted with inputs containing heavy metals such as Pb, Zn, Cu and Cd have showed the high rates of abnormalities (Snelder, 1995; Wilson, 1999). Drainage of organic poisons (e.g. DDT) from farm lands to water habitats will severely increase the skeleton abnormality on fishes living in the receiving water bodies (Bengtson *et al.*, 1985). Such skeleton abnormalities not only have direct effect on life style of fishes, but also they have indirect effects on the natural activities and swimming behavior (Sadler, 1990). Revise laws to monitor the effluents of industrial factories and optimize the conditions of Mahisefid propagation at least in the semi-natural conditions are the primary factors in order to decrease stress on the Mahisefid broodstocks. Different ecomorphs of Mahisefid populations have been reported (Abdolhay *et al.*, 2010). Therefore, monitor Mahisefid populations via tagging methods to



**Figure 4.** The pictures of healthy (left) and abnormal (right) Mahisefid samples

decrease inbreeding will be very important in the near future (unpublished data).

It was primarily confirmed an abnormal population of Mahisefid in the Caspian Sea. The abnormal fishes had different body component ratios (such as SLr, SNLr, PVr etc). Although, diverse abiotic (diverse pollutants) and biotic (inbreeding) parameters have been proposed as affecting factors but it needs to a multi disciplinary approach to conserve Mahisefid populations.

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