

Journal of Coastal Life Medicine

journal homepage: www.jclmm.com



Original article

doi: 10.12980/jclm.4.2016J6-33

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Morphological differences among the *Garra rufa* populations (Cyprinidae) in Tigris River system of Southeast TurkeyTark Cicek¹, Serbest Bilici^{2*}, Erhan Ünlü¹¹Department of Biology, Section of Hydrobiology, Faculty of Science, Dicle University, 21000, Diyarbakır, Turkey²Department of Animal Science, Faculty of Agriculture, Sırnak University, 73000, Sırnak, Turkey

ARTICLE INFO

Article history:

Received 4 Feb 2016

Received in revised form 19 Feb, 2nd

revised form 20 Feb 2016

Accepted 6 Apr 2016

Available online 6 Jul 2016

Keywords:

Garra rufa

Tigris River

Morphological

Discriminant analysis

Cyprinidae

ABSTRACT

Objective: To determine morphometric and meristic variations between *Garra rufa* (*G. rufa*) samples obtained from different locality in Tigris River.**Methods:** Transformed morphometric characters were subjected to discriminant analysis and according to grouping model, number of discriminant function and morphologic variation between populations with respect to their importance of explaining total variation were determined.**Results:** Success rate of classifying the groups according to the results of discriminant analysis of morphometric characters of *G. rufa* individuals, belonging to seven different localities of Tigris and Euphrates river system revealed as 56.7%. Savur stream group showed different distribution from the other groups. Success rate of classifying the groups according to the discriminant analysis of meristic characters of *G. rufa* individuals appeared as 56.32%.**Conclusions:** Devegeçidi Dam Lake and Kulp stream groups were the ones which showed the most different distributions in the discriminant analysis. Between locality groups of *G. rufa* individuals belonging to Cyprinidae family, meristic and especially morphometric variations were significantly found in the consequent of discriminant analysis

1. Introduction

It is very well known that morphological characteristics may show high flexibility in response to variations in environmental conditions[1-3]. Meristic characters are sensitive to the environment and especially temperature when they complete their full development[4]. Natural factors also may affect the meristic counts[4]. It has been found that prenatal and post-fertilization effects (temperature, salinity and other environmental factors)

significantly reduce meristic differences in the number of fin bones[4-6]. In general, individuals developed at low water temperatures produce more meristic counts compared to those developed at high temperature[7].

Stream fishes have wider pelvic, pectoral, anal and dorsal fins to be used in a stream for stability and maneuverability[8,9]. Caudal peduncles of stream fishes are wider and stronger with more muscle in a lesser depth[10]. Pectoral fins of stream fishes are positioned deeper compared to lake fishes to improve their adaptation to stream and help them swim stronger and steadier[11].

The hierarchical model of genetic difference reflecting the suitable habitat distribution is sensitive to speciation caused by the discrepancies in the isolation of remote or separate sub-populations. In a large scale, allopatric speciation which took place in isolated areas can occur due to the differences

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Foundation Project: Supported by Dicle University with DUPAB-07-02-016 numbered project.

The journal implements double-blind peer review practiced by specially invited international editorial board members.

accumulated during continuous isolation of drainages.

Migratory individuals carry genetic material between populations and gene transfer can thus restrict the adaptive classification of the populations in nature[12-14]. Natural habitats of annual fishes are natural lakes that can be easily isolated from each other. In this way, disruptions of gene transfers may occur very often between populations. In addition, annual fishes have short generation times (< 1 year in nature) and high metabolic rates[15].

Garra rufa (Heckel, 1843) (*G. rufa*) and *Garra variabilis* (*G. variabilis*) are well separated from each other clearly based on geometric morphometric methods and *G. variabilis* shows more metric and meristic variation than *G. rufa* throughout the localities[16,17]. According to Qayoom *et al.*, *Garra gotyla gotyla* have isometric growth pattern from Kashmir[18].

It is inevitable for a specie that shows a wide dispersion to show some genetical variations with the ecological conditions that they live in the river systems. Some sets were constructed for irrigation and hydroelectricity on Tigris River system. These sets generated the isolated parts in the river basin. Therefore, we tried to determine morphometric and meristic variations between *G. rufa* samples by discriminant analysis that are caught from asunder or disconnected seven different localities and show a wide dispersion in Tigris and Euphrates river system.

2. Materials and methods

G. rufa samples were obtained from seven different localities (Sinek stream, Devegeçidi dam lake, Göksu stream, Savur stream, Kulp stream, Kayser stream and Batman stream) by using gill net, cover net and electro-shocker. Samplings were made between September 2007 and April 2008. Fish samples were brought to 5%–6% formaldehyde inside the laboratory. Determined samples were preserved in 70% alcohol. In order to determine the morphological characters of the fish samples, measurements of morphometric characters and counting of meristic characters were made.

A total of 26 morphological variances which were about morphometric characters were measured by sensitive 0.01 mm electronic compass and in this measurement, truss network method was used (Figures 1 and 2)[19-23]. About countable meristic characters, totally 13 different variances were used: numbers of dorsal opined fin, dorsal furcated fin ray, ventral spined fin ray, ventral frucrated fin's ray, anal opined fin ray, anal branched fin ray, pectoral fin in left spined ray, pectoral fin in left furcated ray, pectoral fin in right spined ray, pectoral fin in right furcated ray, number of gill arch spine, lateral line in left number of scale and lateral line in right number of scale.

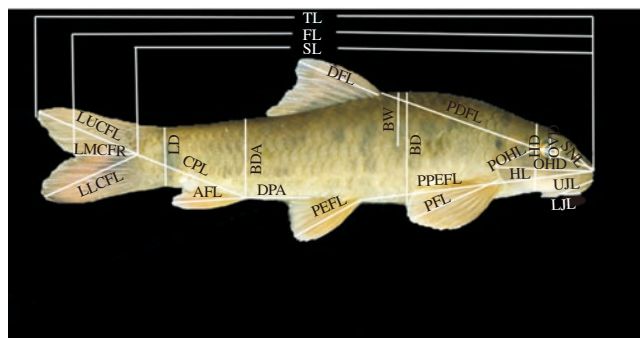


Figure 1. Morphometric measurements worked on *G. rufa*.

TL: Total length; FL: Fork length; SL: Standard length; SNL: Snout length; OHD: Horizontal ocular diam; OVD: Vertical ocular diam; UJL: Upper lip length; LJL: Lower lip length; HL: Head length; HD: Head height; POHL: Postocular head length; PFL: Pectoral fin length; BD: Body height; BW: Body width; DFL: Dorsal fin length; PDFL: Predorsal length; PEFL: Pelvic Fin Length; PPEFL: Prepelvic length; DPA: Distance between pelvik and anal fin; BDA: Body height in anal level; AFL: Anal fin length; CPL: Caudal pedunculus length; LD: Body height in caudal pudunculus area; LUCFL: Upper lab length of caudal fin; LMCFR: Caudal fin's fork's length; LLCFL: Length of lower lab of caudal fin.

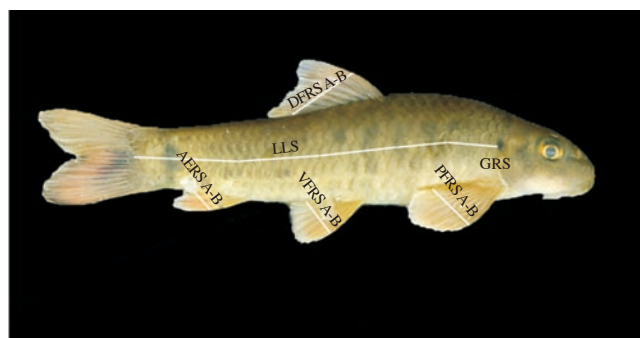


Figure 2. Meristic characters worked on *G. rufa*.

DFRS(A): Dorsal ray score (spine); DFRS(B): Dorsal ray score (branched); VFRS(A): Ventral ray score (spine); VFRS(B): Ventral ray score (branched); AFRS(A): Anal ray score (spine); AFRS(B): Anal ray score (branched); PFRSA(L): Pectoral ray score (left part branched); PFRSA(R): Pectoral ray score (right part spine); PFRSB(R): Pectoral ray score (right part branched); GRS: Number of gill rakers (under operculum); LLS(L): Lateral line score (left part); LLS(R): Lateral line score (right part).

In order to determine morphologic variations between *G. variabilis* populations by calculating all the morphometric characters SL, we tried to eliminate the variations that were derived from length. Transformed morphometric characters were subjected to discriminant analysis and according to grouping model, number of discriminant function and morphologic variation between populations with respect to their importance of explaining total variation were determined. Features that provided the classifications and their effective functions were determined. In two dimensions, based on two different discriminant functions, the place of discriminant functions were determined. The features of classification and their influential functions were determined using stepwise regression analysis. With the help of canonical discriminant function, the limit maps of the groups in a two

dimensional platform were created. Among the distinguished groups, the place of medium group (group centers) was detected [20,22]. Similar applications about discriminant analysis were also applied for countable meristic characters. Morphometric and meristic variations between *G. variabilis* populations were shown on plot charts. Furthermore, morphometric differences between populations were analyzed with ANOVA and *F*-test.

3. Results

In order to investigate the morphologic differences of *G. rufa* populations, 158 *G. rufa* individuals were analyzed (25 individuals from Sinek stream, 4 from Devegeçidi dam lake, 29 from Göksu, 2 from Savur stream, 28 from Kulp stream, 33 from Kayser stream and 37 from Batman stream). The standard heights of the samples were between 77–201 cm and the height difference between populations was no statistically significant ($P > 0.05$). The standard height belonging to populations and morphometric characters that were calculated as the percentage of standard height were given in Table 1. In terms of morphometric characters of *G. variabilis*, between the variations of LUCFL/LLCFL, SL/OHD, OVD/OHD, SL/PFL, BD/LD, PFL/PEFL, SL/HD, PFL/AFL, SL/OVD, PFL/DFL, HL/HD, UJL/LJL, SL/UJL, SL/LUCFL, SL/LLCFL, SL/LMCFR, SL/DFL, FL/SL, SL/PEFL, TL/SL, SL/LD, SL/POHL vs. SL/BD, SL/DPA, SL/AFL, SL/CPL, SL/

SNL, SL/PPEFL, SL/HL, SL/BW, SL/BDA, SL/PDFL, SL/LJL, BD/BDA, significant differences were seen ($P < 0.05$).

From of morphometric characters, SL/OHD, SL/HD, SL/PPEFL, SL/DPA, SL/AFL, SL/LUCFL, UJL/LJL were more determinant to reveal the variations. This variation was originated from all seven locality groups. In addition to this, between other variations, no differences were found ($P > 0.05$).

From 13 countable meristic characters belonging to populations, four of them were different from the others among populations (Table 2). The mentioned difference was based on GRS, LLS(L), LLS(R) vs. DFRSB characteristics ($P < 0.05$). However, LLS(L), LLS(R) and DFRSB variations were not determinant in this analysis despite showing significant correlations. In addition to this, no differences were found between other variations.

According to the result of discriminant analysis which was applied to the transformed morphometric characters that were obtained from 158 *G. rufa* individuals belonging seven different populations, six discriminant function corresponded to 99.9% of total variation. Hence, six functions were considered that 1) separation group generated 39.1% (canonical correlation = 0.662) of total variation, 2) separation group generated 33.3% (canonical correlation = 0.631) of total variation, 3) separation group generated 16.2% (canonical correlation = 0.493), 4) separation group generated 6.7% (canonical correlation

Table 1
Morphometric characters of different *G. rufa* populations in Tigris River (%).

Morphometric characters	Sinek stream	Devegeçidi dam lake	Göksu stream	Savur stream	Kulp stream	Kayser stream	Batman dam lake
TL/SL	1.25 ± 0.07	1.26 ± 0.03	0.20 ± 0.01	0.30 ± 0.02	1.25 ± 0.03	1.24 ± 0.04	1.24 ± 0.50
FL/SL	1.15 ± 0.06	1.15 ± 0.03	1.16 ± 0.01	1.15 ± 0.02	1.15 ± 0.01	1.14 ± 0.02	1.15 ± 0.04
SL/SNL	8.24 ± 1.77	8.37 ± 0.12	8.36 ± 0.39	8.00 ± 0.47	8.00 ± 0.55	8.35 ± 1.50	7.96 ± 0.67
SL/OHD	19.47 ± 2.25	16.64 ± 1.16	18.67 ± 1.42	23.51 ± 2.75	20.19 ± 1.90	19.20 ± 3.90	20.96 ± 2.09
SL/OVD	19.34 ± 2.15	17.20 ± 0.95	20.04 ± 1.56	23.89 ± 4.65	21.13 ± 1.40	20.50 ± 4.30	21.51 ± 2.12
SL/UJL	14.02 ± 1.56	13.31 ± 2.44	13.05 ± 1.35	15.70 ± 1.79	15.15 ± 4.80	12.80 ± 2.50	13.71 ± 2.37
SL/LJL	9.03 ± 1.20	8.58 ± 1.07	8.87 ± 0.80	8.93 ± 0.42	9.43 ± 1.20	9.77 ± 1.90	9.67 ± 0.68
SL/HL	4.38 ± 0.26	4.88 ± 1.35	4.35 ± 0.16	4.45 ± 0.24	4.31 ± 0.15	4.44 ± 0.80	4.33 ± 0.69
SL/HD	6.83 ± 0.38	10.04 ± 3.95	6.89 ± 0.30	6.70 ± 0.38	6.98 ± 0.28	6.95 ± 1.17	7.15 ± 0.91
SL/POHL	11.85 ± 1.18	13.19 ± 0.96	12.03 ± 0.90	12.59 ± 0.08	11.65 ± 0.70	12.10 ± 2.70	12.16 ± 1.18
SL/PFL	4.61 ± 0.36	5.26 ± 1.15	4.50 ± 0.48	4.97 ± 0.43	4.71 ± 0.75	4.80 ± 0.90	5.04 ± 0.63
SL/BD	4.72 ± 0.43	4.28 ± 0.20	5.08 ± 2.32	3.70 ± 0.22	4.61 ± 0.31	4.57 ± 0.72	4.95 ± 0.52
SL/BW	7.78 ± 0.60	7.56 ± 0.55	7.84 ± 0.53	6.67 ± 0.38	7.87 ± 0.79	7.83 ± 1.43	8.30 ± 0.66
SL/DFL	4.75 ± 0.41	4.64 ± 0.37	4.78 ± 0.27	5.11 ± 0.07	4.99 ± 0.32	5.04 ± 0.90	5.11 ± 0.54
SL/PDFL	2.06 ± 0.12	1.97 ± 0.04	2.06 ± 0.06	2.08 ± 0.10	2.00 ± 0.08	2.11 ± 0.40	2.09 ± 0.07
SL/PEFL	5.23 ± 0.43	4.93 ± 0.82	5.13 ± 0.29	5.48 ± 0.28	5.04 ± 0.28	5.13 ± 1.05	5.44 ± 0.48
SL/PPEFL	1.86 ± 0.13	1.86 ± 0.08	1.87 ± 0.04	1.75 ± 0.02	1.79 ± 0.07	1.86 ± 0.33	1.84 ± 0.06
SL/DPA	4.20 ± 0.32	4.32 ± 0.28	9.99 ± 0.25	3.89 ± 0.12	4.22 ± 0.41	4.62 ± 0.77	4.27 ± 0.48
SL/BDA	6.19 ± 1.14	5.65 ± 0.12	6.29 ± 2.21	5.58 ± 0.01	5.96 ± 0.40	5.79 ± 0.88	5.91 ± 0.30
SL/AFL	5.61 ± 0.54	5.36 ± 0.47	5.48 ± 0.40	6.02 ± 0.22	5.49 ± 0.35	5.49 ± 1.07	6.08 ± 0.50
SL/CPL	6.18 ± 0.81	6.66 ± 0.65	6.37 ± 0.56	6.02 ± 0.21	6.66 ± 0.45	6.74 ± 1.17	6.44 ± 0.24
SL/LD	7.59 ± 1.30	7.48 ± 0.36	7.84 ± 0.44	7.56 ± 0.23	7.59 ± 0.40	7.81 ± 1.36	7.87 ± 0.42
SL/LUCFL	3.93 ± 0.41	3.79 ± 0.55	4.11 ± 0.26	4.50 ± 0.13	4.25 ± 0.25	4.32 ± 0.85	4.32 ± 0.32
SL/LMCFR	6.66 ± 1.08	6.36 ± 0.40	6.47 ± 0.53	7.18 ± 0.13	7.09 ± 0.63	7.14 ± 1.37	7.12 ± 0.65

All values were expressed as mean ± SD.

Table 2Meristic characters of different *G. rufa* populations in Tigris River.

Meristic characters	Sinek stream	Devegeçidi dam lake	Göksu stream	Savur stream	Kulp stream	Kayser stream	Batman barrage
DFSR-A	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00
DFSR-B	7.92 ± 0.28	8.00 ± 0.00	7.90 ± 0.31	8.00 ± 0.00	8.00 ± 0.00	7.76 ± 0.44	8.00 ± 0.00
VFRS-A	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00	3.00 ± 0.00
VFRS-B	5.00 ± 0.00	5.00 ± 0.00	5.00 ± 0.00	5.00 ± 0.00	5.00 ± 0.00	5.00 ± 0.00	5.00 ± 0.00
AFRS-A	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00
AFRS-B	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00
PFRSA-L	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00
PFRSB-L	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00
PFRSA-R	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00
PFRSB-R	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00	12.00 ± 0.00
GRS	29.08 ± 0.64	35.25 ± 1.50	30.38 ± 0.94	30.00 ± 0.00	30.61 ± 1.17	30.82 ± 1.24	38.65 ± 1.99
LLS-L	33.44 ± 1.36	33.75 ± 0.50	33.55 ± 1.50	33.50 ± 2.12	32.93 ± 1.15	30.06 ± 1.50	34.08 ± 1.28
LLS-R	33.60 ± 1.80	36.75 ± 1.26	33.41 ± 1.27	32.50 ± 0.71	32.79 ± 1.03	32.21 ± 1.27	32.86 ± 5.01

All values were expressed as mean ± SD.

= 0.343), 5) separation group generated 3.5% (canonical correlation = 0.256) and 6) separation group generated 1.1% (canonical correlation = 0.148) of the rest variation (canonical discriminant).

In the statistical check of discriminant functions, even there were seven localities, one discriminant function was obtained. This function was significant. Only one discriminant function was 100% of total variation (canonical correlation = 0.940).

In the analysis made according to some morphometric characters of 158 *G. rufa* samples which were brought from seven localities, success rate of separating locality groups in terms of morphometric characters was 56.7%.

3.1. Evaluation of the results of discriminant analysis for morphometric characters

A total of 14 of 25 samples brought from Sinek stream stayed in their own group, one of them was transferred to Savur stream, five of them were transferred to Göksu, two of them were transferred to Kulp, one of them was transferred to Kayser and two of them were transferred to Batman stream group. Possibility of the samples brought from Sinek stream in their own group in terms of studied characters was 56%, $P = 0.562$.

A total of two of four samples brought from Devegeçidi dam lake stayed in their own group, one of them was transferred to Sinek stream and the other were transferred to Göksu stream. Possibility of the samples brought from Devegeçidi dam lake in their own group in terms of studied characters was 50%, $P = 0.50$.

A total of 16 of the 29 samples brought from Göksu stream stayed in their own group; four of them were in Sinek stream, two

of them were in Kulp stream, two of them were in Kayser stream and five of them were in Batman stream group. Possibility of the samples brought from Göksu stream in their own group in terms of studied characters was 55.2%, $P = 0.552$.

While one of the two samples brought from Savur stream stayed in its own group, the other were in Batman stream group. Possibility of the samples brought from Savur stream in their own group in terms of studied characters was 50%, $P = 0.50$.

A total of 11 of the 28 samples brought from Kulp stream stayed in their own group, four of them were in Göksu, six of them were in Kayser and seven of them were in Batman stream group. Possibility of the samples brought from Kulp stream in their own group in terms of studied characters was 39.3%, $P = 0.393$.

A total of 23 of the 33 samples brought from Kayser stream stayed in their own group, five of them were in Batman stream, one of them was in Sinek stream, three of them were in Göksu stream and the last one was in Kulp stream group. Possibility of the samples brought from Kayser stream in their own group in terms of studied characters was 69.7%, $P = 0.697$.

A total of 24 of 37 samples brought from Batman stream stayed in their own group, one of them was in Sinek stream, one of them was in Devegeçidi dam lake, one of them was in Göksu stream, one of them was in Savur stream, four of them were in Kulp stream and other five of them were in Kayser stream group. Possibility of the samples brought from Batman stream in their own group in terms of studied characters was 64.9%, $P = 0.649$.

In the analysis made according to the meristic characters belonging to *G. rufa* samples brought from three different regions, success rate of separating localities groups in terms of meristic characters was 56.32% (Figure 3).

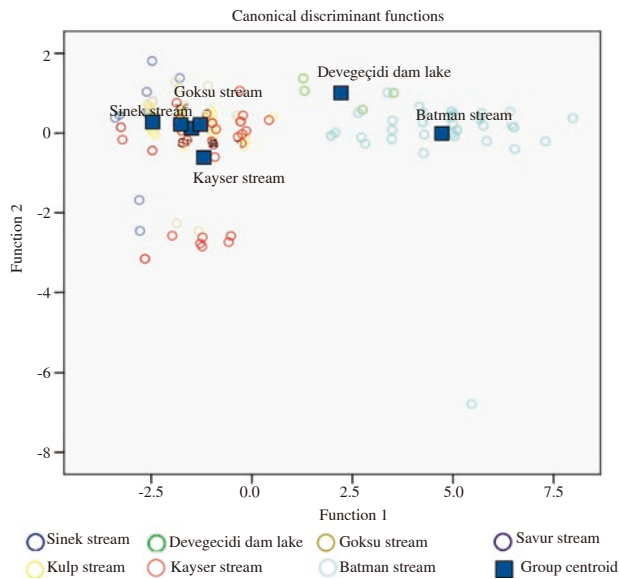


Figure 3. Results of discriminant analysis belonging to morphometric characters.

3.2. Evaluation of the results of discriminant analysis for meristic characters

A total of 19 of 25 samples brought from Sinek stream stayed in their own group, six of them were transferred to Göksu stream. Possibility of the samples brought from Sinek stream in their own group in terms of studied characters was 76%, $P = 0.76$.

A total of two of four samples brought from Devegeçidi dam lake stayed in their own group, two of them were transferred to Batman stream group. Possibility of the samples brought from Devegeçidi dam lake in their own group in terms of studied characters was 50%, $P = 0.50$.

A total of 12 of 29 samples brought from Göksu stream stayed in their own group, five of them were transferred to Sinek stream and 12 of them were transferred to Kayser stream group. Possibility of the samples brought from Göksu stream in their own group in terms of studied characters was 41.4%, $P = 0.414$.

A total of two samples brought from Savur did not stay in their own group and they were transferred to Batman stream group. Possibility of the samples brought from Savur stream in their own group in terms of studied characters was 0%, $P = 0.0$.

While none of 28 samples brought from Kulp stream stayed in their own group, six of them were transferred to Sinek stream, seven of them were transferred to Göksu stream and 15 of them were transferred to Kayser stream group. Possibility of the samples brought from Kulp stream in their own group in terms of studied characters was 0%, $P = 0.00$.

A total of 22 of 33 samples brought from Kayser stream stayed in their own group, six of them were in Sinek stream and other were in Göksu stream group. Possibility of the samples brought from Kayser stream in their own group in terms of studied characters was 66.7%,

$P = 0.667$.

A total of 34 of 37 samples brought from Batman stream stayed in their own group, three of them were transferred to Devegeçidi dam lake group. Possibility of the samples brought from Batman stream in their own group in terms of studied characters was 91.9%, $P = 0.919$.

In the discriminant analysis that was made by using meristic characters for *G. rufa*, success rate of classifying the groups according to the variations that showed in groups was 56.3% (Figure 4).

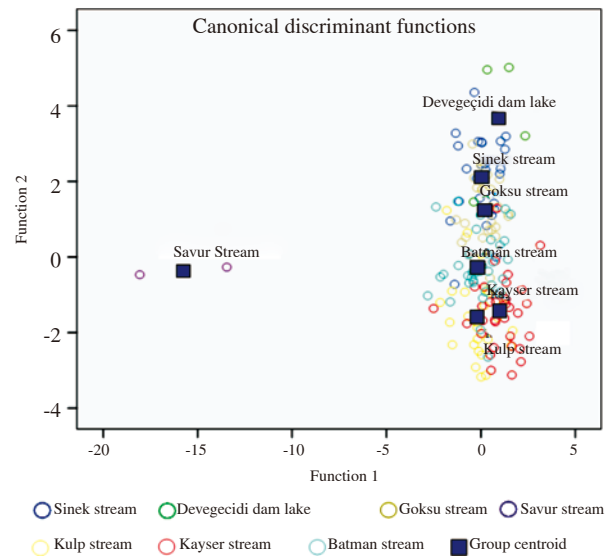


Figure 4. Results of discriminant analysis belonging to meristic scores.

4. Discussion

In the discriminant analysis that was made by using morphometric characters for *G. rufa*, success rate of classifying groups according to the variations that showed in groups was 56.7% (Figure 3). If we look at the canonical discriminant function, we see that Savur, Göksu and Kulp streams show very similar distributions. The other ones who have similar distributions are Kayser and Sinek stream. Batman stream group seems to be very different from the other groups. We see that Devegeçidi dam lake group is the one which resembles mostly with Batman stream group. In second canonical discriminant function, we see that group distributions are close to each other in general. We can say that generally, the differences between the groups are in the first discriminant function. Although, the groups that are close to each other on the distribution graph are geographically far from each other, we can say that the populations in these groups have similar habitat specifications. It is sayable that the reason why there is low success rates for classifying the groups may be using one meristic characters (GRS) only.

In the first canonical discriminant analysis, Savur stream group showed different distribution than the other groups. For Savur stream, such a conclusion may base on the habitat characters and

the lack of samples are obtained from localities. If we check the second canonical discriminant analysis, we see that Devegeçidi dam lake and Kulp stream are the ones which show most unlikely distributions. Geographical distance and isolation mechanisms deriving from barrage sets may be the reasons of showing different distributions. Again we see that Kulp stream and Kayser stream groups that are geographically close and have similar water characters, show very similar distributions. We see that Batman stream group shows similar distribution to these groups. The other groups which have closer distributions are Sinek and Göksu stream. Despite using seven morphometric characters here (SL/OHD, SL/HD, SL/PPEFL, SL/DPA, SL/AFL, SL/LUCFL, UJL/LJL), success rate of classifying the groups is low (57.6%). Depending on having low success rate of classifying the groups, it is seen that morphometric characters that are used in discriminant analysis are insufficient to have higher success rates for classifying the groups. Although locality groups are different, they usually showed similar distributions. Therefore, we can associate with the idea of having similar habitat conditions for the populations of each group.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgments

This study was supported by Dicle University with DUPAB-07-02-016 numbered project.

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