

**MORPHOLOGICAL DIFFERENTIATION OF ISKANDARIA
KUSCHAKEWITSCHI (HERZENSTEIN, 1890) POPULATIONS DISTRIBUTED IN
THE CHORTOQSOY AND OLTARIQSOY RIVERS**

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ABSTRAKT

This study presents a comparative analysis of the morphological differentiation of *Iskandaria kuschakewitschi* populations distributed in the Chortoqsoy and Oltariqsoy rivers. A total of 15 specimens from the Chortoqsoy population and 19 specimens from the Oltariqsoy population were examined. Morphometric measurements were obtained following the methodology proposed by Kottelat and Freyhof (2007), and statistical analyses were performed using Student's t-test. The results revealed significant differences in 14 out of 33 morphometric characters between the two populations. The most pronounced differences were observed in head length, predorsal and postdorsal lengths, fin lengths, and caudal peduncle parameters. Coefficients of variation indicated a higher level of morphological variability in the Oltariqsoy population. The detected differences may be associated with variation in ecological conditions, hydrological regime, and microhabitat characteristics of the studied river systems. The findings provide an important scientific basis for further ecological, morphological, and taxonomic studies of local populations of *Iskandaria kuschakewitschi* in the Fergana Valley.

Keywords: *Iskandaria kuschakewitschi*, morphometric analysis, population differentiation, Chortoqsoy River, Oltariqsoy River, coefficient of variation, Nemacheilidae, morphological variability, endemic fish, Fergana Valley.

Introduction

Annotatsiya

В данной работе проведён сравнительный анализ морфологической дифференциации популяций *Iskandaria kuschakewitschi*, распространённых в реках Чортоксой и Алтыарыксой. В исследовании изучено 15 экземпляров из популяции Чортоксося и 19 экземпляров из популяции Алтыарыксося. Морфометрические показатели рыб определялись по методике Kottelat и Freyhof (2007), статистическая обработка данных проводилась с использованием критерия Стьюдента. Результаты показали наличие достоверных различий по 14 из 33 морфометрических признаков между двумя популяциями. Наиболее выраженные различия отмечены по длине головы, преддorsальной и постдorsальной длине, длине плавников и параметрам хвостового стебля. Значения коэффициента вариации свидетельствуют о более высокой

морфологической изменчивости популяции Алтыарыксоя. Выявленные различия, вероятно, связаны с особенностями экологических условий, гидрологического режима и микробиотопов исследуемых рек. Полученные результаты представляют важную научную основу для комплексного экологического и таксономического изучения локальных популяций *Iskandaria kuschakewitschi*.

Ключевые слова: *Iskandaria kuschakewitschi*, морфометрический анализ, популяционная дифференциация, Чортоксой, Алтыарыксой, коэффициент вариации, Nemacheilidae, морфологическая изменчивость, эндемичные рыбы, Ферганская долина.

Introduction

The capacity of populations to adapt and evolve as independent biological entities under different environmental conditions is primarily determined by the level of individual exchange and gene flow among them. Restriction of gene flow or increasing geographic isolation among populations may lead to significant phenotypic and genetic differentiation within a species. At the same time, changes in habitat conditions intensify selective pressures, thereby promoting the formation of local adaptive processes in certain populations. As a result, some organisms exhibit high adaptability under complex ecological conditions and undergo local adaptive divergence (Kawecki & Ebert, 2004). Such differences serve as important scientific criteria for distinguishing populations, assessing their ecological status, and developing effective management strategies. For this reason, morphological approaches based on body shape and meristic characters have long been used in fish population identification (Heincke, 1898) and still retain their scientific significance today. Despite the rapid development of molecular-genetic and biochemical methods used to detect neutral genetic variation, morphological characters continue to be one of the most important approaches for differentiating fish populations and identifying population units (Swain & Foote, 1999). In particular, morphometric and meristic characters are recognized as the most widely used morphological criteria in comparative studies of fish populations. Morphometric characters represent continuous measurements reflecting different aspects of body shape, whereas meristic characters are countable repetitive anatomical structures formed during embryonic or larval development and remaining relatively stable throughout ontogenesis (Turan, 2004). Furthermore, the analysis of morphometric differences and coefficients of variation among populations is of considerable importance for evaluating their ecological status and adaptive characteristics (Azamov et al., 2024).

The genus *Iskandaria* Prokofiev, 2009 is represented by two species in the river systems of Uzbekistan: *Iskandaria pardalis* (Turdakov, 1941) and *Iskandaria kuschakewitschi* (Herzenstein, 1890) (Qayumova, 2022). One of these benthic loaches, *Iskandaria pardalis*, is an endemic species of the Amu Darya basin and was originally recorded from the Dushanbadarya River, a right tributary of the Kofarnihon River in Tajikistan (Fayziyeva & Sheraliyev, 2025). In contrast, *I. kuschakewitschi* is considered an endemic species of the Fergana Valley and is of particular scientific interest. It was first reported from the Marg‘ilonsoy River (Berg, 1949; Amanov, 1985; Sheraliyev & Kayumova, 2022; Fayziyeva

& Sheraliyev, 2025). Subsequent studies revealed that this species forms small populations not only in the northern and southern mountain rivers of the valley, but also in artificial irrigation systems such as the Great Fergana Canal and the South Fergana Canal (Sheraliyev & Kayumova, 2022). In recent years, records of this species from the Qamchiqsoy, Kosonsoy, and So‘x rivers indicate an ongoing expansion of its distribution range (Qayumova, 2025; Qayumova et al., 2025; Qayumova et al., 2026). However, the degree of gene flow and population connectivity among populations inhabiting ecologically distinct northern and southern river systems of the valley remains insufficiently studied. Studies focused on the morphological differentiation of *I. kuschakewitschi* populations have intensified since 2024 (Qayumova et al., 2024; Qayumova & Begmatova, 2024). Nevertheless, there is still a need for comparative evaluation of morphological differences, variability, and morphometric characters among populations inhabiting ecologically contrasting river systems. Therefore, the present study aimed to comparatively assess the variability and degree of morphological differentiation of *Iskandaria kuschakewitschi* populations distributed in the Chortoqsoy and Oltiariqsoy rivers.

Materials and Methods

Specimens of *I. kuschakewitschi* were collected from the Chortoqsoy and Oltiariqsoy rivers during the summer, autumn, and winter seasons of 2024–2025. Fish sampling was carried out using nets measuring 2–3 m in length and 1.5–2 m in width. The collected specimens were initially fixed in 10% formalin solution under field conditions. After three days of fixation, the samples were transferred to 75% ethanol for long-term preservation. Morphometric measurements were conducted in laboratory conditions using an electronic caliper with an accuracy of 0.01 mm. All measurements were taken from the left side of the fish according to the methodology proposed by Kottelat and Freyhof (2007). Biometric analyses of variability in morphometric characters and interpopulation differences were performed following the methods described by G. Lakin (1990). Statistical significance of differences was evaluated using Student’s t-test at the 1% significance level ($P > 0.01$). All statistical analyses were performed using MS Excel 2019. The following abbreviations were used in the study: TL- total length; SL - standard length; HL - head length; BDD - body depth at dorsal-fin origin; BWD- body width at dorsal-fin origin; PRD - predorsal length; PSD - postdorsal length; PRV - preventral length; PRA- preanal length; PrAn - preanus length; DFL- dorsal-fin length; DFBL - dorsal-fin base length; AFL - anal-fin length; AFBL - anal-fin base length; PFL - pectoral-fin length; VFL - pelvic-fin length; CFL - caudal-fin length; CPL - caudal peduncle length; CPD- caudal peduncle depth; CPW - caudal peduncle width; PVD - distance between pectoral and pelvic fins; VAD - distance between pelvic and anal fins; AnAD - distance between anus and anal fin; HDN- head depth measured at the nape region; HDE - head depth measured at the eye region; HMW - maximum head width; SNL - snout length; ED - eye diameter; IOW- interorbital width; POL- postorbital length; MW - mouth width; MBL - maxillary barbel length; IRBL - inner rostral barbel length; ORBL - outer rostral barbel length; D - dorsal fin; A - anal fin; P - pectoral fin; V- pelvic fin; min - minimum value; max - maximum value; M - arithmetic mean; m- standard error of the mean; SD - standard deviation;

CV- coefficient of variation; t - Student's t-distribution; Tf - calculated t-value; Tst - tabulated t-value.

Results and Discussion

A mathematical and statistical analysis of the morphometric characters of *I. kuschakewitschi* (Herzenstein, 1890) specimens collected from the Chortoqsoy and Oltiariqsoy rivers was carried out. In total, 15 individuals from the Chortoqsoy population and 19 individuals from the Oltiariqsoy population were examined. The obtained results revealed morphometric differences between the two populations in body length, body proportions, fin positioning, caudal peduncle parameters, and head morphology. Statistical analyses of the morphometric characters of *I. kuschakewitschi* populations inhabiting the Chortoqsoy and Oltiariqsoy rivers are presented in Table 1.

	Chortoqsoy population (n=15)					Oltiariqsoy population (n=19)					t-test
	Min	Max	M±m	CD	V	Min	Max	M±m	CD	V	
TL	42,07	69,27	55,33	-	-	42,91	108,44	66,66	-	-	-
SL	35,05	58,28	46,01	-	-	35,07	92,41	55,30	-	-	-
SL%											
HL	21,51	25,32	22,92±0,41	0,92	4,0	20,17	24,07	21,91±1,37	1,19	5,4	tφ>tst
BDD	12,45	15,32	13,81±0,32	0,71	5,2	3,08	17,62	14,73±0,42	3,06	20,8	Tφ<tst
BWD	11,02	13,59	12,43±0,33	0,74	6,0	12,13	15,21	13,34±0,81	0,94	7,1	Tφ<tst
PRL	50,58	56,25	53,61±0,66	1,47	2,8	49,78	52,91	51,64±0,68	0,95	1,8	tφ>tst
PDL	33,52	42,18	36,04±0,85	1,90	5,3	31,79	38,79	34,94±0,59	1,81	5,2	tφ>tst
PVL	49,10	54,03	51,86±0,71	1,59	3,1	48,83	52,90	51,49±0,36	1,15	2,2	Tφ<tst
PAL	71,18	78,01	74,20±0,81	1,82	2,4	68,21	74,88	72,14±0,4	1,52	2,1	tφ>tst
PAnL	66,22	72,34	69,28±0,69	1,53	2,2	65,73	73,64	69,54±0,28	1,53	2,2	Tφ<tst
DFL	13,05	17,65	15,54±0,50	1,13	7,2	12,46	17,52	14,27±0,34	1,31	9,2	tφ>tst
DFBL	8,54	12,78	11,31±0,59	1,31	11,6	10,09	12,68	11,42±0,28	0,81	7,1	Tφ<tst
AFL	11,19	15,40	14,00±0,41	0,92	6,6	10,32	13,56	12,22±0,51	0,89	7,3	tφ>tst
AFBL	6,16	8,05	7,31±0,20	0,44	6,1	6,41	8,56	7,28±0,20	0,63	8,7	Tφ<tst
PFL	14,12	17,97	15,66±0,49	1,09	6,9	12,98	16,47	14,76±0,23	0,76	5,1	tφ>tst
VFL	12,72	16,13	14,27±0,43	0,96	6,8	11,84	14,14	13,05±0,44	0,63	4,9	tφ>tst
CFL	19,08	22,85	20,77±0,53	1,18	5,7	18,48	23,98	21,27±0,55	1,45	6,8	Tφ<tst
CPL	10,62	14,57	12,10±0,45	1,00	8,3	16,53	20,11	18,18±0,21	1,14	6,3	tφ>tst
CPD	5,98	7,16	6,62±0,17	0,37	5,7	5,79	7,64	6,60±2,42	0,46	6,9	Tφ<tst
CPW	2,13	2,59	2,37±0,06	0,13	5,6	3,01	5,29	3,81±1,40	0,52	13,6	Tφ<tst
PVD	24,83	32,79	29,86±0,85	1,91	6,4	28,48	32,33	30,88±3,31	0,99	3,2	Tφ<tst
PAD	19,65	24,22	21,96±0,56	1,25	5,7	18,30	23,55	20,20±1,70	1,22	6,0	tφ>tst
VAD	3,34	5,04	4,40±0,25	0,56	12,7	0,97	2,60	1,93±0,95	0,47	24,2	tφ>tst

CPL/C PD	1,59	2,12	1,83±1,13	0,14	7,6	2,20	3,36	2,77±0,15	0,33	11,8	Tφ<tst
HL%											
HD	41,31	52,26	46,64±1,07	2,52	5,4	31,75	57,11	50,92±0,94	5,42	10,6	Tφ<tst
HD(E)	36,02	46,89	40,79±1,41	2,40	5,9	40,77	52,59	46,55±1,60	3,13	6,7	Tφ<tst
HW(E)	49,73	61,97	56,87±1,39	3,16	5,6	37,99	72,20	62,40±1,58	7,41	11,9	Tφ<tst
SNL	36,78	47,74	41,77±0,51	3,11	7,4	28,87	46,93	40,90±1,09	3,79	9,3	Tφ<tst
ED	13,56	17,05	15,28±0,91	1,13	7,4	10,61	18,56	14,42±0,98	2,11	14,7	Tφ<tst
IOW	23,83	30,41	28,05±1,54	2,04	7,3	8,41	16,13	13,12±0,15	2,09	16,0	tφ>tst
POL	36,02	49,36	41,93±1,81	3,44	8,2	29,27	44,83	36,65±1,60	3,58	9,8	tφ>tst
MW	19,92	33,62	26,62±1,36	4,05	15,2	19,72	32,72	23,55±0,94	3,53	15,0	tφ>tst
MBL	17,16	28,57	24,22±0,92	3,04	12,6	16,59	27,80	22,47±0,95	2,43	10,8	Tφ<tst
IRBL	12,99	19,75	16,24±1,06	2,05	12,6	12,30	21,11	17,26±1,70	2,19	12,7	Tφ<tst
ORBL	15,78	25,24	21,18±0,06	2,36	11,2	15,89	23,11	20,56±0,15	2,15	10,5	Tφ<tst

The total length of individuals from the Chortoqsoy population ranged from 42.07 to 69.27 mm, with a mean value of 55.33 mm. Standard length varied between 35.05 and 58.28 mm, averaging 46.01 mm. In the Oltiariqsoy population, total length ranged from 42.91 to 108.44 mm, with a mean of 66.66 ± 8.49 mm, whereas standard length varied between 35.07 and 92.41 mm, averaging 55.30 ± 7.33 mm. The broader range of total and standard lengths observed in the Oltiariqsoy population may be associated with the coexistence of individuals belonging to different age or growth stages, as well as with the relatively heterogeneous environmental conditions of the habitat. Body proportions calculated relative to standard length showed similar values for some characters and considerable differences for others between the two populations. In the Chortoqsoy population, head length constituted on average $22.92 \pm 0.41\%$ of standard length, whereas in the Oltiariqsoy population this value was $21.91 \pm 1.37\%$. The coefficient of variation for head length was 4.0% in Chortoqsoy and 5.4% in Oltiariqsoy, indicating that this character remains relatively stable in both populations. Body depth at the dorsal-fin origin ranged from 12.45 to 15.32% in the Chortoqsoy population, with an average of 13.81%, whereas in the Oltiariqsoy population it ranged from 3.08 to 17.62%, averaging $14.73 \pm 0.42\%$. The coefficient of variation for this character was 5.2% in Chortoqsoy and 20.8% in Oltiariqsoy, indicating stronger intrapopulation variability in body depth within the Oltiariqsoy population. Such increased variability may be associated with differences in water flow velocity, microhabitat diversity, substrate composition, and feeding conditions. Body width also showed slightly higher values in the Oltiariqsoy population. The average BWD value was 12.43% in Chortoqsoy and $13.34 \pm 0.81\%$ in Oltiariqsoy. In both populations, coefficients of variation for this character were low to moderate (6.0% and 7.1%, respectively), indicating relative stability of body width, although some interpopulation differences may reflect ecological adaptation. Predorsal and postdorsal lengths, which characterize body segmentation proportions, also exhibited diagnostic significance. In the Chortoqsoy population, predorsal length ranged from 50.58 to 56.25%, averaging 53.61%, whereas in the Oltiariqsoy population it ranged from 49.78 to

52.91%, with a mean of $51.64 \pm 0.68\%$. Postdorsal length averaged 36.04% in Chortoqsoy and $34.94 \pm 0.59\%$ in Oltiariqsoy. In both populations, predorsal length exceeded postdorsal length, indicating that the anterior part of the body is relatively elongated in *Iskandaria kuschakewitschi*. Moreover, the slightly larger predorsal and postdorsal values observed in the Chortoqsoy population may indicate a comparatively more elongated body segmentation pattern. Fin-related characters are important indicators of ecological adaptation in the two populations. In the Chortoqsoy population, mean dorsal-fin length was 15.54%, anal-fin length 14.00%, pectoral-fin length 15.66%, pelvic-fin length 14.27%, and caudal-fin length 20.77%. In the Oltiariqsoy population, the corresponding values were $14.27 \pm 0.34\%$, $12.22 \pm 0.51\%$, $14.76 \pm 0.23\%$, $13.05 \pm 0.44\%$, and $21.27 \pm 0.55\%$, respectively. These results indicate that dorsal, anal, pectoral, and pelvic fins are slightly longer in the Chortoqsoy population, whereas the caudal fin is relatively longer in the Oltiariqsoy population. Such differences may reflect adaptations associated with swimming performance, near-substrate locomotion, and resistance to water currents under different river conditions. Pronounced differences were observed in caudal peduncle characters. In the Chortoqsoy population, caudal peduncle length averaged 12.10%, whereas in the Oltiariqsoy population it reached $18.18 \pm 0.21\%$. Caudal peduncle depth showed similar values in both populations (6.62% and $6.60 \pm 2.42\%$, respectively). However, caudal peduncle width averaged 2.37% in Chortoqsoy and $3.81 \pm 1.40\%$ in Oltiariqsoy. Similarly, the CPL/CPD ratio was 1.83 in Chortoqsoy and 2.77 ± 0.15 in Oltiariqsoy. These findings suggest that individuals from the Oltiariqsoy population possess a relatively longer and more developed caudal peduncle, which may be associated with adaptation to water current dynamics and benthic lifestyle.

Distances between fins also reflected differences in body proportions between populations. The distance between pectoral and pelvic fins averaged 29.86% in Chortoqsoy and $30.88 \pm 3.31\%$ in Oltiariqsoy. The distance between pelvic and anal fins was 4.40% in Chortoqsoy and $1.93 \pm 0.95\%$ in Oltiariqsoy. Notably, the coefficient of variation for the VAD character reached 24.2% in the Oltiariqsoy population, indicating high intrapopulation variability, whereas in Chortoqsoy it was 12.7%, representing moderate variability. Head morphology also demonstrated substantial interpopulation differences. In the Chortoqsoy population, head depth averaged 46.64%, head depth at the eye region 40.79%, and maximum head width 56.87%. In the Oltiariqsoy population, these values were $50.92 \pm 0.94\%$, $46.55 \pm 1.60\%$, and $62.40 \pm 1.58\%$, respectively. These results indicate that individuals from the Oltiariqsoy population possess a relatively deeper and broader head morphology. Such variation may be associated with differences in feeding conditions, substrate type, and microhabitat structure. Snout length showed similar values between populations (41.77% in Chortoqsoy and $40.90 \pm 1.09\%$ in Oltiariqsoy). Eye diameter averaged 15.28% in Chortoqsoy and $14.42 \pm 0.98\%$ in Oltiariqsoy. However, a substantial difference was observed in interorbital width: 28.05% in Chortoqsoy versus $13.12 \pm 0.15\%$ in Oltiariqsoy. The higher value recorded in the Chortoqsoy population indicates morphological differentiation in the anterior head region and eye placement. Postorbital length was also greater in Chortoqsoy (41.93%) than in Oltiariqsoy ($36.65 \pm 1.60\%$). Mouth width and barbel lengths are important traits associated with feeding and substrate detection in benthic loaches. In the Chortoqsoy population, mouth width

averaged 26.62%, whereas in the Oltiariqsoy population it was $23.55 \pm 0.94\%$. Maxillary barbel length averaged 24.22% in Chortoqsoy and $22.47 \pm 0.95\%$ in Oltiariqsoy. Inner rostral barbel length averaged 16.24% and $17.26 \pm 1.70\%$, while outer rostral barbel length averaged 21.18% and $20.56 \pm 0.15\%$ in the Chortoqsoy and Oltiariqsoy populations, respectively. Although several values were larger in the Chortoqsoy population, inner rostral barbels were slightly longer in Oltiariqsoy individuals. These differences may be related to variation in food resources, substrate composition, and benthic microorganism communities between the two rivers. Overall, the coefficient of variation results demonstrated that the two populations differ in the degree of morphological stability. In the Chortoqsoy population, the coefficients of variation for most characters were low or moderate, indicating relatively stable morphological structure within this population. In contrast, the Oltiariqsoy population exhibited higher variability in characters such as TL, SL, BDD, VAD, IOW, and ED. This suggests that morphological variability is more strongly expressed in the Oltiariqsoy population, possibly due to greater ecological heterogeneity, variability of the hydrological regime, or anthropogenic influences. *Iskandaria kuschakewitschi*. In addition, morphometric characters of the two populations of *Iskandaria kuschakewitschi* were comparatively analyzed using Student's t-test at the 1% significance level (Table 1). The comparative analysis included 33 morphometric characters. The results revealed statistically significant differences ($t_f > t_{st}$) in 14 morphometric characters between the Chortoqsoy and Oltiariqsoy populations, whereas no significant differences ($t_f < t_{st}$) were detected in 19 characters. Significant differences were identified in HL, PRL, PDL, PAL, DFL, AFL, PFL, VFL, CPL, PAD, VAD, IOW, POL, and MW. In contrast, no significant differences were observed in BDD, BWD, PVL, PAnL, DFBL, AFBL, CFL, CPW, CPL/CPD, HD, HD(E), SNL, ED, MBL, IRBL, and ORBL. These findings indicate that the degree of differentiation between the two populations is not particularly high and suggest that the individuals developed under relatively similar environmental conditions. Thus, the Chortoqsoy and Oltiariqsoy populations differ in body size, body depth, fin lengths, caudal peduncle parameters, and head morphology. These differences may represent phenotypic responses formed during the adaptation of *I. kuschakewitschi* to different river environments. At the same time, the observed morphometric differences do not exclude the possibility of ecological isolation and potential genetic differentiation between the populations. However, because many characters remained relatively similar between the two populations, further molecular-genetic analyses are required to clarify the actual population-genetic status of the Chortoqsoy and Oltiariqsoy populations.

Conclusion

In the present study, a comparative analysis of the morphological characteristics and variability ranges of *I. kuschakewitschi* was conducted based on specimens collected from the Chortoqsoy and Oltiariqsoy rivers. Chortoqsoy represents a small mountain river located in the northern part of the Fergana Valley, whereas Oltiariqsoy belongs to the southern river system of the region. The hydrological and ecological conditions of these water bodies differ considerably, which may contribute to the relative isolation of populations and their

development under distinct ecological environments. The obtained results demonstrated the presence of morphological variability within the populations of this species. Therefore, regular investigation of morphometric variability among populations inhabiting different regions is important for assessing their population status, ecological adaptation, and degree of phenotypic differentiation. Furthermore, future analyses based on molecular-genetic markers would make it possible to determine whether the observed morphometric differences are solely the result of phenotypic plasticity or are also supported by genetic differentiation. *Iskandaria kuschakewitschi*. Thus, the present study provides an important preliminary scientific basis for comprehensive ecological, morphological, and taxonomic investigations of local populations of *I. kuschakewitschi* inhabiting the water bodies of the Fergana Valley.

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