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Morphometrics and polymorphism in populations of *Rana macrocnemis* Boulenger, 1885 in the Azerbaijan part of the Major Caucasus (Anura: Ranidae)

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Abstract

The polymorphism and morphometrics of the *Rana macrocnemis* Boulenger, 1885 taken from 2 populations distributed in the Azerbaijan part of the Major Caucasus were studied during in the breeding season (early May to late July) between 2010 and 2014. Thirty nine specimens (25 females, 12 males and 2 juveniles) of *R. macrocnemis* were captured, measured and released into their natural habitat. Statistical analyses showed considerable differences between two populations for measured characters. SVL, FL, TL, T1L and IMTL measurements in *R. macrocnemis* specimens picked from 2 populations in Major Caucasus showed that females in the Guba population are bigger than males in the same population and females in the Zagatala population.

Keywords: Morphology, polymorphism, *Rana macrocnemis*, Guba, Zagatala, Azerbaijan

1. Introduction

Among Western Palaearctic amphibians, the genus *Rana* is the most speciose. Most species are western members of the Palaearctic subgenus *Rana*, the so-called brown frogs. Also known as a “mountain frog”, its altitudinal distribution ranges from 1,000 to 2,300 m. In contrast to the semi-aquatic water frogs, brown frogs are largely terrestrial, with an aquatic larval stage [1]. Brown frogs of the complex *Rana macrocnemis* demonstrate various degrees of differentiation between the two widespread forms, *macrocnemis.str.* and *camerani*, in different parts of Anatolia, the Caucasus Isthmus and Northern Iran [2]. Different views on the taxonomic position of the mountain frogs living in Anatolia and the Caucasus were presented in the recent years. Tarknishvili *et al.* (1999) who morphologically and biometrically compared 14 mountain frog populations from Georgia has concluded that *R. macrocnemis* and *R. camerani* are subspecies of *R. macrocnemis*, viz., *R. m. macrocnemis* and *R. m. camerani* [3]. On the other hand, Picariello *et al.* (1999) studied the S1satellite DNA and morphology of mountain frogs and stressed that the three nominal mountain frog species, *R. macrocnemis*, *R. camerani* and *R. holtzi*, should be considered as a single species, which should be named *R. macrocnemis*, according to the priority rules of zoological nomenclature [4, 5, 6, 7, 8].

Within *R. macrocnemis*, three subspecies are distinguished on morphological grounds: *R. m. macrocnemis* Boulenger, 1885, *R. m. pseudodalmatina* Eiselt and Schmidtler, 1971, and *R. m. tavasensis* Baran and Atatür, 1986. *R. m. tavasensis* is known only from a single creek at the Akdag Mountain in southern Turkey at an elevation of 1,650 m a.s.l. and the distribution of *R. m. pseudodalmatina* is confined to the Hyrcanian corridor of Northern Iran and Southern Azerbaijan [9, 10].

The *Rana macrocnemis* Boulenger, 1885, is included in the “least concern” (LC) category in the IUCN Red List, but it is reported that the populations are tending to decrease [11]. *Rana macrocnemis* is a very common species in the Azerbaijan [12].

Information about the morphology, distribution, reproduction, population structure and dynamics of the Caucasian populations of the species was compiled by Tarknishvili and Gokhelashvili [3, 13].

In this study, it was aimed to determine the morphology and polymorphism of the Guba and Zagatala population of *R. macrocnemis* [Figure 1].

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Fig 1: Two types of dorsal pattern as observed in the specimens of *Rana macrocnemis* Boulenger, 1885 from the Major Caucasus Mountains (Major Caucasus Natural Region, Azerbaijan): from Zagatala district (left); from Guba district (right).

2. Materials and Methods

The study was conducted at two stations in the mountains of the Azerbaijani a part of Caucasus Major. Field surveys were conducted at afternoon and evening. Thirty-nine mountain frog specimens examined in this study were caught from the banks of small streams and temporary ponds located in the meadowland formed by spring water and waterfall in the Major Caucasus Mountains, Gebizdere of Zagatala district (41° 40' 43") at an altitude of 950 meters and Khinalig of Guba district (41° 19' 54") at an altitude of 2100 meters [Figure 2, 3]. We were collected 12 females and 2 juveniles from Balakan and 12 males and 13 females from Guba during 2010-2014 years. The adult specimens were distinguished by the presence/absence of a callus on the thumb.

Color photographs were made and the color-pattern characteristics of the specimens were noted when they were alive. SVL - Snout-Vent Length, FL - Femur Length, TL - Tibia Length, T1L, First toe Length, IMTL – Inner Metatarsal Tubercle Length were measured according to Bannikov et. al. (1977), using a caliper (accuracy 0.01 mm) and the ratios SVL/T, FL/TL, TL/IMTL and T1L/IMTL were computed [14]. The morphometric data of males and females were compared using the independent-sample t test. Sexual dimorphism was considered significant at $p < 0.05$.



Fig 2: Map showing the study area in Major Caucasus Mountains (part of Azerbaijan).



Fig 3: Habitat types of *Rana macrocnemis* Boulenger, 1885 in Gudyalchay riverside, at Khinalig (Guba, Azerbaijan).

3. Results and discussion

The granulated skin was observed in all 25 specimens collected in the territory of Guba district. These folds are well visible in 1 female and 3 male specimens. There was a swelling on the first digit of the foreleg in all of the male specimens. The ground color of head, dorsum and dorsal side of legs is greenish brown in all specimens. It should be noted that the ground color of the specimens at the time of their catches in nature was greenish brown. However in 15-20 minutes they adapted to the environment and the color became lighter. Four different patterns can be distinguished in terms of the shape and size of the dorsal spots. In collected 7 females and 4 males dorsal side of the body is distinguished by well seen many large and small spots. Two females and 4 males have many large and a few small spots on the dorsal side of the body, 3 females and 2 males are characterized by a few large and small spots. There are a few large and many small spots in 1 female and 3 males. Black spots are registered in the gular and neck sections on the ventral side in 2 females and 4 males. Only neck section is spotted in 4 females. In 7 females and 8 males the ventral side is spotless. Inguinal region is pink. From 14 specimens collected in Zagatala district 4 ones (12 adults and 2 juveniles) characterized by granulated skin. Two from 14 specimens bear a lot of large and small spots on dorsal side. The similar spots were few in 6 specimens. Two specimens have a few large spots and a lot of small ones on dorsal side. Another 3 ones have a lot of large and a few small spots. One specimen is spotless on dorsal side. The ventral side in 9 specimens is spot less, the neck and abdomen in 5 specimens bear spots [Table 1]. Inguinal region is orange-pink. As Baran (2007) and Tarknishvili (1999) also noted the sexual dimorphism by color and spot forms was not found [15, 16, 17, 18].

Table 1: Dorsal, ventral pattern and skin characteristics of *Rana macrocnemis* Boulenger, 1885 from the Khinalig (Guba, Azerbaijan) compared with information from the Gebizdere (Zagatala, Azerbaijan): M- a lot of large spots on dorsal side, hM- a few number of small spots on dorsal side, P-a lot of small spots on dorsal side, hP- a few number of small spots on dorsal side, R-existence of granules on dorsal side, Nc- neck with spots, Ac-neck without spots, Nv- ventral side with spots, Av-ventral side without spots, *-pattern less.

Character	Guba district		Zagatala district	
	Ind.	%	Ind.	%
M	15	62,5	5	38,46
hM	9	37,5	8*	61,53
P	15	60	8	57,14
hP	10	40	6	42,85
R	4	16	-	0,00
Nc	10	40	5	35,71
Ac	15	60	9	64,28
Nv	19	76	5	35,71
Av	6	24	9	64,28

There is no significant difference in sexual dimorphism in any morphometric parameters of the Guba district specimens according to independent-sample *t* test (the measurement values of males and females were combined). All body measurements, ratios and descriptive statistical data are given for both sexes collectively in Table 2. Snout-vent length ranged from 48,40 to 73,20 mm ($x = 63,44$ mm) in male specimens, 46,60 to 84,40 mm ($x = 68,34$ mm) in female specimens, femur length from 24,70 to 37,80 mm ($x = 32,73$ mm) in male, from 26,50 to 40,00 mm ($x = 34,84$ mm) in female specimens, tibia length from 26,90 to 39,10 mm ($x = 34,67$ mm) in male, from 27,90 to 41,40 mm ($x = 35,46$ mm) in female specimens, first toe length from 4,80 to 8,30 mm ($x = 6,33$ mm) in male, and from 2,60 to 8,90 mm ($x = 6,52$ mm) in female specimens, inner of metatarsal tubercle from 2,10 to 3,60 mm ($x = 2,79$ mm) in male, and from 1,90 to 4,70 mm ($x = 3,22$ mm) in female specimens. Some body measurements for specimens collected in Zagatala region are presented in Table 2. Snout-vent length ranged from 41,10 to 78,10 mm ($x = 60,10$ mm), femur length from 25,00 to 37,60 mm ($x = 31,68$ mm), tibia length from 26,90 to 38,40 mm ($x = 33,56$ mm), first toe length from 4,30 to 7,60 mm ($x = 6,01$ mm), inner of metatarsal tubercle from 2,00 to 3,90 mm ($x = 2,91$ mm) in female specimens. All specimens collected in Zagatala region were females, so the sexual dimorphism was not studied.

In conclusion, SVL, FL, TL, TIL and IMTL measurements in *R. macrocnemis* specimens picked from 2 populations in Major Caucasus showed that females in the Guba population are bigger than males in the same population and females in the Zagatala population (Table 2, 3).

Table 2. Morphometric measurements (mm) and ratios of the adult (12 male and 13 female) samples of *Rana macrocnemis* Boulenger, 1885 from the Khinalig (Guba, Azerbaijan) including descriptive statistics. n – sample size, M – arithmetic mean, SD – standard deviation, SE – standard error of the mean, SVL – snout-vent length, FL – femur length, TL – tibia length, TIL, first toe length, IMTL – inner metatarsal tubercle length.

Character, ratio	M		Range		SD		SE	
	13♀	12♂	13♀	12♂	13♀	12♂	13♀	12♂
SVL	68,34	63,44	46,60-84,40	48,40-73,20	10,00	2,77	8,78	2,54
FL	34,84	32,73	26,50-40,00	24,70-37,80	3,96	1,10	4,76	1,37
TL	35,46	34,67	27,90-41,40	26,90-39,10	3,95	1,10	4,53	1,31
TIL	6,52	6,33	2,60-8,90	4,80-8,30	1,53	0,42	1,12	0,32
IMTL	3,22	2,79	1,90-4,70	2,10-3,60	0,81	0,23	0,42	0,12
SVL/T	1,99	1,83	1,67-2,77	1,71-1,92	0,31	0,09	0,07	0,02
FL/TL	0,95	0,94	0,05-1,33	0,90-0,98	0,30	0,08	0,03	0,01
TL/IMTL	11,44	12,57	5,94-17,74	9,69-15,63	3,26	0,90	1,87	0,54
TIL/IMTL	2,16	2,30	0,55-3,53	1,75-3,46	0,72	0,20	0,46	0,13

Table 3: Morphometric data (SVL, FL, TL, TIL, IMTL and resultant ratios; for explanation see legend to Table 1) of *Rana macrocnemis* Boulenger, 1885 from the Khinalig (Guba, Azerbaijan) compared with information from the Gebizdere (Zagatala, Azerbaijan). F+M – males and females mixed sample. M (SD) – mean value (standard deviation).

Character, Ratio	<i>R. macrocnemis</i> (Gebizdere, Zagatala district)	<i>R. macrocnemis</i> (Khinalig, Guba district)
	n=12 (F) M (SD)	n=25 (13F+12M) M (SD)
SVL	60,10(10,44)	65,99(9,57)
FL	31,68(3,45)	33,83(4,40)
TL	33,56(3,45)	35,08(4,17)
TIL	6,01(1,02)	6,43(1,32)
IMTL	2,91(0,56)	3,02(0,68)
SVL/T	1,78(0,14)	1,91(0,24)
FL/TL	0,94(0,04)	0,95(0,21)
TL/IMTL	11,87(2,35)	11,98(2,69)
TIL/IMTL	2,15(0,63)	2,23(0,60)

However females in the Zagatala population are larger than females in Guba population according to TL/IMTL (M=11,87) ration.

Specimens picked from Zagatala population were females only, so it is difficult to say which of specimens of two populations is larger. Judging by the color and spot, the back in the individuals of the Guba population is more spotted. Specimens from Zagatala populations are less spotted and even one specimen is spotless. The skin in 4 specimens (3 adults and 1 juvenile) from Zagatala population and in 4

specimens (1 female, 3 males) from Guba population is granulated. This indicates that the skin in females is more granular than in males. Furthermore, the ventral side of specimens from both populations was mainly spotless.

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