

## *Rana macrocnemis* BOULENGER, 1885, from the Emir Mountains in western inner Anatolia (Turkey)

(Anura: Ranidae)

*Rana macrocnemis* BOULENGER, 1885 von den Emir Bergen  
im westlichen Inneranatolien (Türkei)

(Anura: Ranidae)

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### KURZFASSUNG

Die vorliegende Studie untersucht Braunfrösche aus den Emir Bergen (Provinz Afyonkarahisar) im Inneren von Westanatolien (Türkei) hinsichtlich morphologischer (Morphometrie, Färbung und Zeichnung) und serologischer Merkmale, letztere anhand elektrophoretischer Auftrennung der Blut-Plasmaproteine mittels Polyacrylamid-Gel-Scheibenelektrophorese.

Danach entsprechen die Individuen der Emir Berge morphologisch und in der elektrophoretischen Auftrennung ihrer Plasmaproteine *Rana macrocnemis* BOULENGER, 1885.

### ABSTRACT

In the present study, mountain frogs from the Emir Mountains (Province of Afyonkarahisar) in western inner Anatolia (Turkey) were examined both morphologically i.e., morphometry and color-pattern, and serologically by analyzing the blood-plasma proteins using polyacrylamide gel disc electrophoresis.

Based on the analyses, the mountain frog population of the Emir Mountains represents *Rana macrocnemis* BOULENGER, 1885.

### KEY WORDS

Amphibia: Anura: Ranidae; *Rana macrocnemis*, taxonomy, morphology, polyacrylamide gel disc electrophoresis, densitometry; Sarıkız, Emir Mountains, Province of Afyonkarahisar, Anatolia, Turkey

### INTRODUCTION

The authors put a brief history of Anatolian mountain frog systematics at the beginning to explain the reasons for their studies. Until recently, various researchers studied the systematic position of the mountain frogs *Rana macrocnemis* BOULENGER, 1885, *Rana holtzi* WERNER, 1898 and *Rana camerani* BOULENGER, 1886 living in Anatolia and the Caucasus Mountains addressing morphological, biometrical, biochemical and molecular aspects (BOULENGER 1898; WERNER 1902; LANTZ & CYRÉN 1913; BODENHEIMER 1944; MERTENS 1952; BAŞOĞLU & HELLMICH 1958, 1970; EISELT 1965; BARAN 1969; BARAN & ATATÜR 1986; ARIKAN et al. 2001; VEITH et al. 2003; ÇEVİK et al. 2006) and various views had been set forth. Initially, *R. holtzi* and *R. camerani* were considered identical by BOULENGER (1898) and WERNER (1902), as were *R. mac-*

*rocnemis* and *R. camerani* by LANTZ & CYRÉN (1913) and BODENHEIMER (1944), whereas, others (e.g., DELWIG 1928) treated the latter two taxa as geographical races of a single species. Subsequent studies accepted that three different species (*R. macrocnemis*, *R. camerani* and *R. holtzi*) lived in Anatolia (MERTENS 1952; BAŞOĞLU & HELLMICH 1958, 1970; EISELT 1965; BARAN 1969; BARAN & ATATÜR 1986). Accordingly, in a detailed study by BARAN (1969), the populations of Karagöl-Çinigöl (Bolkar Mountains, Taurus Massif) were included in *R. holtzi*, the populations in the Uludağ Mountain and some mountains in western Anatolia in *R. macrocnemis*, and the population of Mt. Erciyes in central Anatolia and of Mt. Nemrut and the Province of Ardahan in eastern Anatolia in *R. camerani*. In addition, the populations of Mt. Abant, Mt.

Murat and Mt. Akdağ were discussed in view of their differences and similarities to the three species concerned, and attention was drawn to their disputable taxonomic positions. The morphology, geographical distribution and taxonomic position of Anatolian mountain frogs other than *R. holtzi* were evaluated again (BARAN & ATATÜR 1986) by including a large number of specimens collected from 30 new localities and the specimens obtained from previously known localities. As a consequence, these authors put the populations of Mt. Abant, Mt. Murat and some mountains in western and eastern Anatolia to *R. macrocnemis*, described a new subspecies (*R. macrocnemis tavasensis*) from Mt. Akdağ (Province of Denizli, western Anatolia) and included some populations in central, eastern and northern Anatolia in *R. camerani*.

Different views on the taxonomic position of the mountain frogs living in Anatolia and the Caucasus were presented in the recent years. TARKHNISHVILI et al. (1999) who morphologically and biometrically compared 14 mountain frog populations from Georgia found that *R. macrocnemis* and *R. camerani* represented subspecies of *R. macrocnemis*, viz., *R. m. macrocnemis* and *R. m. camerani* (TARKHNISHVILI & GOKHELASHVILI 1999). On the other hand, PICARIELLO et al. (1999) studied the S1 satellite 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 a single species, which should bear the name *R. macrocnemis*, according to the priority regulations of zoological nomenclature. ARIKAN et al. (2001) could not detect any difference at species level between the populations of the high plateaus of Çamlıyayla (Mersin) and Körmenlik

(Aladağlar) in the central Taurus Mountains, identified as *R. macrocnemis* and the population of Karagöl-Çinçinöl as *Rana holtzi* by BARAN (1969). VEITH et al. (2003a, 2003b) suggested that *R. macrocnemis macrocnemis*, *R. camerani* and *R. holtzi* are conspecific, and that other populations of the collective species "*Rana macrocnemis*" (now *Rana tavasensis* BARAN & ATATÜR, 1986 and *Rana pseudodalmatina* EISELT & SCHMIDTLER, 1971) represent species of their own outside of this clade. A recent morphological, serological and morphometrical study on Anatolian mountain frogs by ÇEVİK et al. (2006), established the presence of only two different species (*R. macrocnemis* and *R. holtzi*) living in Anatolia and considered *R. camerani* reported from Mt. Erciyes a synonym of *R. macrocnemis*, a view that was adopted by BARAN et al. (2007).

Detailed information on the taxonomic positions of the mountain frog specimens collected from new localities in Anatolia was recently provided by BARAN et al. (2007). This study revealed that *R. holtzi*, known only from two localities in the Bolkar Mountains, also lived on Mt. Eğrigöl (Bolkar Mountains, Çamlıyayla, Mersin), thereby showing a wider distribution in this region than previously known. Furthermore, new localities of *R. macrocnemis* were identified from the Provinces of Konya (Ereğli, Seviçova) and Elazığ (Maden, Örtülü Village), where Örtülü Village marks the southern border of the taxon's distribution-range.

In the present study, mountain frog specimens from the Emir Mountains, Province of Afyonkarahisar, in western inner Anatolia were morphologically and serologically examined and compared with information from the literature.

## MATERIALS AND METHODS

Twenty-four mountain frog specimens examined in this study were caught in the afternoon from the banks of small streams and temporary ponds located in the meadowland formed by spring water and melting snow in the Emir Mountains at Sarıkız (38°48'N, 31°10'E), Province of Afyonkara-

hisar, at an altitude of 2,010 meters (Figs. 1, 2). For information on plant cover and geology of the Emir Mountains see KURT (2002). The materials presently kept in the Department of Zoology of the Celal Bayar University (CBU, Manisa) under the collection numbers CBU 6/2011, 1-7 (males), 8-



Fig. 1: Habitat of *Rana macrocnemis* BOULENGER, 1885 in the Emir Mountains (Province of Afyonkarahisar, western Anatolia, Turkey).

Abb. 1: Lebensraum von *Rana macrocnemis* BOULENGER, 1885 im Emir Gebirge (Provinz Afyonkarahisar, Westanatolien, Türkei).

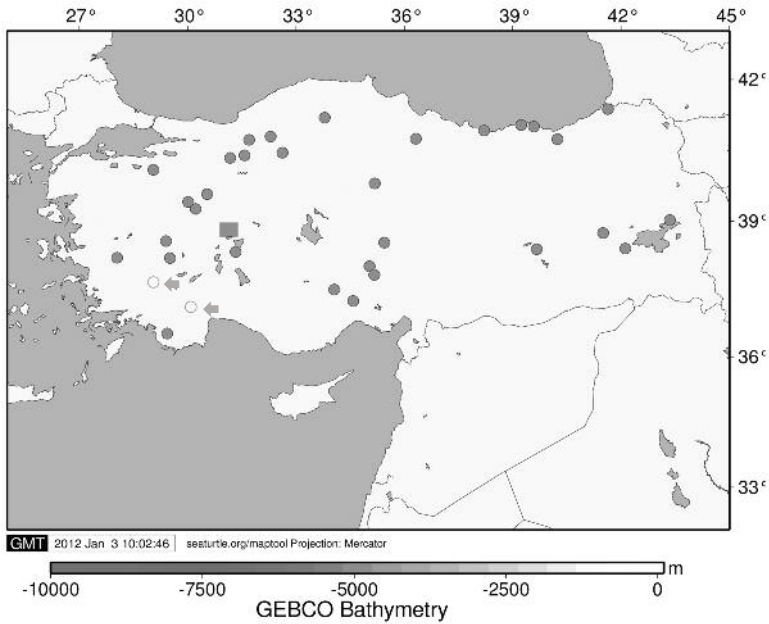


Fig. 2: Records of “*Rana macrocnemis*” in Turkey. ● - *Rana macrocnemis* BOULENGER, 1885; ○ ◄ - *Rana tavasensis* BARAN & ATATÜR, 1986; ■ - the new locality of *R. macrocnemis* in the Emir Mountains (Province of Afyonkarahisar, western Anatolia, Turkey). Data according to BARAN & ATATÜR (1986).

Abb 2: Fundorte von “*Rana macrocnemis*” in der Türkei. ● - *Rana macrocnemis* BOULENGER, 1885; ○ ◄ - *Rana tavasensis* BARAN & ATATÜR, 1986; ■ - der neue Fundort von *R. macrocnemis* im Emir Gebirge (Provinz Afyonkarahisar, Westanatolien, Türkei). Daten aus BARAN & ATATÜR (1986).

14 (females), and 15-24 (juveniles) were collected on June 10, 2011, by M. AFSAR and B. AFSAR, and ultimately will be incorporated into the collection of the Zoology Museum of the Ege University, İzmir (ZDEU). The adult specimens were sexed 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. Snout-vent length (SVL), head length (HL), head width (HW), internarial distance (ID), femur length (FL), tibia length (TL) and metatarsal tubercle length (CAL) were measured according to TERENT'EV & CHERNOV (1965) and BARAN (1969), using a caliper (accuracy 0.01 mm) and the ratios HL/SVL, HW/SVL, ID/SVL, IO/SVL, FL/SVL, TL/SVL and CAL/SVL were computed. The morphometric data of males and females were compared using the independent-sample t test. Sexual dimorphism was considered significant at  $p < 0.05$ .

Blood samples were taken from six adult specimens (three males and three females). Using a hematocrit capillary tube, blood was obtained by puncture of the cardiac ventricle from animals that had been anesthetized with ether. The blood was centrifuged at 600 g for five minutes and the plasma was stored at  $-20^{\circ}\text{C}$ . Electropho-

retic protein separation was carried out at a laboratory temperature of  $20\text{-}25^{\circ}\text{C}$  by processing  $5\ \mu\text{l}$  of the plasma sample in a Canalco 1200 electrophoresis bath with a Gelman Deluxe power supply. Glass gel columns ( $4.7\ \text{mm} \times 75\ \text{mm}$ ) were utilized. Amperage per column was adjusted to 0.85 mA initially and gradually increased to 1.75 mA (36 mA, 370 volts for 12 gel columns) until the tracking dye (bromphenol blue) moved about 53 mm from the start. The polyacrylamide disc electrophoretic analysis of the blood-plasma proteins was made according to ÖZETİ & ATATÜR (1979), who applied a slightly modified method of DAVIS (1964). Accordingly, a buffer system containing the stacking gel (2.5 %, pH 6.7) at the top and the mixture of the separation gel (7.5 %, pH 9.0) and tris-glycine (pH 8.3) at the bottom was used. The separation gels were stained with 0.5 % Amido Black (Naphthol Blue Black 10-B) and then the extra stain was passively discharged in 7 % acetic acid baths. The images of the stained gels were taken with a Nikon Coolpix P1 digital camera; the qualitative evaluation of the gels was made directly from the electropherograms. The densitometric curves of the separated proteins were obtained at 500 nm by means of a Gelman ACD-15 model 39430 densitometer.

## RESULTS

### Morphological analysis

The skin is granulated and the dorso-lateral fold present in all 24 specimens. These folds are well visible in 4 male and 3 female specimens and less evident in 3 male and 4 female, and all 10 juvenile specimens. When the extended hind leg is bent forward, in the adult specimens, the tibiotarsal joint projects the tip of the snout in 5 male and 5 female specimens and reaches the tip of the snout in 2 male and 2 female specimens. There is 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 7 male, 4 female and 10 juvenile specimens, while it is pale pinkish brown in 3 female specimens. There are green spots on the

dorsal ground in juveniles and darker spots in different shades of black and green in adults. As the ground color is dark, these spots are not distinct in 3 male and 1 female specimens. However, they can easily be distinguished in 4 male, 6 female specimens and all 10 juveniles. Two different pattern types can be distinguished in terms of the shape and size of the dorsal spots. The pattern types on the dorsal side of the specimens are represented in Fig. 3. The spots are small and symmetrical in 2 males and 3 females but large and asymmetrical in 5 male and 4 female specimens. A pale vertebral band is present in 3 male specimens and absent in 4 males, 7 females and the 10 juveniles. While there is a continuous V-shaped spot on the back immediately behind the head in 2 male and 4 female specimens,

Table 1: Morphometric measurements (mm) and ratios of the adult (seven male, and seven female) sample of *Rana macrocnemis* BOULENGER, 1885 from the Emir Mountains (Province of Afyonkarahisar, western Anatolia, Turkey) including descriptive statistics. *n* – sample size, *M* – arithmetic mean, *SD* – standard deviation, *SE* – standard error of the mean, *SVL* – Snout-Vent Length, *TL* – Tibia Length, *HL* – Head Length, *HW* – Head Width, *ID* – Internarial distance, *CAL* – Metatarsal Tubercle Length, *FL* – Femur Length..

Tab 1: Morphometrische Meßdaten (mm) und Quotienten der adulten (7 Männchen, 7 Weibchen) Stichprobe von *Rana macrocnemis* BOULENGER, 1885 aus den Emir Bergen (Provinz Afyonkarahisar, Westanatolien, Türkei) mit beschreibenden Statistiken. *n* – Stichprobenumfang, *M* – arithmetisches Mittel, *SD* – Standardabweichung *SE* – Standardfehler des Mittelwertes, *SVL* – Kopf-Rumpflänge, *TL* – Tibialänge, *HL* – Kopflänge, *HW* – Kopfbreite, *ID* – Nasenlochabstand, *CAL* – Länge des Metatarsaltuberkels, *FL* – Femurlänge.

Character, Ratio / Merkmal, Quotient	<i>n</i>	<i>M</i>	Range / Spannweite	<i>SD</i>	<i>SE</i>
<i>SVL</i>	14	63.42	48.35-77.01	10.16	2.71
<i>HL</i>	14	19.66	15.45-25.32	3.42	0.91
<i>HW</i>	14	20.86	15.9-25.7	3.56	0.95
<i>ID</i>	14	4.76	3.78-5.78	0.62	0.16
<i>FL</i>	14	33.01	23.1-40.2	6.26	1.67
<i>TL</i>	14	37.00	28.21-44.18	6.10	1.63
<i>CAL</i>	14	3.15	2.15-4.10	0.61	0.16
<i>HL/SVL</i>	14	0.30	0.29-0.33	0.01	0.003
<i>HW/SVL</i>	14	0.32	0.31-0.35	0.01	0.002
<i>FL/SVL</i>	14	0.51	0.47-0.56	0.02	0.006
<i>TL/SVL</i>	14	0.58	0.56-0.62	0.01	0.005
<i>CAL/SVL</i>	14	0.04	0.04-0.05	0.003	0.001
<i>(TL/SVL) x 100</i>	14	58.33	56-62	1.97	0.52

Table 2: Morphometric data (*SVL*, *HL*, *HW*, *FL*, *TL*, *CAL* and resultant ratios; for explanation see legend to Table 1), vertebral stripe, pattern and skin characteristics of *Rana macrocnemis* BOULENGER, 1885 from the Emir Mountains (Province of Afyonkarahisar, western Anatolia, Turkey) compared with information from the literature for *Rana holtzi* WERNER, 1898 and *R. macrocnemis*. F+M - Males and Females mixed sample. *M* (*SD*) - mean value (Standard Deviation).

Tab. 2: Meßdaten (*SVL*, *HL*, *HW*, *FL*, *TL*, *CAL* und Quotienten, Erklärung siehe Legende zu Tab. 1), Vertebralstreifen sowie Zeichnung und Beschaffenheit der Haut von *Rana macrocnemis* BOULENGER, 1885 aus den Emir Bergen (Provinz Afyonkarahisar, Westanatolien, Türkei) im Vergleich mit Literaturabgaben über *Rana holtzi* WERNER, 1898 und *R. macrocnemis*. F+M - Männchen und Weibchen gemischte Stichprobe. *M* (*SD*) - Mittelwert (Standardabweichung).

	<i>R. macrocnemis</i> (Emir Mountains, Afyon) Present study <i>n</i> = 14 (F+M)	<i>R. holtzi</i> (Eğrigöl, Bolkar Mountains) BARAN et al. (2007) <i>n</i> = 25 (F+M)	<i>R. macrocnemis</i> (Seviçova, Konya) BARAN et al. (2007) <i>n</i> = 38 (F+M)	<i>R. holtzi</i> (Bolkar Mountains) ÇEVİK et al. (2006) <i>n</i> = 48 (F+M)	<i>R. macrocnemis</i> (Uludağ, Bursa) ÇEVİK et al. (2006) <i>n</i> = 36 (F+M)
Character, Ratio	<i>M</i> ( <i>SD</i> )	<i>M</i> ( <i>SD</i> )	<i>M</i> ( <i>SD</i> )	<i>M</i> ( <i>SD</i> )	<i>M</i> ( <i>SD</i> )
<i>SVL</i>	63.42 (10.16)	45.20 (5.37)	49.00 (4.94)	44.50 (5.25)	55.60 (4.42)
<i>HL</i>	19.66 (3.42)	16.45 (2.14)	18.64 (2.04)	14.50 (4.39)	17.63 (4.26)
<i>HW</i>	20.86 (3.56)	17.64 (2.01)	18.24 (2.18)	17.70 (2.36)	20.01 (1.60)
<i>FL</i>	33.01 (6.26)	23.44 (2.80)	25.45 (2.97)	22.90 (2.94)	29.80 (2.52)
<i>TL</i>	37.00 (6.10)	24.04 (2.55)	26.58 (2.91)	24.70 (3.22)	32.30 (2.64)
<i>CAL</i>	3.15 (0.61)	2.46 (0.36)	2.73 (0.50)	2.60 (0.36)	3.50 (0.36)
<i>HL/SVL</i>	0.31 (0.01)	---	---	0.33 (0.016)	0.32 (0.012)
<i>HW/SVL</i>	0.32 (0.01)	---	---	0.40 (0.018)	0.36 (0.016)
<i>SVL/FL</i>	1.93 (0.09)	1.94 (0.11)	1.93 (0.08)	---	---
<i>SVL/TL</i>	1.72(0.01)	1.88(0.01)	1.85(0.02)	---	---
<i>HL/HW</i>	0.99 (0.05)	0.93 (0.06)	1.02 (0.05)	---	---
<i>TL/FL</i>	1.13 (0.04)	1.03 (0.06)	1.05 (0.06)	---	---
<i>SVL/(FL+TL)</i>	0.91 (0.03)	0.95 (0.03)	0.94 (0.04)	---	---
Vertebral stripe	present in 3 (21 %)	absent in all	present in 13 (32 %)	absent in all	present in 2 (6 %)
Skin	granulated	smooth	granulated	smooth	granulated
Dorsal maculations	without lighter colored rims	encircled by lighter colored rims	without lighter colored rims	encircled by lighter colored rims	without lighter colored rims

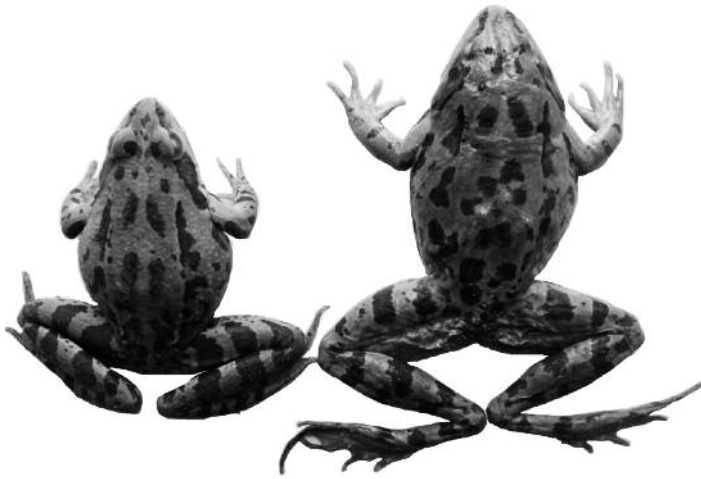


Fig. 3: Two types of dorsal pattern as observed in the specimens of *Rana macrocnemis* BOULENGER, 1885 from the Emir Mountains (Province of Afyonkarahisar, western Anatolia, Turkey).

Abb. 3: Die beiden Typen des dorsalen Zeichnungsmusters, die bei *Rana macrocnemis* BOULENGER, 1885 aus dem Emir Gebirge (Provinz Afyonkarahisar, Westanatolien, Türkei) beobachtet wurden.

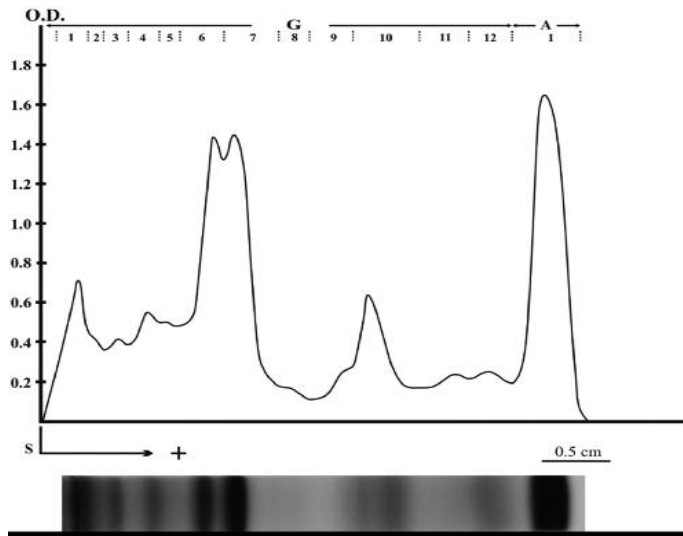


Fig. 4: The gel photograph showing the electrophoretic analysis of the blood-proteins obtained from a male *Rana macrocnemis* BOULENGER, 1885 (CBU 6/2011, No 3) representing the population of the Emir Mountains (Province of Afyonkarahisar, western Anatolia, Turkey) with the densitometric curve above. O.D. - Optical density, S - Start, A<sub>(1)</sub> - Albumin, G<sub>(1-12)</sub> - Globulins.

Abb. 4: Das Elektrophoresegel mit den aufgetrennten Proteinen des Bluts einer männlichen *Rana macrocnemis* BOULENGER, 1885 (CBU 6/2011, No 3) als Vertreterin der Population des Emir Gebirges (Provinz Afyonkarahisar, Westanatolien, Türkei); darüber die densitometrische Kurve. O.D. - optische Dichte, S - Start, A<sub>(1)</sub> - Albumin, G<sub>(1-12)</sub> - Globuline.

the sides of the V are separated by a vertebral band in 2 males. There are black spots in the gular and neck sections on the ventral side in 2 males and 3 females. The venter is spotless in all specimens. Small black dots are present on the dorsal side in 2 males and 3 females. The dorso-lateral folds have a slightly lighter color than the ground color in six adults (4 males and 2 females), whereas they are the same color as the ground in the other adult specimens and juveniles. The inguinal region is yellow. No obvious sexual dimorphism was detected in the color pattern as already noted by BARAN (1976).

Since the independent-sample t test did not detect significant sexual dimorphism in any morphometric parameters of the Emir Mountains specimens, 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 1. Snout-vent length ranged from 54.57 to 72.54 mm ( $\bar{x}$  = 63.50 mm) in male specimens, 48.35 to 77.01 mm ( $\bar{x}$  = 63.42 mm) in female specimens, head length from 16.47 to 23.31 mm ( $\bar{x}$  = 63.50 mm) in male, from 15.45 to 25.32 mm ( $\bar{x}$  = 19.66 mm) in female specimens, head width from 17.40 to 24.80 ( $\bar{x}$  = 20.83

mm) in male, from 15.90 to 25.70 mm ( $\bar{x}$  = 20.90 mm) in female specimens, internarial distance from 3.76 to 5.78 mm ( $\bar{x}$  = 4.77 mm) in male, from 3.96 to 5.68 mm ( $\bar{x}$  = 4.76 mm) in female specimens, femur length from 26.90 to 40.20 mm ( $\bar{x}$  = 33.30 mm) in male, from 23.10 to 39.80 mm ( $\bar{x}$  = 32.72 mm) in female specimens, tibia length from 31.72 to 44.18 mm ( $\bar{x}$  = 37.39 mm) in male, from 28.21 to 43.74 mm ( $\bar{x}$  = 36.61 mm) in female specimens, length of metatarsal tubercle from 2.63 to 3.71 mm ( $\bar{x}$  = 3.24 mm) in male, and from 2.15 to 4.10 mm ( $\bar{x}$  = 3.07 mm) in female specimens.

#### Electrophoretic analysis

Since neither sex-dependent nor individual variation was visually detected in the densitometric curves of the blood-plasma proteins, the corresponding gel photograph and densitometric curve from a male specimens' (CBU 6/2011, No 3) electrophorogram are considered representative of all the individuals analyzed (Fig. 4). Accordingly, the blood-plasma proteins comprised 13 fractions or fraction groups (one in the albumin region and 12 in the region of globulins).

## DISCUSSION

Most body measurement values of *R. holtzi* such as SVL, FL, TL, HL, HW and CAL are smaller than those of the other mountain frog species in Anatolia; there is only little divergence among the latter regarding their body ratios (comp. Table 2) and different from *R. macrocnemis* the skin of *R. holtzi* is smooth on the dorsal side, there is no vertebral band and a ring, lighter than the ground color, surrounds each dorsal spot (BARAN 1969; ÇEVİK et al. 2006; BARAN et al. 2007; BAŞKALE et al. 2012).

In the adult population of the Emir Mountains examined in this study, the measurement values of SVL, FL, TL, HL, HW and CAL exceeded those in *R. holtzi* and largely paralleled the values given for *R. macrocnemis* (Table 2). However, the ratio SVL/TL ( $M = 1.72$ ) was smaller than indicated for *R. holtzi* from Eğrigöl (1.88) and *R. macrocnemis* from Seviçova (1.85)

by BARAN et al. (2007) and for *R. holtzi* from the Bolkar Mountains by ARIKAN et al. (2001). Apart from this, the frogs correspond with *R. macrocnemis* by their granulated skin, the high rate of a vertebral band present, and the absence of a pale ring that surrounds the dorsal spots.

DESSAUER (1956), CHEN (1967), FERGUSON (1980) and ARIKAN et al. (1998, 1999), who studied the blood-plasma proteins of amphibians and reptiles with biochemical methods, found that factors such as age and physiological and environmental conditions were able to cause quantitative differences in the composition of blood-plasma proteins within a given species, whereas hereditary variation (i.e., different systematic status) was the only factor leading to qualitative differences. The population of the Emir Mountains strongly resembles *R. macrocnemis* when the blood-plasma proteins are

qualitatively compared by electrophoretic analysis. Nevertheless, the Emir Mountains frogs were distinguished by quantitative differences in the globulins, when compared with the Uludağ frogs; this is however not thought to be of systematic significance.

In conclusion, the mountain frog of the Emir Mountains was found to be in good agreement with *Rana macrocnemis* regarding its morphological characteristics and electrophoretic pattern of the blood-plasma proteins.

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