

### Hematology of *Triturus karelinii* (STRAUCH, 1870), from Gallipoli, Turkey

In Turkey, the Southern Crested Newt, *Triturus karelinii* (STRAUCH, 1870), is found in the north and west of the country, from sea level to 2,100 m a.s.l. (BAŞOĞLU et al. 1994; BARAN & ATATÜR 1998). Studies on its taxonomical status and distribution (BAŞOĞLU et al. 1994; BARAN & ATATÜR 1998), age structure (OLGUN et al. 2005), and ecology (OLGUN et al. 2005; MERMER et al. 2008) are available, whereas hematological information is confined to blood cell size and morphology (ATATÜR et al. 1998; ARIKAN & ÇICEK 2010). The purpose of this study was to provide data on cell counts, hemoglobin contents, hematocrit, mean cell volume, mean cell hemoglobin and mean cell hemoglobin concentration of *T. karelinii*.

Seven female specimens collected on April 26, 2010, from the vicinity of Gelibolu-Çanakkale (NW Turkey) at the altitude of sea level during the species' breeding season were anaesthetized with ether prior to cardiac ventricular puncture. Blood samples of the live specimens were obtained in the laboratory within one day after capture using heparinized hematocrit capillaries.

Erythrocytes (RBC, [per mm<sup>3</sup>]) and leucocytes (WBC, [per mm<sup>3</sup>]) were counted manually with a Neubauer hemocytometer. Erythrocytes were diluted in standard Hayem's solution, whereas for leucocytes the preparation method of JERRETT & MAYS (1973) was used, applying aliquots of 1/5000 neutral red solution and 12 % formaline in 0.07 % physiological saline. The latter method is a slight modification of

Blain's method (STURKIE 1954). Hematocrit value (HCT, [%]) was determined by the micro-hematocrit method. The tubes were spun in a micro-hematocrit centrifuge for 5 minutes at 13,000 rpm and the packed cell volume was calculated with a hematocrit reader. Hemoglobin concentration (Hb, [g/dl]) was measured colorimetrically with a Sahli hemoglobinometer. In this apparatus, 100 % corresponds to 14.5 g Hb / dl blood (TANYER 1985). The derived values 'mean cell volume of an erythrocyte' (MCV =  $HCT / 100 * RBC$ , [fl]), 'mean cell hemoglobin' (MCH =  $Hb / RBC$ , [pg]) and 'mean cell hemoglobin concentration' (MCHC =  $Hb * 100 / HCT$ , [%]) were calculated according to WINTROBE (1933). Descriptive statistics were computed using SPSS (ver. 10.00) statistical package. Sample size was seven in all counts and calculations.

Results are summarized in Table 1. In the blood samples, mean erythrocyte and leucocyte counts per mm<sup>3</sup> were 202,133±14,964.9 and 2,092±80.46, respectively. Hemoglobin concentration was 9.70±1.50 g/dl, mean hematocrit value was 24.50±3.50 %, mean cell volume of an erythrocyte was estimated to be 1,055.92±31.03 fl, mean cell hemoglobin was 417.19±7.24 pg, and mean cell hemoglobin concentration was calculated to be 39.52±0.48 %.

In poikilothermic vertebrates, the use of erythrocyte counts as an indicator of physiological conditions should be regarded reliable only if large samples are taken. This is necessary since the number of erythrocytes per mm<sup>3</sup> is regulated by feedback mechanisms which work at a high level of precision in animals that maintain constant body temperatures and have high rates of tissue metabolism, which does not apply to amphibians and reptiles (HUTCHINSON & SZARSKI 1965). The number of erythrocytes per mm<sup>3</sup> in amphibian circulating blood shows a wide individual variation and considerable interspecies differences (HUTCHINSON & SZARSKI 1965; SZARSKI & CZOPEK 1966) as well as a dependence on weight, age, sex, habitat conditions and season (ARVY 1947; GONIAKOWSKA 1973; ZHUKOVA & KUBANTSEV 1979; SINHA 1983; RUIZ et al. 1989; WOJTASZEK et al. 1997).

ATATÜR et al. (1998), who studied *Lissotriton vulgaris* (LINNAEUS, 1758),

Table 1: Hematological parameters of circulating blood of *Triturus karelinii* (STRAUCH, 1870), from Gallipoli, Turkey. RBC - red blood cell count, WBC - white blood cell count, Hb - hemoglobin, HCT - hematocrit, MCV - mean cell volume of erythrocyte, MCH - mean cell hemoglobin, MCHC - mean cell hemoglobin concentration, SD - standard deviation.

Species	RBC (per 1 mm <sup>3</sup> ) Mean ± SD (Min-Max)	WBC (per 1 mm <sup>3</sup> ) Mean ± SD (Min-Max)	Hb (g/dl) Mean ± SD (Min-Max)	HCT (%) Mean ± SD (Min-Max)	MCV (fl) Mean ± SD (Min-Max)	MCH (pg) Mean ± SD (Min-Max)	MCHC (%) Mean ± SD (Min-Max)
<i>Triturus karelinii</i> (present study)	202133 ± 14964.9 (166600-273200)	2092 ± 80.46 (1800-2266)	9.7 ± 1.5 (8.20-11.20)	24.50 ± 3.50 (21.00-28.00)	1055.92 ± 31.03 (1024.89-1086.96)	417.19 ± 7.24 (409.96-424.43)	39.52 ± 0.48 (39.05-40.00)
<i>Lyciasalamandra fazilae</i> (Tok et al. 2009)	106000 ± 22032.34 (73000-140000)	2140 ± 376.43 (1600-2870)	5.00 ± 0.20 (4.80-5.20)	36.66 ± 1.15 (36.0-38.0)	3281.2 ± 492.25 (2714.3-3600.0)	451.01 ± 94.84 (342.86-520.00)	13.65 ± 0.92 (12.63-14.44)
<i>Lissotriton vulgaris</i> (TOSUNOĞLU et al. 2008)	173670 ± 2780 (120000-220000)	1948 ± 278.39 (1360-2300)	-	-	-	-	-
<i>Ommatotriton ophryticus</i> (TOSUNOĞLU et al. 2011)	110000 ± 21.189 (80000-150000)	-	8.10 ± 1.03 (7.0-9.8)	30.37 ± 3.02 (26.0-36.0)	2852.98 ± 638.13 (2000-3750)	771.27 ± 128.14 (546.66-975.0)	27.52 ± 3.93 (22.22-33.84)
<i>Neurergus strauchii</i> (ARIKAN et al. 2003)	127000 ± 9000	2330 ± 1269	-	-	-	-	-

*Salamandra salamandra* (LINNAEUS, 1758), *Triturus karelinii*, *Ommatotriton ophryticus* (BERTHOLD, 1846), *Lyciasalamandra luschni* (STEINDACHNER, 1891) and *Mertensella caucasica* (WAGA, 1876), mentioned individual variation concerning erythrocyte size but not sexual dimorphism.

The erythrocyte count of *T. karelinii* was highest in comparison with other Turkish salamandrid species shown in Table 1. Leucocyte counts vary depending on species, season, sex, nutritional and physiological conditions such as health and breeding status) (ARIKAN 1989; ROUF 1969; WOJTA-SZEK & ADAMOWICZ 2003). In *T. karelinii*, the number of leucocytes varied within a wide range between 1,800 and 2,266 per mm<sup>3</sup>, which was similar to the other salamandrid species shown in Table 1.

Among the Turkish salamandrid comparative species, *T. karelinii* showed the highest values of RBC, Hb and MCHC and the lowest of HCT, MCV and MHC. Values of RBC, Hb and MCHC were lowest in *Lyciasalamandra fazilae* (Table 1). In conclusion, the hematological parameters of the Southern Crested Newt appear fairly different to other urodelan species mentioned in the literature.

REFERENCES: ARIKAN, H. (1989): *Rana ridibunda* (Anura, Ranidae) populasyonlarının kan hücrelerinin sayısı bakımından incelenmesi.- Turkish Journal of Zoology, Ankara; 13 (2): 54-59. ARIKAN, H. & OLGUN, K. & ILGAZ, Ç. & BARAN, İ. & KUMLU TAŞ, Y. (2003): Erythrocyte size and number in *Neurergus strauchii* (Urodela: Salamandridae).- Russian Journal of Herpetology, Moskva; 10 (2): 163-166. ARIKAN, H. & ÇİÇEK, K. (2010): Morphology of peripheral blood cells from various species of Turkish herpetofauna.- Acta Herpetologica, Firenze; 5 (2): 179-198. ARVY, L. (1947): Le dimorphisme sexuel sanguin chez *Rana temporaria* L. et *Bufo vulgaris* L.- Comptes Rendus des Séances de la Société de Biologie, Paris; 141: 457-459. ATATÜR, M. K. & ARIKAN, H. & MERMER, A. (1998): Erythrocyte sizes of some urodeles from Turkey.- Turkish Journal of Zoology, Ankara; 22: 89-91. BARAN, İ. & ATATÜR, M. K. (1998): Turkish herpetofauna (amphibians and reptiles) Ankara (Republic of Turkey Ministry of the Environment), pp. 214. BAŞOĞLU, M. & ÖZETİ, N. & YILMAZ İ. (1994): Türkiye amfibileri [The amphibians of Turkey].- Ege Üniversitesi Fen Fakültesi Kitaplar Serisi, Bornova-İzmir; 151: 1-221. GONIAKOWSKA, L. (1973): Metabolism, resistance to hypotonic solutions, and ultrastructure of erythrocytes of five amphibian species.- Acta Biologica Cracoviensia, Krakow; (Series Zoologia) 13: 225-236. HUTCHISON, H. V. & SZARSKI, H. (1965): Number of erythrocytes in some amphibians and reptiles.- Copeia, Washington; 1965 (3): 373-375. JERRETT, D. P. & MAYS,

C. E. (1973): Comparative hematology of the Hell-bender, *Cryptobranchus alleganiensis* in Missouri.- Copeia, Washington; 1973 (2): 331-337. MERMER, A. & AYAZ, D. & ÇİÇEK, K. (2008): Abundance of syntopic newts, *Triturus karelinii* (STRAUCH, 1870) and *Triturus vittatus* (GRAY, 1835), in Uludağ National Park (Bursa, Turkey).- Turkish Journal of Zoology, Ankara; 32: 59-64. ÖLGÜN, K. & ÜZÜM, N. & AVCI, A. & MIAUD, C. (2005): Age, size and growth of the southern crested newt *Triturus karelinii* (STRAUCH, 1870) in a population from Bozdag (Western Turkey).- Amphibia-Reptilia, Leiden; 26: 223-230. ROUF, M. A. (1969): Hematology of the leopard frog, *Rana pipiens*.- Copeia, Washington; 1969 (4): 682-687. RUIZ, G. & ROSENMAN, M. & VELOSO, M. (1989): Altitudinal distribution and blood values in the toad, *Bufo spinulosus* WIEGMANN.- Comparative Biochemistry and Physiology, New York; (A) 94: 643-646. SINHA, R. C. (1983): Haematological studies on the prewintering and wintering frog, *Rana esculenta*.- Comparative Biochemistry and Physiology, New York; (A) 74: 311-314. STURKIE, P. D. (1954): Avian physiology. 2nd edition. Ithaca, N.Y. (Comstock Publishing Associates. Co., Inc.), pp. 58. SZARSKI, H. & CZOPEK, G. (1966): Erythrocyte diameter in some amphibians and reptiles.- Bulletin de l'Académie Polonaise des Sciences, Varsovie; (Cl. II. - Série des Sciences biologique) 14 (6): 433-437. TANYER, G. (1985): Hematoloji ve laboratuvar [Hematology and laboratory]. Ankara (Ayyıldız Matbaası A.Ş.), pp. 442. TOK, C. V. & TOSUNOĞLU, M. & AYAZ, D. & ÇİÇEK, K. & GÜL, Ç. (2009): Hematology of the Lycian Salamander, *Lyciasalamandra fazi-lae*.- North-Western Journal of Zoology, Oradea; 5 (2): 321-329. TOSUNOĞLU, M. & TOK, C. V. & GÜL, Ç. & GÜLDALI, G. (2008): The blood cells of West Anatolian *Lissotriton vulgaris* (LINNAEUS, 1758).- Herpetozoa, Wien; 21 (1/2): 87-91. TOSUNOĞLU, M. & TOK, C. V. & ÖLGÜN, K. & ÖZDEMİR, N. & GÜL, Ç. (2011): Hematology of the northern banded newt, *Ommatotriton ophryticus* (Amphibia: Urodela) from north Anatolia.- Russian Journal of Herpetology, Moskva; 18: 59-64. WINTROBE, M. M. (1933): Variations in the size and hemoglobin content of erythrocytes in the blood of various vertebrates.- Folia Haematologica, Leipzig; 51: 32-49. WOJTASZEK, J. & ADAMOWICZ, A. (2003): Haematology of the fire-bellied toad, *Bombina bombina* L.- Comparative Clinical Pathology, London; 12: 129-139. WOJTASZEK, J. & BARANOWSKA, M. & GLUBIAK, M. & DZUGAJ, A. (1997): Circulating blood parameters of the water frog, *Rana esculenta* L. at pre-wintering stage.- Zoologica Poloniae, Wrocław, Warsaw; 1-4: 117-126. ZHUKOVA, T. I. & KUBANTSEV, B. S. (1979): Changes in the blood composition of salientians during hibernation.- The Soviet Journal of Ecology, New York; 9: 379-382.

KEY WORDS: Amphibia: Urodela: Salamandridae; *Triturus karelinii*, hematological parameters, Turkey

SUBMITTED: July, 13, 2012

AUTHORS: Murat TOSUNOĞLU <mtosun@comu.edu.tr >, Çiğdem GÜL (corresponding author <gulcigdem@comu.edu.tr > <gulcigdem17@hotmail.com.tr >, İbrahim UYSAL <uysalibrahim@windowslive.com >, Çanakkale Onsekiz Mart University, Faculty of Arts and Sciences, Department of Biology, Terzioğlu Campus, 17100 Çanakkale, Turkey