Journal of Applied Biological Sciences 3(3):77-79, 2009
ISSN: 1307-1130, www.nobel.gen.tr

Blood Serum Proteins of *Myotis myotis* (Borkhausen, 1797) and *Myotis blythii* (Tomes, 1857) (Chiroptera: Vespertilionidae)

Nursel AŞAN BAYDEMİR* İrfan ALBAYRAK Tarkan YORULMAZ Sinan İNCİ
University of Kırıkkale, Faculty of Science and Arts, Department of Biology, 71451 Yahşihan, Kırıkkale, TURKEY

*Corresponding Author
e-mail: nurselasan@yahoo.com

Recevied : August 23, 2009
Accepted : October 17, 2009

Abstract

This study is based on the globulin and albumin proteins, determined using SDS-PAGE, of two sibling bat species *Myotis myotis* (Borkhausen, 1797) (Greater Mouse-eared Myotis) and *M. blythii* (Tomes, 1857) (Lesser Mouse-eared Myotis) distributed in Turkey. No difference is found in the globulin and albumin protein bands. It was concluded that blood serum proteins could not be enough diagnostic character for separating *Myotis myotis* from *M. blythii*.

Key words: *Myotis myotis*, *Myotis blythii*, SDS-PAGE, Turkey

INTRODUCTION

The genus *Myotis* is represented by 103 species in the Holoarctic Region [1, 2]. The taxonomic status of two taxa of this genus, *Myotis myotis* (Borkhausen, 1797) (Greater Mouse-eared bat) and *Myotis blythii* (Tomes, 1857) (Lesser Mouse-eared bat), are stil controversial.

Myotis myotis (Borkhausen, 1797) is represented by *M. myotis myotis* distributed in Turkish Thrace, and *M. myotis macrocephalicus* distributed in Mediterranean Region. In addition, *M. blythii* (Tomes, 1857) is represented by *M. blythii oxygnathus* and *M. blythii* omari [3].

Although taxomic, karyologic or biologic studies of two taxa were achieved from Turkey [4-7] no studies have been achieved on the SDS-PAGE patterns of blood serum proteins.

Fig. 1. Two sibling species *Myotis myotis* (A) and *Myotis blythii* (B)

The objective of this paper is to describe the blood serum proteins, using SDS-PAGE, of both sibling species.

MATERIAL AND METHODS

Study area: Edirne (41° 40′ N 26° 33′ E), Kırklareli (41° 44′ N 27° 13′ E), Bolu (40° 47′ N 32° 11′ E), Kırıkkale (39° 40′ N 33° 36′ E), Tokat (40° 16′ N 36° 17′ E), Artvin (41° 07′ N 42° 03′ E), Şanlıurfa (37° 01′ N 37° 59′ E), Diyarbakır (38° 16′ N 39° 45′ E), Batman (37° 42′ N 41° 26′ E), Mersin (36° 38′ N 33° 26′ E), Kilis (36° 43′ N 37° 06′ E), Gaziantep (37° 03′ N 37° 22′ E).

This study is based on 34 (29 ♂♂ and 5 ♀♀) *Myotis myotis* and 9 (6 ♂♂ and 3♀♀) *M. blythii* specimens collected from different localities of Turkey between the years 2003 and 2006 (Fig. 1).

Blood was taken by cardiac puncture with a dispersal 2 ml syringe, from anesthetized specimen. After blood clotting the sera, mixed with 0.0625 M Tris Cl, pH 6.8, 2 % SDS, 10 % Glycerol, 5% 2-Mercaptoethanol and 0.01% bromphenol blue, were boiled for 3-3.5 minutes and preserved at -80°C until electrophoresis. SDS-PAGE was performed according to the method of Laemmli [8]. Separating (7.5 %) and stacking (4 %) gels were prepared according to the method of Sambrook et al. [9]. SIGMA Protein Molecular Weight Marker (MW-SDS-200) was applied. Samples and the marker of 30 µl were applied to the gel. Electrophoresis was carried out using a MBT80EL model double vertical slab gel electrophoresis apparatus. 10 V/cm and 20 V/cm were applied to the stacking and separating gel, respectively. The gel was run about 4.5 hours and then stained with 0.25 % Coomasie Brillant Blue R-250 over the night. After destaining in a mixed washing solution of 45 ml methanol: 10 ml acetic acid: 45 ml dH2O, gel was photographed. Blood serum proteins were evaluated as globulin and albumin regions.

The albumin region is subdivided into prealbumin (PA), albumin (A) and postalbumin (PsA) zones.

The stuffed and skinned specimens as well as karyotype preparations are deposited in the zoology museum of Department of Biology, University of Kırıkkale.

RESULTS

Blood serum proteins of *Myotis myotis* and *M. blythii* using SDS-PAGE were first examined by this study. Female and male specimens are evaluated together, no differences were found between the sexes (Fig. 2).

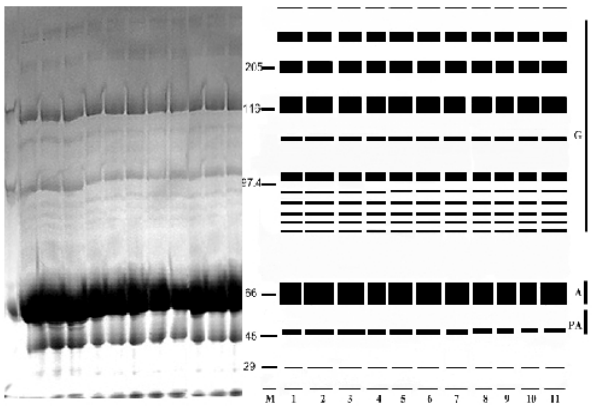

Fig. 2. Blood serum proteins of *Myotis myotis* (6-11) and *Myotis blythii* (1-5). G= Globulin, A= Albumin, PA= Prealbumin. 1. Mersin (KKU04003 ♂), 2. Diyarbakır (KKU05008 ♂), 3. Gaziantep (KKU06012 ♂), 4. Kırıkkale (KKU04019 ♂), 5. Diyarbakır (KKU05009 ♀), 6. Edirne (KKU04032 ♂), 7. Kilis (KKU06016 ♂), 8. Kırıkkale (KKU04017♂), 9. Tokat (KKU05030 ♂), 10. Kırıkkale (KKU04018 ♀), 11. Artvin (KKU04040 ♂), M=Marker

There were 10 bands in the globulin region, one band in albumin and three bands in the prealbumin zones. In contrast, no band was found in the postalbumin zone. In both species, the bands in the line of marker protein 205.000 D and 66.000 D were stained very weakly. The bands between the line of 97.400 D and 66.000 D of the specimens 8-11 were more detectable than the others.

DISCUSSION

Morphology, chromosomal banding, protein electrophoresis, protein sequencing, immunology, DNA hybridization and DNA sequencing techniques are used for determining the relationship levels between the taxa [10].

In Turkey, many electrophoretic studies on the blood serum proteins of various rodents, were achieved [11-17]. To date, no electrophoretic study on blood serum proteins of bats has not been done yet. This study represents the first records of blood serum proteins of two sibling vespertilionid bats.

The electrophoretic patterns expected to be a useful diagnostic character for separating the two taxa with this study. In conclusion, similarities in the electrophoretic band patterns of blood serum proteins supported the claim; in addition to identical karyotype and morphology, electrophoretic aspects of blood serum proteins also could not be a diagnostic character for separating greater mouse-eared bat from lesser mouse-eared bat.

ACKNOWLEDGEMENT

This study was supported by the Research Fund of Kırıkkale University (BAP-03.03.04.04).

REFERENCES

[1] Simmons NB., 2005. Order Chiroptera. In: Mammal Species of the World. A Taxonomic and Geographic Reference. Third edition (eds. Wilson DE, Reeder DM). pp. 500-518. The Johns Hopkins University Press, Baltimore.

[2] Stadelmann, B, Lin LK, Kunz TH, Ruedi M., 2007. Molecular phylogeny of New world Myotis (Chiroptera, Vespertilionidae) inferred from mitochondrial and nuclear DNA genes, Molecular Phylogenetics and Evolution, 43: 32-48.

[3] Aşan A., 2001. Taxonomic status of *Myotis myotis* (Borkhausen, 1797) and *Myotis blythii* (Tomes, 1857) in Turkey (Mammalia: Chiroptera). Ph D Thesis, Ankara University Graduate School of Natural and Applied Sciences, Ankara, 170 pp.

[4] Albayrak İ, Aşan N., 1998. Geographic variations and taxonomic status of Myotis myotis (Borkhausen, 1797) in Turkey (Chiroptera: Vespertilionidae). Turk. J. of Zoology 22: 267-275.

[5] Albayrak İ, Aşan N., 1999. Distributional status of the bats from Turkey (Mammalia: Chiroptera). Commun. Ac. Sci. Univ. Ank. Series C, 17: 59-68.

[6] Albayrak İ, Aşan N., 2001. The Structure of baculum in *Myotis myotis* and *Myotis blythii* (Chiroptera: Vespertilionidae). Turk. J. of Zool., 25 (3): 229-233.

[7] Aşan N, Albayrak I., 2006. A study on the breeding biology of some bat species in Turkey. Turk. J. of Zool., 30: 103-110.

[8] Laemmli UK., 1970. Cleavage of structural proteins during the assembly of the head of bacteriophage T4. Nature, 227: 680-685.

[9] Sambrook J, Fritsch EF, Maniatis T., 1989. Molecular cloning. A laboratory Manual. Cold Spring Harbor Laboratory Press, New York.

[10] Zima J., 1982. Chromosomal homology in the complements of bats of the family Vespertilionidae II. G-band karyotypes of some *Myotis*, *Eptesicus* and *Pipistrellus* species. Folia Zool., 31: 31-36.

[11] Verimli R, Çolak E, Yiğit N, Sözen M, Özkurt Ş., 2000 a. Electrophoretic aspects of blood serum proteins of *Apodemus mystacinus* and *Apodemus agrarius* (Mammalia: Rodentia) in Turkey. Turk J. Zool., 24: 225-229.

[12] Verimli R, Yiğit N, Çolak E, Sözen M., 2000

b. Blood serum proteins of the genus *Mesocricetus* Nehring, 1898 (Mammalia:Rodentia) in Turkey. Turk J. Biol., 24:855-858.

[13] Verimli R, Çolak E, Yiğit N, Sözen M, Özkurt Ş., 2001. Blood serum proteins of *Apodemus flavicollis* and *Apodemus hermonensis* (Mammalia: Rodentia) in Turkey. Turk J. Biol., 25: 89-92.

[14] Çolak R, Yiğit N, Çolak E, Gattermann R, Neumann K., 2002. SDS-PAGE patterns of blood serum proteins in some species of the genus *Meriones* (Mammalia:Rodentia). Turk. J. Zool. 26: 177-181.

[15] Yiğit N, Verimli R, Çolak E, Sözen M, Özkurt Ş., 2001. Blood serum proteins of *Rattus rattus* and *Rattus norvegicus* (Mammalia: Rodentia) in Turkey. Turk. J. Biol. 25:83-88.

[16] Yiğit, N., Çolak, E., Çolak, R., Özkan, B. and Özkurt, Ş. 2003. On the Turkish populations of *Dryomys nitedula* (Pallas, 1779) and *Dryomys laniger* Felten and Storch, 1968 (Mammalia: Rodentia). Acta Zoologica Academiae Scientiarum Hungaricae. 49: 147-158.

[17] İyigün C, Çolak R., 2004. An electrophoretic study on esterase and blood serum proteins of the water vole, *Arvicola terrestris* (L., 1758) (Mammalia: Rodentia), in Kırşehir province. Turk. J. Biol. 28:47-53.