Turk J Zool
26 (2002) 177-181

Research Article

SDS-PAGE Patterns of Blood Serum Proteins in some Species of the Genus *Meriones* (Mammalia: Rodentia)

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Received: 02.10.2001

Abstract: The blood serum proteins of *Meriones meridianus*, *Meriones crassus*, *Meriones persicus* and *Meriones tristrami*, all found in Turkey, along with domestic *Meriones unguiculatus*, which originated in Mongolia, were examined for the first time by SDS-PAGE. In the globulin zone, seven to ten bands were identified, one band in the post-albumin and albumin zones and one or two bands in the pre-albumin zone of five species of the genus *Meriones*. There was no diagnostic difference in the pattern of serum protein bands between species.

Key Words: SDS-PAGE, Serum, *Meriones*, Turkey

Meriones (Mammalia: Rodentia) Cinsinin Bazı Türlerinin Kan Serum Proteinlerinin SDS -PAGE Özellikleri

Özet: Türkiye'de yayılış gösteren *Meriones meridianus*, *Meriones crassus*, *Meriones persicus* ve *Meriones tristrami* ile Moğolistan kökenli evcil *Meriones unguiculatus*' un kan serum proteinleri SDS -PAGE tekniği ile ilk kez incelendi. *Meriones* cinsinin çalışılan beş türünde globulin bölgesinde yedi – on band, post-albumin ve albumin zonlarında bir bant ve pre-albumin zonunda ise bir – iki bant tespit edildi. Bu cinsin türlerinin serum protein bantları arasında herhangi bir diagnostik farklılık bulunmadı.

Anahtar Sözcükler: SDS - PAGE, Serum, *Meriones*, Türkiye

Introduction

The dominant genus of gerbils is *Meriones*, which is confined to the Palearctic region (1). According to Harrison and Bates (2), the genus *Meriones* is represented by seven species in the Middle East. Of these species, *Meriones meridianus*, *Meriones libycus*, *Meriones crassus*, *Meriones persicus*, *Meriones tristrami* and *Meriones vinogradovi* have been reported in Turkey in many taxonomic, morphological and biological studies (3-12). *Meriones unguiculatus*, another species of the genus *Meriones*, is naturally distributed in Mongolia and adjacent countries (1,13). Up to now, there have been no electrophoretic studies on blood serum proteins of the genus *Meriones*. Brand and Ryckman (19) have pointed out the taxonomic importance of blood serum proteins for the genus Peromyscus. Apart from these, there have only been a few studies of Turkish rodents (28-31). This is a preliminary paper and the first report of work on blood serum proteins of *M. meridianus*, *M. crassus*, M. unguiculatus, *M. persicus* and *M. tristrami*. Furthermore, this study also aims to assess whether the electophoretic aspects of blood serum proteins are of taxonomic importance for the genus *Meriones*.

Materials and Methods

SDS-PAGE (sodium dodecyl sulphate polyacrylamide gel electrophoresis) analysis was performed on live specimens caught in 8 different localities in the distribution and record areas of *M. meridianus* (n= 14), *M. crassus* (n= 6), *M. persicus* (n=3) and *M. tristrami* (n =13) in Turkey (Fig. 1), along with four specimens of *M. unguiculatus* obtained from a pet shop. Globulin (G) and albumin (A) regions with subzones were assayed in all specimens. The globulin region was considered without separating into subzones, but the albumin region was subdivided into post-albumin (PsA), albumin (A) and prealbumin (PA) zones. Blood was taken by cardiac puncture from the animals, which had been anaesthetized with ether. After blood clotting, the separated sera were

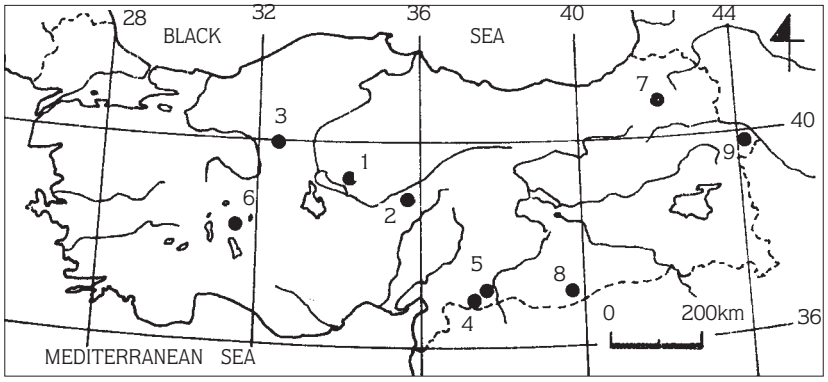

Fig. 1. The localities of the specimens examined. 1. Kırşehir, 2. Kayseri, 3. Ankara, 4. Kilis, 5. Nizip, 6. Yeşilköy (*M. tristrami*), 7. Oltu/Erzurum (*M. persicus*), 8. Çaylık/Şanlıurfa (*M. crassus*), 9. Aralık/Iğdır (*M. meridianus*).

centrifuged at 12,000 rpm for 3 min. The sera were mixed with a sample buffer as described by Laemmli (32). The final concentration of sera was adjusted to 5%. Samples were boiled for 3 min and stored at -70ºC until electrophoresis. Electrophoresis was carried out using a Consort E 863 model vertical slab gel electrophoresis apparatus. SDS-polyacrylamide denaturing gels (separating gels (7.5%) and stacking gels (4%)) were prepared as described by Sambrook et al. (33). The amount of protein loaded to the gel was semi-quantitatively determined according to Esen's method (34). Electrode buffer solution was made up of 0.025 M Tris, 0.192 M Glycine and 0.1% SDS at pH 8.3 (33). A sample of 15 µl and Molecular Weight Marker (Sigma MW- SDS – 200, carbonic anhydrase: 29000, egg albumin: 45000, bovine albumin: 66000, phosphorylase B: 97400, β-galactosidase: 116000, myosin: 205000) were applied to gels in the experiments. Constant voltage (8 V/cm) was applied to stacking gel. After the tracing dye attained the separating gel, the voltage was adjusted to 15 V/cm. After electrophoresis, gels were stained with 0.25% Coomassie Brilliant Blue R250 (CBB) in a solution of methanol, water and glacial acetic acid (45:45:10), and destained in the same solution without CBB.

Results and Discussion

M. meridianus was first recorded in Aralık province (Iğdır), and its distribution is confined to this area of Turkey (10). Serum proteins of 14 specimens of *M. meridianus* captured from Aralık/Iğdır were examined by SDS-PAGE electrophoresis (Figs. 1, 2). Male and female specimens were evaluated together as there are no differences between the sexes. Globulin (G) and albumin (A) regions were assayed in 14 specimens. Nine or ten bands were determined in the globulin region, and this variation resulted from the patterns of the first three or four weak bands. The bands in the globulin region were usually stained weakly. However, there was a strong band almost between the marker proteins of 205000 D and 116000 D. Post-albumin and albumin zones were monomorphic and strong in all specimens examined. One phenotype had a fast PA band, a second had a slow PA band, and a third phenotype also had two bands of equal density. This phenotype was probably in heterozygous state (Fig. 2). *M. crassus* is distributed in south-east Turkey (10) (Fig. 1). We discovered one electrophoretic pattern in blood serum protein with nine bands in the globulin region. The strong band approximately between the marker proteins of 205000 D and 116000 D was also found in this species. PsA and A zones were similar to those of the previous species, but the PA zone had 2 bands. One of these bands was fast and strongly stained, but the other was slow and weaker (Fig. 2). *M. unguiculatus* is not found in Turkey. This species, known as the Mongolian gerbil, is distributed across central Asia (13). Blood serum proteins of four specimens of *M. unguiculatus* from a laboratory colony were assayed by SDS-PAGE. Nine bands without variation were observed in the globulin region of the specimens examined. The band patterns in the globulin region were similar to those of *M. crassus*. PA and A zones were the same as those of previous species, but the PsA band was markedly slower than that of other species of the genus *Meriones*. The PA zone had 2 bands (Fig. 2). *M. persicus* is confined to a single locality around Oltu/Erzurum (6) (Fig. 1). Two different patterns of blood serum protein in the globulin region with 7-8 bands were observed in the specimens of *M. persicus*. The variation resulted from patterns of the first three weak bands. Of the specimens, one had only

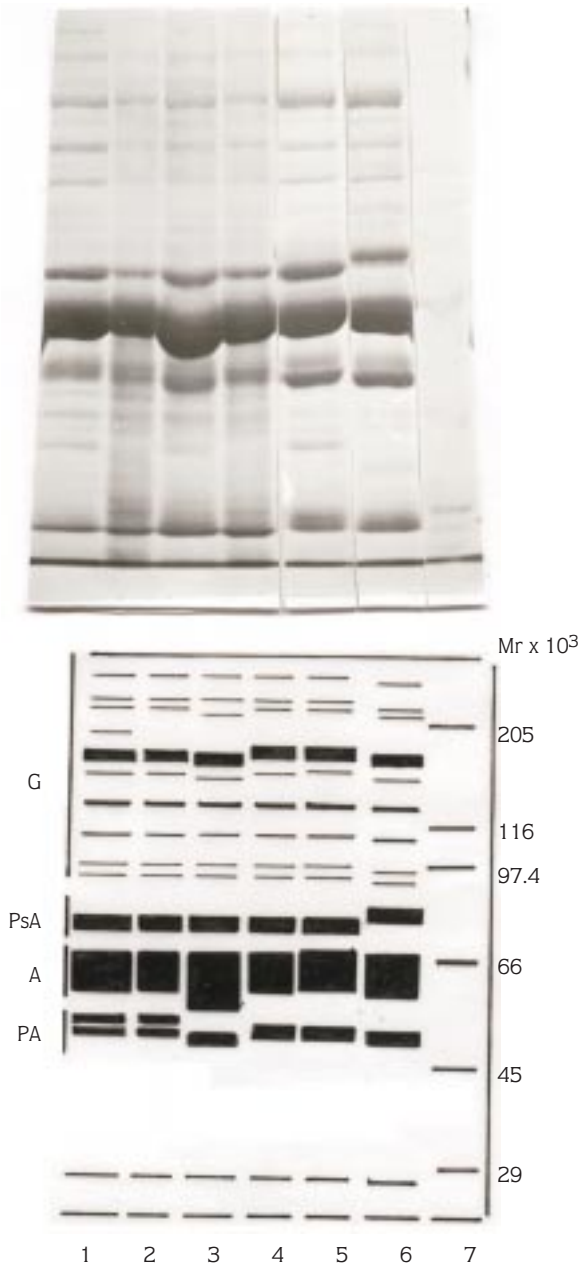

Fig. 2. SDS-PAGE patterns of serum proteins of *M. meridianus* (1-4), *M. crassus* (5) and *M. unguiculatus* (6). G= Globulin, PsA= post-albumin, A= albumin, PA= pre-albumin, Mr= marker (7).

two weak bands, but the other two had three weak bands in the globulin region (Fig. 3). The strong band in the globulin region was almost in the marker proteins of 205000 D. Additionally, the PsA, the A and the PA zones were found to be the same as those of *M. crassus*. The first band in the PA zone was weaker than the second (Fig. 3). *M. tristrami* is a common species, which occupies

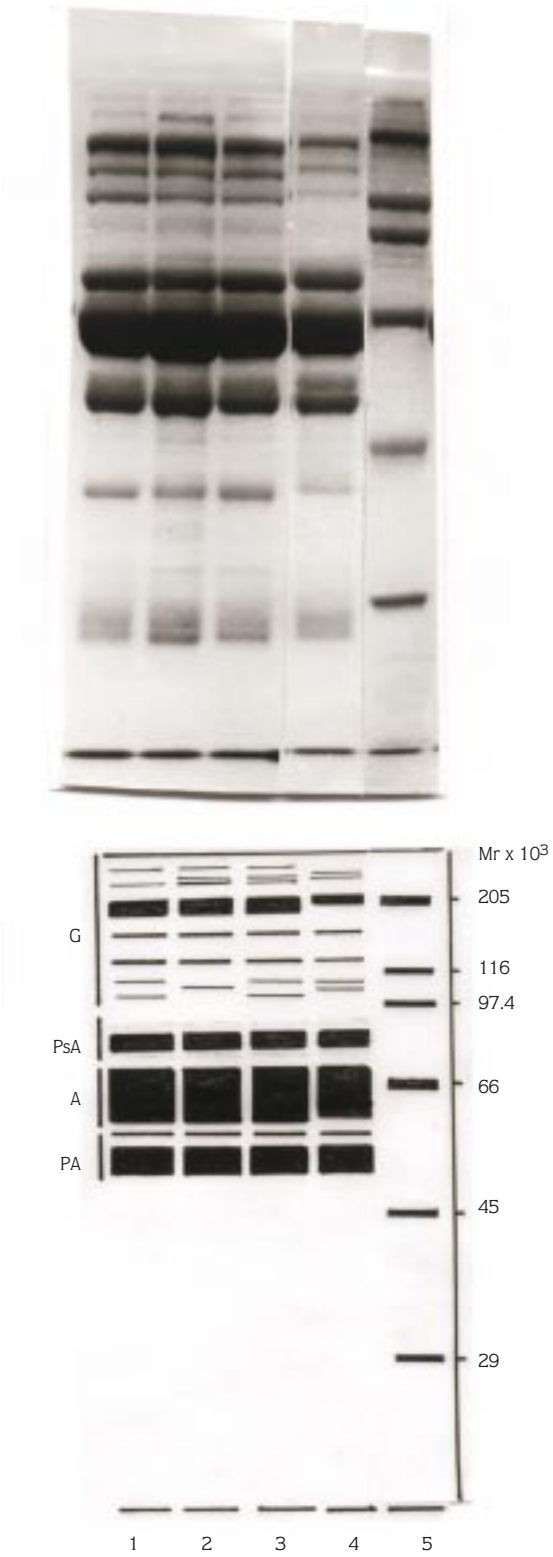

Fig. 3. SDS-PAGE patterns of serum proteins of *M. persicus* (1-3) and *M. tristrami* (4). G= Globulin, PsA= post-albumin, A= albumin, PA= pre-albumin, Mr= marker (5).

Table 1. The subgroups and their bands of the four main blood serum proteins of the genus ***Meriones*** along with a comparison of other species from Turkey (G= Globulin, PsA= post-albumin, A= albumin, PA= pre-albumin).

The subgroups of serum protein/Species	G	PsA	A	PA	References
Meriones meridianus	9 - 10	1	1	1 -2	This study
Meriones crassus	9	1	1	2	This study
Meriones unguiculatus	9	1	1	2	This study
Meriones persicus	7 - 8	1	1	2	This study
Meriones tristrami	7	1	1	2	This study
Mesocricetus brandti	7	1	1	2	Verimli et al. (2000a)
Mesocricetus auratus	7	1	1	2	Verimli et al. (2000a)
Apodemus mystacinus	7 - 8	1	1	2 - 4	Verimli et al. (2000b)
Apodemus agrarius	9	1	1	2	Verimli et al. (2000b)
Rattus rattus	8 - 12	1 - 2	1	1 - 4	Yigit et al. (2001)
Rattus norvegicus	8 - 11	1- 2	1	3 - 4	Yigit et al. (2001)
Apodemus flavicollis	6 - 7	1	1	2	Verimli et al. (2001)
Apodemus hermonensis	7 - 9	1	1	2	Verimli et al. (2001)

the steppe areas of Turkey (9) (Fig. 1). We identified only one different type of blood serum protein pattern in the specimens of *M. tristrami*. Seven bands were observed in the globulin region of the specimens from Kırşehir, Kayseri, Ankara, Kilis, Nizip and Yeşilköy (Fig. 3). The strong band in the globulin region almost corresponded to the marker proteins of 205000 D in *M. persicus* (Fig. 3). There was a weak band in front of this strong band that was constantly present in the blood serum pattern of *M. tristrami*. Post-albumin and albumin zones were monomorphic and strong in all the specimens examined. A very weak band constantly appeared between the PsA zone and the A region, but this was judged to be a smear (Fig. 3). The pre-albumin zone consisted of two bands. The slow one was weaker than the fast one.

Verimli et al. (28, 29) and Yigit et al. (30) performed electrophoretic studies on the blood serum proteins of *Mesocricetus brandti, Mesocricetus auratus, Apodemus mystacinus, Apodemus agrarius, Apodemus hermonensis, Apodemus flavicollis,* and *Rattus rattus and Rattus norvegicus* respectively. They detected a single band in the A zone and variations in other zones such as G, PsA and PA (Table 1). Nagase et al. (21) also reported the patterns of PsA and A bands of analbuminemic Sprague–Dawley rats. These findings are consistent with our results. Freguedakis-Tsolis et al. (24) determined two bands in the A zone of *A. flavicollis* and *A. mystacinus*, and one band in that of *A. sylvaticus* as well as one band in the PA of both species. According to Freguedakis-Tsolis and Chondropoulus (25), there are two bands in the PA zone of *Mus musculus*, one band in *A. flavicollis* and no band in *Pitymys atticus*. These authors also reported one band in the A zone of *M. musculus* and *P. atticus* and two bands in *A. flavicollis*. Brown and Welser (18) and Jensen and Rasmussen (20) found three bands with different mobilities and 1-2 bands with five different mobilities in the A zone of the genus *Peromycus*, respectively. In contrast, species of the genus *Meriones* had only a single band in the A zone, as reported for other species from Turkey (28-30). However, the patterns of PA in the genus *Meriones* have been similarly reported for other rodent species by Reuter and Kennes (16), Gemmeke (23), Freguedakis-Tsolis et al. (24), Freguedakis-Tsolis and Chondropoulus (25), Verimli et al. (28,29) and Yigit et al. (30). Brand and Ryckman (19) revised the taxonomic relationship of *Peromyscus eremicus, P. guardia* and *P. interparietalis* using morphologic and electrophoretic data. They stated that the protein patterns showed consistent inter-population differences, which tend to support the current interpretation of the taxonomy of this genus, and the serum protein data indicated a closer relationship between *P. eremicus* and *P. interpariatelis*. Unlike these, according to comparisons of the SDS-PAGE pattern of the genus *Meriones*, no post-albumin and albumin polymorphisms were observed in the four species studied. However, *M. unguiculatus* has a slow band in the PsA zone. In conclusion, the patterns of serum protein bands investigated failed as a diagnostic indicator of the genus *Meriones* in Turkey, unlike those reported for the genus *Peromyscus* by Brand and Ryckman (19).

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