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Electrophoretic comparison of blood- Serum proteins of Ellobius Fisher, 1814. (Mammalia: Rodentia) in Iran

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Abstract
Globulin and Albumin blood-serum proteins of 48 Ellobius specimens which were collected from north-east (Mashhad and Bojnord), west (Kurdistan, Hamadan, Qazvin, and Zanjan), and north-west (Pir-ahmad-kandi, Kelisa-kandi, and Nadou villages) of Iran have been examined using SDS-PAGE method. In residual region of Globulin protein (G), specimens from north-east and west group and north-west group had created 5 and 11 electrophoretic bands, respectively; and in residual region of Albumin protein (A) all specimens created only one band and north-west group created 4 electrophoretic bands. Although these electrophoretic band differences could led to diagnosis of these two Ellobius specimens, but this difference along with morphological and *Karyological* properties of these specimens, can help identifying the distribution of Ellobius fuscocapillus in north-east and west and Ellobius lutescens in north-west of the Iran.

Keywords: Albumin, Globulin, Iran, Ellobius fuscocapillus , Ellobius lutescens

INTRODUCTION

Mole voles, genus *Ellobius*, are Palearctic region animals distributed from East Anatolia to Mongolia.Gromov and Baranova stated[1,2,3,4]. That the genus *Ellobius* has existed since the Mid-Pleistocene[5,6]. Pleistocene remains of *Ellobius spp*. were found from Konya-Akşehir-Dursunlu in Turkey [7,8].The species *E. fuscocapillus*, *E. lutescens*, *E. talpinus*, *E. tancrei* and *E. alaicus* exist at the present time[9,10].These species exhibit on allopatric distribution.*E. fuscocapillus* is distributed in Iran [11,12,13]. Afghanistan and Pakistan;[14] *E. talpinus* in Ukraine[15,16], Kazakhstan, Turkmenia, Uzbekistan and Afghanistan [17,18]; *E. tancrei* is in Kyrgyzystan, Tajikistan and Mongolia; *E.alaicus* is in Kyrgyzystan (this species is endemic for Kyrgyzstan) and *E. lutescens* in Iran [19], Armenia, Azerbaijan and Anatolia [20,21]. *Ellobius lutescens* reaches the western limit of its distribution area from Van-Hakkari province in East Anatolia [22,23], and was first described by Thomas in 1897, based on six specimens collected from Qazvin and Zanjan(Fig. 1).The taxonomical, karyological, morphological and some biological peculiarities were studied by Moradi.M and Kivanç.E.[24]. Although the presence of this species is known in East and north- west of Iran, its exact distribution area and ecological peculiarities have not yet been documented in detail. The aim of this study was therefore to determine the distribution boundaries and some ecological peculiarities.

MATERIALS AND METHODS

Electrophoretic analysis was performed on 48 live specimens of *Ellobius fuscocapillus* (n=18) and *Ellobius lutescens* (n=30) (Fig.1).

Blood was taken by cardiac puncture from the animals anaesthetized with ether. After blood clotting the separated sera were centrifuged at 12000 rpm for 3 min. The sera were mixed with a sample buffer containing 0.0625 MN Tris Cl, PH 6.8, 2%SDS, 10% Glycerol, 5% 2-Mercaptoethanol and 0.01% Bromphenol Blue (Laemmli, 1970) and the sera was adjusted to 5% in the mixture. Samples were boiled for 3 min and stored at -70ºC until electrophoresis. Amount of protein loaded to gel was qualitatively determined by the method of Esen (1978). Samples of 10 to 15 µL were applied to gels in different experiments. Electrophoresis was carried out using 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 Sam brook et al. (1989). Electrophoresis buffer contains 0.025 M Tris, 0.192 M Glycine, 0.1% SDS at PH 8.3 (Sam brook et al., 1989). Molecular Weight Marker (Sigma MW-SDS-200) consists of carbonic anhydrase (29.000 D), egg albumin (45.000D), bovine albumin (66.000 D), phosphory lase B (97.400 D), β-galactosidase (116.000 D), myosin (205.000 D).

Constant voltage (8 V cm^{-1} . After electrophoresis, gels were stained with 0.25% Coomassie Brillant Blue R250 in 90 ML of methanol: water (1:1 v/v) and 10 mL glacial acetic acid and destained in methanol: eater: acetic acid (45: 45: 10) (Sam brook et al., 1989).

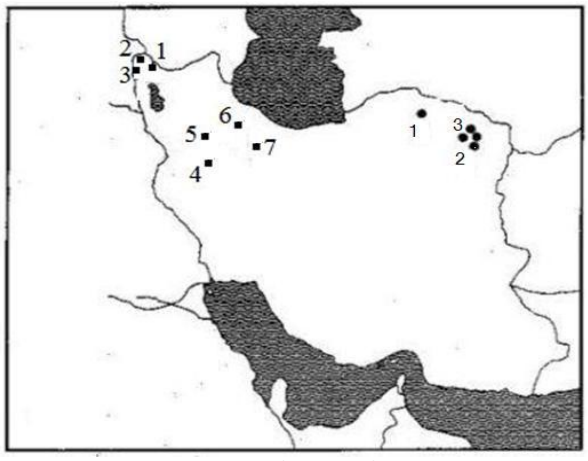

Fig. 1: Map showing localities of *Ellobius fuscocapillus* [●](1.Bojnurd,2.Mashhad,3.Toroq)and *Ellobius lutescens* [■](1.Pirahmad kandi , 2.Kelisa kandi, 3.Naderloo Velage, 4.Hamadan 5.Kurdestan, 6.Zanjan, 7Qazvin.) and 10. Marker

RESULTS

In residual region of Globulin protein (G), specimens from north-east group and west, north-west group had created 5 and 11 electrophoretic bands, respectively; and in residual region of Albumin protein (A) all north-east specimens created only one band (Fig 2). But west and north-west group created 4 electrophoretic bands. Although these electrophoretic band differences could led to diagnosis of these two *Ellobius* specimens, but this difference along with morphological and Karyological properties of these specimens, can help identifying the distribution of *Ellobius fuscocapillus* in north-east and *Ellobius lutescens* in west and north-west of the Iran.

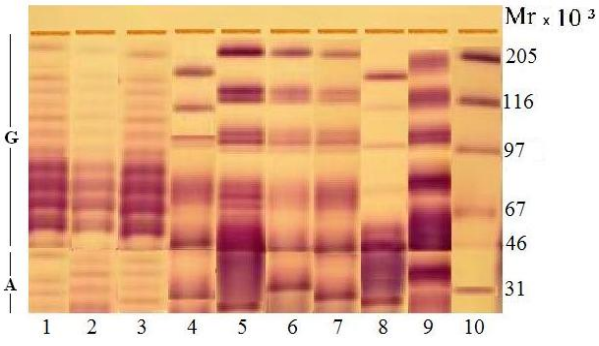

Fig. 2. SDS-PAGE zymogram of blood-serum proteins of *Ellobius fuscocapillus*

(1.Bojnurd,2.Mashhad,3.Toroq)and *Ellobius lutescens*(1.Pirahmad kandi, 2.Kelisa kandi, 3.Naderloo Velage, 4.Hamadan 5.Kurdestan, 6.Zanjan, 7Qazvin.) and 10. Marker

DISCUSSION

We analysed specimens from north-east Iran and found 4 bands in globulin zone, 1-2 bands in post albumin zone, 2 bands in pre-albumin zone and 2-3 bands in fast zone. In *Ellobius fuscocapillus*, 9-11 bands in glubin zone, 1-2 bands in post albumin zone, 3 bands in prealbumin zone and 2-3 bands in fast zone in *Ellobius lutescens* (Fig 3). These findings showed that pre-albumin distinguished *Ellobius fuscocapillus* from *Ellobius lutescens*. Also, we recorded firstly *Ellobius fuscocapillus* from Iran.

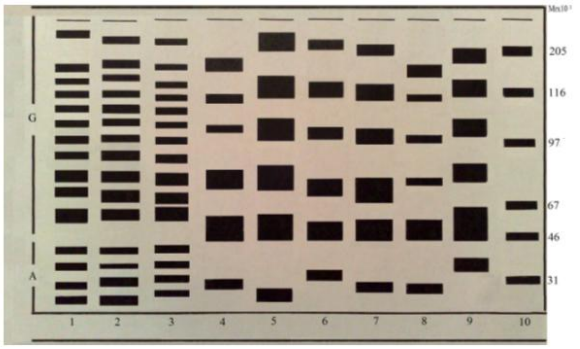

Fig. 3. SDS-PAGE patterns of blood-serum proteins of *Ellobius fuscocapillus*

(1.Bojnurd,2.Mashhad,3.Toroq)and *Ellobius lutescens*(1.Pirahmad kandi, 2.Kelisa kandi, 3.Naderloo Velage, 4.Hamadan 5.Kurdestan, 6.Zanjan, 7Qazvin.) and 10. Marker

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