A serological comparison of the populations of the Levantine Viper, *Macrovipera lebetina* (Linnaeus, 1758) in Cyprus and Southern Turkey

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Abstract: The present study compares the blood-serum proteins of populations of the Levantine viper, *Macrovipera lebetina* L. from Cyprus and southern Anatolia by polyacrylamide gel electrophoresis and densitometry analysis methods. There were discernible differences found between the two populations for the blood serum protein features compared. By considering these differences, it is concluded that the southern Anatolian population of *Macrovipera lebetina* should not be considered as the nominate subspecies *M. l. lebetina*, which lives in Cyprus.

Key Words: *Macrovipera lebetina lebetina*, *M. l. obtusa*, blood-serum proteins, Cyprus, Turkey.

Introduction

The Levantine viper, *Macrovipera lebetina*, has a rather extensive geographical range in central Asia, the Middle East and northern Africa (Algeria & Tunis) (Nilson & Andren 1988, Nilson et al. 1988, Böhme & Wiedl 1994, Göçmen et al. 1996, Schleich et al. 1996, David et al. 1999, Atatür & Göçmen 2001).

There has been considerable confusion and disagreement among authors with regard to the taxonomic status of *M. lebetina*. Asiatic and Russian *lebetina* were attributed to the subspecies *obtusa* for a long time by several authors (Chernov 1944, 1959, Terentjev & Chernov 1949, Bannikov et al. 1977, Başoğlu & Baran 1980, Joger 1984, Nilson & Andrén 1988, Chickin & Szczerbak 1992, Leviton et al. 1992, Baran & Atatür 1998, Tok et al. 2002), but there was no general consensus about its definition. The populations from the easternmost parts of the range of *M. lebetina*, previously regarded as belonging to the subspecies *turanica* (Chernov in Terentjev & Chernov, 1949), were described as *chernovi* by Chikin & Szczerbak (1992). Different authors argued over the validity of the other subspecies *turanica*, *euphratica*, *peilei*

and *chernovi* from Asia, and, for example, Joger (1984) considered only two valid subspecies: *lebetina* of Cyprus and *obtusa* of Asia (except Cyprus).

Although Anatolian *Macrovipera lebetina* were attributed to the subspecies *obtusa* for a long time (Baran 1976, Başoğlu & Baran 1980, Baran & Atatür 1998, Budak & Göçmen 2005), some authors (Billing & Schaetti 1984, Broodmann 1987, Ossenegg 1989, Mulder 1995) stated that the *M. lebetina* specimens from the southern parts of Anatolia closely resemble those of Cyprus (*M. l. lebetina*). Billing & Schätti (1984), Broodmann (1987) and Mulder (1995) stated that the specimens from the southern parts of Anatolia closely resemble those of Cyprus (*M. l. lebetina*) regarding head shape, color pattern, and number of ventrals. However, Arıkan et al. (2005) and Göçmen et al. (2006) comparatively examined populations from both localities regarding morphology, hemipenes and electrophoresis pattern of venom proteins. They found that significant differences between the two populations for some aspects of general morphology, hemipenial morphology and venom proteins. Therefore, they concluded that these populations are taxonomically distinct.

The present study aims to further examine the southern Turkey population from a serological point of view in order to clarify whether it should be considered as the nominate subspecies *M. l. lebetina* which lives in Cyprus.

Materials and Methods

For elecrophoresis, we used a total of 8 (4 ♂♂ and 4 ♀♀) adult *Macrovipera lebetina* specimens collected in northern Cyprus (2 ♂♂ and 2 ♀♀ from Kyrenia, Nicosia and Karpas Districts) and southern Turkey (2 ♂♂ and 2 ♀♀ from Kilis, Şanlıurfa and Diyarbakır Provinces). Blood samples were obtained from the postorbital sinuses via heparinized haematocrit tubes (MacLean et al. 1973). For electrophoretic study, 5 µl of blood-serum proteins were separated in polyacrylamide gel disk electrophoresis using a Canalco Model 1200 disk electrophoresis apparatus. Detailed procedure was explained by Arıkan et al. (1998, 2005). Qualitative evaluations of the gels were done directly from the electropherograms and the densitometric curves of the separations were created by means of a Gelman ACD-15 Model 39430 densitometer scanning at 500 nm.

Results

All specimens examined were sexually mature and no obvious difference was recorded in serum protein pherograms (in densitometric curves) between the sexes in either Cypriot or southern Turkish populations. Consequently, the sexes of each population were pooled for further evaluation. The protein distribution pattern of southern Turkey and Cypriot specimens representing of the *M. lebetina* populations studied is given in Figure 1 and 2, along with their densitometric tracing curves.

In qualitative comparisons between the specimens from northern Cyprus and southern Turkey, the electropherograms clearly differed in glo-

bulin zones. In Cypriot samples (Fig. 1), the number of discernible globulin fractions or fraction groups (zones) was 11, while in samples from southern Turkey they number 10 (Fig.2). Besides, the total fraction number of the blood-serum proteins of the Cypriot *M. lebetina* specimens comprises 12 fractions or fraction groups but 11 in the southern Turkey specimens when we include a common albumin fraction or fraction group. Additional quantitative differences (in the values of optic densities) were seen in each globulin fraction or fraction groups (fig. 1-2).

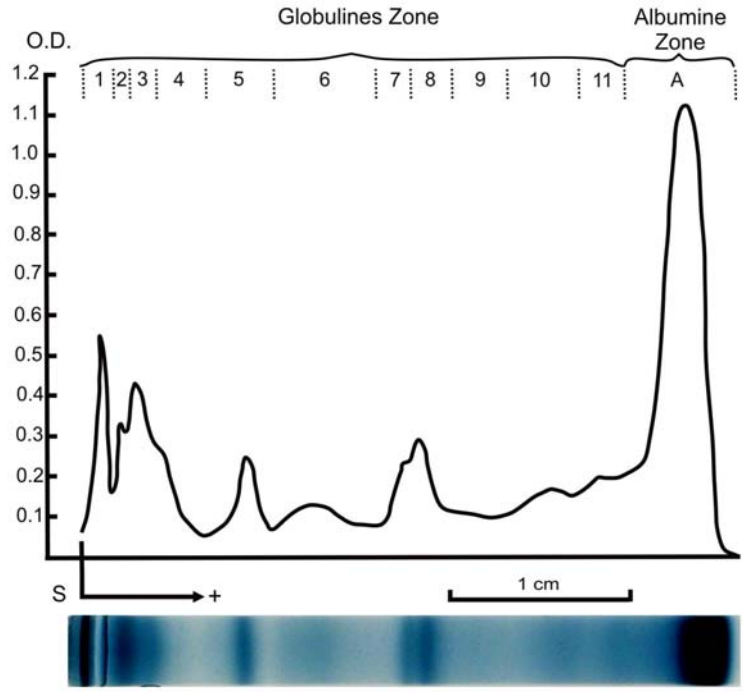

Figure no.1 Representative electropherogram (gel photograph) showing the electrophoretic separation of the blood-serum proteins of the Cypriot *Macrovipera lebetina*, together with its densitometric tracing curve. O.D.: Optical density, S: Start (junction between the spacer and separation gels).

Discussion

Many researchers have underlined the taxonomic importance of the density, speed and fraction numbers obtained from electrophoretic separation of blood-serum proteins of amphibians and reptiles (Dessauer & Fox 1956, Chen 1967, Ferguson 1980, Arıkan et al. 1998). The qualitative differences of fractions could be caused by genetic variations, whilst

quantitative differences could reflect age, gender, environmental and physiological factors (Ferguson 1980); therefore, qualitative differences are important for taxonomic studies. In this study, the obtained data from the electrophoresis of blood serum proteins indicate that *M. lebetina* populations from southern Anatolia and Cyprus show significant differences qualitatively.

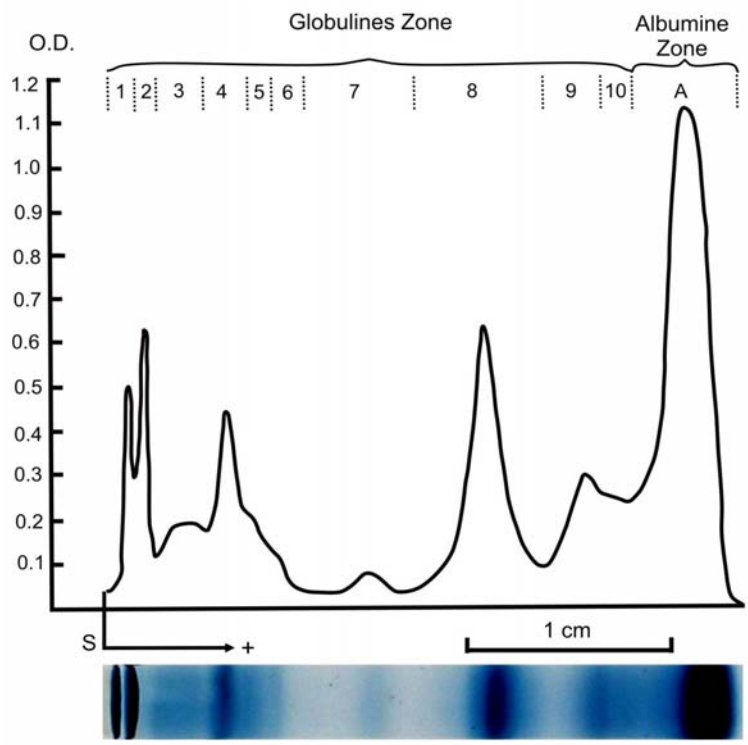

Figure no.2 Representative electropherogram (gel photograph) showing the electrophoretic separation of the blood-serum proteins of the southern Turkish *Macrovipera lebetina*, together with its densitometric tracing curve (For further explanations, see legend to Fig. 1).

Arıkan et al. (2005) and Göçmen et al. (2006) have compared the general morphology, hemipenial morphology and venom protein pattern of Cypriot and southern Turkey population and they concluded that the southern Turkey population should not be included *M. lebetina lebetina.* In this study, we also conclude that these populations are taxonomically distinct in the viewpoint of blood serum proteins. Consequently, the popu-

lation from southern Anatolia should not be allocated to the nominate subspecies *M. l. lebetina*, which lives in Cyprus, as previously suggested by Billing & Schätti (1984), Broodmann (1987), Ossenegg (1989) and Mulder (1995).

In the relevant literature, the population of *M. lebetina* from NE Anatolia has long been referred to as *M. l. obtusa* (Mertens 1952, Eiselt & Baran 1970, Baran 1976, Nilson et al. 1988, Nilson & Andrén 1988, Joger et al. 1997, Atatür & Göçmen 2001, Tok et al. 2002). As suggested by Billing and Schätti (1984), Broodmann (1987), Ossenegg (1989) and Mulder (1995), the population existing in southern Anatolia is distinctly different at the subspecific level from the population in NE Anatolia, which should be resurrected as *M. l. euphratica* Martin, 1838 from the synonym list of the species, since its type locality was given as "shores of Euphrates".

In summary, our findings on blood serum proteins indicate that the southern Turkey population might be quite distinct from the Cypriot population at a subspecific level; and while we were unable to compare with the specimens from northeastern Turkey populations, according to the relevant literature, it is possible to suggest that they also belong to distinctly different subspecies. We thus accept, provisionally, considering the limited number of our specimens for comparison, the subspecific status of the southern Anatolian population as *Macrovipera lebetina* ssp.

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