Arch. Biol. Sci., Belgrade, 61 (4), 719-722, 2009 DOI:10.2298/ABS0904719T

ESTERASE VARIATION IN TURKISH WHITE-TOOTHED SHREWS (CROCIDURA): RECORD OF A TRIMERIC ESTERASE

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Abstract — This study focuses on esterase variation of the genus *Crocidura* in Turkey. A total of 248 white-toothed shrews were analyzed by means of cellulose acetate gel electrophoresis. Liver tissue and alfa naphthyl acetate were used to investigate esterase variation in Turkish white-toothed shrews. A different esterase banding pattern was found in one *Crocidura* individual. This phenotype had four anodally migrated bands on cellulose acetate gel. The *Crocidura* individual displaying the given phenotype was identified as *Crocidura suaveolens*. The different esterase banding pattern observed in this study is considered to be a result of the trimeric structure of esterase in the lesser white-toothed shrew (*Crocidura suaveolens*).

Key words: *Crocidura suaveolens,* variation, esterase, Turkey

UDC 599.363(560):575

INTRODUCTION

Living organisms have a great number of enzymes, which are extremely large biomolecules important for metabolic events and polymorphism in organisms (Bugg, 2004). Enzymes may often possess subunit structures such as monomers, dimers, trimers, and tetramers in animals and other organisms. Of these subunit structures, the trimeric structure has not been extensively reported in vertebrates, including mammals (Richardson et al., 1986; Manchenko, 2003).

The enzyme esterase (EST) (EC 3.1.1.1), known as an important hydrolase enzyme in animals (Bugg, 2004), is polymorphic and generally composed of one or two subunits (Richardson et al., 1986; Hoelzel, 1998; Manchenko, 2003). It was reported by Kendal (1983) that the esterases are characterized by substrate specification, tissue distribution, and electrophoretic mobility. The enzyme esterase was found to be trimeric in some organisms, including man, rat, pig, guinea pig, and the common shrew (Hopkinson et al., 1976; Heymann, 1980; Oehm et al., 1982; Searle, 1986). So far, there are no data available indicating the existence of trimeric forms in white-toothed shrews.

Although esterase variation has been investigated in some rodents of Turkey, there is not much information available on Turkish insectivorous species (Vogel et al., 1986; Fillipucci and Simson, 1996; Macholan, 1996; Verimli et al., 2000; Macholan et al., 2001a, 2001b; İyigün and Çolak, 2004; Çolak et al., 2005; Kankiliç et al., 2005). Vogel et al. (1986) conducted biochemical analysis for some populations of Turkish white-toothed shrews, but they did not examine esterase enzyme in their study.

In the present study, we investigated whether or not esterase variation exists in the Turkish white-toothed shrews.

MATERIAL AND METHODS

A total of 248 *Crocidura* samples collected from Turkey were examined for esterase variations. The samples of *Crocidura* were captured from the field

with mouse traps and were humanely killed in the field and/or the laboratory. Liver tissues were used for enzyme electrophoresis. Such tissues were sampled from freshly killed white-toothed shrews (*Crocidura*). All tissues were washed thoroughly in physiological saline (0.9% NaCl) (Searle, 1986) and frozen on dry ice in the field or at -80°C in the laboratory. Tissues frozen in the field were transferred to -80°C. With these tissues, tissue homogenization, enzyme electrophoresis, and staining for esterase were performed according to the method published by Richardson et al. (1986). The samples of liver tissue were run on cellulose acetate plates (94x76- and 60x76-mm Titan III plates, Helena Lab) in a cooled electrophoresis unit.

RESULTS

Esterase variation of *Crocidura* samples collected from Turkey was investigated by means of cellulose acetate gel electrophoresis, alfa naphthyl acetate being used to stain samples of liver homogenates. All samples migrated anodally on cellulose acetate gels. Of 248 samples, only one, which was collected from Samsun in the Black Sea region of Turkey, displayed a different banding pattern for esterase.

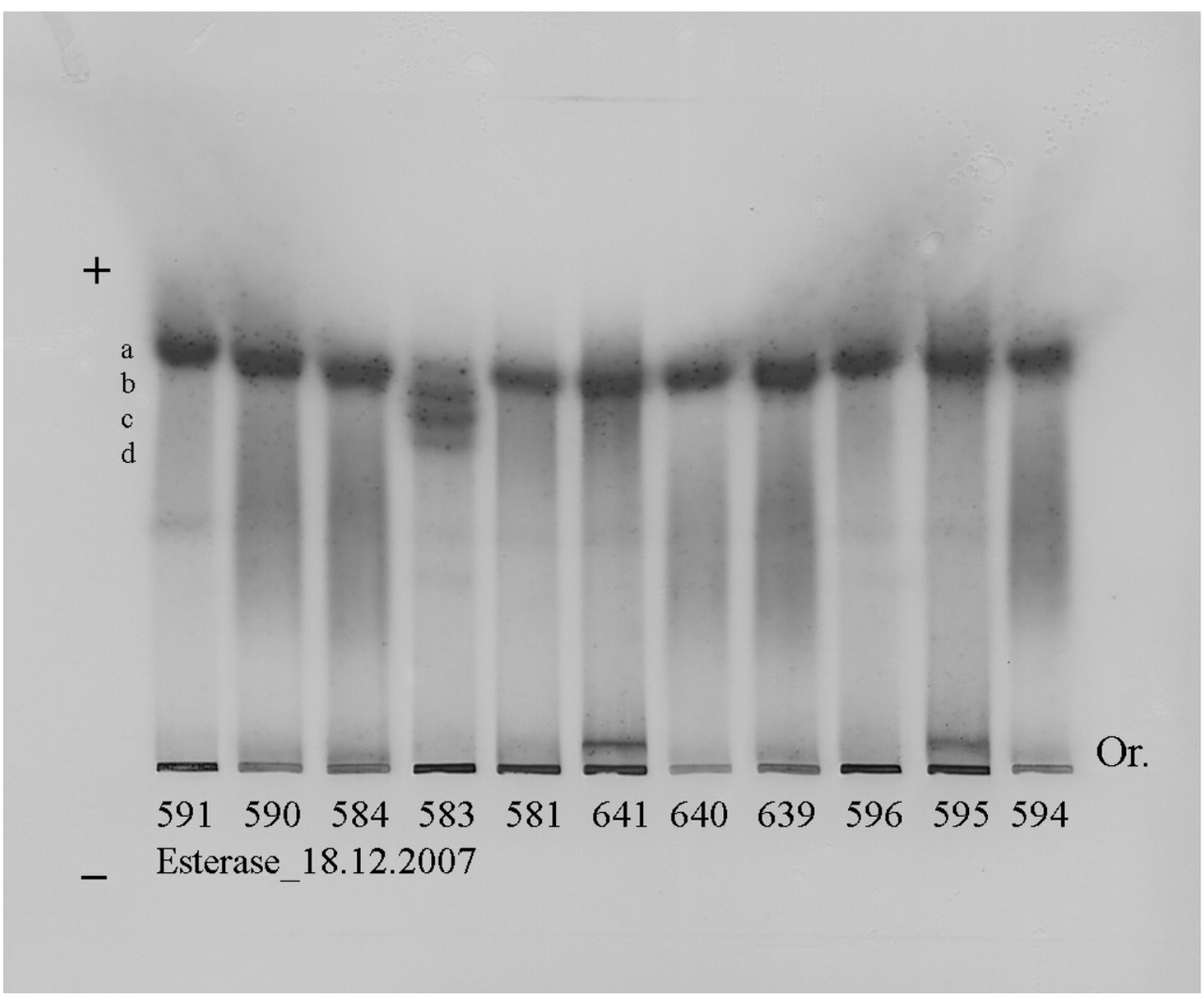

Fig. 1. Photograph of a cellulose acetate gel showing two different phenotypes (single band and four bands) of esterase in Turkish *Crocidura*.

This individual was recognized as *Crocidura suaveolens* according to Tez (2000) and Krystufek and Vohralik (2001). The phenotype observed in *C. suaveolens* was with four bands that migrated anodally on the gel (sample 583, Fig. 1). The remaining 247 *Crocidura* samples screened were observed to have a single band.

DISCUSSION

Studies of enzyme variation are an important tool for investigating genetic variability of world mammal species. In line with this, there are various enzyme studies on soricids as well (Catzeflis et al., 1985; Searle, 1985, 1986; Vogel et al., 1986, 2004; Ruedi et al., 1990; Macholan, 1996; Ruedi, 1996; Wojcik et al., 1996a, 1996b; Driskell and Feldhamer, 2003). There are also some studies reporting enzyme variations in Turkish mammals (Vogel et al., 1986; Filippucci and Simson, 1996; Macholan, 1996; Verimli et al., 2000, Macholan et al., 2001; İyigün and Çolak, 2004; Çolak et al., 2005; Kankiliç et al., 2005; Çolak, 2006; Yiğit et al., 2007). However, there are very few publications on enzyme variation in Turkish shrews (Vogel et al., 1986; Macholan, 1996).

Detailed investigation of esterase variation in mammals has been conducted by various workers (Hunt et al., 1973; Peters, 1982; Kendal, 1983; Verimli et al., 2000; İyigün and Çolak, 2004; Çolak et al., 2005). The most interesting results were obtained in mouse, rat, guinea pig, pig, common shrew, and man (Hopkinson et al., 1976; Heymann, 1980; Oehm et al., 1982; Searle, 1986). The results obtained in these mammals revealed a trimeric esterase.

Mouse, rat, guinea pig, pig, and man are not soricids. However, the common shrew, *Sorex araneus*, is a soricid, as is white-toothed shrew (*Crocidura*). Searle's study (1986) is of interest to us because he reported that the enzyme esterase has six phenotypes in *Sorex araneus*. Searle found that esterase in *Sorex areneus* is trimeric in regard to its subunit numbers.

Heymann (1980) reported a trimeric structure of esterase in human, pig, rat, and guinea pig liver, as well as pig kidney. Searle (1986) used the kidney of common shrews and alfa-naphthyl propionate as substrate. We used the liver of white-toothed shrews and alfa naphthyl acetate. Up to now, a trimeric esterase has not been reported in soricids, except by Searle (1986). In the present study, the quaternary structure of esterase in one *Crocidura* sample identified as *C. suaveolens* (like *Sorex araneus*, a member of the family Soricidae) may be a trimeric structure. This result is in keeping with results on *S. araneus* from Britain reported by Searle (1986). Thus, a trimeric structure arose in different substrates and tissues. As stated by Searle (1986), "this trimeric structure may be an ancient character belonging to mammals".

Acknowledgments - This work was supported by the Scientific and Technical Research Council of Turkey (TÜBİTAK) (Project No. 107T157) and the Research Fund of Erciyes University (Project Nos. E.U.FBA-07-15 and E.U.FBY-07-82).

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