Taxonomical and Karyological Features of *Microtus hartingi* (Barrett-Hamilton, 1903) (Mammalia: Rodentia) with Some Biological and Ecological Features

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Abstract.- This study is based on 27 *Microtus hartingi* (Barrett-Hamilton, 1903) specimens collected from Isparta province between March and September 2010. The specimens were caught by kill and live trapping and they were skinned and stuffed. Some behavioral features of the live sample fed in the laboratory condition were observed. Its karyotypical features were also determined. Specimens were divided into two age groups as juvenile and adult depending on molar wearing, the clearance of the sagittal crest in the skull, pregnancy and nursing status. Diagnostic characters, habitat, fur color, hair morphology, feeding and reproductive behavior, karyology, bacular features in all examined specimen were assessed. It was found that the diploid number (2n), the fundamental number (NF) and the number of autosomal arms (NFa) were 54, 56 and 52, respectively. Consequently, this species known as *Microtus guentheri* previously in Isparta was determined to be actually *Microtus hartingi* on the basis of data presented in this work.

Key words: Chromosome numbers, cytotaxonomy, karyotype, Microtus hartingi, vole.

INTRODUCTION

Rodentia is the largest order of living encompassing 2277 species Mammalia, as recognized herein, or approximately 42% of worldwide mammalian biodiversity (Wilson and Reeder, 2005). The genus Microtus (Rodentia: Cricetidae) consists of 65 extant species, making it one of the Rodentia genera with the highest number of species (Lemskaya et al., 2010). The genus *Microtus* is represented with nine species in Turkey; Microtus anatolicus, M. arvalis, M. daghestanicus, M. dogramacii, M. guentheri, M. levis, M. majori, M. socialis and M. subterraneus (Çolak et al., 1997, Kefelioğlu and Kryštufek, 1999; Kryštufek and Kefelioğlu, 2001; Yiğit and Çolak, 2002; Musser and Carleton, 2005; Mitsainas et al., 2010).

There are different systematic reshufflings within *Microtus* genus. Blackler (1916) described as *Microtus lydius* specimens morphologically different from *Microtus guentheri* and collected from İzmir. Ellerman and Morrison-Scott (1951) regarded *M. lydius* as a subspecies of *M. guentheri*. In contrast, Corbet (1978) and Harrison and Bates (1991) considered *M. guentheri* to be a synonym of *Microtus socialis*. Corbet (1978) also regarded *M. lydius* as a subspecies of *M. socialis*. Apart from these taxonomic remarks, *M. guentheri* was recorded from various localities in Anatolia by Neuhäuser (1936), Ellerman (1948), Osborn (1962), Lehmann (1966), Steiner and Vauk (1966), Çağlar (1967), Lay (1967), Felten *et al.* (1971), Kock *et al.* (1972), Morlok (1978), Kefelioğlu (1995), Çolak *et al.* (1997), and Kefelioğlu and Kryštufek (1999).

Since the primary food sources of rodents are wild plants and agricultural products, they are regarded as agricultural pests. Chemical substances used for controlling the species in agricultural areas are known to be harmful to birds and mammals (Rustamani *et al.*, 2005). Kumerloeve (1980) noted high reproductive potential of the species belonging to the subfamily Microtina, and stated that its populations fluctuate depending on the availability of food. Some types of rodents are known as carries of plague, typhoid, typhus and tularemia (Corbet and Southern, 1977).

Numerous karyological surveys have been done on the genus *Microtus*, enhancing the available cytogenetic and karyological data (Fredga and Bergström, 1970; Mazurok *et al.*, 2001; Şekeroğlu *et al.*, 2011; Yavuz *et al.*, 2011; Albayrak *et al.*,

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2012; Yiğit *et al.*, 2012; Azkur *et al.*, 2013; Zima *et al.*, 2013; Arslan and Zima, 2014). However, the phylogenetic relationships among *Microtus* and its closest relatives are uncertain, and difficulties remain both in delimiting species and defining subgenera (Zagorodnyuk, 1990; Musser and Carleton, 1993; Jaarola *et al.*, 2004). The most common chromosome number of the genus is vary between 2n = 17 and 62, one of the highest rates of karyotypic change in mammals (Maryama and Imai, 1981; Zima and Král, 1984; Modi, 1987; Zagorodnyuk, 1990; Jaarola *et al.*, 2004).

The aim of this study is to identify biological, karyological and taxonomical characteristics, to assess the economic importance and to contribute to resolving systematic problems of *Microtus hartingi* (Barrett-Hamilton, 1903) having taxonomic conflict.

MATERIALS AND METHODS

This study was based on 27 *Microtus hartingi* specimens collected from field works between March and September 2010 in Isparta (Fig. 1).



Fig. 1. Geographic locations of specimens studied.

Specimens were captured by using live trap and Sherman traps in agricultural fields, roadsides, meadows and fruit gardens. After taking its weights and the standard external measurements according to Thomas (1905), the specimens were skinned, stuffed and prepared as conventional museum type sample.

Peanuts, beans and cheese were provided to one of the samples for determining their food preferences for 4 months (Fig. 2).



Fig. 2. The specimen of *Microtus hartingi* in laboratory.

Specimens were divided into two age groups as juvenile and adult based on sagittal crest of the skull, degree of teeth wear, fur color and some reproductive features (embryos, pregnancy and lactation). The significance test of differences between the means of males and females was made according to Parker (1979) and the results were given in the Table I. All the samples were deposited in mammalian collection of the Department of Biology, Faculty of Science, Süleyman Demirel University.

The diagnostic characters, habitats, some feeding and breeding behaviors, karyotype, hair morphology, fur color of specimens examined and collection localities were recorded. Fur color and guard hair samples were defined and prepared according to Ridgway (1886) and Day (1966), respectively. In addition, bacula and karyology of the specimens were analyzed according to Lidicker (1968), Ford and Hamerton (1956) and Patton (1967). Seven slides were prepared and least 10 metaphases were analyzed and photographed. The diploid number (2n), fundamental number (NF), and fundamental autosomal number (NFa) along with the shape of autosomes and sex chromosomes were also determined.

RESULTS

Karyological and habitat features of *Microtus hartingi* were determined in this study. In addition, the external and cranial measurements and weights 27 adult individuals were presented in detail.

Diagnostic characters

In adults: tail length 21-28, condylobasal length 27.3-29.9; zygomatic width 16.0-17.7; skull height 11.1-12.2; mandible length 16.7-18.7; baculum length 2.54-3.15; baculum proximal width 1.05-1.55, and baculum distal width 0.32-0.49 mm. Head and body length is up to about 19.2 mm to 23.5% of tail length. General color was light brown with grayish yellow tint.

Habitat

It was determined that the species, very sensitive to heat and thirst has lived in regions up to 940-1250 m above sea level. The animals were frequently observed in agricultural areas (especially wheat fields), fruit gardens and roadsides. They form colonies in steppe areas with herbaceous wild grasses (Poaceae), plains, fallow or unimproved agricultural lands. However, it was observed that the species withdraw to surroundings of these habitats if there is habitat destruction or famine (Fig. 3). *Microtus hartingi* was frequently encountered in apple gardens but not observe in cherry gardens.

Nests

They often live in tunnels in soil dug by them. Nests are mostly located around field boundaries. M. hartingi built a nest from weeds when the soil was not enough. In laboratory, we observed that it still had not dig for a few days after addition of more soil in territoryum. However, disturbed individual dug the soil and hidden. Then, it carried the herbaceous plants to put in the cage in rooms. Moreover, we observed a lot of nests particularly in fallow fields during field works. Some of these nests had tilted; others had perpendicular entrance-exit holes and only one entrance hole. The plants around entrances were generally eaten. Therefore, there were bare areas at least 1 cm in diameter around entrances. On the other hand, individuals might hide the entrances

with remaining crop straws in plowed fields. In field and laboratory, we observed that stool was released into the nest and then thrown out of the nest in batches.



Fig.3. Habitats of *Microtus hartingi*; rural area (a) and cultivated area (b)

Fur color

The dorsal color of adults was light brown with light grayish yellow tint, and ventral color was very light gray with yellowish white tint. Dorsal hairs were in pale smoky black tones until the last portion. The last portion, starting with yellowish brown, ends with two brownish black bands. Dorsal of the tail was the same color of dorsal of the body while the ventral color was clear pale yellowish brown. Dorso-medial region of the body was relatively darker than dorso-lateral color longitudinally. Dorsal color mixed with ventral color around the abdominal region without a distinctive line.

Hair morphology

Guard hairs of the body of the species were found to be annular type (Fig. 4).



Fig. 4. Morphology of annular type hair of *Microtus hartingi*.

Feeding behavior

M. hartingi typically feeds on seeds of crop products by cutting stalks and exposing seeds in the study area. Their major nutrients are grains like wheat and barley, as well as roots and stems of herbaceous. The individual in laboratory was given to eat barley, wheat, sunflower seeds, walnuts, peanuts, almonds together with a variety of herbaceous plants and cheese. It pulled the wheat stalks into its nest and consumed them using its front limbs to hold it. There was no vegetation around the areas where the nests were dense.



Fig. 5. Embryos of Microtus hartingi.

Reproductive features

In April, two individuals weighting 50 **gr** and 55 **gr** carried 8 embryos were caught in Gönen and Keçiborlu, respectively. In May, a 68 **gr** female caught in Kayı village had 8 embryos, another one weighting 52,5 **gr** had 6 embryos. A 69 **gr** female caught in Keçiborlu had 5 embryos and another female caught in Atabey district weighting 72 **gr** had 7 embryos (Fig. 5). It was observed that females have 4 nipples and males have a prominent scrotum.

Karyological features

M. hartingi specimens were karyologically analyzed. We determined that chromosome number of the species was 2n=54, the fundamental number 56 and the number of autosomal arms 52. Also, it was found that the species had 26 pairs of acrocentric autosomes aligned from the largest to the smallest in chromosome set. The X chromosome was large acrocentric as well as the Y chromosome small acrocentric (Fig. 6).



Fig. 6. Metaphase chromosomes (A) and karyogram (B) of \bigcirc *Microtus hartingi*.

Measurements

Some measurements of 27 adult (14 $\Im \Im$, 13 $\Im \Im$) *Microtus hartingi* were presented in Table I.

Baculum features

Proximal part of the baculum was onion shaped and constituted the widest part of baculum. Middle part of the baculum was relatively thinner. The distal part ends with forming a slight knob. Three finger shaped extensions in distal part resemble candles on a candlestick (Fig. 7). Baculum sizes of 12 adult males were presented in Table II.

Table I	External and cranial measurements, and
	weights of 27 adults Microtus hartingi (14 3, 13
	\bigcirc ; the number of specimens (n), range (r),
	mean (m) and standard deviation (± sd)

Measurements	n	R	Mean±SD
Total length	4	130-154	145.5±10.72
Head and body length	4	109-126	119.5±7.59
Hindfoot length	27	19-24	21±1.24
Ear length	4	12-13	12.7±0.5
Weight	27	28.5-72.0	49.5±11.24
Condylobasal length	22	27.3-29.9	28.5±0.79
Occipitonasal length	21	27.7-30.3	28.8±0.77
Basilar length	22	24.0-26.7	25.3±0.74
Palatal length	23	13.0-15.5	14.3±0.60
Palatal foramina length	23	4.5-5.5	5.0±0.30
Diastema length	23	8.2-9.3	8.7±0.34
Nasal length	22	7.6-8.7	8.2±0.34
Zygomatik width	16	16.0-17.7	16.9±0.45
Interorbital width	22	3.5-3.9	3.7±0.11
Skull height	19	11.1-12.2	11.5±0.32
Maxillary toothrow length	23	5.9-7.0	6.4±0.26
Mandibulary toothrow length	23	5.9-7.1	6.5±0.28
Mandible length	23	16.7-18.7	17.7±0.53
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Fig. 7. View of baculum (a, dorsal; b, ventral; c, lateral).

Specimen examined and its collection localities

Isparta: Atabey, Gönen road, 3 (1 \circlearrowright , 7 April 2010; 1 \circlearrowright , 22 May 2010, 1 \heartsuit , 23 May 2010; Eğirdir, Sevinçbey village, 1 (1 \circlearrowright , 29 May 2010), Bademli village, 1 (1 \circlearrowright , 16 June 2010); Gelendost, 3 (3 \circlearrowright , 26 July 2010); Senirce town near Gönen, 7 (3 \heartsuit , 7 April 2010; 1 \circlearrowright , 3 \heartsuit , 18 April 2010); Keçiborlu Dinar crossroads, 7 (3 \circlearrowright , 1 \heartsuit , 18 April 2010; 1 \circlearrowright , 1 \heartsuit , 12 May 2010, 1 \heartsuit , 31 May 2010); Kuleönü town, 2 (1 \circlearrowright , 1 \heartsuit , 4 April 2010), Kayı village 3 (2 \heartsuit , 1 May 2010, 1 \circlearrowright , 9 August 2010).

Table II.- Bacular measurements (mm) of *Microtus hartingi* (ろう); number of samples (n), range (r), mean (m) and standard deviation (±sd)

n	r	m± sd
11	3.58-4.68	4.18±0.26
12	2.54-3.15	2.82 ± 0.14
11	0.32-0.49	0.42±0.05
12	1.05-1.55	1.33±0.17
	n 11 12 11 12	n r 11 3.58-4.68 12 2.54-3.15 11 0.32-0.49 12 1.05-1.55

DISCUSSION AND CONCLUSION

We determined that the species mostly live forming colonies in steppe areas with herbaceous wild grasses, plains, fallow or unimproved agricultural lands. They were frequently encountered in apple gardens but not observe in cherry gardens. Colak et al. (1998) observed that the species live in colonies in steppes, plains, fallowed or tilled agricultural lands, around these lands as well as fruit gardens. Yavuz et al. (2008) reported that they encountered the species in agricultural lands and roadsides around these lands. Our findings are consistent with these authors.

According to our findings, external and cranial measurements of the species were completely not suitable for the original definition of *M. guentheri*. Internal and external characteristics of morphometric data of *M. guentheri* were very similar as compared with Neithammer (1978) and Yavuz *et al.* (2008) except for the number of offspring (5-8).

Internal and external measurements of *M. hartingi* were consistent with those Felten *et al.* (1971), Kefelioğlu (1995) and Sözen *et al.* (1999).

We determined that head, body and tail length of our specimens were larger than M. *hartingi*, and also its hind foot, ear and condylobasal length were smaller as compared to Çağlar (1967). In addition; cephalothorax (head and body), tail and ear length were shorter, but hind leg length was nearly the same with Çolak *et al.* (1997). External measurements of Yiğit and Çolak (2002) were comprised the upper limits of our measurements, as well.

Diploid chromosome count of the samples collected from Isparta was 2n=54 as determined by Niethammer (1978), Çolak *et al.* (1997), Yiğit and

Colak (2002) and Chassovnikarova et al. (2008). Number of fundamental chromosome arms was determined 56. This count was different from Niethammer (1978). In addition, number of autosomal chromosome arms was found 52. This data was similar to that of Colak et al. (1998) and Yiğit and Çolak (2002). There were 26 pairs of acrocentric autosomal chromosomes arranged from largest to smallest. X chromosome was the largest and Y chromosome was the smallest acrocentric. However, Colak et al. (1997) stated that X chromosome was medium sized acrocentric and Chassovnikarova et al. (2008) identified that one of the X chromosome pairs of females was submetacentric and the other one was acrocentric, and in males X chromosome was submetacentric.

Gözütok (2010) concluded that *M. guentheri* was different from *M. hartingi* with regard to baculum, morphometric measurements and fur color in a study on *Microtos* genus involving Isparta province. Comparing the literatures information with available data during our studies, we have decided that our samples were *M. hartingi*.

In this study, we determined that fundamental chromosome arm count and morphometric measurements are not consistent with *M. guentheri*. This data however, correspond well with *M. hartingi* recorded in Greece by Ognev (1964). *Microtus hartingi* is therefore being recorded for the first time in Isparta province. This study will help resolve taxonomic problems ongoing for a century within *Microtus* genus.

ACKNOWLEDGEMENTS

The authors are grateful to the Department of Scientific Research Project Management of Süleyman Demirel University for the financial support by the project SDUBAP (2487). They are also grateful to Dr. Serdar Gözütok (Bolu Abant İzzet Baysal University) for helping in field and laboratory studies.

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(Received 13 February 2015, revised 24 March 2015)