

## A Study on *Mus domesticus* Ruddy, 1772 and *Mus macedonicus* Petrov and Ruzić, 1983 (Mammalia: Rodentia) Distributed along the Line of Ankara, Bolu and Zonguldak\*

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**Abstract:** A total of 105 specimens of the genus *Mus* collected from Ankara, Bartın, Bolu, Düzce and Zonguldak in Turkey were biometrically evaluated. The isocitrate dehydrogenase enzyme system along with karyotypes of 37 specimens were analysed. The zygomatic index and the index of head plus body length/tail length were diagnostic characters for *Mus domesticus* and *Mus macedonicus*. Starch gel electrophoresis gave 2 loci (*Idh-1* and *Idh-2*) for the isocitrate dehydrogenase enzyme system. Two alleles (*Idh-1*<sup>100</sup> and *Idh-1*<sup>105</sup>) of the *Idh-1* locus were determined in *M. domesticus* and *M. macedonicus*. *M. domesticus* and *M. macedonicus* have the same karyological values  $2n = 40$ ,  $NFa = 38$  and  $NF = 40$ .

**Key Words:** *Mus*, Isocitrate dehydrogenase, Karyotype, Morphometry, Turkey

### Ankara, Bolu ve Zonguldak Hattında Yayılış Gösteren *Mus domesticus* Ruddy, 1772 ve *Mus macedonicus* Petrov ve Ruzić, 1983 (Mammalia: Rodentia) Üzerine Bir Çalışma

**Özet:** Ankara, Batın, Bolu, Düzce ve Zonguldak'tan toplanan 105 *Mus* örneği değerlendirildi. Bu örneklerden 37 tanesinin izositrat dehidrogenaz enzim sistemi özellikleri ve karyotipleri analiz edildi. Zygomatik indeksin (ZI), baş ve beden uzunluğu/kuyruk uzunluğu (H+B/T) indeksi *Mus domesticus* ve *Mus macedonicus* için ayırıcı özellikte olduğu saptandı. Nişasta jel elektroforezi izositrat dehidrogenaz enzim sisteminde iki lokusu (*Idh-1* ve *Idh-2*) ortaya çıkardı. Hem *M. domesticus* hem de *M. macedonicus*'ta *Idh-1* lokusunda iki alel (*Idh-1*<sup>100</sup> ve *Idh-1*<sup>105</sup>) tespit edildi. *M. domesticus* ve *M. macedonicus*  $2n = 40$ ,  $NFa = 38$  ve  $NF = 40$  değerlerine sahiptir.

**Anahtar Sözcükler:** *Mus*, İzositrat dehidrogenaz, Karyotip, Morfometri, Türkiye

### Introduction

Molecular genetics allows for the identification of *M. musculus*, *M. spretus*, *M. abbotti* and *M. spicilegus* in Europe (Bonhomme et al., 1984; Bonhomme et al., 1989; Britton-Davidian, 1990; Frisman et al., 1990). Morphological criteria were recorded by Marshall and Sage (1981), Orsini et al. (1983), Marshall (1986), Auffray et al. (1990), Macholán and Zima (1994), Macholán (1996) and Mezhzherin et al. (1998) for the recognition of *Mus* species. In Turkey, *Mus musculus musculus* L., 1758; *Mus musculus domesticus* Ruddy, 1772; *Mus musculus praetextus* Brants, 1827; *Mus musculus brevisrostris* Waterhouse, 1837; *Mus musculus wagneri* Eversmann, 1848, and *Mus musculus spicilegus* Petenyi, 1882 were determined (Schwarz and Schwartz,

1943; Ellerman, 1948; Ellerman and Morrison-Scott, 1951; Hoogstraal, 1959; Stainer and Vauk, 1966). Petrov and Ruzić (1983) described *Mus macedonicus* from Yugoslavia, and Harrison and Bates (1991) and Musser and Carleton (1993) included Turkey in the distribution area of *M. macedonicus*. Kryštufek and Macholán (1998) recorded *M. macedonicus* from Burdur, Konya and Manisa. According to Boursot et al. (1993), *M. domesticus* lives in Turkey. Auffray et al. (1990) recorded zygomatic index and head plus body length/tail as discriminatory parameters for *M. m. domesticus* and *M. "spretoides"* in Israel.

After analysing many loci, Thaler et al. (1981) and Mezhzherin et al. (1998) stated that the isocitrate dehydrogenase (*Idh-1*) locus separated *M. domesticus*

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and *M. musculus* in Europe and the Caucasus, respectively. Bonhomme et al. (1983) described carbonic anhydrase (Ca) as the diagnostic locus for *M. m. domesticus* in Eastern Europe. The studies mentioned above showed that there are no isozymic and morphometric studies on the genus *Mus* in Turkey. However, the karyotype was described by Gündüz et al. (2000) for *M. m. domesticus* and *M. macedonicus* in Turkey.

The aim of this study is to contribute to the taxonomy, distribution, karyology and morphometry of *M. domesticus* and *M. macedonicus* in Turkey.

### Materials and Methods

This study is based on 142 specimens collected from Ankara, Bartın, Bolu, Düzce and Zonguldak in Turkey (Table 1). In order to determine variations in *Mus* species, we established an area extending from Central Anatolia, with a dry and cold climate, to Zonguldak in the western Black Sea region with rainy and humid climate.

The isocitrate dehydrogenase enzyme system was electrophoresed from 37 specimens. *Idh-1* and *Idh-2* loci were analysed from muscle, and were kept in a deep freeze (-80 °C). Starch gels and buffers were prepared in accordance with the method described by Selander et al. (1971) and Hillis and Moritz (1990). The staining procedure followed the method described by Ayala et al. (1972). Twenty-one specimens were karyotyped using the method described by Ford and Hamerton (1956). Identifications of all specimens were based on distinguishing characters recorded by Marshall and Sage (1981), Orsini et al. (1983), Marshall (1986), Auffray et

al. (1990), Macholán and Zima (1994), Macholán (1996) and Mezhzherin et al. (1998). ZI (width of malar process anterior part/ width of upper part of zygomatic arch) and H+B/T (the index of head length plus tail length/tail length) were determined on skulls. Measurements of 105 adult specimens were used for biometric analysis. Twenty-three measurements (mm) and 2 ratios were taken (to the nearest 0.01 mm) from the skull of each specimen. Five populations belonging to the 2 species were analysed using NTSYS-II (Version 1.80, Rohlf, 1994). A set of 23 cranial and 6 body characters along with 2 ratios (Table 2) were scored and used to generate phylogenetic trees: total length (TL), head and body length (H+B), tail length (T), hind foot (HF), ear (E), weight (W), zygomatic breadth (ZB), interorbital constriction (IC), condylobasal length (CBL), occipito-nasal length (ONL), basilar length (BL), nasal length (NL), nasal width (NW), facial length of braincase (FLB), braincase length (BCL), mastoid breadth (MB), height of braincase with tympanic bulla (HBB), height of braincase without tympanic bulla (HB), occipital width (OW), braincase width (BW), diastema (D), palatal length (PL), foramina incisive length (FI), tympanic bulla length (TB), mandible Length (ML), length of upper toothrow alveoli (LUTa), length of upper toothrow crown (LUT), length of lower toothrow alveoli (LLTa), length of lower toothrow crown (LLT), width of malar process of anterior part/width of upper part of the zygomatic arch (ZI), and H+B/T.

### Results

**Distribution:** *M. macedonicus* was recorded from Ankara and Bolu, and *M. domesticus* from Ankara, Bolu, Bartın and Zonguldak (Figure 1).

Table 1. Localities and numbers of specimens examined.

Species	Morphometric Evaluation	Electrophoretic Evaluation
<i>M. domesticus</i>	Ankara: Center 16, Gölbaşı 3, Sarayköy 7, Bala 11, Bartın 11, Bolu: Abant 10, Düzce 5, Zonguldak 11	Ankara 5, Bartın 4, Bolu 7, Düzce 2, Zonguldak 11
<i>M. macedonicus</i>	Ankara: Bala 1, Gölbaşı 7, 30 km West 5, Sarayköy 6, Şereflikoçhisar 6, Bolu 6	Ankara 4, Bolu 4

Table 2. Measurements (mm) of cranial and body characters along with ZI and H+B/T indexes of *M. domesticus* and *M. macedonicus*.

Characters	Mus domesticus										Mus macedonicus														
	Ankara					Bolu					Zonguldak-Bartın					Ankara					Bolu				
	Mean	Min	Max	SD	Mean	Min	Max	SD	Mean	Min	Max	SD	Mean	Min	Max	SD	Mean	Min	Max	SD	Mean	Min	Max	SD	
TL	167.7	124	210	16	156.7	132	187	12.8	156	130	176	15.1	145.6	109	174	18.3	143.8	115	178	23.7					
H+B	80.7	61	120	9.7	78.3	61	92	8.2	73.5	58	86	9.5	80.4	57	98	9.7	70.8	59	79	7.1					
T	84.7	58	105	8.6	78.4	69	95	6.5	82.4	72	96	6.3	65.3	35	85	10.6	73	56	100	17.8					
HF	18.3	15	22	1.25	16.9	12	20	0.5	17.4	16	19	1.03	17.4	13	20	1.8	17.4	15	20	1.5					
E	14.7	11	17	1.37	13.2	10	19	1.9	13.9	11	17	1.5	13.8	12	16	1.16	14	13	17	1.3					
W (g)	14.5	8.5	25	3.5	12.7	7.5	24	3.4	12.7	6	19	3.9	14.8	9	22	3.76	11.4	4	18	5.3					
H+B/T	0.95	0.73	1.0	0.1	0.9	0.78	1.0	0.09	0.88	0.78	1	0.07	1.19	0.94	1.46	0.1	1	0.96	1.2	0.1					
ZI	0.36	0.25	0.45	0.07	0.3	0.25	0.46	0.07	0.38	0.3	0.46	0.2	0.63	0.5	0.83	0.08	0.66	0.6	0.8	0.06					
ZB	10.8	9.4	12.2	0.5	10.4	9.4	12.4	0.63	10.4	9	11.4	0.6	11.2	10	12.4	0.5	11	10.2	11.7	0.4					
IC	3.8	3.5	4.1	0.1	3.8	3.6	4	0.1	3.8	3.7	4	0.07	3.84	3.6	4	0.09	3.7	3.5	3.9	0.1					
CBL	20	17	22.7	1.08	19.2	17.2	23	1.2	19.3	16.6	20.9	1.1	29.3	18.8	21.6	0.7	19.7	18.4	20.9	0.9					
ONL	21.4	18.8	23.9	1.01	20.2	18	24	1.1	20.7	19	21.9	0.7	22	20.5	23.8	0.9	21.1	20.2	22.3	0.6					
BL	18.2	15.5	21.6	1.13	17.8	16	21.6	1.1	17.9	15.5	19.8	1.09	18.6	17.2	20.3	0.7	18	16.9	19.2	0.7					
NL	7.7	6.6	8.6	0.4	7.4	6.6	9	0.4	7.5	6.6	8.5	0.5	8.1	7.2	9.2	0.5	7.5	6.8	8.2	0.4					
NW	2.6	2.2	3.3	0.2	2.4	1.7	2.7	0.2	2.46	2.1	2.7	0.1	2.7	2.2	3	0.1	2.14	2.1	2.2	0.04					
FUB	10.2	8.9	11.7	0.5	9.8	9	11.2	0.5	10	9	11.2	0.5	10.8	10	11.9	0.4	10.2	9.9	10.6	0.2					
BCL	11.2	10	12.7	0.5	10.7	9.6	12.4	0.5	10.9	9.9	11.7	0.5	11.2	10.1	12	0.4	10.5	10	11.2	0.3					
MB	5.8	5.1	6.7	0.2	5.8	5.4	6.6	0.3	5.7	5.5	6.1	0.2	5.6	5.2	6.3	0.2	5.5	5.1	5.9	0.2					
HBB	7.6	6.8	8.2	0.2	7.2	6.6	8	0.2	7.1	6.6	7.7	0.2	7.4	7	7.8	0.2	7.5	7.2	7.9	0.2					
HB	6.5	5.6	7.2	0.3	6.3	5	7	0.3	6.3	5.8	7.4	0.4	6.5	6.3	7	0.1	6.7	6.5	7	0.1					
OW	9.3	8.4	10.1	0.3	9.1	8.3	9.9	0.3	9	8.2	9.5	0.3	9.1	8.3	9.9	0.3	9	8.5	9.4	0.3					
BW	9.9	9.3	10.8	0.2	9.7	9	10.3	0.2	9.8	9.3	10.1	0.2	10.1	9.9	10.5	0.2	9.9	9.7	10.1	0.3					
D	5.3	4.5	6.6	0.4	5.1	4.4	5.6	0.3	5.4	5.0	6.1	0.3	5.73	5	6.6	0.3	5.1	4.5	5.6	0.4					
PL	9.2	8.1	11.2	0.5	8.6	7.7	9.9	0.5	8.9	8.3	9.9	0.4	9.6	9	10.2	0.3	9	8.3	9.5	0.1					
FI	4.9	3.26	5.2	0.3	4.8	4.3	5.6	0.3	4.9	4.73	5.2	0.2	5.1	4.73	5.78	0.3	4.84	4.73	5.2	0.1					
TB	3.7	3.5	4.3	0.1	3.7	3.5	4.1	0.1	3.6	3.15	4	0.2	3.8	3.5	4.3	0.1	3.9	3.7	4	0.3					
ML	11.7	10.4	13.5	0.1	11.3	10.1	13.3	0.6	11.7	10.8	12.2	0.4	12.2	11.6	13	0.5	11.2	10.6	11.7	0.09					
LU <sup>Ta</sup>	3.4	2.8	4	0.2	3.4	2.8	3.9	0.2	3.4	3.15	3.7	0.1	3.6	3.15	4.3	0.5	3.5	3.41	3.7	0.09					
LUT	2.8	2.63	3.5	0.1	2.8	2.63	3.15	0.1	2.7	2.63	2.8	0.1	3.03	2.63	3.15	0.1	3.1	2.89	3.4	0.1					
LL <sup>Ta</sup>	3.0	2.63	3.5	0.1	2.9	2.63	3.5	0.2	2.8	2.63	3.1	0.1	3.19	2.88	3.7	0.2	3.05	2.89	3.4	0.1					
LLT	2.8	2.6	3.5	0.1	2.8	2.63	3.5	0.1	2.7	2.36	3.1	0.2	3.02	2.63	3.5	0.1	2.93	2.8	3.1	0.1					

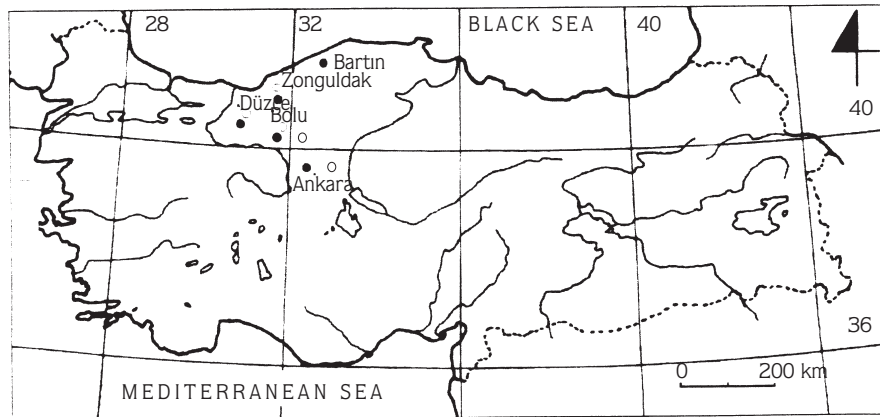


Figure 1. Map showing Turkish sampling localities of *Mus domesticus* (●) and *Mus macedonicus* (○).

**Morphometry:** ZI and H+B/TL distinguished the 2 species. According to measurements of 29 characters along with ZI and H+B/TL (Table 2), *M. domesticus* is divided into 2 subgroups; the first group contains the Bolu and Zonguldak populations, and the second group consists of the Ankara population. *M. macedonicus* is divided into the Ankara and Bolu populations (Figure 2). ZI and H+B/TL seem to be discriminative between the 2 species. On the ZI and H+B/TL parameters *M. domesticus* and *M. macedonicus* were perfectly separated (Figure 2,3).

### Isozymes

**Isocitrate dehydrogenase 1 (*Idh-1*):** We determined 2 alleles of the *Idh-1* locus in 37 specimens of the genus *Mus*. The slow common allele and fast allele were denoted as 100 (*Idh-1*<sup>100</sup>) and *Idh-1*<sup>105</sup>, respectively. There was *Idh-1*<sup>105</sup> in 16 out of 29 specimens collected from 5 localities and *Idh-1*<sup>100</sup> in 13 specimens of *M. domesticus*. Two out of 8 specimens of *M. macedonicus* captured from 2 localities had *Idh-1*<sup>105</sup> and 6 had *Idh-1*<sup>100</sup>. All specimens belonging to 2 species were homozygote for

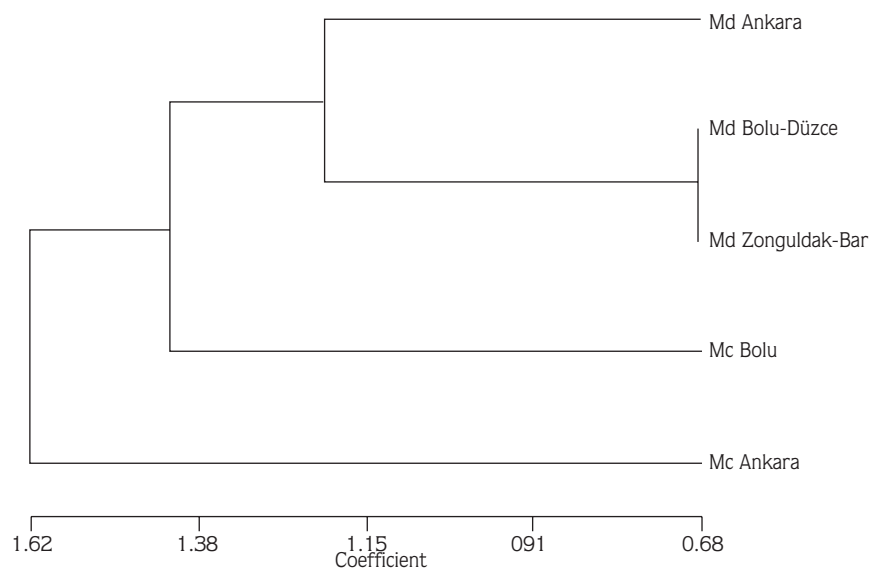


Figure 2. UPGMA dendrogram of 5 populations of *M. domesticus* (Md) and *M. macedonicus* (Mc) based on measurements of 31 characters, using NTSYS-*pc*.

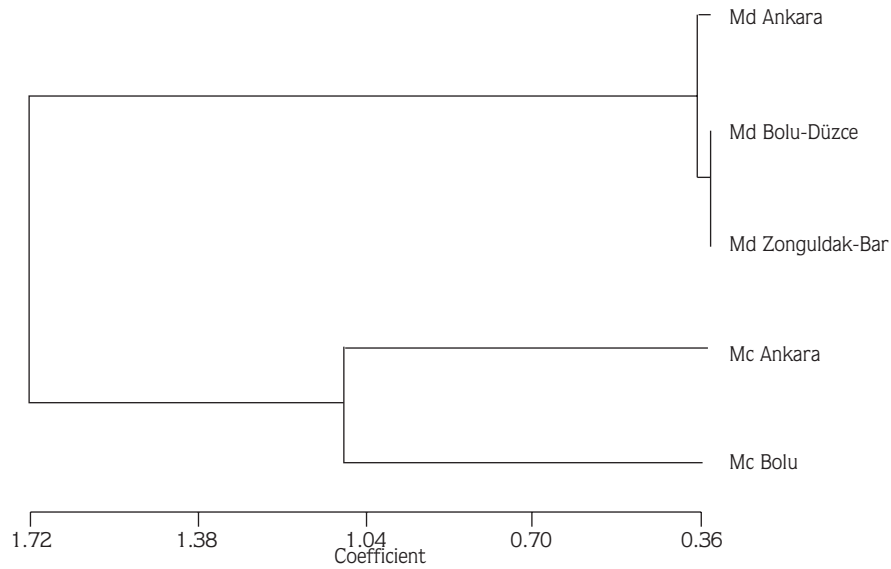


Figure 3. UPGMA dendrogram of 5 populations of *M. domesticus* (Md) and *M. macedonicus* (Mc) based on ZI and H+B/TL, using NTSYS-*pc*.

both loci. According to the comparison of populations of both species, the Ankara population of *M. domesticus* has the highest frequency (0.80) of *Idh*<sup>105</sup>, the Düzce specimens (n = 2) of *M. domesticus* have only the *Idh*<sup>100</sup> allele, and the Ankara and Bolu populations have the highest frequency (0.75) of common alleles (Table 3).

**Isocitrate dehydrogenase 2 (*Idh-2*):** This locus is a mitochondrial form of isocitrate dehydrogenase; it remained in the origin, and fixed for the same allele in both species.

**Karyology:** *M. domesticus* and *M. macedonicus* have the same karyotypic values. The diploid number of chromosomes (2n) is 40, the number of autosomal chromosomes (NFa) is 38, and the fundamental number of chromosomes (NF) is 40. The autosomal set contains 19 pairs of acrocentrics. The X and the Y chromosome are acrocentric, and of the same size (Figure 4 and 5).

## Discussion

We compared the ZI values given by Orsini et al. (1983) and Auffray et al. (1990) for some *Mus* species with those determined in the present study (Table 4). The ZI values given by Orsini et al. (1983) for *M. m. domesticus*, *M. spicilegus*, and *M. m. musculus* are different from those in this study. Our ZI values ranged from 0.25 to 0.46 in 3 populations of *M. domesticus*, and it varied from 0.63 to 0.83 in the 2 populations of *M. macedonicus*. The ZI value for *M. domesticus* in this study was smaller than that of *M. m. domesticus* from Israel, but similar to that of *M. m. domesticus* from Bulgaria and Greece. Harrison and Bates (1991) stated that the anterior part of the molar process in *M. musculus* is narrower than the upper part of the zygomatic arches. This description is consistent with Turkish *M. domesticus* specimens. Furthermore, the ZI pattern described by Harrison and Bates (1991) for *M.*

Table 3. Allele frequencies of the *Idh-1* locus in *Mus domesticus* (Md) and *Mus macedonicus* (Mc). n = number of specimens.

Species	Alleles	Ankara (Md; n = 5, Mc; n = 4)	Bolu (Md; n = 7, Mc; n = 4)	Düzce (n = 2)	Zonguldak (n = 11)	Bartın (n = 4)
<i>Mus domesticus</i>	<i>Idh</i> <sup>100</sup>	0.20	0.34	1.00	0.45	0.75
	<i>Idh</i> <sup>105</sup>	0.80	0.66	-	0.55	0.25
<i>Mus macedonicus</i>	<i>Idh</i> <sup>100</sup>	0.75	0.75			
	<i>Idh</i> <sup>105</sup>	0.25	0.25			

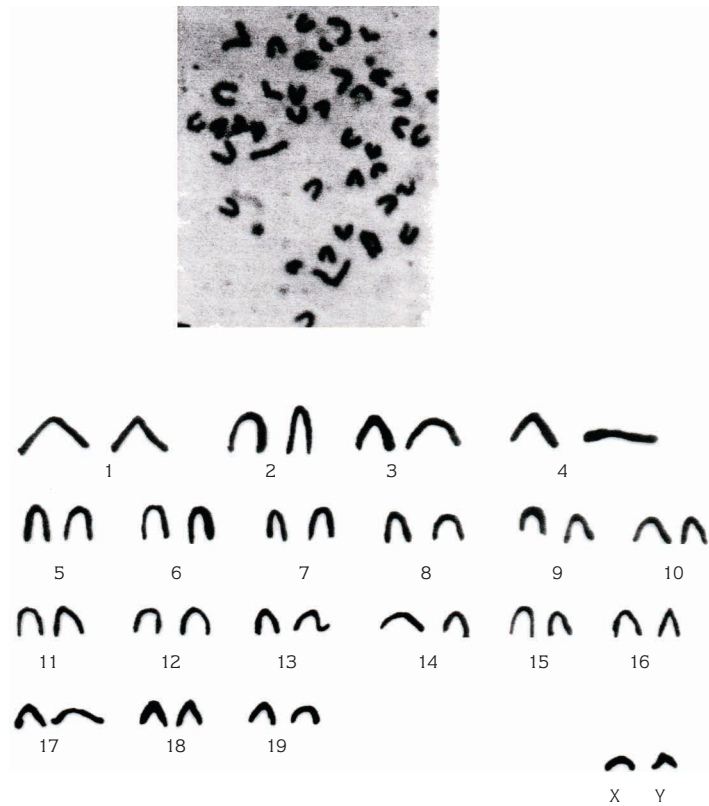


Figure 4. Karyotype of male *M. domesticus* from Zonguldak. a. metaphase plate, b. idiogram.

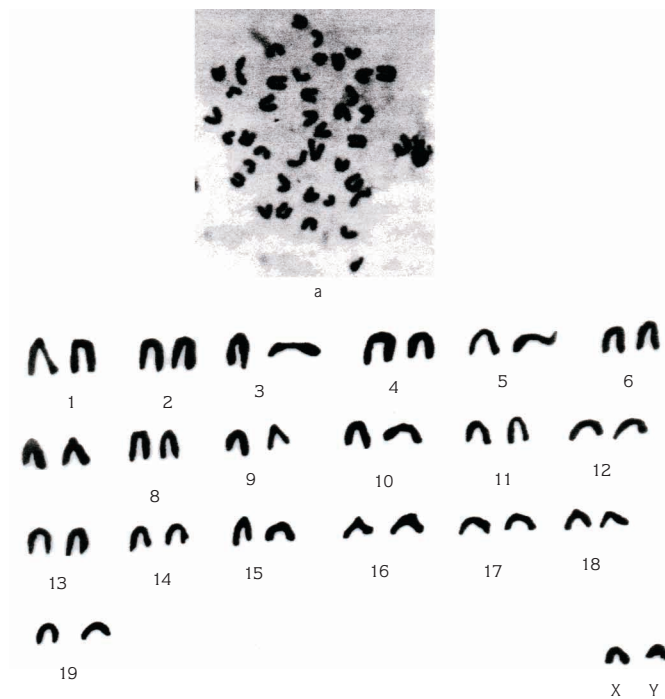


Figure 5. Karyotype of male *M. macedonicus* from Ankara. a. metaphase plate, b. idiogram.

Table 3. A comparison of average values ZI and H+B/T of the genus *Mus*.

Species	Country	ZI	H+B/T	References
<i>M. d. domesticus</i>	Greece	0.48		Orsini et al. 1983
<i>M. d. domesticus</i>	Israel	0.52	1.03	Orsini et al. 1983
<i>M. domesticus</i>	Turkey	0.25-0.46	0.73-1.0	Present study
<i>M. m. domesticus</i>	Greece, Bulgaria	0.47	1.07	Orsini et al. 1983
<i>M. spicilegus</i>	Greece	0.74		Orsini et al. 1983
<i>M. spicilegus</i>	N. Bulgaria	0.80		Orsini et al. 1983
<i>M. spicilegus</i>	Hungary	0.44		Present study
<i>M. spicilegus</i>	N. Austria	0.78		Orsini et al. 1983
<i>M. m. musculus</i>	N. Bulgaria	0.48		Orsini et al. 1983
<i>M.m. musculus</i>	Austria	0.45		Orsini et al. 1983
<i>M. macedonicus</i>	Turkey	0.50-0.80	0.76-1.46	Present study
<i>M. "spretoides"</i>	Israel	0.80	1.28	Auffray et al. 1990
<i>M. "spretoides"</i>	Bulgaria, Greece	0.74	1.49	Auffray et al. 1990

*macedonicus* is consistent with that of *M. macedonicus* in Turkey.

Auffray et al. (1990) determined H+B/T values for *M. m. domesticus*, and *M. "spretoides"* from Bulgaria, Greece, and Israel (Table 4). Table 4 shows that the H+B/T value of *M. domesticus* in the present study is consistent with that of *M. m. domesticus* in Israel, Bulgaria and Greece, and that the H+B/T value of *M. macedonicus* in Turkey is lower than that of *M. "spretoides"*.

According to Thaler et al. (1981), Bonhomme et al. (1984), Bonhomme et al. (1989), Frisman et al. (1990) and Britton-Davidian (1990), the slow allele of *Idh-1* is characteristic for *M. domesticus*, *M. praetextus* and *M. castaneus*. In contrast to previous studies, we have determined the fast allele of *Idh-1* in *M. domesticus* in Turkey. Mezhzherin et al. (1998) stated that specimens of *M. musculus* from Poland, Ukraine and Daghestan had only a fast allele of *Idh-1*, and only a slow allele in a population of *M. praetextus* from Syria. According to Mezhzherin et al. (1998), there is a slow allele (0.19) and fast allele (0.81) and slow allele (0.19) in a population of *M. praetextus* from Azerbaijan, a slow allele (0.52) and a fast allele (0.48) in a population of *M. domesticus* from Armenia, and only fast allele in population of *M. domesticus* from Germany. Mezhzherin et al. (1998) first recorded the slow allele of *Idh-1* (0.371) in a population

of *M. domesticus* from Transcaucasia. Awasthi et al. (1999) recorded abundant heterozygotes and 5 alleles of *Idh-1* in populations of *M. musculus* from India. Findings in this study showed that *M. musculus* is not found in the area studied. In this study, both fast and slow alleles of *Idh-1* were determined in the pair *M. domesticus* and *M. macedonicus* in Turkey. Therefore, *Idh-1* is not a diagnostic locus between *M. domesticus* and *M. macedonicus*.

A diploid number of  $2n = 40$  was given by Zima and Král (1984) for *M. musculus* from Europe, by Giagia et al. (1987) for *M. m. domesticus* in Greece, by Zima and Macholán (1989) for *M. m. musculus* and *M. m. domesticus* from Europe, by Bulatova et al. (1991) for *M. musculus* and *M. abbotti* from Azerbaijan, and by Ivanitskaya et al. (1996) for *M. macedonicus* from Israel. A standard karyotype of 40 chromosomes was described by Gündüz et al. (2000) for *M. musculus domesticus* and *M. macedonicus* from Kayseri and Samsun. These karyological values are consistent with the values in this study.

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