Karyotypes of the Mole Rats, genus *Nannospalax* (Palmer, 1903) (Spalacidae: Rodentia) populations in eastern Anatolia, Turkey

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The Spalacidae are Southeast European and East Mediterranean blind rodents, highly adapted for life underground. Their taxonomy needs a modern revision including chromosomal data as well as morphology. Mole rats of the family Spalacidae range over Turkey and approximately 30 karyotypes of Nannospalax complex inhabit. The diploid number of chromosome of Nannospalax ranges from 36 to 62. Also, fundamental number of chromosomal arms, NF values vary from 66 to 92 while the fundamental number of autosomal arms, NFa ranging from 62 to 88. Unfortunely, karyological studies of the Nannospalax populations on the territory of Turkey on the whole are far from being satisfactory. Karyological studies of this group may yield further chromosomal forms in Turkey, which has a wide range of climatic and biotic conditions and the boundaries of the distribution region of the known species might be determined. In this study, chromosomal forms of Nannospalax in East Anatolia (Erzurum, Kars and Ağrı province) were investigated. The materials of 16 specimens of Nannospalax were collected at 8 different localities in the region. Preparations of mitotic chromosomes were obtained from bone marrows by means of the general air-drying technique. Skins and skulls of specimens have been deposited at the Dicle University, Science Faculty Biology Department. We have identified two chromosomal forms of Nannospalax in East Anatolia of which have diploid chromosome numbers (2n) are 2n = 48 and 2n = 50.

Key words: Spalacidae, Nannospalax, Karyotype, Chromosomal polymorphism, Turkey.

INTRODUCTION

In the last three decades, chromosomes have become an important tool for the classification and taxonomy of organisms, see, for example, Savic and Nevo (1990). Although standard karyotypes are of limited value in assessing chromosomal evolution and phylogenetic relationships at lower taxonomic levels, such information can be useful for clarifying systematic relationship, such as detecting cryptic species or confirming species status. Chromosomal analysis is one of convenient tools in explanation of speciation and taxonomic problems. Standard karyotypes also help in determining general karyotypic trends within taxa. The modern geographic distribution of family Spalacidae, known as mole rats, are the Balkans, around Black and Caspian Seas, Anatolia, Caucasus, Middle East adjacent to the Mediterranean cost of North Africa. This subterranean rodent has been studied intensively to reveal karyological structures (Soldatovic and Savic 1978, Savic and Nevo

1990). The diploid number (2n) of chromosomes ranges from 36 to 62, and the fundamental number of chromosomal arms (NF) varies between 60 - 124 (Savic and Nevo 1990, Nevo 1999; Musser and Carleton, 2005). Therefore, taxonomic treatment of the family remains a challenge (Nevo 1999).

The mole rats, *Nannospalax*, Palmer 1903 represent an unresolved systematic and taxonomic problem. The systematics of the genus was highly confused. Recently, the genus *Nannospalax* has been extensively studied by karyologic approaches (Yüksel 1984; Coşkun et al. 2006, and references therein). These studies allowed for the recognition and description of new species.

Karyological studies on *Nannospalax* populations from this region were started in 1959 by Matthey (1959) who recorded 2n = 48, NF = 64 from the Caucasus (Armenia Talin region) and Lyapunova et al. (1974) identified the population of mole rats from Pambak-Armenia as *Mesospalax nehringi*; their karyotype was 2n = 50, NF = 72. Later, Nevo et al. (1995) reported the diploid number of chromosomes of specimens from Erzurum and Kars (Sarıkamış) as 2n = 50 and regarded as *Spalax leucodon*. Sözen et al. (2000) found 2n = 50, NF = 72 in the population from Kars (Susuz), Erzurum and Ardahan. Then, 2n = 48 and 50 karyotype of *Nannospalax* reported by Coşkun (2003) from Eastern Anatolia. Also, Coşkun and Kaya (2012) confirmed the diploid number of chromosomes of this genus as 2n=48.

This paper presents additional information on the karyology of the two populations of *Nannospalax* from eight localities of Eastern Anatolia, based on the published accounts and observations in the region.

MATERIAL AND METHODS

Between 2007 and 2008 sixteen mole-rats were trapped in their burrows in 8 different localities in the Eastern Anatolia (Fig. 1), during many expeditions. The sampling area is characterized by substantial habitat and climatic diversity. Voucher specimens are deposited at the Department of Biology, Science Faculty, University of Dicle-Turkey.

Bone marrow metaphase chromosomes were obtained as described by (Lee and Elder 1980). Slides were conventionally stained with 4% Giemsa-solution. Well-spread metaphases were recorded using a camera attached to a microscope. Karyotypes were prepared from the best metaphases. Chromosomes were paired by eye, using the position of the centromere and chromosome size. The fundamental number of chromosome arms (NF) and autosomal number of chromosome arms (NFa) was computed by counting bi-armed autosomes as two arms and acrocentric autosomes as one arm. To allow comparison between the different karyotypes, the NF and the NFa that are listed refer to the female karyotype.

RESULTS

The areas and localities from which the material was obtained are shown in the figure 1. The karyotypes of *Nannospalax* from the East Anatolia are shown in figure 2. In according to karyotype analysis, two different *Nannospalax* populations were described from the East Anatolia. Diagnostic features for the different karyotypes are described below and summarized in table 1. The table gives a comparative survey of all populations analysed to date of the species *Nannopalax* from the region. The approximately geographical distribution areas of each chromosomal form are shown in figure 1.

2n= 48 POPULATION (FROM AĞRI PROVINCE LOCALITY NO: 19, 20, 23 IN FIGURE 1)

The karyotypes of specimens from Ağrı populations have 2n = 48, NF = 68, and NFa = 64. Their karyotype consists of nine pairs meta/submetacentrics, and 14 pairs of acrocentric autosomes. The X chromosomes are large sized submetacentric and the Y chromosome was one of small acrocentric (Fig. 2A).



FIGURE 1 - Sampling localities and geographical distribution of chromosomal forms in the region. 1. Ardahan-Hanak-Altaş village, 2. Ardahan 10km E, 3. Ardahan-Göle-Yiğitkonağı village, 4. Ardahan-Göle-Boğatepe village, 5. Ardahan-Çıldır-Akçakale village, 6. Kars-Susuz, 7. Kars-Arpaçay, 8. Erzurum-Narman-Koçkaya village, 9. Kars-Selim, 10. Kars-Digor-Gülheyran village, 11. Kars-Digor 8.km, 12. Erzurum-Sarıkamış, 13. Erzurum-Horasan 5km E, 14. Erzurum-Köprüköy-Ilıcasu village, 15. Erzurum 20km E, 16. Erzurum 20km E, 17. Erzurum-Pasinler, 18. Iğdır-Tuzluca-Kazkoparan village, 19. Erzurum-Hınıs 15km N, 20. Ağrı-Tutak 5km S, 21. Ağrı-Hamur-Küpkıran village, 22. Ağrı-Taşlıçay-Yanalyol village, 23. Ağrı-Patnos-Sarısu village, 24. Van-Çaldıran, 25. Van-Erciş, 26. Muş-Malazgirt, 27. Erzurum-Çat-Yaylasuyu village, 28. Kars-Arpaçay.

The karyotype of the *Nannospalax* specimens collected from Erzurum-Hinis and Ağri-Patnos province which are called Ağri population, differs in the number of chromosomal arms (NF and NFa values) from those specimens collected by Coşkun et al. (2009) from Muş- Malazgirt and Van-Erciş (Table 1). The karyotypes resulted identical to those described by Matthey (1959) from Talin-Armenina and Coşkun (2003) from Ağrı and Van-Çaldıran,

2n = 50 Population (from Kars and Erzurum, province; Locality no: 8, 10, 13, 14, 27)

The karyotypes of this population comprised 2n = 50 chromosomes. The autosomal complement consisted of nine meta/submetacentric and fifteen acrocentric pairs. The X chromosome was large sized metacentric, the Y chromosome was one of small acrocentric. The number of chromosomal arms is NF = 70 and number of autosomes NFa = 66 (Fig. 2B).

The karyotypes resulted identical to those described by Nevo *et al.* (1995), Coşkun (2003) and Ulutürk et al. (2009) from Ardahan, Kars and Erzurum, but different Sözen et al. (2000) in Eastern Anatolia (Table 1).



FIGURE 2. Standard karyotypes of *Nannospalax* chromosomal forms, 2n = 48 in Ağrı population and 2n = 50 from Kars and Erzurum populations.

DISCUSSION

Mehely (1909) arranged all taxa into a genus *Spalax* in his monograph. But, according to Gromov and Baranova (1981), Topachevskii (1969) and Pantalayev (1998) Spalacidae have two distinct genera, *Nannospalax* and *Spalax*.

TABLE 1. Chromosomal data for the genus *Nannospalax* from East Anatolia. 2*n*: diploid number; m/sm: number of pairs of metacentric and submetacentric chromosomes; a/st: number of pairs of acrocentric and subtelocentric chromosomes; X: X chromosome, Y: Y chromosome, NF(a): (autosomal) fundamental number (female).

Form		Locality	2n	Autosome		NFa	NF	Gonosomes		References
				m/sm	а			Х	Y	References
2n = 48	Ağrı	Taşlıçay-Yanalyol	48	9	14	64	68	sm	a	Coşkun 2003
		Hamur-Киркітап								
		Patnos-Sarısu								This Study
		Tutak-5 km S								
	Van	Çaldıran								Coşkun 2003
		Erçiş		11 9	12	68	72 68			Coşkun et al. 2009
	Muş	Malazgirt								
	Iğdır	Tuzluca-Kazkoparan			14	64				Coşkun and Kaya 2012
	Erzurum	Hınıs- 15 km N								This study
2n = 50	Kars	Sarıkamış	50	9	15	66	70	sm a		Nevo et al. 1995
		Susuz		10	14	68	72		Sözen et al. 2000	
		Digor		9	15	66	70		a	Coşkun 2003
		Selim								
		Arpaçay								
		Arpaçay								Ulutürk et al.2009
		Digor-Gülheyran								This study
	Erzurum	20 km E		9	15	66	70	sm	a	Nevo et al. 1995
		20 km E		9	14	68	72			Sözen et al. 2000
		Pasinler								Coşkun 2003
		Horasan- 5 km E								This study
		Köprüköy-Ilıcasu								
		Narman-Koçkaya								
		Çat-Yaylasuyu								
	Ardahan	10 km E		10	14	68	72	sm a		Sözen et al. 2000
		Göle-Boğatepe		9	15	66	70		a	Coşkun 2003
		Hanak_Altaş								Ulutürk et al.2009
		Çıldır-Akçakale								
		Göle-Yiğitkonağı								

Species of *Spalax* include high diploid and fundamental numbers, no acrocentric chromosomes in their karyotypes those in *Nannospalax* have low diploid and fundamental numbers, (Zima and Kral, 1984 and Savic and Nevo 1990). Karyologically there is good reason to distinguish two forms within Spalacinae, viz., *Microspalax* [2n = 48-60, NF = 64 (?)-98] and *Spalax* (2n = 60-62, NF = 120-124) (Lyapunova et al., 1974). The genus name *Nannospalax* is used in this paper.

Satunin (1898) regarded the mole rats of the Eastern Anatolia as representing one species. He described *Spalax nehringi* from Ararat Valley, Tendürek River at Kazkoparan in the region. Later, many authors (Ognev 1947, Mursaloğlu 1979, and Kıvanç 1988) considered *Spalax nehringi* as varieties of *Spalax leucodon*.

Cytogenetical studies have shown that *Nannospalax* consist of different allopatric chromosome forms (2n = 48; NF = 68, NFa= 64 and 2n = 50; NF = 70, NFa= 66) without hybrid zones in territory of Eastern Anatolia. Each of the karyotype forms exhibits a well defined geographical range, separated mostly by the Aras River. It seems justified to consider the nominate species, *Nannospalax nehringi* Satunin 1898 as a valid species name for Ağrı population which have identical karyotype (2n = 48, NF = 68, and NFa = 64) (Fig. 2A). In spite of that, these populations are very similar morphologically and are identical karyologically.

According to these results, Ağrı populations (2n = 48a; NF = 68, NFa = 64) and the Muş-Malazgirt and Van-Erciş populations defined by Coşkun et al. (2009) (2n = 48b, NF = 72, and NFa = 68) represent two different chromosomal forms of the species *N. nehringi*, even though there are no significant geographic barriers between these two populations. However, each karyotype form occupies a well-defined geographical range, hence some ecological barriers may limit its spreading (Fig.1).

According to the karyotype analyses in this study, however, the Kars and Erzurum population (2n = 50; NF = 70, NFa = 66) of *Nannospalax* could even be considered a separate species. Chromosome forms in this group can accept represent good biological species, each adapted different ecogeographical regions.

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