Short communication

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Distribution of C-heterochromatin and Nucleolar Organizer Regions in the Karyotype of Marbled Polecat, *Vormela peregusna* (Carnivora: Mustelidae)

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Abstract: Banded chromosomes of a male marbled polecat, *Vormela peregusna*, from the Konya province in Turkey were studied. Conventional chromosome staining, Ag-NOR staining and C-banding were used to reveal the detailed morphology of the karyotype. The karyotype included eight metacentric, four submetacentric , five subtelocentric and one acrocentric autosomal pair (NFa = 70). The X chromosome was a medium-sized metacentric and the Y chromosome was a small submetacentric (NF = 74). C-heterochromatin regions were observed in large blocks forming the whole long arms of three metacentric and two submetacentric autosomal pairs, in telomeric regions of the short arms of two subtelocentric autosomal pairs, and in centromeric areas of six autosomes, including two metacentric, three subtelocentric and one acrocentric pair. The X chromosome appeared to be uniformly C-negatively stained and the short arm and centromeric area of the Y chromosome were entirely C-heterochromatic. The Ag-NOR regions were found within the large C-heterochromatic blocks on the long arms of two large metacentric and two large submetacentric autosomal pairs. Distinct differences in the karyotype morphology assessed according to the number of large C-heterochromatic blocks and nucleolar organizer regions were found between own findings and the previously published data.

Key words: chromosomes, Ag-NOR staining, C-banding, Turkey

Introduction

The marbled polecat, *Vormela peregusna* (Güldenstaedt, 1770) is a mustelid species distributed in steppes and semi-deserts of south-east Europe, Middle East, Caucasus, Central Asia, Siberia, Mongolia, northern China and Far East (Gorsuch, Lariviere 2005). This rare species was recorded from some regions in Turkey and the distribution range includes northern Anatolia along the Black Sea coast and eastern parts of Asiatic Turkey (Özkurt *et al.* 1999, Aulagnier *et al.* 2009).

The conventionally stained karyotype of the marbled polecat was described by Raicu, Duma (1971) in Romania, Dzuyev, Tchamokov (1976) in the Caucasus, Peshev, Al-Hossein (1989) in Syria, and Özkurt *et al.* (1999, 2000) studied two specimens from north-western Anatolia and south-eastern Anatolia in Turkey, respectively. The only detailed cytogenetic study using differential staining of chromosomes of this species was published by Graphodatsky *et al.* (1982). In this paper, the C- and G- banding patterns and the NORs distribution were reported in a male from captivity. The origin of the individual studied is not clear and we can only assume that it originated from western Siberia.
Information on differentially stained chromosomes and detailed structure of the karyotype is therefore lacking in marble polecat populations from the Western Palaearctic. The aim of this paper is to perform a chromosomal banding analysis of the karyotype in a specimen from Turkey with the use of C-banding and

Ag-NOR staining and to reveal possible differences in comparison with the published data.

Materials and Methods

Cytogenetic analyses were performed in one male specimen of the marbled polecat from Konya (37° 52′ N, 32° 35′ E). Karyotype preparations were made from bone marrow cells after colchicine treatment (Ford, Hamerton 1956). Air-dried preparations were stained conventionally by Giemsa. Constitutive heterochromatin and nucleolus organizer regions (NORs) were detected by the techniques of C-banding (Sumner 1972) and Ag-NOR staining (Howell, Black 1980), respectively. Altogether, 20 microscopic slides were prepared and well-spread metaphase plates were examined, including seven conventionally stained complements, seven C-banded complements, and seven Ag-NOR stained complements which were photographed and analysed. The nomenclature system introduced by Hsu, Benirschke (1967-1977) was used for classification of chromosomes according to the centromere position, and the metacentric, submetacentric, subtelocentric and acrocentric elements were distinguished. The voucher specimen (skin and skull) is deposited at Selçuk University, Biology Department, Faculty of Science, Konya, Turkey.

Results

The karyotype of the male marbled polecat examined from Konya consists of 38 chromosomes including four large, one medium-sized and three small metacentric autosomal pair (nos. 1-8), two large and two medium-sized submetacentric autosomal pair (nos. 9-12), five subtelocentric autosomal pairs of varied size (nos. 13-17), and one small acrocentric autosomal pair (no. 18) (NFa = 70). Secondary constrictions were observed in the long arms of the autosomal pairs no. 1, 2, 9 and 10. The X chromosome was medium-sized submetacentric, and the Y chromosome was submetacentric and the smallest element in the complement (NF = 74) (Fig. 1).

Large blocks of C-heterochromatin forming whole long arms were observed in three large metacentric and two large submetacentric autosomal pairs (nos. 1, 2, 3, 9, 10). Other autosomal pairs (nos. 6, 8, 14, 16-18) possessed distinct centromeric dark C-bands. Tiny telomeric C-positive bands were apparent in the short arms of the subtelocentric autosomal pairs no. 14 and 16. The X chromosome appeared to be uniformly and C-negatively stained and the short arm and the centromeric area of the Y chromosome were positively C-heterochromatic. The C-banded karyotype is illustrated in Fig. 2.

The Ag-NOR regions were detected in two large metacentric and two large submetacentric autosomal pairs (nos. 1, 2, 9, 10), and the NORs were localised in similar interstitial position on the long arms (Fig. 3). These AgNOR-bearing long arms were stained C-positively.

Discussion

The published studies on the karyotype of the marbled polecat reported in the complement 38 chromosomes including 16 or 17 biarmed and two or one acrocentric autosomal pairs (Raicu, Duma 1971, Dzuyev, Tembotov 1976, Peshev, Al-Hossein 1989, Özkurt *et al.* 1999, 2000). It is difficult to identify possible reasons of this variation in the morphology of one autosomal pair but it is possible that it originated after alternative assessment of the centromeric position in the small subtelocentric autosome no. 17 which could be considered acrocentric by some authors (see Özkurt *et al.* 1999, 2000). The X chromosome was uniformly determined is a medium-sized submetacentric; the Y chromosome was described as small biarmed or dot-like element.

The diploid chromosome number determined in the marbled polecat is similar as in many other species belonging to the Mustelidae family (Graphodatsky *et al.* 1976, 1977, Zima, Král 1984). However, the karyotypic features of the marbled polecat revealed by chromosome banding are rather specific. Particularly unusual is the very large amount of C-heterochromatic regions forming whole arms on five autosomal pairs. This extensive heterochromatin amplification event is apparently responsible for the high DNA content reported in the marbled polecat (Graphodatsky *et al.* 1977). Most of the karyologically studied mustelid species had only one autosomal pair possessing a whole heterochromatic arm (Graphodatsky *et al.* 1977). High amount of C-heterochromatin was reported only in the Altai weasel (*Mustela altaica*) and the weasel (*M. nivalis*) (Graphodatsky *et al.* 1977, Mandahl, Fredga 1980). In the weasel, distinct geographic variation in the number of whole C-heterochromatic arms was recorded (Zima, Graphodatsky 1985, Zima, Cenevová 2002). In the marbled polecat, the comparison of our finding and that of Graphodatsky *et al.* (1982) indicates that the basic pattern of the distribution of C-heterochromatin in the whole arms may be stable in various geographic populations. However, there are distinct differences in other details of the C-heterochromatin distribution. Graphodatsky *et al.* (1982) recorded another C-heterochromatic block forming the short arm of one of the medium-sized

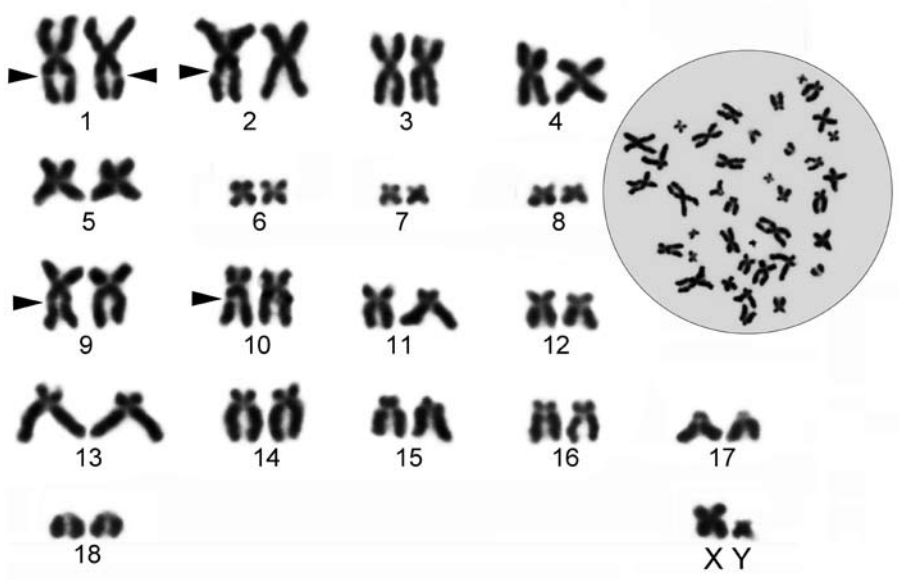

Fig. 1. Metaphase spread and karyotype of *Vormela peregusna*. Arrows indicate the position of the secondary constriction

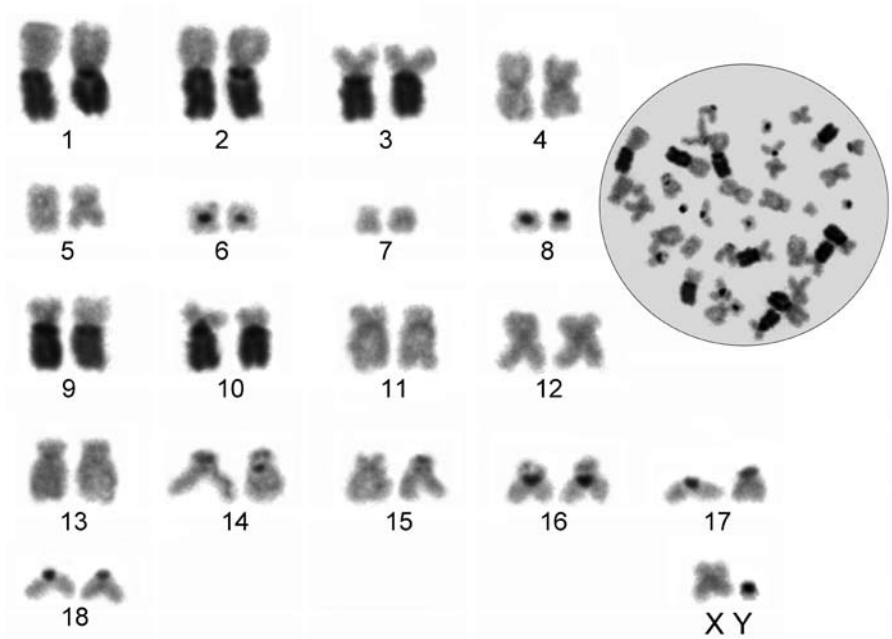

Fig. 2. Metaphase spread and C-banded karyotype of *Vormela peregusna*

submetacentric autosomes (this autosome is similar in size and centromeric position to the autosome no. 11 in Figs. 1 and 2). This C-heterochromatic short arm was not observed in cells of the male examined in this study. Graphodatsky *et al.* (1982) further recorded C-positive telomeric regions on the short arms of four submeta- and subtelocentric autosomal pairs but only two autosomal pairs possessing such telomeric dark bands were demonstrated in this study. Finally, Graphodatsky *et al.* (1982) characterized the dot-like Y chromosome as completely C-heterochromatic but the long arm of the submetacentric Y chromosome appeared euchromatic in the complement of our specimen.

The secondary constriction is usually situated in a single acrocentric autosomal pair in most of the mustelid species studied (Graphodatsky, Radjabli 1980). The marbled polecat represents a remarkable exception, with secondary constrictions of Ag-NORs localized within long arms of several autosomal pairs. Graphodatsky *et al.* (1982) reported five secondary constrictions or Ag-NORs in the studied male marbled polecat, and all the NORs were localized inside the C-heterochromatic long arms. Our study of a specimen from Turkey revealed the NORs in only four pairs and their presence was not confirmed on the long arm of the autosome no. 3. Peshev, Al-Hossein (1989) recorded the secondary

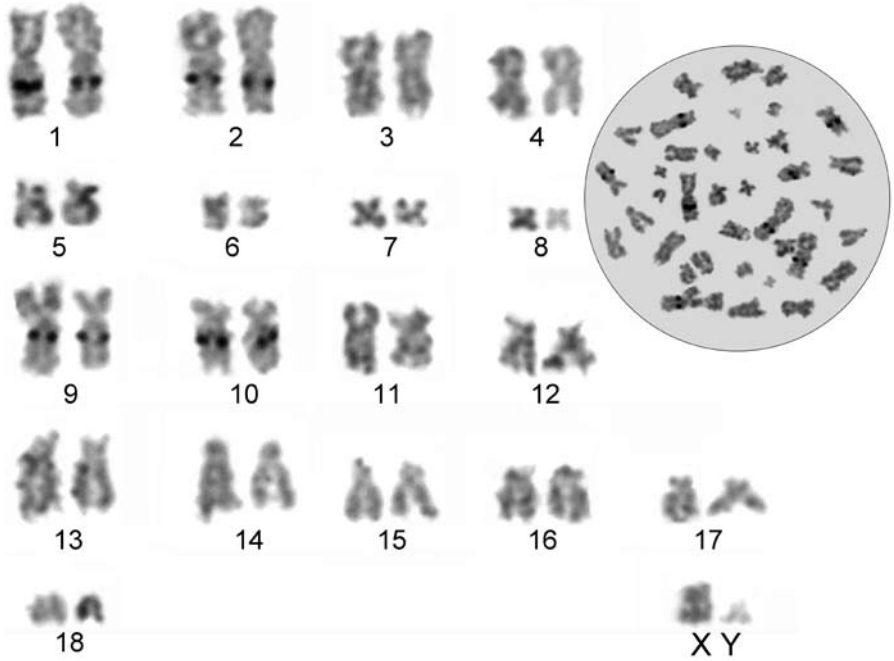

Fig. 3. Silver-stained metaphase spread and karyotype of *Vormela peregusna*

constrictions on three autosomal pairs in the complement of specimens from Syria. Graphodatsky *et al.* (1982) observed variation in the number of observed Ag-NORs between the cells examined. We cannot exclude therefore that the differences between individual records may be influenced by such variation and also by the low number of specimens examined in each study.

References

Aulagnier S., P. Haffner, A. J. Mitchell-Jones, F. Moutou, J. Zima 2009. Mammals of Europe, North Africa and the Middle East. A&C Black Publishers, London, 272 pp.

Dzuyev R. I., P. N. Tchamokov 1976. The chromosome complements of *Mustela nivalis* L. and *Vormela peregusna* Güld. from the Caucasus. – *Animal Faunistics, Ecology and Conservation in the Northern Caucasus,* **3**: 142-146. (in Russian).

Ford C. E., J. L. Hamerton 1956. A colchicine, hypotonic citrate, squash sequence for mammalian chromosomes. – *Stain Technology,* **31**: 247-251.

Gorsuch W. A., S. Lariviere 2005. *Vormela peregusna*. – *Mammalian Species,* **779**: 1-5.

Graphodatsky A. S., S. I. Radjabli 1980. Nucleolar organizer regions in chromosomes of nine mustelid species. – *Doklady AN SSSR,* **255**: 1487-1489. (in Russian, English summary).

Graphodatsky A. S., Yu. G. Ternovskaya, D. V. Ternovsky 1982. Differential staining of chromosomes of *Vormela peregusna* (Carnivora, Mustelidae). – *Zoologicheskii Zhurnal,* **61**: 464-467. (in Russian, English summary).

Graphodatsky A. S., D. V. Ternovsky, A. A. Isayenko, S. I. Radjabli 1977. Constitutive heterochromatin and DNA content in some mustelid species (Carnivora, Mustelidae). – *Genetika,* **13**: 2123-2128. (in Russian, English summary).

Graphodatsky A. S., V. T. Volobouev, D. V. Ternovsky, S. I. Radjabli 1976. Chromosomal G-banding in seven species of Mustelidae (Carnivora). – *Zoologicheskii Zhurnal,* **55**: 1704-1711. (in Russian, English summary).

Howell W. M., D. A. Black 1980. Controlled silver staining of nucleolar organizer regions with a protective colloidal developer: a 1-step method. – *Experientia,* **36**: 1014-1015.

Hsu T. C., K. Benirschke 1967-1977. An atlas of mammalian chromosomes, Vol. 1-10. Springer Verlag, Berlin-Heidelberg-New York.

Mandahl N., K. Fredga 1980. A comparative chromosome study by means of G-, C-, and NOR-banding of the weasel, pygmy weasel and the stoat (*Mustela*, Carnivora, Mammalia). – *Hereditas,* **93**: 75-83.

Özkurt S., M. Sözen, N. Yiğit, E. Çolak 1999. A study on *Vormela peregusna* Guldenstaedt, 1770 (Mammalia: Carnivora) in Turkey. – *Turkish Journal of Zoology,* **23**:141-144.

Özkurt S., M. Sözen, N. Yiğit, E. Çolak, R. Verimli 2000. On colouration and karyology of the marbled polecat, *Vormela peregusna*, in Turkey. – *Zoology in the Middle East,* **21**:13-18.

Peshev D. T., K. Al-Hossein 1989. Karyology and biochemical characteristics of the polecat (*Vormela peregusna syriaca* Pockock) (Carnivora: Mustelidae) from Syria. – *Acta zoologica bulgarica,* **38**: 54-57.

Raicu P., D. Duma 1971. Chromosome complement of *Vormela peregusna* ssp. *euxina* (order Carnivora, fam. Mustelidae). – *Mammalian Chromosomes Newsletter,* **12**: 81-82.

Sumner A. T. 1972. A simple technique for demonstrating centromeric heterochromatin. – *Experimental Cell Research,* **75**: 304-306.

Zima J., E. Cenevová 2002. Coat colour and chromosome variation in central European populations of the weasel (*Mustela nivalis*). – *Folia Zoologica,* **51**: 265-274.

Zima J., A. S. Graphodatsky 1985. C-heterochromatin arm variation in the weasel, *Mustela nivalis* (Mustelidae, Carnivora). – *Folia Zoologica,* **34**: 125-132.

Zima J., B. Král 1984. Karyotypes of European mammals III. – *Acta Scientiarum Naturalium, Brno,* **18**: 1-51.

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