Karyotype of NIH, C57BL/6 and Razi strains of laboratory mice (*Mus musculus*)

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Abstract

The laboratory mouse is recognized as the pre-eminent model for genetic research. Awareness of chromosomal patterns of experimental animals increases their value for a variety of different fields of study. We aimed to study mitotic chromosome preparations from NIH, C57BL/6 and Razi strains of mice, which are outbred, inbred and partially inbred laboratory mice respectively. Bone marrow cells were prepared from 36 male and female mice, 12 from each strain, and stained by use of Giemsa staining and G-banding methods. Karyotyping of the samples showed that there was no difference in chromosomal numbers among the three mice strains, also the metaphase preparations of their diploid cells contained 40 chromosomes ($2n = 40$) and all chromosomes were telocentric. However, some differences in band tonality and the size of chromosomes were seen.

Introduction

All of the *Mus musculus* subspecies, including *M. m. domesticus*, *M. m. musculus*, *M. m. castaneus*, and *M. m. bactrianus* and closely related species *M. spretus*, *M. spicilegus* and *M. macedonicus*, have the same standard karyotype with 20 pairs of chromosomes, including 19 autosomal pairs and one pair of sex chromosomes. All of the 19 autosomes as well as the X chromosome appear to be telocentric, with a centromere at one end and a telomere at the other (Silver, 2001). Chromosomes can be distinguished on the basis of banding patterns with the use of various staining protocols (IAEA, 2001). A mild trypsin treatment was followed by Giemsa staining to produce dark Giemsa-stained bands called G-bands that alternate with Giemsa-negative bands called R bands for reverse G-bands. R, Q, and T banding are visualised by staining methods based on the same principal of chromatin denaturation or mild enzymatic digestion followed by staining with a DNA-binding dye (Craig and Bickmore, 1993). In general, all of these different protocols produce the same patterns of bands and interbands observed with Giemsa staining, although in some cases the dark and light regions are reversed.

Since 1967, there have been numerous reports of wild-caught mice with karyotypes that contain fewer than 20 pairs of chromosomes. The first report described a karyotype with 13 pairs of chromosomes, seven metacentrics and six telocentrics in mice captured from the Valle di Poschiavo in southeastern Switzerland (Gropp *et al.*, 1972). It was assumed that animals with such a different karyotype could not possibly be members of the *M. musculus* species, but belonged to the *Mus poschiavinus* species and were referred to tobacco mouse. Further studies of wild house mice by other investigators have led to the discovery of additional non-standard karyotypes in house mice from other regions of Europe, South America and Northern Africa (Adolph and Klein, 1981; Wallace, 1981; Searle, 1982). It seems that all of the non-standard karyotypes have arisen by simple fusion events, each of which resulted in the attachment of two standard mouse chromosomes at their centromeres (Adolph and Klein, 1981). Other cytogenetic techniques, spectral karyotyping (SKY), related multiplex fluorescence *in situ* hybridization (M-FISH), are chromosome-specific multicolor, FISH techniques improved the characterization of aberrant chromosomes that contain DNA sequences not identifiable using conventional banding methods. However, Gimsa staining and the G-banding method are rapid techniques (Mirabzadeh, 2001) and are an economical way to study mouse karyotypes. This study leads us to improve our knowledge of three different strains laboratory mice that are commonly used for biomedical research and the production of biological products.

Material and Methods

Bone marrow samples from 36 male ($n = 18$) and female ($n = 18$) mice, 12 each of outbred NIH, inbred C57BL/6 and partially inbred Razi strains, respectively, were prepared according to the Deanna and Robbins method (1991). Mitotic metaphase

chromosomes were prepared and stained with Giemsa and the G-bandings method. At least 50 metaphase samples were drawn for each mouse using a Zeiss camera lucida microscope. The nomenclature used for the description of the chromosome morphology is that proposed by Levan *et al.* (1964) and Guerra (1986).

Results

All 36 mice from the three strains had 40 chromosomes ($2n = 40$) with telocentric morphology. The phenotypes and karyotypes of the male and female mice of the studied strains are illustrated in Figures 1-3.

Discussion

The results obtained from this research have allowed us to illustrate the karyotypes of mouse strains of outbred NIH, partial inbred Razi and inbred C57BL/6 mice. The chromosome numbers agree with those published previously (Mirabzadeh, 2001; Silver, 2001). The numbers of diploid chromosomes in three strains of mice were similar in that they may belong to same ancestor of *Mus musculus*. Analysis of karyotypes showed that, in general, the chromosomes were telocentric (Mirabzadeh, 2001; Manna *et al.*, 1974) and of similar size (Goleman *et al.*, 1996; Padilla-Nash *et al.*, 2006) in all of the mice. They formed a homogeneous group and differed mainly in the length of the Y chromosome (Levan *et al.*, 1962; Nesbitt and Francke, 1973). With Giemsa staining, the centromeres stain very densely; this finding has been reported in a previous study (Nesbitt and Francke, 1973). The G-bandings showed that many of the major bands contained minor bands (Nesbitt and Francke, 1973; Cowell, 1984) and homologous chromosomes were paired according this banding pattern. The X chromosome is one of longest chromosomes could be easily paired (Cowell, 1984; Levan *et al.*, 1962; Mirabzadeh, 2001); the Y chromosome was constantly dark and the centromeric chromatin was not obvious (Cowell, 1984; Nesbitt and

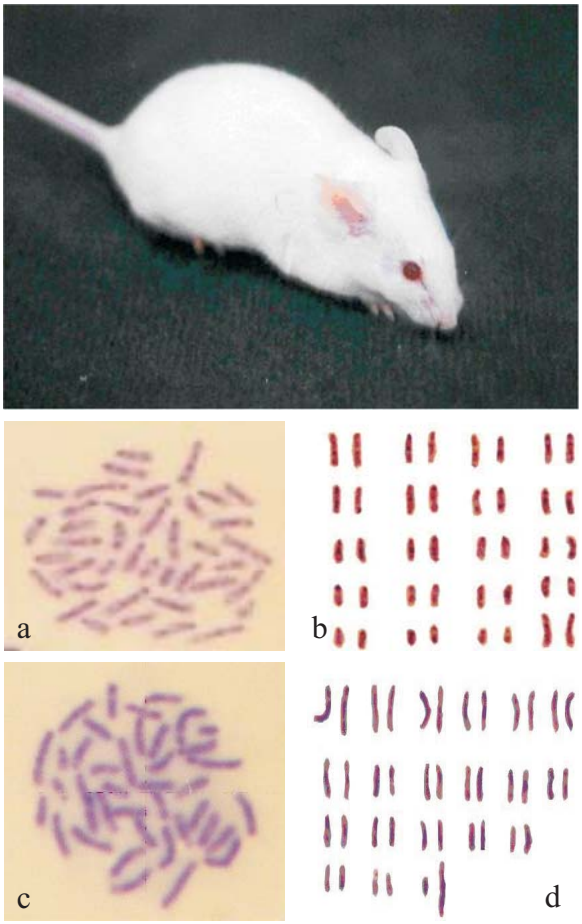

Figure 1: Phenotype of the NIH, outbred mouse. (a) Karyotype of a female NIH mouse. (b) Homologous chromosomes of a female NIH mouse. (c) Karyotype of a male NIH mouse. (d) Homologous chromosomes of a male NIH mouse.

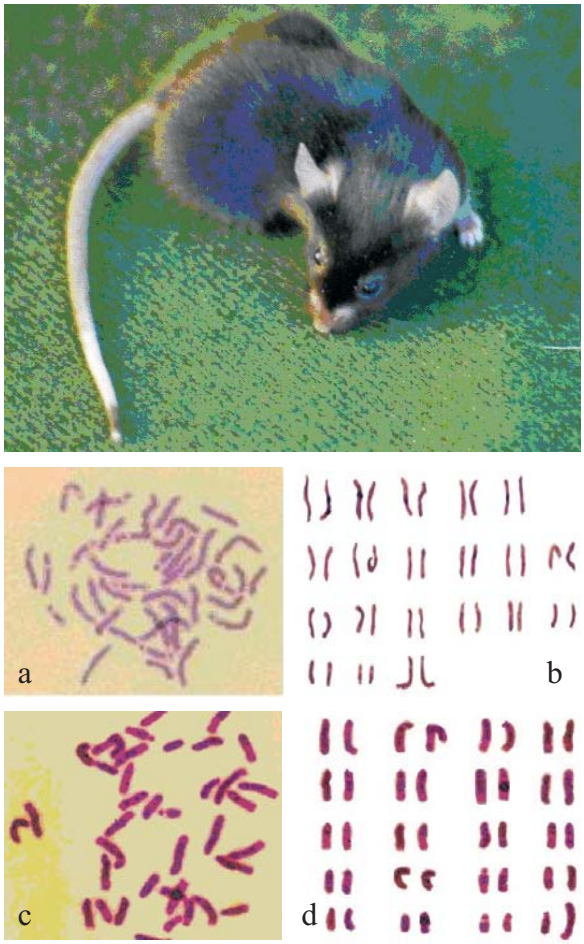

Figure 2: Phenotype of C57BL/6 inbred mice. (a) Karyotype of a female C57BL/6 mouse. (b) Homologous chromosomes of a female C57BL/6 mouse. (c) Karyotype of a male C57BL/6 mouse. (d) Homologous chromosomes of a male C57BL/6 mouse.

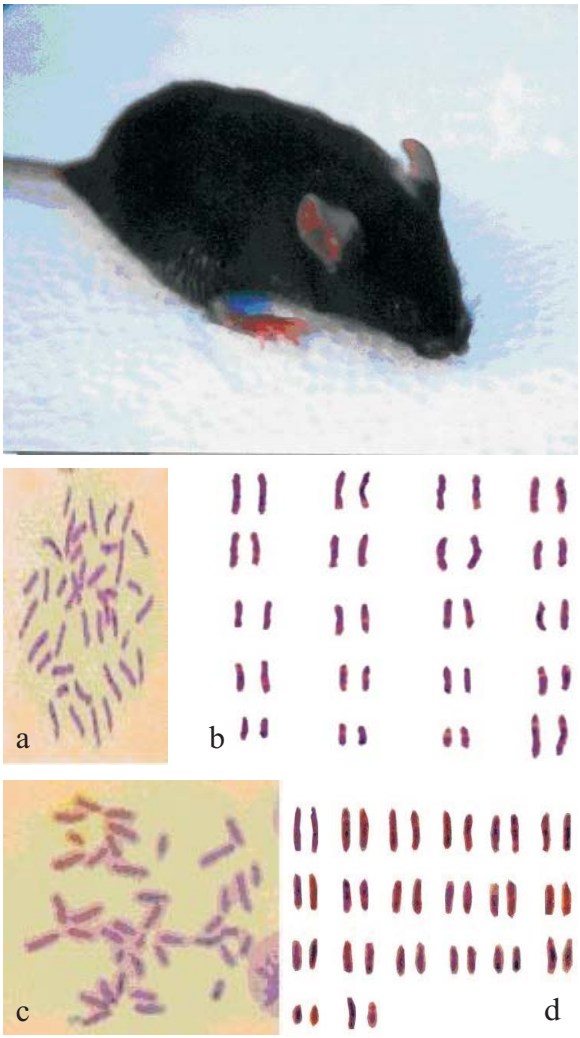

Figure 3: Phenotype of Razi, partial inbred mice. a) Karyotype of a female Razi mouse. (b) Homologous chromosomes of a female Razi mouse. (c) Karyotype of a male Razi mouse. (d) Homologous chromosomes of a male Razi mouse.

Francke, 1973; Sawyer *et al.*, 1987; Mirabzadeh, 2001).

In conclusion, the findings of this study provide information about the karyology of outbred, inbred and partial inbred mice that are bred in conventional colonies in Iran and could be used as a reference patterns for researchers and breeders.

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