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Standard karyotype of the domestic horse (*Equus caballus*)

Committee for standardized karyotype of *Equus caballus*. The Second International Conference for Standardization of Domestic Animal Karyotypes, INRA, Jouy-en Josas, France, 22nd–26th May 1989

C. L. RICHER, M. M. POWER, L. R. KLUNDER, R. A. McFEELY and M. G. KENT

Département d'anatomie, Faculté de médecine, Université de Montréal, Montréal, Canada

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The following decisions concerning the banded karyotype of the horse (*Equus caballus*) were made at the second International conference for Standardization of Domestic Animal Karyotypes, held at Jouy-en Josas, France, 22nd–26th May 1989: (1) numbering of the chromosomes was modified to correspond to an arrangement into only two groups (the non-acrocentrics and the acrocentrics) within which the autosomes are placed according to length alone; (2) a more compact karyotype arrangement was adopted: chromosomes 1 to 5 on the first row, 6 to 10 on the second, 11 to 13, and, at the far right, X and Y on the third row, 14 to 19 on the fourth row, chromosomes 20 to 25 on the fifth, and 26 to 31 on the sixth row; (3) the NOR-bearing horse chromosomes were identified as numbers 1, 28 and 31.

Claude-Lise Richer, Département d'anatomie, Faculté de médecine, Université de Montréal, C.P. 6128, Succ. "A", Montréal, Québec, H3C 3J7, Canada

For horse, no other standard karyotype has been internationally accepted since the Reading Conference (Proceedings of the First International Conference for the Standardization of Banded karyotypes of Domestic Animals, 1980). However, little uniformity was observed in karyotypes of published papers, and dissatisfaction was widely expressed mainly concerning the three following problems: (1) impossibility to verify with calculations of the centromeric indices, especially of longer chromosomes, the complicated four group classification proposed at Reading; (2) conflict with journal editors concerning the karyotype placement which required more space than necessary; and (3) difficulty in identifying the smallest chromosomes (26 to 31) by using the karyotype and the description of Reading. This report presents the new classification and karyotype arrangement adopted, and also karyotypes, at early metaphase and prometaphase (equivalent to approximately 400 and 600 bands per genome) in solid stain, and after C-, G-, and R-banding. Finally the nomenclature for NOR-bearing chromosomes which was agreed upon is presented.

Methods

The C-banding used was the CBG (C-bands by Barium hydroxide and Giemsa) of SUMNER (1972); the G-banding was the GTG (G-bands by Trypsin and Giemsa) of SEABRIGHT (1971); and the R-banding, the RBG (R-bands by BrdU incorporation and Giemsa staining after UV irradiation) adapted from LATT et al. (1976) and PERRY and WOLFF (1974). Correspondence between G- and R-bands was ascertained by relating them through sequential Q- and R-banding, since the identical localization of Q- and G-bands is well recognized. Staining of the nucleolar organizer regions was done according to a technique derived from that of BLOOM and GOODPASTURE (1976), and chromosome identification was obtained by sequential banding by RBG and NOR.

Chromosome measurements to determine overall length were made on conventionally stained chromosomes not subjected to prior chemical or thermal treatment, nor to DNA analog substitution; their size was approximately within the 250-band range (POWER 1987).

Decisions made at the meeting

A. Numbering of the chromosomes

This question was addressed with great caution, because of the concern to keep, if possible, the continuity with previously published works. However, the classification into metacentrics, submetacentrics, subtelocentrics and telocentrics could not be scientifically supported by measurements of chromosomes longer than those analysed at Reading, and this resulted in continuously renewed discussions, far from universal acceptance, and lack of uniformity in publications. Taking this into consideration, as well as (1) the often discussed proposition, made by HANSEN (1984), to divide the chromosomes into only two groups, and (2) the agreement arrived at in the Bristol meeting of 1988 that the name "acrocentric" should be used, instead of "telocentric", particularly since electron micrography of the horse chromosomes had clearly shown the presence of short arms (ROMAGNANO 1986), it was finally adopted that the chromosomes should be arranged into only two groups: the non-acrocentrics and the acrocentrics, and, that within these two groups, the autosomes should be arranged according to length alone.

Correspondence between the new nomenclature and the old one is presented in Table 1. This modification in chromosome number, as far as can be ascertained, does not affect the number of any chromosome for which an anomaly or a gene localization has been published.

B. Placement of the karyotype

A more compact karyotype arrangement was adopted. It is the following:

first row:	chromosomes 1 to 5
second row:	chromosomes 6 to 10
third row:	chromosomes 11 to 13, and, at the far right, X and Y
fourth row:	chromosomes 14 to 19
fifth row:	chromosomes 20 to 25
sixth row:	chromosomes 26 to 31.

Table 1. Comparison of the horse chromosome numbers in the new nomenclature (Jouy Meeting, 1989) with the numbers proposed at the Reading Conference in 1976

New number	Old number	Length
1	1	68.41
2	5	45.94
3	2	44.25
4	13	40.28
5	6	37.79
6	3	36.71
7	7	35.66
8	4	35.26
9	9	33.86
10	8	32.62
11	10	24.61
12	12	21.13
13	11	20.27
14 to 31	same	

C. Identification of the NOR-bearing chromosomes

It was adopted that the NOR-bearing horse chromosomes are numbers 1, 28, and 31.

Results and discussion

Four karyotypes are presented: (A) solid-Giemsa stained, (B) CBG-banded, (C) GTG-banded, and (D) RBG-banded. The first three, which are combined, are at early metaphase, approximately in the 400-band range (Fig. 1), while the BrdU-substituted and R-banded one is at the prometaphase stage, in the 550-band range (Fig. 2).

The particularities of C-bands in the horse are: (A) positive staining of all centric and paracentric regions except for chromosome 11, in which this is not usually observed; (B) individual variations in the length of the centric and paracentric regions of chromosomes 1, 12, and 13, and in the length of the heterochromatic segment in the long arm of chromosome Y; and (C) an additional positive band in the proximal third of the long arm of the X chromosome. It may be useful to note that this C-positive feature of the X, which is much used for sex determination, may not always be clearly apparent, especially on long chromosomes. It also seems necessary to mention that the polymorphism of these regions in chromosomes 12 and 13 may, in some individuals, alter the relative lengths.

Concerning the NOR-bearing chromosomes, identified after sequential RBG- and NOR-banding, it should be mentioned that the six NOR-bearing

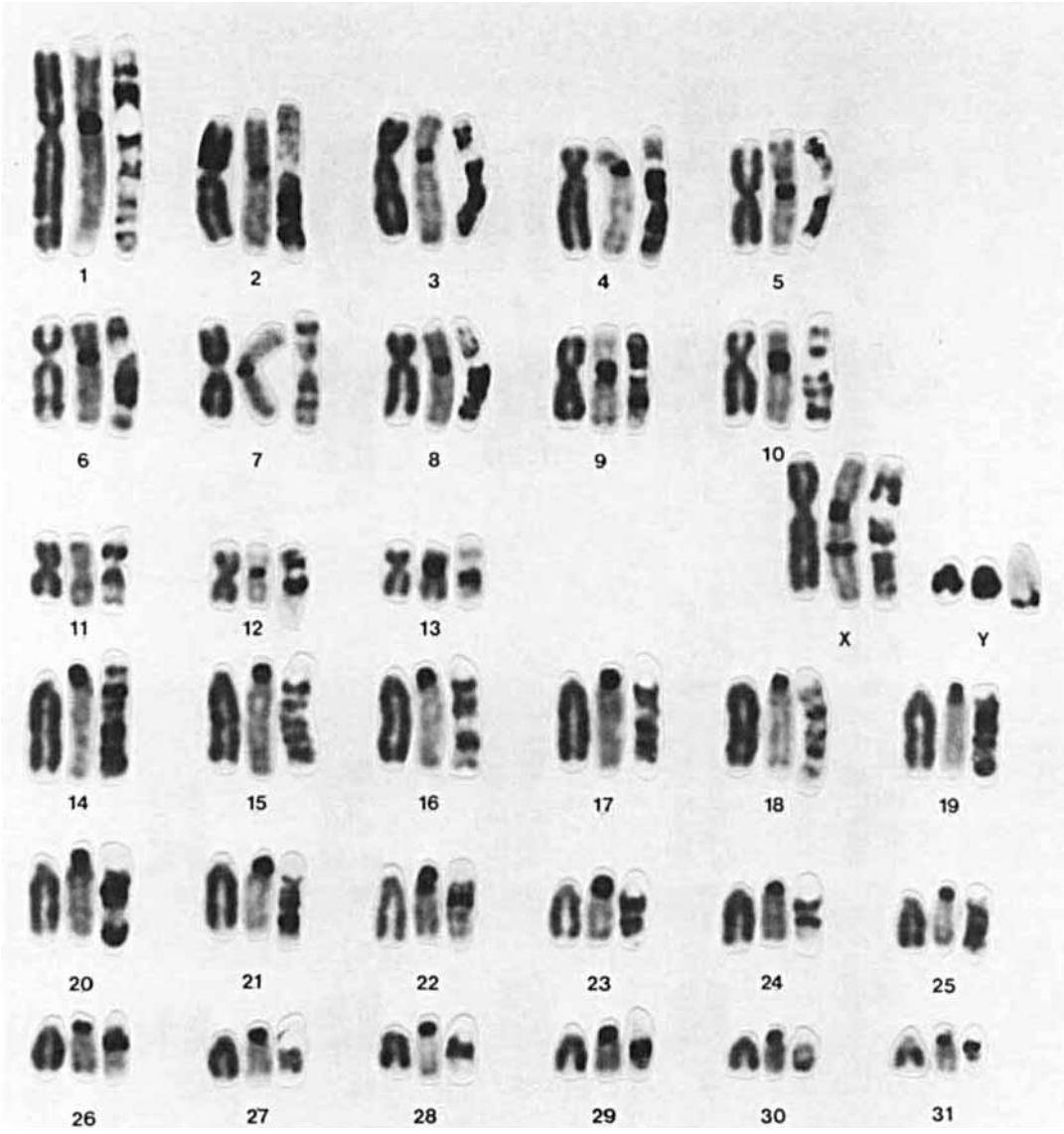

Fig. 1. Combined haploid karyotypes of (from left to right) unbanded (Giemsa-stained), C-banded, and GTG-banded horse chromosomes at the level of 400 bands. (Courtesy L. R. Klunder, R. McFeely, M. M. Power and C. L. Richer.)

chromosomes (pairs 1, 28, and 31) do not always react positively to the NOR technique (LAU et al. 1978); most often five are observed and sometimes only four are positive. The chromosomes of pair number 28 are those which respond variably to the technique.

G- and R-bands serving as landmarks for horse chromosomes have not yet been listed. Also, even though G- and R-band idiograms have been proposed, the committee has not yet adopted them.

Conclusion

Before description of landmarks and adoption of idiograms, the present karyotypes should help stan-

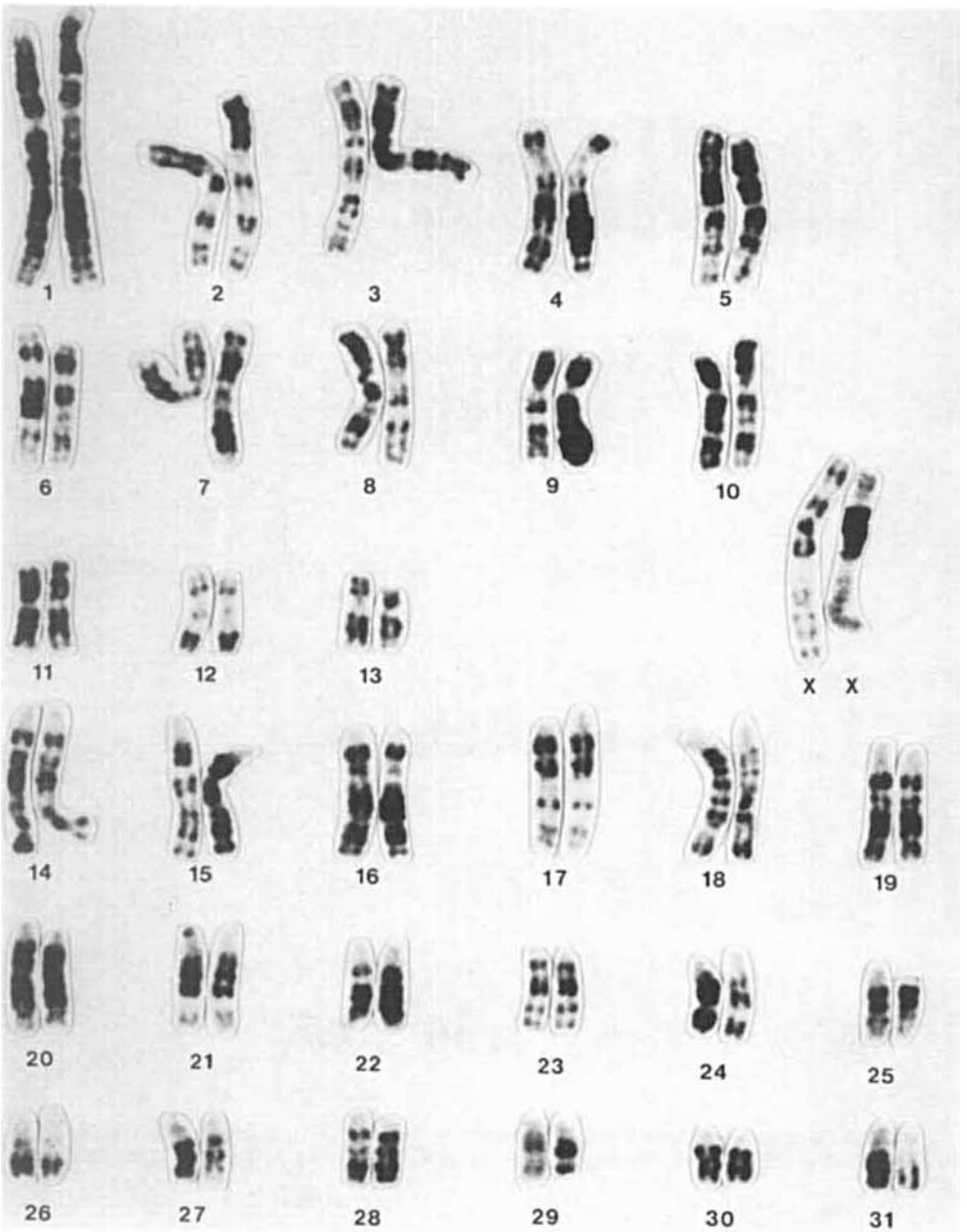

Fig. 2. RBG-banded horse karyotype at the level of 550 bands. The inactive X is on the right. (Courtesy M. M. Power.)

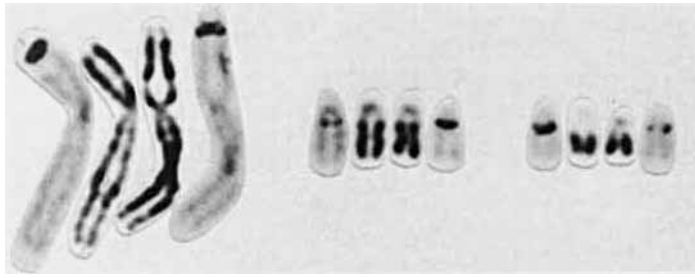

Fig. 3. RBG-banded and NOR-stained chromosomes number 1, 28, and 31. (Courtesy C. L. Richer.)

dardize karyotyping of horse chromosomes. The committee hopes that the new nomenclature and karyotype placement, as well as the identification of the NOR-bearing chromosomes, will prove useful for cytogenetic analyses of horse chromosomes.

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