

Zusammenfassung. Aufgrund des Fluoreszenzbandenmusters können die einzelnen Chromosomen der parasitischen Fliege *Voria ruralis* identifiziert und unterschieden werden. Die diploide Chromosomenzahl beträgt 12, und die Geschlechtschromosomen sind vorwiegend heterochromatisch. Nach der Fluoreszenzfärbung mit Quinacimmustard wurden ein Bandenpolymorphismus

eines Chromosomenpaares und morphologische Abweichungen des X-Chromosoms in 8 von 9 Weibchen beobachtet.

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Identification of Chromosomes Involved in the 9 Robertsonian Fusions of the Apennine Mouse with a 22-Chromosome Karyotype

CAPANNA, CIVITELLI and CRISTALDI¹ have recently discovered in the Central Apennines a population of house mice that may be classified as *Mus musculus* and are characterized by a 22-chromosome karyotype (Figure 1a). This karyotype consists of 18 metacentric chromosomes, obtained by the centric fusion of 18 pairs of acrocentric autosomes of the standard 40-chromosome mouse karyotype, plus a pair of small acrocentric autosomes, as well as the pair of heterochromosomes.

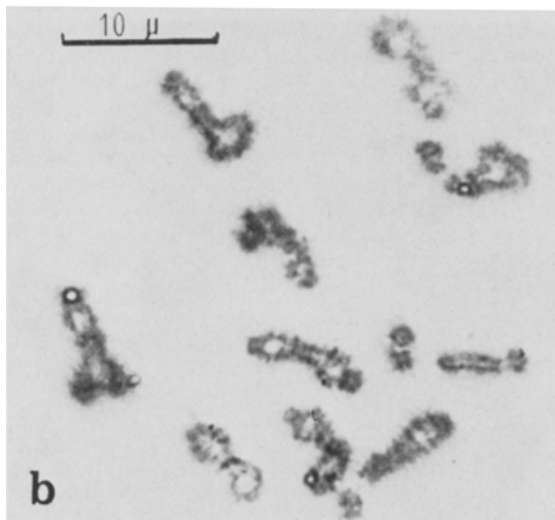
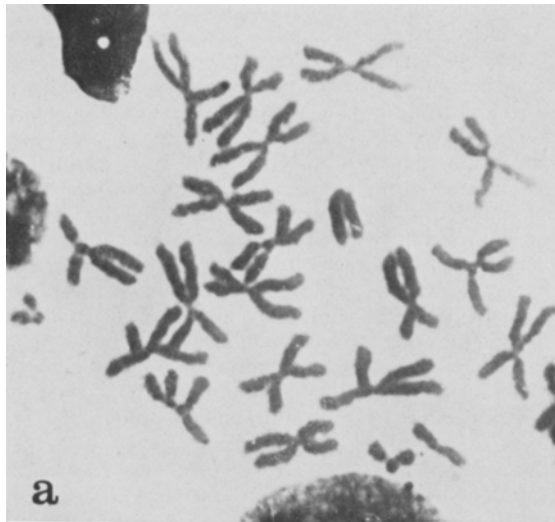
The population of 22-chromosome mice has a rather large area of distribution which includes part of the north-

east basin of the Tiber and stretches as far as the Adriatic coast. The southwest boundaries of this area have been ascertained by the finding of both 40-chromosome border populations and by a number of natural hybrid populations intermediate between these two karyological situations².

A survey of the meiotic diakineses in laboratory hybrids obtained by cross-breeding 40- and 22-chromosome mice has revealed the existence of 9 trivalents. The latter clearly display a correspondence between the autosomal arms of the metacentrics of the 22-chromosome karyotype and the single chromosomes of the 40-chromosome karyotype (Figure 1b).

It thus seemed worth while making an accurate identification of the elements involved in the Robertsonian fusion processes and comparing the arrangement of the mutation events in our Apennine material with the arrangement of those in other natural populations of *Mus musculus*³ and in *Mus poschiavinus* Fatio⁴. In our view, this approach has two interesting features. The first, and most obvious one, is the possibility of characterizing cytologically material that may be very useful in *in vitro* cytogenetic research both because of the low diploid number, which is in complete contrast with the high number of chromosome markers and because the species in question is *Mus musculus*, i.e. a species whose standard karyotype characters have been identified perfectly and whose linkage groups related to each chromosome are also well known⁵. The second feature is the possibility of adding further cytological data to the problem of the chromosomal 'polymorphism' of *Mus musculus* and its evolution.

For this purpose, primary cultures of fibroblasts were prepared in the cell culture laboratory of the Study Centre on Evolutionary Genetics of the National Research Council, starting from the kidneys of 2 hybrid female individuals obtained by crossing a 40-chromosome female with a 22-chromosome male in the laboratory. The *in vitro* cell lines had a 31-chromosome karyotype consisting of 20 chromosomes from the maternal set and 11 from



¹ E. CAPANNA, M. V. CIVITELLI and M. CRISTALDI, *Rc. Accad. naz. Lincei*, ser. 8, 54, 981 (1973).

² E. CAPANNA, M. V. CIVITELLI and M. CRISTALDI, *Theriologia*, Vladivostok, in press.

³ A. GROPP, H. WINKING, L. ZECH and H. MÜLLER, *Chromosoma* 39, 265 (1972).

⁴ A. GROPP, U. TETTENBORN and E. VON LEHMANN, *Cytogenetics* 9, 9 (1970).

⁵ Committee on Standardized Genetic Nomenclature for Mice, *J. Heredity* 63, 69 (1972).

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Fig. 1. a) Somatic metaphase from bone marrow of a 22-chromosome Apennine *Mus musculus*; b) first meiotic diakinesis of a hybrid mouse obtained crossbreeding a 22-chromosome male with a 40-chromosome female.

Species	Localities	2n	Metacentrics from Robertsonian translocations	References
<i>Mus musculus</i>	Central Apennine Italy	22	Rb (6.1) 1 Rma Rb (8.3) 2 Rma Rb (11.7) 3 Rma Rb (15.4) 4 Rma Rb (17.5) 7 Rma Rb (18.2) 6 Rma Rb (9.10) 5 Rma Rb (14.12) 8 Rma Rb (16.13) 9 Rma	Present paper
<i>Mus poschiavinus</i>	Val Poschiavo Rhaetian Alps	26	Rb (3.1) 1 Bnr Rb (6.4) 2 Bnr Rb (13.11) 4 Bnr Rb (15.5) 3 Bnr Rb (17.16) 7 Bnr	GROPP et al. 1972 ³
<i>Mus musculus</i>	Val Mesolecina Rhaetian Alps	28	Rb (3.1) 1 Rov = 1 Bnr Rb (14.2) 2 Rov Rb (11.10) 3 Rov = 9 Bnr Rb (12.4) 4 Rov	GROPP et al. 1972 ³
<i>Mus musculus</i>	Chiavenna Rhaetian Prealps	35	Rb (17.16) 7 Bnr Rb (11.10) 8 Bnr	GROPP et al. 1972 ³
<i>Mus musculus</i>	Albula Rhaetian Alps	38	Rb (12.4) 9 Bnr	GROPP et al. 1972 ³
<i>Mus musculus</i>	Laboratory strain	39	Rb (16.6) 1 Ald	LEONARD and DEKNUDT 1967 ⁹
<i>Mus musculus</i>	Laboratory strain	39	Rb (19.9) 163 H	EVANS et al. 1967 ¹⁰
<i>Mus musculus</i>	Laboratory strain	39	Rb (17.8) 1 Jem	BARANOV and DYBAN 1971 ¹¹
<i>Mus musculus</i>	Laboratory strain	39	Rb (5.19) 1 wh	WHITE and TIJO 1967 ¹²

the paternal set. For the characterization of the T-G banding pattern, the technique suggested by SEABRIGHT⁶ was followed. The results of the T-G banding analysis are shown in Figure 2 while the Table gives the arrangement of the acrocentrics in the 9 Robertsonian fusions of the 22-chromosome Apennine mouse. In this Table, the autosomal acrocentrics of *Mus musculus* identified by the T-G banding developed by WURSTER⁷ have been numbered from 1 to 19 in accordance with the classification of the Committee on Standardized Genetic Nomenclature for Mice⁸. The 9 metacentrics of the 22-chromosome mouse set have been given a symbol as recommended by the Committee on Standardized Genetic Nomenclature for Mice⁸; Rb stands for Robertsonian translocation, the 2 numbers in brackets for the chromosomes involved in this translocation; the progressive number refers to the karyotype element and the symbol Rma means Rome.

The Table also compares the Robertsonian fusions of the present Apennine material with those of *Mus poschiavinus* and of the other mountain populations of *Mus musculus*. The result of the banding is indicative *per se* of definite classification of the acrocentrics of the standard karyotype of the mouse in the autosomal arms of the Robertsonian metacentrics. As stated in the introductory part, this also allows the chromosome markers to be accurately characterized.

As for the comparison of the arrangement of the acrocentric autosomes of the standard karyotype of mouse in forming the Robertsonian metacentrics of the Apennine material of ours, with that occurring in the tobacco mouse (*Mus poschiavinus*) and in the mountain populations of *Mus musculus*, two evidences come out: 1. the non homology of the chromosomes having undergone Robertsonian transformation in the Apennine population is evident on comparing them with the metacentrics of *Mus poschiavinus*; 2. there does not seem to be any correspondence, not even between the Robertsonian transformations that characterize the karyotype of the *Mus musculus* populations from Apennine and Rhaetic Alps.

It follows that, like every mutational event, Robertsonian translocation is accidental. While it is true that *Mus musculus* tends to undergo centric fusions with relative ease as has been demonstrated also be the frequency with which Robertsonian events occur in laboratory strains⁹⁻¹² and in in vitro cell lines¹³, there is as yet no proof of the existence of preferential sites of centric fusion or of chromosomes prone to this kind of mutational event.

For the moment, only cautious hypotheses, as those already discussed elsewhere² by some of us (CAPANNA and CRISTALDI), can be put forward to explain the mechanisms leading to the formation of populations which are homozygous by Robertsonian metacentrics in restricted mountain areas. According to this model, the numerous isolations made possible by the compartmentalization of a mountain district favour inbreeding. As a result, genetic

⁶ M. SEABRIGHT, Lancet 2, 971 (1971).

⁷ D. H. WURSTER, Cytogenetics 11, 379 (1972).

⁸ Committee on Standardized Genetic Nomenclature for Mice, Meetings of 2nd, 9th and 20th November 1973 at Harwell, personal communication.

⁹ A. LEONARD and G. H. DEKNUDT, Nature, Lond. 214, 504 (1967).

¹⁰ E. P. EVANS, M. L. LAYON and M. DAGLISH, Cytogenetics 6, 105 (1967).

¹¹ V. S. BARANOV and A. P. DYBAN, Ontogenez 2, 164 (1971).

¹² B. J. WHITE and J. H. TIJO, Hereditas 58, 284 (1967).

¹³ S. OHNO, E. T. KOVACS and R. KINOSHITA, J. natn. Cancer Inst. 24, 1187 (1960).

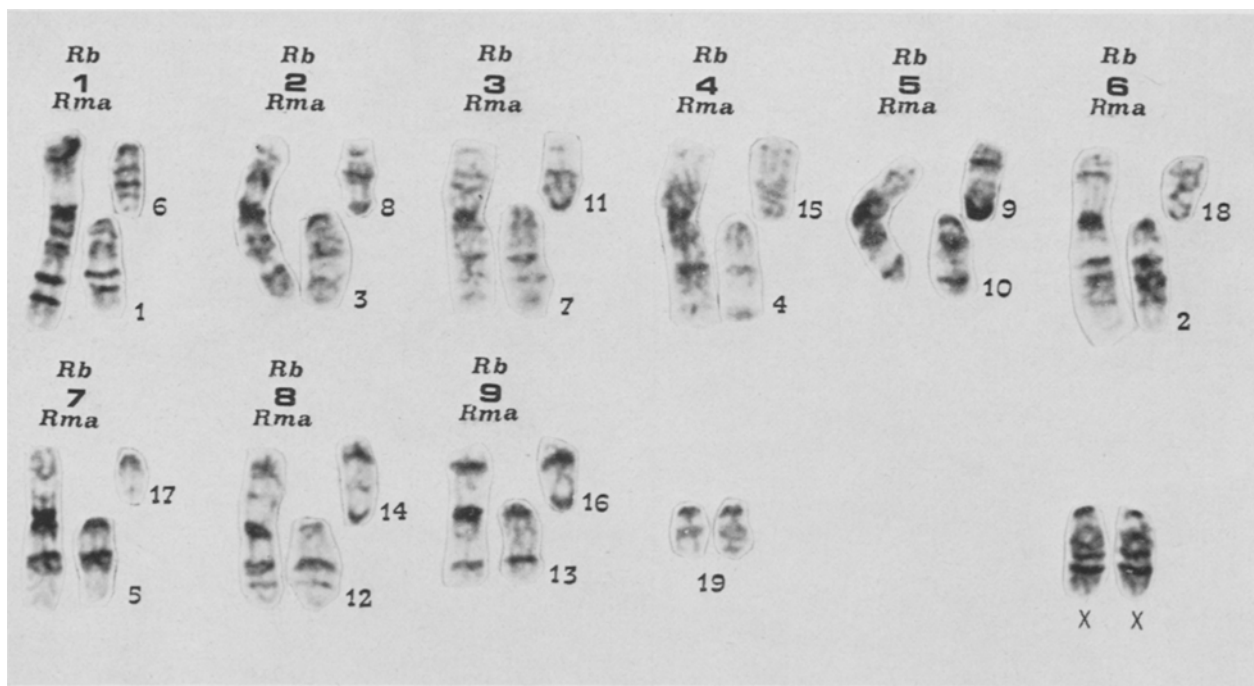


Fig. 2. The Trypsin-Giemsa-banding pattern of the karyotype of a hybrid mouse in which the haploid sets of both parental strains – i.e. 22- and 40-chromosomes – are shown.

drift allows the homozygous condition for chromosomes having undergone Robertsonian translocations to be set up in small isolated populations. The opposite is true in a densely anthropized cultivated plainland environment, or in towns, or in any case, where human activities favour large-scale exohybridization phenomena. Here any Robertsonian translocation mutants are at an immediate disadvantage as regards their reproductive rate because of their gametic aneuploidy¹⁴, and are thus quickly eliminated from the natural populations. Therefore, in order to exist in a natural population, a centric fusion must succeed in reaching a homozygous situation, a condition which is offered by isolation at the end of a valley in a mountainous district.

Riassunto. Le nove traslocazioni Robertsoniane, responsabili della trasformazione del cariotipo standard del topo nel cariotipo a 22 cromosomi caratteristico dei *Mus musculus* dell'Appennino Centrale, sono state caratterizzate individuando, attraverso la tecnica del T-G-banding, gli elementi acrocentrici coinvolti nel

processo di fusione centrica. Le fusioni Robertsoniane presenti nel popolamento appenninico sono differenti da quelle realizzate sia in *Mus poschiavinus* sia in *Mus musculus* di altre popolazioni Alpine.

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¹⁴ E. CAPANNA, *Chromosomes Today* 5, in press.

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Lactate Dehydrogenase Isozymes in Xiphophorin Fish Melanoma Conditioned by the Locus *Sd*.*

In the Xiphophorin fish the formation of melanomas occurs by hybridization- or mutation-conditioned depression of specific loci^{1,2}. Biochemical investigations on these genetically defined neoplasms should be able to add information on mechanisms of tumor formation. For the present study lactate dehydrogenase (LDH, EC 1.1.1.27) was chosen as reference enzyme. The LDH-isozyme pattern of various types of tumors is known to be different from that of normal tissues, as has been reviewed by CRISS³. In human malignant melanoma, however, no appreciable differences were detected in the LDH-isozymes between the malignant and the normal tissue⁴.

This investigation was undertaken to find out whether any changes in the LDH-isozyme pattern occur during the

* Supported by Deutsche Forschungsgemeinschaft through Sonderforschungsbereich 103, Zellenergetik und Zelldifferenzierung (Marburg).

¹ F. ANDERS, *Experientia* 23, 1 (1967).

² A. ANDERS, F. ANDERS and K. KLINKE, in *Genetics and Mutagenesis of Fish* (Ed. J. H. SCHRÖDER; Springer Verlag, Berlin/Heidelberg/New York 1973), p. 33.

³ W. E. CRISS, *Cancer Res.* 31, 1523 (1971).

⁴ R. PRASAD, M. M. ROMSDAHL, C. R. SHAW, D. M. MUMFORD and J. L. SMITH JR., *Cancer Res.* 34, 1435 (1974).