cally different from lichens growing in areas where the atmosphere is not polluted is well established (*3, 4, 7*). This study has demonstrated that *Parmelia sulcata*, a corticolous lichen, shows physiological as well as morphological abnormalities when growing near industrial centers, with both rate and pattern of photosynthesis being affected.

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Chromosomes of American Marsupials

Abstract. *Studies of the chromosomes of four American marsupials demonstrated that* Caluromys derbianus *and* Marmosa mexicana *have a diploid number of 14 chromosomes, and that* Philander opossum *and* Didelphis marsupialis *have a diploid number of 22. The karyotypes of* C. derbianus *and* M. mexicana *are similar, whereas those of* P. opossum *and* D. marsupialis *are dissimilar. If the 14-chromosome karyotype represents a reduction from a primitive number of 22, these observations suggest that the change has occurred independently in the American and Australasian forms.*

The order Marsupialia is represented by two major living groups, one in the American continents and the other in Australasia. The American group consists of 69 species (*1*), and the Australasian group consists of about 160 species (*2*). The studies on the chromosomes of the Australasian species are now fairly comprehensive, and they show that the diploid number ranges from 10 to 24, there being a bimodal distribution with peaks at 14 and 22. In contrast, the information on American species is totally unrepresentative; so far only three species have been studied and these all have a diploid number of 22 (*3, 4*). Here we report, first, that a diploid number of 22 is not an invariant property of American marsupials and that a diploid number of 14 occurs in at least two species, namely *Caluromys derbianus* (woolly opossum) and *Marmosa mexicana* (murine opossum), and second, that although several species may have a diploid number of 22 their karyotypes may vary. The latter point is illustrated by a comparison of the karyotypes of *Didelphis marsupialis* (common opossum) and *Philander opossum* (four-eyed opossum).

We have studied the chromosomes of five woolly opossums (three male and two female), four four-eyed opossums (three male and one female), one male murine opossum, and two common opossums (one male and one female). Although only a few individuals of each species were observed, the results are reported in view of the rarity of much of the material. Apart from the common opossums, all specimens were trapped alive in Nicaragua and flown to Philadelphia for observation. The common opossums were trapped in the environs of Philadelphia.

In all species, except the common opossum, chromosome preparations were made from one or more tissues; namely, bone marrow, lung, kidney, skin, and peritoneum. Bone marrow preparations were made directly (*5*). The other tissues were cultured by the method of Basrur, Basrur, and Gilman (*6*), with either Eagle's medium and 15 percent fetal calf serum or medium 1066 with 10 percent lactalbumin hydrolyzate (0.5 percent) and 20 percent inactivated calf serum. In the case of the common opossum, the preparations were made from short-term cultures of peripheral blood leucocytes (*7*), a modified BGJ culture medium (*8*) being used (0.22 percent sodium bicarbonate); the preparations were incubated at 34°C, since this temperature more closely approximates the mean body temperature of the opossum (*9*).

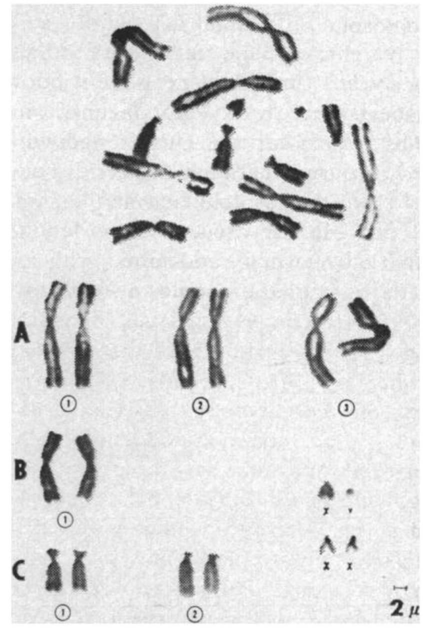

Fig. 1. Karyotype of a cell cultured from the tunica vaginalis from a male woolly opossum (*10*). Sex chromosomes XX from a female wooly opossum are included.

Figure 1 shows a metaphase spread and the karyotype from a male woolly opossum (*10*). Consistent results were obtained within and among animals with respect to chromosome number and karyotype. The diploid number is clearly 14. The karyotype is composed of three pairs of large sub-metacentric autosomes (group A), one pair of medium-metacentric autosomes (group B) and two pairs of medium sub-telocentric autosomes (group C). The X

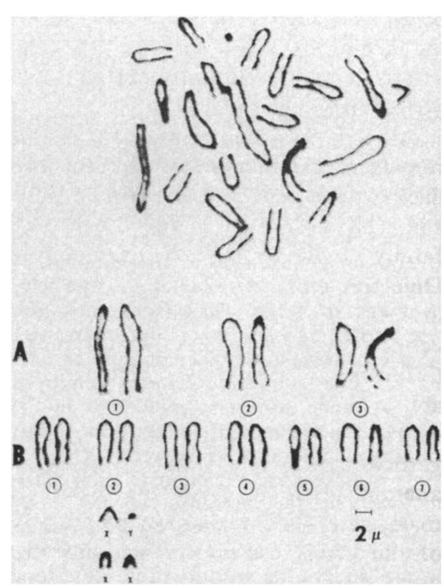

Fig. 2. Karyotype of a cell cultured from the peritoneum of a male four-eyed opossum. The sex chromosomes XX from a female four-eyed opossum are included.

chromosome is a small acrocentric and the Y chromosome is a very small telocentric. Thus the sex is controlled genetically by an XY/XX mechanism.

The results for the murine opossum were consistent between cells, and suggest that the diploid number is 14 and that the karyotype is very similar to that of the woolly opossum.

The metaphase spread and karyotype from a male four-eyed opossum (Fig. 2) indicate that the diploid number for this species is 22. However, the karyotype is different from that of the common opossum. The four-eyed opossum has three pairs of large telocentric autosomes (group A) and seven pairs of medium telocentric autosomes (group B). The X chromosome is a small telocentric and the Y chromosome is a very small telocentric, sex being controlled by an XY/XX mechanism. Our observations on the karyotype of the common opossum, which confirm those of Shaver (*4*), show that there are six pairs of subtelocentric autosomes, and four pairs of telocentric autosomes. The X chromosome is a medium sub-metacentric and the Y chromosome is a small telocentric.

Our observations show that the Didelphidae are not characterized by the uniform diploid number of 22 assumed by Sharman (*2*), and suggest that the variation in diploid number among American marsupials may be as great as that observed in Australasian marsupials, the numbers 14 and 22 predominating in both groups. Moreover, our results indicate that within the group of American species characterized by 22 chromosomes the karyotype may nevertheless vary.

Among the many questions which remain is that of whether the Didelphidae are a closely related group. Simpson (*11*) stated that only one subfamily of the Didelphidae, namely, the Didelphinae, is still extant. However, in 1955 Reig (*12*) presented osteological evidence that the living genera *Caluromys*, *Dromiciops*, and *Glironia* are in fact surviving members of the subfamily Microbiotheriinae usually regarded as extinct. Other comparative anatomical studies have shown that the male and female reproductive organs of the genus *Caluromys* have features quite distinctive from those of *Didelphis* and many similar forms. These features are the median vagina (*13*), the cleft glans penis (*14*), and the morphology of the spermatozoa (*15*). The little evidence available at present indicates that although one genus of the possibly extant Microbiotheriinae has a diploid number of 14 chromosomes, it may not be unique in this aspect since *Marmosa* also has a diploid number of 14 chromosomes. However, the diploid number of chromosomes and the karyotypes of *Dromiciops* and *Glironia* must be studied before further comments can be made on this matter. In addition, detailed studies are required on the chromosomes of the genus *Marmosa*, a very extensive group of the didelphids which are taxonomically divided into five subgeneric categories (*16*).

At present we have no information on the chromosomes of the family Caenolestidae which lives in the Andes Mountains of South America (*17*). This very rare family is of particular interest from an evolutionary point of view because of its alleged affinities with the superfamily Phalangeroidea of Australasia (*18*), which is characterized by a range of chromosome numbers (*2*).

Sharman and Barber (*19*) suggested that during the evolution of marsupials the chromosome numbers have been reduced by some type of Robertsonian chromosomal change. Various plausible patterns of translocation can be postulated whereby 22 chromosomes can be reduced to 14. However, the suggestion that a diploid number of 22 is the primitive number of chromosomes for marsupials can no longer be sustained by the argument that American marsupials, considered evolutionarily primitive on other grounds, are characterized by a diploid number of 22. If the Robertsonian change has occurred in marsupials, then it has probably occurred independently in the American and Australasian stock.

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Sex-Linkage of Glucose-6-Phosphate Dehydrogenase in the Horse and Donkey

Abstract. *Distinctly different electrophoretic patterns of red cell glucose-6-phosphate dehydrogenase were resolved from the hemolyzates of horse and donkey erythrocytes. Examination of their reciprocal hybrids, mules and hinnies, showed that the red cells of female mules and female hinnies contain both horse and donkey G-6-PD; the male mule with an X chromosome from its horse mother contained pure horse G-6-PD, whereas the male hinny with the donkey X chromosome contained pure donkey G-6-PD. These findings on the male reciprocal hybrids suggest X-linkage.*

The production of glucose-6-phosphate dehydrogenase (G-6-PD) in erythrocytes is governed in humans by a gene linked to the X chromosome, and genetic variants detectable by starch-gel electrophoresis are known (*1, 2*). Usually the presence of wild-type alleles is revealed by finding their deficient mutants; chance plays a great role in detection of such mutants. For instance, electrophoretic variants of