

CHROMOSOMAL HOMOLOGY AND DIVERGENCE BETWEEN SIBLING SPECIES OF DEER MICE: *PEROMYSCUS MANICULATUS* AND *P. MELANOTIS* (RODENTIA, CRICETIDAE)

IRA F. GREENBAUM<sup>1</sup>, ROBERT J. BAKER<sup>1</sup> AND J. HOYT BOWERS<sup>2</sup>

Received February 14, 1977. Revised July 11, 1977

From a chromosomal standpoint, speciation in the *Peromyscus maniculatus* complex has been accompanied by considerable change in the number of arms in the autosomal complement. This report is concerned with a determination of the chromosomal homologies and types of chromosomal rearrangements between two members of this complex, *P. melanotis* and *P. maniculatus*, as determined by G- and C-band comparisons. This study was undertaken to document the level of chromosomal evolution that is characteristic of some sibling species as well as to provide some insight into the evolutionary history of these two species.

The diploid number for all species of *Peromyscus* thus far described is 48. Numerous authors have, however, documented extensive chromosomal variation dealing with the number of chromosome arms in *Peromyscus* (Hsu and Arrighi, 1966; Ohno et al., 1966; Singh and McMillan, 1966; Sparkes and Arakaki, 1966; Hsu and Arrighi, 1968; Arakaki et al., 1970; Te and Dawson, 1971; Bradshaw and Hsu, 1972; Lee et al., 1972; Bowers et al., 1973; Pathak et al., 1973; Arrighi et al., 1976; Schmidly and Schroeter, 1974; Murray and Kitchin, 1976). Hsu and Arrighi (1968) described the karyotype for 19 species of *Peromyscus* and reported both inter- and intraspecific variation of 56-96 in the total number of chromosome arms. Bradshaw and Hsu (1972) documented chromosomal variation in 15 subspecies of

*P. maniculatus* and suggested that where dual modes were evident such as in *P. m. rufinus* of southeastern Arizona that these populations might be representative of another species (e.g., *P. melanotis*). Bowers et al. (1973) and Bowers (1974) have shown by karyotypic, electrophoretic, and breeding data that this is indeed the case.

It has been most convenient and practical to describe chromosomal variation in *Peromyscus* in terms of the number of biarmed versus the number of acrocentric elements in the autosomal complements, although in some cases it is difficult to separate the two categories. Bradshaw and Hsu (1972) chose to describe variation in *P. maniculatus* in terms of the number of biarmed elements in the autosomal complement, whereas Bowers et al. (1973) chose to employ the number of autosomal acrocentrics for their descriptions. We have chosen the latter for our discussion. A summary of the current view of chromosomal variation in *P. maniculatus* is presented in our discussion.

#### MATERIALS AND METHODS

All preparations were from fibroblast tissue-cultured cells initiated from ear biopsies. Cultures were grown at 37 C in Ham's F-10 medium supplemented with 11% fetal calf serum. Velban (Lilly) was used as a mitotic inhibitor (0.03-0.1%, for 15-30 min). The cells were subsequently incubated in a hypotonic solution (approximately 27% growth medium in distilled water) for 15 min. After multiple washes in Carnoy's fixative (3 methanol: 1 acetic acid) the cell suspension was dropped into 40% acetic acid on cleaned slides and air dried.

<sup>1</sup>Department of Biological Sciences and The Museum, Texas Tech University, Lubbock, Texas 79409.

<sup>2</sup>Department of Biology, Wayland Baptist College, Plainview, Texas 79702.

Slides for G-banded chromosomal preparations were incubated overnight at 68 C. Slides were then treated in 0.05% trypsin (in Hanks balanced salt solution, pH 8.0) for 1–4 min, rinsed in baths of Hanks solution, 70% ethanol, 95% ethanol and stained in 2% Giemsa in 0.1 M phosphate buffer (pH 6.8) for 7 min.

The C-band technique was performed on chromosomal preparations which were allowed to sit overnight at room temperature. Slides were immersed in 0.2 N HCl for 20 min, treated in 5% Ba(OH)<sub>2</sub> at 46 C for 2–10 min, rinsed first in the 0.2 N HCl and then in distilled water, incubated in either 1X or 6X SSC at 60 C for 45 min, and stained in 2% Giemsa in 0.1 M phosphate buffer for 10 min.

The G-banded chromosomes (Figs. 1–5) are numbered and arranged according to the proposed standardized karyotype for *Peromyscus* (Committee, 1977). The C-banded karyotypes (Figs. 1b and 2b) are tentatively arranged to correspond with the G-banded figures.

Specimens examined were as follows: *P. maniculatus bairdii* (3) Iowa: Bremer Co.; 1 mi S, 0.5 mi E Plainfield (these individuals were laboratory F<sub>1</sub>s of wild trapped individuals from the above locality), *P. melanotis* (1) Mexico: Durango; 29.1 mi W El Salto (laboratory F<sub>1</sub> of wild trapped individuals), (1) Mexico: Durango; 29.6 mi W El Salto (laboratory F<sub>2</sub> of wild trapped individuals).

## RESULTS

*The P. maniculatus karyotype.*—Specimens of *P. maniculatus bairdii* used in this study had a diploid number of 48. The number of acrocentric elements varied from nine to 10 (Fig. 1). This intra-specific variation resulted from polymorphism in two pairs of autosomes. As detected by G-banding, pair 6 was homomorphic for the biarmed condition in two of the specimens examined (Fig. 1a), and heteromorphic in the other specimen, one

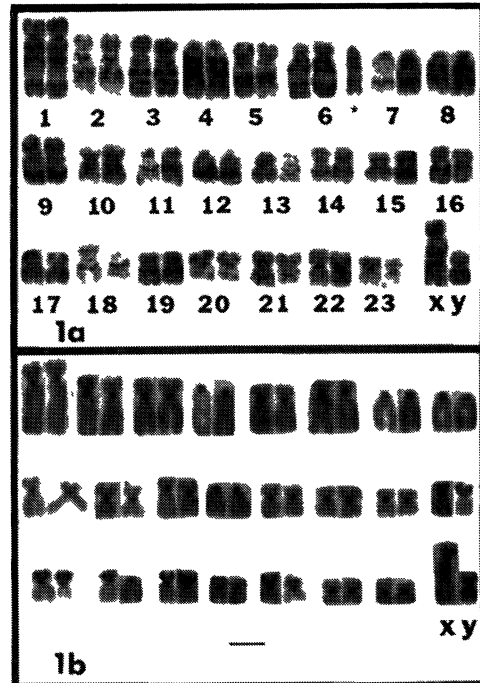


FIG. 1. a) G-banded and b) C-banded karyotypes of *Peromyscus maniculatus*. \* Indicates acrocentric condition for pair 6. Scale equals 10 microns in all figures.

homologue being biarmed and the other acrocentric (Fig. 1a\*). While this chromosome cannot be specifically distinguished from other large biarmed chromosomes in C-banded cells, it is clear that the short arms are not heterochromatic because none of the large biarms have heterochromatic arms (Fig. 1b).

Chromosome pair 18 in *P. maniculatus* was heteromorphic in two of the specimens examined (Fig. 1a) and homomorphic in the third. The long arm of the biarmed chromosome appears to be homologous to an acrocentric element in the G-banded preparation (Fig. 1a). Examination of the C-bands reveals that the short arm of the biarmed condition in this pair is heterochromatic (Fig. 1b). One of the three *P. maniculatus* analyzed was heteromorphic for both pairs involved in the observed polymorphism.

The X chromosome in the *P. maniculatus* examined was a large submetacentric with a G-banding pattern of two trypsin resistant bands (Fig. 1a). C-banding revealed that the short arm of the X is heterochromatic (Fig. 1b). The Y chromosome of the *P. maniculatus* studied was a medium-sized metacentric with no distinct G-bands (Fig. 1b).

The C-banded karyotype of *P. maniculatus* presented in Fig. 1b reveals that nine of the medium-sized banded chromosomes have short arms which are heterochromatic. The specimen with eight acrocentric autosomes correspondingly had 10 elements with heterochromatic short arms. In most of these cases the heterochromatin appears telomeric and is separated from the centromere by a small region of chromosomal material which does not stain darkly in our preparations. The visibility of these supposedly euchromatic regions varied in our material due to the degree of contraction and quality of the C-band preparation.

*The P. melanotis karyotype.*—Both specimens of *P. melanotis* examined had a diploid number of 48 (Fig. 2). The number of acrocentric autosomes could be interpreted as being either 28 or 30, depending upon whether pair 12 is considered to be banded or acrocentric. Although this pair appears to be submetacentric, it is possible that using standard karyotypic preparations this pair of chromosomes would appear more acrocentric. It is only in our C-banded preparation (Fig. 2b) that we were readily able to distinguish the short arm in this chromosome pair. This condition may well explain the one specimen of *P. melanotis* (*P. maniculatus rufinus* I. by Bradshaw and Hsu, 1972) reported as having 28 acrocentric (18 banded) autosomal elements. As determined by both G- and C-banded karyotypes no chromosomal polymorphism was detected in *P. melanotis*.

The X chromosome of *P. melanotis* is a large submetacentric and the Y is a me-

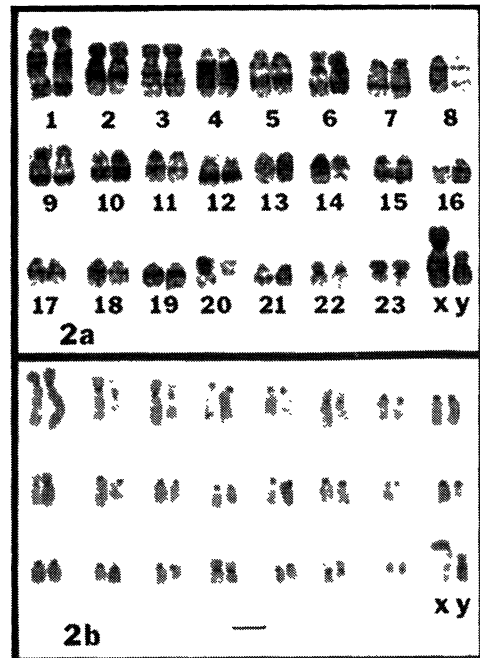


FIG. 2. a) G-banded and b) C-banded karyotypes of *Peromyscus melanotis*.

dium sized submetacentric chromosome (Fig. 2). The X chromosome has a larger short arm than in *P. maniculatus*. The euchromatic long arm of the X chromosome of *P. melanotis* has the same G-banded pattern as that seen in *P. maniculatus* (Fig. 3). The Y chromosome of *P. melanotis* differs from that of *P. maniculatus* by showing a trace of trypsin resistant G-band just proximal to the centromere in the long arm (Figs. 2a and 3).

C-banding revealed relatively little heterochromatin in the *P. melanotis* karyotype (Fig. 2b). Centromeric heterochromatin can be seen associated with all of the autosomal elements. Of the banded chromosomes, only the X has a heterochromatic short arm. The Y chromosome is entirely heterochromatic although there apparently is a somewhat darker C-band staining region just proximal to the centromere in the long arm corresponding to the G-band in this same region.

*Interspecific homology.*—Figures 3–5

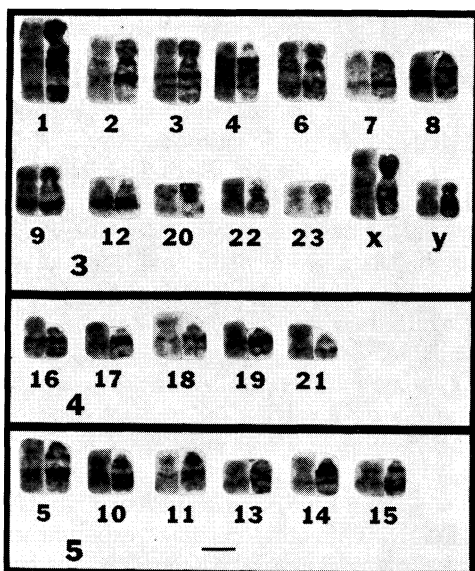


FIG. 3. Composite partial karyotype comparing the G-banded chromosomes of *P. maniculatus* and *P. melanotis*, showing the elements unchanged between the two species. In each pair the chromosome on the left is from *P. maniculatus* and the one on the right is from *P. melanotis*.

FIG. 4. Composite partial karyotype comparing the G-banded chromosomes of *P. maniculatus* and *P. melanotis*, showing those elements which differ by the presence (in *P. maniculatus*) or absence (in *P. melanotis*) of heterochromatin. Arrangement of chromosomes is as in Fig. 3.

FIG. 5. Composite partial karyotype comparing the G-banded chromosomes of *P. maniculatus* and *P. melanotis*, showing those elements which differ by a pericentric inversion. Arrangement of chromosomes is as in Fig. 3.

are composite karyotypes comparing G-banded chromosomes of *P. maniculatus* and *P. melanotis*. In the composite figures each matched pair has a *P. maniculatus* chromosome on the left and what we believe to be the homologous element of *P. melanotis* on the right. Despite the apparent differences in the gross karyotypes of these two species, with the aid of G- and C-banding techniques it is possible to identify all of the homologous elements between the species.

From study of the G- and C-banded

chromosomes shown in Figs. 1–5 and those from other preparations examined, we have concluded that the karyotypes of *P. maniculatus* and *P. melanotis* are related as described below.

Twelve pairs of autosomes, including five pairs of large biarmed elements, three pairs of medium to small biarmed elements, and four pairs of acrocentric elements are homologous and unchanged in G- and C-banding patterns between the two species (Fig. 3).

Five pairs of medium to small sized biarmed autosomes (Fig. 4) appear to differ only in the presence or absence of heterochromatic short arms. In these five pairs, the *P. maniculatus* chromosomes have heterochromatic short arms, and the respective four pairs of acrocentric elements of the *P. melanotis* karyotype appear to be homologous to the euchromatic long arm.

Six pairs of chromosomes appear to differ by pericentric inversions between the two species (Fig. 5). These pairs are biarmed in the *P. maniculatus* examined and acrocentric in the *P. melanotis*. C-banding reveals that constitutive heterochromatin is confined to the centromeric regions of these chromosomes in both species (Figs. 1b and 2b); short arm addition or deletion, therefore, would not be expected for these pairs. Comparative analysis of the G-banding pattern in these chromosomes (Fig. 5) indicates that if the centromere and short arm of the *P. maniculatus* chromosome is inverted so that the centromere becomes terminal or near terminal, the result is a G-banding pattern similar to that seen in the corresponding acrocentric elements of *P. melanotis* (the reverse process, a pericentric inversion converting an acrocentric to biarm chromosome, would, of course yield the same situation).

Two conditions were detected for pair 6 in *P. maniculatus* (Fig. 1a) as described above. As shown in Fig. 3, the biarmed condition of this pair corresponds to a biarmed element of *P. melanotis*. The rela-

relationship between the acrocentric and biarmed condition within *P. m. bairdii* may involve a pericentric inversion or the presence of undetected heterochromatin (see discussion below).

The other heteromorphic pair detected in *P. maniculatus* (pair 18, Fig. 1) is an intraspecific case of the type of variation depicted in Fig. 4. The biarmed condition of this chromosome in *P. maniculatus* has a heterochromatic short arm (Fig. 1b) and the acrocentric correlate is apparently homologous to the long arm of this chromosome (Fig. 1a). In *P. melanotis* only the acrocentric condition occurs (Fig. 2a) and it corresponds to the acrocentric condition in *P. maniculatus*.

The X chromosomes of *P. maniculatus* and *P. melanotis* are compared in Fig. 3. The X in *P. maniculatus* is subtelocentric, whereas in *P. melanotis* it is submetacentric. In both species the short arm of the X is heterochromatic. Based on the G-banded preparations, the euchromatic long arms of the X chromosomes appear homologous (Fig. 3). The Y chromosomes in the two species (Fig. 3), although somewhat different in the position of the centromere, are relatively similar. In both species, the Y is heterochromatic based on C-band staining.

#### DISCUSSION

As currently understood (Bowers et al., 1973), karyotypic variation in *P. maniculatus* ranges from 20 acrocentric autosomes in *P. m. oreas*, *P. m. rubidus* and *P. m. gambelii* (Bradshaw and Hsu, 1972) to four acrocentric autosomes in one specimen of *P. m. austerus* from Thurston Co., Washington. Bowers et al. (1973) reported karyotypes for 40 specimens of *P. melanotis* (including specimens previously considered *P. maniculatus rufinus* from southeastern Arizona) with a diploid number of 48 which contained 30 acrocentric autosomes. Bradshaw and Hsu (1972) reported a karyotype with 28 acrocentric autosomes for one specimen of *P. mela-*

*notis* (reported as *P. maniculatus rufinus* I.). We also believe that the two specimens reported by Bradshaw and Hsu (1972) as *P. m. blandus* from Saltillo having 30 acrocentric autosomes were specimens of *P. melanotis*. Again, the classification of biarmed versus acrocentric chromosomes, however, is less important when banding data are available and the limitations of such a classification should be recognized when banding data are not available.

Chromosomal variation in *P. maniculatus bairdii* involving the number of acrocentric autosomal elements (ranging from 8–12) has previously been reported by Ohno et al. (1966) and Bradshaw and Hsu (1972). Through breeding experiments and standard karyology, Ohno and his co-workers described polymorphisms in two pairs of chromosomes. Examination of the figures presented in Ohno et al. (1966) suggests that it is likely that at least one and possibly both of the polymorphisms described by these authors are the same as those examined in this study. The lack of consistency in the relative size designation of the polymorphic chromosomes in their study and ours probably is the result of the difficulty in pairing chromosomes from standard karyotypic preparations. Ohno et al. (1966) suggest pericentric inversions as an explanation for both of these polymorphisms and hypothesize that *Peromyscus* must have evolved a mechanism which permanently isolates the regions of pericentric inversions from crossing over.

The intraspecific variation seen in pair 6 of *P. m. bairdii* does not appear to involve a heterochromatic short arm. Because euchromatic segments are not thought to be readily lost with meiotic impunity, euchromatic material of the short arm of the biarmed chromosome probably has not been lost. It appears from the banding patterns (Fig. 1a) that a pericentric inversion involving this small euchromatic region may be stable in these animals. Normally, crossing over in the

inverted region of such a pericentric inversion heterozygote would result in production of genetically imbalanced gametes and subsequent zygotic loss. This, with time, would result in the populational loss of such a rearrangement. In this case it is hypothesized that three factors might work to reduce the amount of crossing over in the inverted segment: 1) crossing over is repressed in proximity to the centromere, 2) regions adjacent to heterochromatic blocks experience reduced chiasma frequency and 3) inversions tend to decrease the frequency of crossing over in adjacent regions. It is likely, therefore, that crossing over in this segment, if inverted, would be very infrequent. This might enable the pericentric inversion to remain in the population without being selected against by meiotic mechanisms.

An alternative explanation for the variation in this pair of chromosomes is that the segment distal to the dark band on the short arm, proximal to the centromere, has been translocated to another chromosome. Since this segment is small and lacks distinguishing G-band markers, a translocation such as this would be difficult to detect. Such a translocation should result in the occasional formation of trivalents in meiosis, and in the absence of a preferential disjunction mechanism, would result in the production of unbalanced gametes and subsequent zygotic loss.

A third explanation for the variation seen in pair 6 is that the short arm is heterochromatic and deleted (or added) but that our techniques have been unable to demonstrate a differential C-positive banding pattern associated with the segment.

The intraspecific polymorphism detected in pair 18 in our sample of *P. m. bairdii* (Figs. 1a and 1b) does not appear to involve a pericentric inversion. In the biarmed condition of this autosomal pair the short arm is heterochromatic (Fig. 1b) whereas the corresponding acrocentric homologue apparently lacks this heterochromatic block. This variation then ap-

pears to be the result of an addition or deletion of heterochromatin.

Although not detected in this study, it is obvious that an individual of *P. m. bairdii*, homozygous for the acrocentric condition in both autosomal pairs that were polymorphic in our sample, would have 12 acrocentric autosomes. Similarly, an individual homozygous for the biarmed conditions would have only eight acrocentric autosomal elements. We have no way of knowing at this point whether or not the other four autosomal pairs with heterochromatic short arms are polymorphic in *P. m. bairdii*. If all are involved, the potential variation in this subspecies alone would range from 8–18 acrocentric autosomes, nearly the entire range reported for the species.

It is apparent that much of the intraspecific chromosomal variation in *P. maniculatus* involves the presence (or absence) of heterochromatic regions. The presence of a pair of chromosomes heteromorphic for a heterochromatic short arm seemingly had no adverse effects on the specimens we examined. Only in the special case discussed above concerning the variation in pair 6 in which a very small second arm may have undergone inversion and obtained stability in the chromosomal complement, and a case reported by Murray and Kitchin (1976), is heterochromatin not known to be involved in intraspecific autosomal polymorphism thus far studied in *P. maniculatus*. We cannot determine from the data presented by Murray and Kitchin (1976) whether the inversion reported by these authors is homologous to that in our sample.

Several points on the chromosomal evolution in *Peromyscus* have been debated by numerous authors. Hsu and Arrighi (1968), using standard karyology, suggested that interspecific variation was the result of pericentric inversions and translocations. Several authors have discussed the role of heterochromatin in *Peromyscus* chromosome evolution (Bradshaw and

Hsu, 1972; Duffey, 1972; Pathak et al., 1973). Pathak et al. (1973) reported that the chromosomal differences between *P. eremicus* and *P. crinitus* result from differences in heterochromatic segments. These authors and Lawlor (1974) have gone so far as to suggest that all chromosomal variation in this genus results from addition or deletion of heterochromatic arms.

Arrighi et al. (1976) compared G- and C-banding data for *P. eremicus*, *P. crinitus* and *P. leucopus*. In *P. leucopus* the short arms of all of the banded chromosomes are not heterochromatic and interstitial heterochromatin was reported in some of the long arms of banded and acrocentric autosomes. G-banding revealed five pericentric inversion alterations between the chromosomal complements of *P. leucopus* and *P. crinitus*.

Our composite banded karyotypes (Figs. 3-5) show that all of the euchromatic material can be accounted for between *P. maniculatus* and *P. melanotis*. The differences in the karyotypes for these two species can be explained by two types of chromosomal alteration mechanisms: addition (or deletion) of heterochromatic segments, and pericentric inversions. The number of pericentric inversions between *P. crinitus* and *P. leucopus* (Arrighi et al., 1976) in one case and between *P. melanotis* and *P. maniculatus* in another, clearly show that addition (or deletion) of heterochromatic arms is not the sole source of chromosomal evolution in the genus. Karyotypic orthoselection (White, 1973) within the genus *Peromyscus* probably involves both types of these chromosomal rearrangements.

#### SUMMARY

Chromosomal homology and amount and location of heterochromatin, as revealed by G-banding and C-banding analyses, were examined for *Peromyscus maniculatus bairdii* and *P. melanotis*. These procedures enabled determination of interspecific ho-

mology of all euchromatic autosomal segments. Autosomally, the karyotypes of *P. maniculatus bairdii* and *P. melanotis* are related as follows: 12 pairs of chromosomes are unchanged, five pairs differ in the presence (in *P. m. bairdii*) and absence (in *P. melanotis*) of heterochromatic short arms, and six pairs differ by pericentric inversions. The acrocentric condition of two pairs of chromosomes found to be polymorphic in *P. maniculatus* is homologous to the acrocentric condition of the corresponding pairs in *P. melanotis*.

Intraspecific polymorphism in our sample was restricted to two pairs of autosomes in *P. m. bairdii*. One polymorphism involves heteromorphism for a heterochromatic short arm, whereas the other may be either the result of a pericentric inversion, translocation of a small segment, or polymorphism for undetected heterochromatin.

In *P. maniculatus* heterochromatin is centromeric on all chromosomes. Additionally, the short arms of five autosomal pairs are heterochromatic (one pair is reported polymorphic for the heterochromatic short arm). In both species, the short arm of the X and the Y are heterochromatic. The amount of C-band material in *P. melanotis* is reduced relative to that characteristic of the *P. maniculatus* examined. Autosomal heterochromatin in *P. melanotis* is confined to the centromeric regions.

Heterochromatic arms and pericentric inversions are important in the evolution of the karyotype of the genus.

#### ACKNOWLEDGMENTS

Dr. Oscar Ward gave assistance in the numerical designation of the G-banded chromosomes in comparison to the standardized karyotype for *Peromyscus*. We thank R. A. Bass, R. L. Honeycutt, and M. A. Johnson for their assistance. We acknowledge Dr. D. Schlitter and the Carnegie Museum for assistance in obtaining the parental stocks of *Peromyscus maniculatus* from which our specimens were

reared. J. W. Bickham, T. C. Hsu, R. C. Jackson, J. T. Mascarello, J. C. Patton, S. Pathak, and D. J. Schmidly critically reviewed this work. This study was made possible through a Graduate student-Faculty research grant from the Graduate School, Texas Tech University, by National Science Foundation grant DEB 76-20580 and by a grant from the Johnson Fund of the American Philosophical Society to J. Hoyt Bowers.

## LITERATURE CITED

- ARAKAKI, D. T., I. VEOMETT, AND R. S. SPARKES. 1970. Chromosome polymorphism in deer mouse siblings (*Peromyscus maniculatus*). *Experientia* 26:425-426.
- ARRIGHI, F. E., A. D. STOCK, AND S. PATHAK. 1976. Chromosomes of *Peromyscus* (Rodentia, Cricetidae). V. Evidence of pericentric inversions. *Chromosomes Today* 5:323-329.
- BOWERS, J. H. 1974. Genetic compatibility of *Peromyscus maniculatus* and *Peromyscus melanotis*, as indicated by breeding studies and morphometrics. *J. Mamm.* 55:720-737.
- BOWERS, J. H., R. J. BAKER, AND M. H. SMITH. 1973. Chromosomal, electrophoretic, and breeding studies of selected populations of deer mice (*Peromyscus maniculatus*) and black-eared mice (*P. melanotis*). *Evolution* 27:378-386.
- BRADSHAW, W. N., AND T. C. HSU. 1972. Chromosomes of *Peromyscus* (Rodentia, Cricetidae) III. Polymorphism in *Peromyscus maniculatus*. *Cytogenetics* 11:436-451.
- COMMITTEE FOR THE STANDARDIZATION OF CHROMOSOMES OF *PEROMYSCUS*. 1977. Standardized karyotype of the deer mouse, *Peromyscus* (Rodentia). *Cytogenet. Cell Genet.* 19:38-43.
- DUFFEY, P. A. 1972. Chromosome variation in *Peromyscus*: A new mechanism. *Science* 176:1333-1334.
- HSU, T. C., AND F. E. ARRIGHI. 1966. Chromosomal evolution in the genus *Peromyscus* (Cricetidae, Rodentia). *Cytogenetics* 5:355-359.
- . 1968. Chromosomes of *Peromyscus* (Rodentia, Cricetidae) I. Evolutionary trends in 20 species. *Cytogenetics* 7:417-446.
- LAWLOR, T. E. 1974. Chromosomal evolution in *Peromyscus*. *Evolution* 28:688-691.
- LEE, M. R., D. J. SCHMIDLY, AND C. C. HUBEY. 1972. Chromosomal variation in certain populations of *Peromyscus boylii* and its systematic implications. *J. Mamm.* 53:697-707.
- MURRAY, J. D., AND R. M. KITCHIN. 1976. Chromosomal variation and heterochromatin polymorphisms in *Peromyscus maniculatus*. *Experientia* 32:307-309.
- OHNO, S., C. WELLER, J. POOLE, L. CHRISTIAN, AND C. STENIUS. 1966. Autosomal polymorphism due to pericentric inversions in the deer mouse (*Peromyscus maniculatus*) and some evidence of somatic segregation. *Chromosoma, Berl.* 18:177-187.
- PATHAK, S., T. C. HSU, AND F. E. ARRIGHI. 1973. Chromosomes of *Peromyscus* (Rodentia, Cricetidae) IV. The role of heterochromatin in karyotypic evolution. *Cytogenet. Cell Genet.* 12:315-326.
- SCHMIDLY, D. J., AND G. L. SCHROETER. 1974. Karyotypic variation in *Peromyscus boylii* (Rodentia: Cricetidae) from Mexico and corresponding taxonomic implications. *Syst. Zool.* 23:333-342.
- SINGH, R. P., AND D. B. McMILLAN. 1966. Karyotypes of three subspecies of *Peromyscus*. *J. Mamm.* 47:261-266.
- SPARKES, R. S., AND D. T. ARAKAKI. 1966. Intraspecific and interspecific chromosomal polymorphism in *Peromyscus maniculatus* (deer mouse). *Cytogenetics* 5:411-418.
- TE, G. A., AND W. D. DAWSON. 1971. Chromosomal polymorphism in *Peromyscus polionotus*. *Cytogenetics* 10:225-234.
- WHITE, M. J. D. 1973. *Animal Cytology and Evolution*. 3rd Ed. vii + 961 p. Cambridge Univ. Press.