THE UNIVERSITY OF OKLAHOMA

GRADUATE COLLEGE

CHROMOSOME EVOLUTION IN THE GENUS DIPODOMYS AND ITS PHYLOGENETIC IMPLICATIONS

A DISSERTATION

SUBMITTED TO THE GRADUATE FACULTY

in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

BY

A. DEAN STOCK

Norman, Oklahoma

1971

CHROMOSOME EVOLUTION IN THE GENUS DIPODOMYS AND ITS PHYLOGENETIC IMPLICATIONS

APPROVED BY

DISSERTATION COMMITTEE

ACKNOWLEDGEMENTS

The major portion of this study was supported by a National Defense Education Act Title IV Fellowship. I thank Drs. John Keever Greer, Cluff E. Hopla, George J. Goodman, Gary D. Schnell, and James R. Estes for critically reading the manuscript and for their helpful suggestions. I also thank Drs. T. C. Hsu, John Neil Jensen, and especially Howard S. Shellhammer for providing me with specimens and representative karyotypes for some species, and Troy Best, Keeneth Heacock, Bert Ritter, and J. K. Greer for help in the field. Dr. E. Raymond Lee offered helpful suggestions during the early phases of the study. I am especially grateful to Professor Cluff E. Hopla for his support, criticism, and constant encouragement during my tenure at the University of Oklahoma. To my wife and sons I offer this apology for the years spent "in waiting" and thank them for their unceasing support of my endeavors.

TABLE OF CONTENTS

LIST OF TABLES

TABLE

LIST OF ILLUSTRATIONS

Figure

Figure

CHROMOSOME EVOLUTION IN THE GENUS DIPODOMYS AND ITS PHYLOGENETIC IMPLICATIONS

CHAPTER I

INTRODUCTION

The use of karyotypes has recently become increasingly useful in establishing phylogenetic relationships and in helping to solve taxonomic problems. The development of modern cell culture and colchicine hypotonic-citrate techniques has given a high level of reliability to mammalian cytosystematics not possible with previous methods and has resulted in a rapid accumulation of karyological data in most mammalian groups. Within the rodent family Heteromyidae, the subfamily Perognathinae has been studied by Patton (1967), and the diploid number and chromosome morphology is known for most species. According to Patton (1967:36) the evolution of distinctive karyotypes has paralleled the evolution of morphological features in the pocket mice, and his arrangement of the species groups largely supports other arrangements based on aspects of the pelage, baculum, and skull. While the pocket mice have received considerable attention, the members of the sub-

family Dipodomyinae, the kangaroo rats, have received little notice. Other than three inaccurate counts in the literature for D. merriami by Cross (1931, 2N=86) and Matthey (1952, 2N=70; 1956, 2N-68), karyological information has not previously been available.

This research was undertaken to determine the amount and kinds of chromosome variation within the genus Dipoodmys in order to illucidate the relationships between chromosome evolution and the evolution of morphological features in kangaroo rats. The study included (1) determining the karyotype of each of the species of the genus Dipodomys; (2) determining the intraspecific and interspecific chromosomal variation present within the genus and formulating a theory or theories concerning the factors contributing to the observed trends of karyotype evolution; (3) presenting a phylogeny for the genus based on the karyotypic evidence and that available from paleontological studies and more conventional types of mammalian taxonomy.

CHAPTER II

MATERIALS AND METHODS

Animals used in this study (N=287) were live trapped and either processed in the field or transported alive to the University of Oklahoma for processing. All animals examined were saved as conventional museum specimens and deposited in the collection of mammals, Stovall Museum, The University of Oklahoma. Slides of D. nitratoides, D. heermanni, D. venustus, D. elephantinus, and D. ingens were provided by Howard S. Shellhammer of San Jose State College, as slides only, as well as two heermanni and one elephantinus which were sent to me alive. Identification was verified using cranial and external morphology (Hall and Kelson, 1959; Huey, 1951; and Grinnell, 1922).

Metaphase chromosomes of bone marrow cells were prepared by a modification of the Ford and Hamerton (1959) colchicine hypotonic-citrate technique. Some of the modifications suggested by Lee (1969) were incorporated and in combination with other changes, such as the use of hypotonic KCL (1.563%) under certain conditions, yielded greatly improved results. Spleen cells were also utilized on occasion and were processed as with marrow material. The

following description of the technique used applies to both field and lab processed specimens: animals were injected intraperitoneally with 0.05% colchicine solution at 0.01 ml/g body wieght and sacrificed after 1-5 hours but usually less than 3 hours. Bone marrow was flushed from the shafts of tibiae and femurs into 12 ml. centrifuge tubes with pre-warmed (37 C) KCL solution or 1.0% sodium citrate solution. The marrow material was then aspirated by forcing in and out of a 2cc syringe with a 21g needle to suspend cells and break up tissue clumps. The cell suspension was incubated 10-12 minutes depending on the species involved (10 minutes is usually sufficient). After incubation, the suspension was centrifuged for 3-5 minutes with a hand centrifuge at approximately 600 RPMs and the supernate was carefully removed by pipetting; 3-5 ml. of fixitive (3 parts methanol and one part glacial acetic acid) were carefully added without disturbing the cell button. The cells were allowed to fix for 2-4 minutes and any oil droplets or floating debris removed from the surface of the fixitive with a pipette. The cells were then resuspended by gentle bubbling with a pipette and allowed to fix for an additional 30 minutes before making slides or stored in a refrigerator at 4-6 C before making slides. Material was stored for several months at this stage with no noticable ill effects. After fixation the material was re-centrifuged and the supernate removed. Fresh fixitive was care-

fully layered on the cell button without disturbing the cells and left for 2 minutes. This procedure was repeated 2-3 times and after the final wash the cells were resuspended in enough of the fixitive to form a hazy suspension. Usually four slides were made from each animal processed but the remaining material was saved until the slides were examined and additional slides made if necessary. In making slides, 3-4 drops of concentrated cell suspension were pipetted on to chilled slides (refrigerated in 20% alcohol and excess fluid shaken off before making the slide) and the mixture ignited on the slide and blown out before burning was complete. Blaze drying (Scherz, 1962) greatly improved spreading of chromosomes when care was taken to avoid heating of the slide in the process. Blaze drying in the field without cold slides ruined otherwise usuable preparations. Chilling of slides in the field was accomplished with a can of compressed gases of the kind used to freeze small cytological specimens. Often slides were air dried in the field and the remaining material stored in vials of fresh fixative until better slides could be made in the lab. After drying, slides were stained in 15% Giemsa Blood Stain (15 ml. stock stain solution in 85 ml. distilled water) for 10 to 15 minutes and carefully transferred to dehydration baths consisting of two baths of acetone, one of equal parts acetone and xylol and two of xylol (ca. 30 sec. in each bath). Slides then were mounted

in Permount using a coverslip. Some unmounted slides were saved for future analysis in case the stain should fade on the mounted materials. Meiotic material for confirmation of sex chromosome morphology and also mitotic material was obtained from testicular tissues of most species studied. The testes were removed while the animal was being processed for bone marrow cells and whole testes were placed in isotonic saline solution until after marrow processing was completed to fixation. The testes were then transfered to hypotonic KCL or sodium citrate and a central portion cut out and finely minced with scissors. Further processing was the same as for morrow or spleen cells. The need to conserve time in the field and to obtain fixed materials that could be retained and processed at a later date led to the use of the following additional methods. Bone marrow material was stored in fresh fixative in the field and additional slides made in the lab if needed. Often in the field and occasionally in the lab the testes were transferred to isotonic sodium citrate (2.2%) solution, and the tubules removed by cutting the tunica and teasing out the tubules; straightening them out as much as possible to eliminate clumps. The tubules were then placed in 37 C hypotonic KCL or sodium citrate for 10-12 minutes. The incubation time can be lengthened for processing at lower temperatures in the field (ca. 15-20 minutes at 24 C).

Tubules were fixed by placing them directly in 3:1 methanol-acetic acid and stored in a refrigerator until needed. Refrigeration can be dispensed with in the field if an effort is made to keep the material from becoming over-heated. Slides were made by a modification of the micro-pipette method of Meredith (1969) with lower temperatures than those suggested by Meredith. Larger amounts of material were processed by modifying the method of Meredith for blaze-dried slides (Scherz, 1962). In either technique care must be used to prevent over-heating of the material, as even the 40-45 C temperature used makes the cells difficult to stain. After the slides are dry, staining, dehydration, and mounting may be carried out as for bone marrow material except that a longer staining time (with 15% Giemsa) of 20-30 minutes may be required if the warm slide method is used.

While colchicine pretreatment is desirable to increase the number of cells at metaphase, usually it is possible to obtain good material, including both meiotic and mitotic metaphase spreads, even without prior injection of colchicine. Results may be poor if the animal is not in a period of active cell division but seldom does the method fail completely. Animals can be processed in the field and the fixed material transported to the lab for further processing, thus making it possible for collectors in the field, not trained in chromosome work, to preserve valuable

cytological materials. The animal does not have to be "fresh-killed", as good material has been obtained from animals removed from morning checks of snap-trap lines. This also applies to bone marrow material, but better material is obtained if the traps are checked every 1-2 hours, especially during periods of below freezing weather. I have processed rodents of several different genera in this manner and usable material was obtained from specimens dead several hours, when kept fairly cool (below 24° C).

Once suitable material was obtained, a diploid count was determined; at least 10 and usually more than 20 selected metaphase spreads were counted from each specimen examined. Selected spreads were photographed on a 4"x 5" format at an initial enlargement of 1000 X, and prints made at approximately 2000 X. The chromosome prints were cut out and paired with their presumed homologue. Since the lengths of the chromosomes from a given specimen varied considerably due to differences in contraction, no attempt was made to measure them; however, the longer chromosomes of kangaroo rats measure approximately 10 micra and the shortest approximately one micron at the stage of contraction usually selected for a representative karyotype. The paired cut-out chromosomes were arranged according to arm-ratio groups modified slightly from Patton (1967:29) and are as follows: Metacentric, less than 1:1.1; Submetacentric, between 1:1.1 and 1:1.9; Subtelocentric, 1:2

or greater; Acrocentric, with a minute second arm; and Telocentric, with no visible second arm. These last two groups were often included together because of difficulties in distinguishing the presence of a small arm in many preparations. Often it was also difficult to distinguish between Subtelocentric and Acrocentric chromosomes and some slight variation in treatment did result from species to species but not to the extent that resulting interpretation of relationships would have been changed. After the basic karyotype was determined, the number of major autosomal arms or Fundamental Number (FN) of Matthey (1951) was determined. Acrocentric and Telocentric elements were scored as having one arm while those in the remaining groups were scored as having two. At least five matching karyotypes were prepared from each population sampled, and usually at least two karyotypes were preapred from each specimen examined though in a few cases the quality of the preparation allowed only counting.

The following is a list of the specimens examined during this study:

Dipodomys elator Merriam.--Texas: Wilbarger Co., 19 mi. S Vernon (5 males, 5 females).

Dipodomys spectabilis perblandus Goldman.--Arizona: Pima Co., near Tuscon (9 females); Highway 77 and Little Hill Mine Road (1 female).

Dipodomys spectabilis spectabilis Merriam.--New

Mexico: Hidalgo Co., 3 mi. S Animas (3 males, 5 females).

Dipodomys spectabilis baileyi Goldman.--New Mexico: Roosevelt Co., 9 mi. W Tolar (1 male, 1 female); Debaca Co., 1.8 mi. SW Dunlap (1 male); Texas: Culbertson Co., 18 mi. NW Kent (1 male).

Dipodomys nelsoni Merriam.--San Luis Potosi: Rancho Pastoriza, 5,200 ft., 8 mi. SSW Matehuala (2 males, 2 females).

Dipodomys ornatus Merriam.--San Luis Potosi: 1.3 mi. W Bledos, 7,100 ft. (4 males, 9 females).

Dipodomys ordii monoensis (Grinnell).--Nevada: Washoe Co., ½ mi. SE Pyramid Lake (1 female).

Dipodomys ordii fetosus Durrant and Hall.--Nevada: Lincoln Co., Sand Springs, Penyoyer Valley (3 males, 1 female).

Dipodomys ordii richardsoni (J. A. Allen).--Oklahoma: Tillman Co., 2 mi. S Davidson (10 males, 9 females); 5.5 mi. S Grandfield (4 males, 9 females).

Dipodomys ordii oklahomae (Trowbridge and Whitaker).--Oklahoma: Cleveland Co., near South Canadian River at Norman (1 male).

Dipodomys gravipes Huey.--Baja California: 8 mi. N San Quintin (1 male, 1 female); Pacific Coast, 12 mi. N El Rosario (2 males).

Dipodomys stephensi (Merriam).--California: Riverside

Co., 1 mi. W Winchester (17 males, 17 females).

Dipodomys heermanni goldmani (Merriam).--California: San Benito Co., Bear Valley (1 male, 1 female); Panoche Valley (2 males); Pinnacles Nat. Mon. (2 males); Monterey Co., Soledad area (4 males, 2 females).

Dipodomys panamintinus caudatus Hall.--Nevada: Clark Co., 9 mi. W Searchlight (9 males, 10 females).

Dipodomys panamintinus mohavensis (Grinnell).--California: San Bernadino Co., Hesperia (6 males, 2 females).

Dipodomys ingens (Merriam).--California: Fresno Co., Panoche Valley (1 male, 1 female).

Dipodomys deserti aquilus Nader.--Nevada: Washoe Co., ½ mi. SE Pyramid Lake (1 male, 2 females).

Dipodomys deserti deserti Stephens.--Nevada: Clark Co., 3 mi. S Riverton (5 males, 5 females).

Dipodomys agilis perplexus (Merriam).--California: San Bernadino Co., Cajon Wash, ½ mi. SW Devore (10 males, 11 females).

Dipodomys agilis simulans (Merriam).--Baja California: 8 mi. N San Quintin (1 male, 1 female).

Dipodomys agilis plectilis Huey.--Baja California: Pacific Coast, 12 mi. N El Rosario (4 males, 1 female); 27 mi. W San Augustine (2 males).

Dipodomys peninsularis pedionomus Huey.--Baja California: San Augustine (9 males, 5 females).

Dipodomys venustus venustus (Merriam).--California: Santa Clara Co., Loma Prieta Mtn. (3 males, 2 females).

Dipodomys elephantinus (Grinnell).--California: San Benito Co., Pinnacles Nat. Mon. (2 males, 1 female).

Dipodomys microps occidentalis Hall and Dale.--Nevada: Lincoln Co., Sand Springs, Penyoyer Valley (2males).

Dipodomys microps celsus Goldman.--Utah: Washington Co., 3 mi. S Hurricane (1 female); 7 mi. NW St. George (8 males, 7 females).

Dipodomys nitratoides brevinasus Grinnell.--California: Fresno Co., Panoche Valley (1 male).

Dipodomys merriami frenatus Bole.--Utah: Washington Co., 1 mi. NW Toquerville (1 male); 3 mi. S Hurricane (1 male, 2 females); 7 mi. NW St. George (7 males, 2 females).

Dipodomys merriami merriami Mearns.--Nevada: Washoe Co., ½ mi. SE Pyramid Lake (2 males); Clark Co., 3 mi. S Riverton (4 males); 9 mi. W Searchlight (2 males; 2 females). Arizona: Pima Co., 20 mi. SE Tucson (1 male, 1 female); Santa Cruz Co., 5 mi. E Elgin, Babacamori Ranch (1 male, 1 female). New Mexico: Chaves Co., 9 mi. W, 1 mi. S Tolar (1 male, 1 female). San Luis Potosi: Rancho Pastoriza, 8 mi. SSW Matehuala (1 male, 2 females).

Dipodomys merriami quintinensis Huey.--Baja California: 8 mi. N San Quintin (2 males).

Dipodomys merriami semipallidus Huey.--Baja California:

San Augustine (1 female); 27 mi. W San Augustine (1 male, 1 female).

Dipodomys merriami atronasus Merriam.--San Luis Potosi: 1.3 mi. W Bledos (1 male, 1 female).

CHAPTER III

RESULTS

The number of individuals analyzed, sexes, diploid numbers, fundamental numbers, and types of chromosomes from each species investigated are given in Table 1. Representative karyotypes for each species are presented in Figs. 1-22. Sex chromosome morphology was checked with meiotic material in all cases where live males were obtained. General trends of karyotype variation at various levels are summarized below.

Individual variation.--In all specimens examined the chromosome count accepted as the diploid number was obtained in more than 85% of the cells counted. Those counts that varied from the count assumed to be characteristic of the individual were lower and resulted from loss of chromosomes from ruptured cells. Selection of intact cells for counting consistently produced stable counts. Certain chromosomes at different stages of contraction often were found to present different arm-ratios. Once cell division was arrested by colchicine, the chromosomes continued to contract and one arm of a chromosome occasionally contracted more rapidly than the other, thus changing the arm-ratio

Table 1.—Data from karyotypic analysis with all subspecies examined included under one heading if the same. Data from D.a. simulans included with D.a. plectilis.

SPECIES	SEX		CHROMOSOMES							
			2N	AUTOSOMES*				X	Y	FN
				M	SM	ST	A&T			
D. ordii	18	20	72	4	26	5		SM	A-ST	140
D. ornatus	4	9	72		12	22	1	SM	A	138
D. spectabilis spectabilis	3	5	72				35	SM	A	70
D. spectabilis perblandus	0	10	72		4		31	SM		78
D. spectabilis baileyi	3	1	72		12		23	SM	A	94
D. elator	5	5	72		3	3	29	SM	A	82
D. nelsoni	2	2	72	4	21	7	3	SM	A	134
D. gravipes	3	1	70		1.5		32.5	SM	A	71
D. stephensi	17	17	70		5	5	24	SM	A	86
D. heermanni	7	3	64	4	12		15	SM	A	94
D. panamintinus	15	12	64	4	9	4	14	SM	A	96
D. ingens	1	1	64	4	14		13	SM	A	98
D. deserti	6	7	64	3	16	4	8	SM	A-ST	108
D. agilis perplexus	10	11	62	3	19	3	5	SM	A	110
D. agilis plectilis	7	2	60	3	23	3		SM	A	116
D. peninsularis	9	5	60	3	23	3		SM	A	116
D. venustus	3	2	60	3	21	5		SM	A	116
D. elephantinus	2	1	60	3	21	5		SM	A	116
D. microps	10	8	60	3	17	9		SM	A	116
D. nitratoides	1	0	54	3	20	3		SM	A	104
D. merriami	26	13	52	3	17	5		SM	A	100

* Autosome numbers refer to number of homologous pairs. M=metacentric, SM=submetacentric, ST=subtelocentric, A&T=acrocentric and telocentric.

and making it difficult to assign certain chromosome pairs to the proper arm-ratio group.

In most cases several individuals were examined from a given population. Other than sexual dimorphism, no karyotypic variation was found between individuals of the same species from the same locality.

Intraspecific variation.--chromosomal variants within what is currently regarded as a single species was rare in Dipodomys and was found to occur in only three species. In one of these, D. panamintinus, the variation was slight and easily over-looked, and consisted of a size difference in the smallest pair of subtelocentric sutosomes. In D. agilis, the subspecies occurring in southern California and Baja California, D.a. simulans and D.a. plectilis, possessed 60 chromosomes, all of which were bi-armed, while specimens of D.a. perplexus from San Bernadino Co., California, possessed 62 chromosomes and several pairs of uni-armed chromosomes. Apparently the southern subspecies underwent a considerable amount of chromosomal change while isolated from more northern populations. Within D. spectabilis, karyotype variation appeared to be correlated with subspeciation. The few specimens examined of D.s. baileyi from eastern New Mexico and western Texas possessed many more submetacentric and fewer acrocentric chromosomes than did specimens of D.s. perblandus from

Arizona. Specimens of D.s. spectabilis from southwestern New Mexico had even fewer bi-armed chromosomes than did either of the above populations. Examination of more animals from other localities throughout the range of this species must be accomplished to determine if the variation noted is indeed a reflection of subspecific difference.

Interspecific variation.--in the animals studied, the diploid number ranged from 52 to 72, and the Fundamental Number (FN) ranged from 70 to 140. The diploid number was found to be a better guide to relationships than the FN since the latter varied greatly between some populations of the same species.

D. merraimi with 52 chromosomes and D. nitratoides with 54 were closely related and together formed a natural group apart from the other species of the genus. The karyotype of both species was composed entirely of bi-armed autosomes, and 2N reduction from the ancestral diploid number was apparently accomplished by a series of centric translocations or centric fusions.

The next lowest diploid count found in the genus was 60 and was possessed by five members of the Heermanni Group. D. agilis, D. peninsularis, D. venustus, D. elephantinus, and D. microps. The populations of D. agilis that possessed 60 chromosomes had the same karyotype as did D. peninsularis while some populations of D. agilis possessed 62 chromosomes and other karyotypic differences. The

southern subspecies of D. agilis appeared to be genetically closer to D. peninsularis than to other populations of D. agilis farther to the north in California. From the karyotypic data I judged D. peninsularis to be conspecific with the southern forms of D. agilis, and the morphological data from skins and skulls (unpublished data) supported this view. The karyotype of D. venustus was also easily derived from a D. agilis (2N=62) condition and D. venustus and D. elephantinus which shared the same karyotype were closely related to and probably derived from D. agilis. D. venustus and D. elephantinus were also very similar to each other morphologically and may be conspecific. The karyotype of D. microps was similar in many respects to the preceding four but differed enough to indicate an earlier divergence, perhaps from an ancestor in common with D. agilis. The 62 chromosome populations were apparently derived from 64 chromosome populations. Four species were found to possess a 2N of 64, and all four occur in California. Three of these, D. heermanni, D. ingens, and D. panamintinus were closely related and were sufficiently generalized, both morphologically and karyotypically, to be ancestral to the D. agilis line. The fourth species in the group, D. deserti, was highly specialized morphologically and was sufficiently divergent karyotypically to indicate an early derivation from the ancestral stock that produced the other three 2N=64 species. The karyotype of D.

heermanni was the most generalized of the group, and D. ingens and D. panamintinus possessed karyotypes that were similar to and easily derived from that of D. heermanni. Karyotypically, D. heermanni, D. ingens and D. panamintinus were more closely related than were the 60 and 62 chromosome populations of D. agilis. Two other species, D. stephensi and D. gravipes, appeared to be closely related to D. heermanni and its derivatives. These two species, gravipes in Baja California and stephensi in scuth-central California, both possessed 70 chromosomes and were similar both in morphology and karyotype. The karyotype of gravipes was more generalized (fewer pericentric inversions) and probably best represents the ancestral condition for the species of the Heermanni Group.

The species which originated outside of the Great Basin and California-Baja California and which, with the exception of the wide-ranging D. ordii, do not occur in that region present a group that, while being heterogeneous morphologically, all possessed 72 chromosomes. Of these species, those occurring in central and southern Mexico, D. ornatus and D. phillipsi, were quite distinct, and the karyotype of D. ornatus (D. phillipsi not examined karyotypically) indicated that they had been isolated from others of the genus since the early radiation of 72 chromosome forms and were not closely related to any extant group.

D. ordii is another of the 72 chromosome species and possessed a karyotype that was highly modified from that seen in the other 72 chromosome forms in that all autosomes were bi-armed, due to the incorporation of many pericentric inversions, most of which involved a large segment of the chromosome. While possibly having descended from an ancestor in common with the other 72 chromosome species, *D. ordii* did not appear to be closely related to other living species.

The remaining three 72 chromosome species, *D. spectabilis*, *D. elator*, and *D. nelsoni*, were all closely related karyotypically and morphologically. The karyotype of *D. spectabilis spectabilis* had fewer pericentric inversions incorporated into it and hence a greater number of acrocentric and telocentric chromosomes and represented what was thought to be the ancestral karyotype for the genus. The karyotype of *D. elator* differed less from that of *D.s. perblandus* in the number of incorporated pericentric inversions than did that of *D.s. baileyi* and *D. elator* may have derived directly from an early population leading to *D. spectabilis*. The karyotype of *D. nelsoni* contained nearly as many large pericentric inversions as did that of *D. ordii* but was probably derived directly from *D. spectabilis* and does not show any close relationship to *D. ordii*.

Figure 23 illustrates the direction or directions of karyotypic change in the several groups and subgroups.

Fundamental Number (FN) is represented vertically while the diploid number (2N) is represented on the horizontal axis. Points on the graph corresponding to high diploid and FN numbers occur to the upper right of the graph. The Merriami Group is distinctly to the left of the other groups indicating that centric fusions have been important in this group. The slope of the line connecting the members of this group indicates that pericentric inversions have also played a part in producing the present arrangements. The positions of the several members of the Heermanni Group on the graph indicates that centric fusions and also pericentric inversions have occurred, however, the latter mechanism is more conspicuous than in the Merriami Group and is responsible for most of the observed variation between members of each subgroup while the differences between each of these subgroups is largely due to centric fusion. Centric fusion does not appear to have occurred in the Spectabilis, Ordii, and Phillipsi groups, and most of the chromosomal changes seen in these groups are clearly the result of pericentric inversions. Centric fusion then has occurred only in those forms originating in the Great Basin and in California-Baja California. The restriction of diploid numbers below 72 as indicated above seems ample justification for regarding the observed changes as being due to centric fusions rather than to centric fission.

Meiotic metaphase.--a description of the meiotic chromosome complements of all species examined is beyond the scope of this study and, even if included, would yield little additional information of importance in determining phylogenetic relationships. All species where meiotic material was examined possessed a sex chromosome bi-valent in which the Y chromosome was in a terminal (end to end) association with the X chromosome at metaphase I. Examination of these sex bi-valents was useful in determining the general morphology of the sex chromosomes. In all cases these were the same as those chosen from examination of the mitotic complement. The X chromosome is a medium to large submetacentric in all species examined, except that it is nearly metacentric D. nitratoides and D. merriami. The Y chromosome is a medium to small acrocentric or telocentric element in all except D. deserti and D. ordii in which very distinct short arms are present and the Y chromosome appears to be nearly, if not actually, subtelocentric.

Figure 24 illustrates the meiotic complement of D. merriami, which is presented to show the usefulness of the sex bi-valent in determining general sex chromosome morphology.

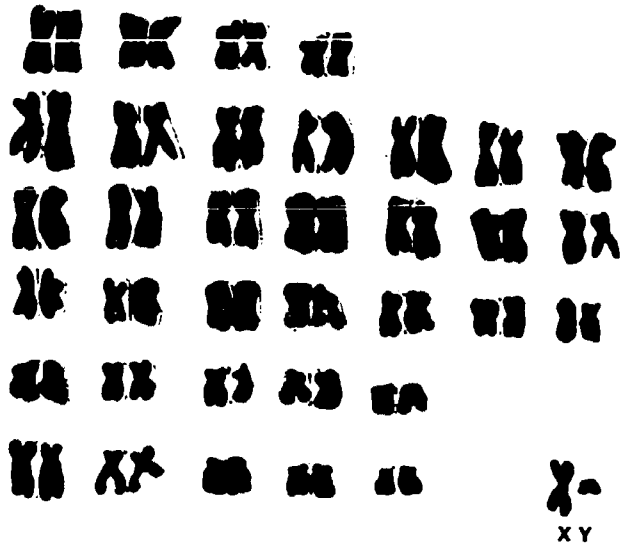


Fig. 1. Representative karyotype of Dipodomys ordii. Tillman Co., Oklahoma.

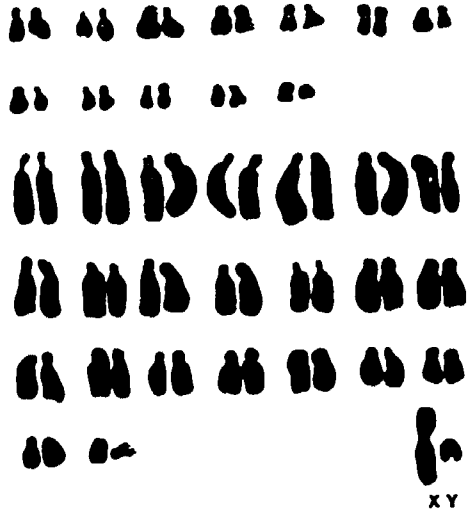
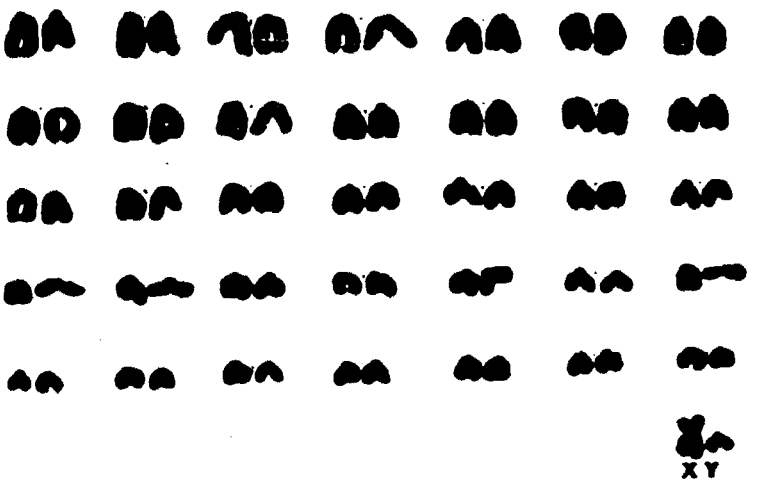


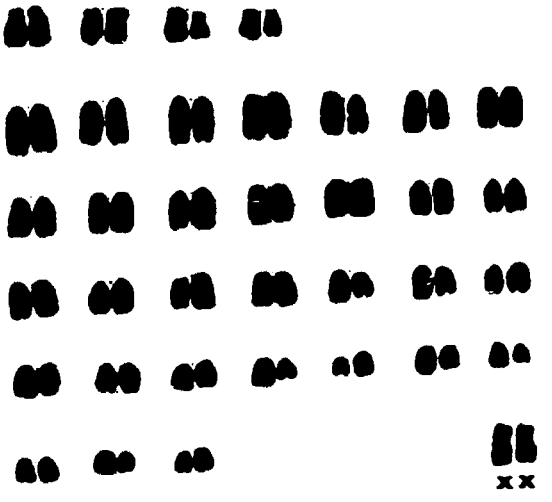
Fig. 2. Representative karyotype of Dipodomys ornatus. San Luis Potosí, Mexico.

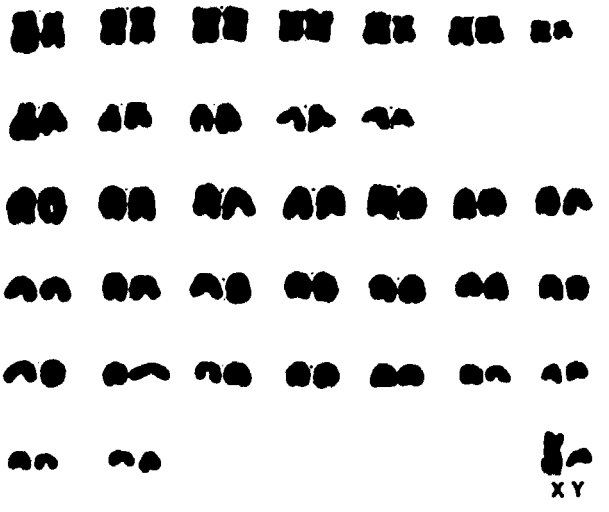
Fig. 3. Representative karyotype of *Dipodomys spectabilis spectabilis*. Hidalgo Co., New Mexico.

Fig. 4. Representative karyotype of *Dipodomys spectabilis perblandus*. Pima Co., Arizona.

Fig. 5. Representative karyotype of *Dipodomys spectabilis baileyi*. Roosevelt Co., New Mexico.


X Y


X X


X Y

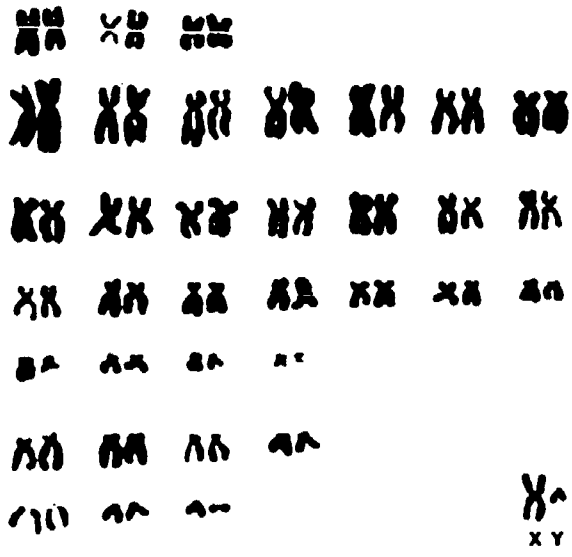


Fig. 6. Representative karyotype of Dipodomys nelsoni. San. Luis Potosi, Mexico.

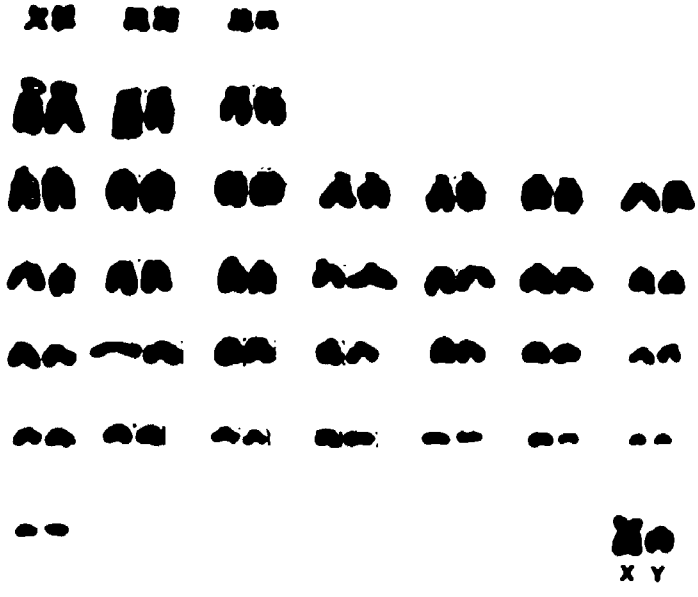


Fig. 7. Representative karyotype of Dipodomys elator. Wilbarger Co., Texas.

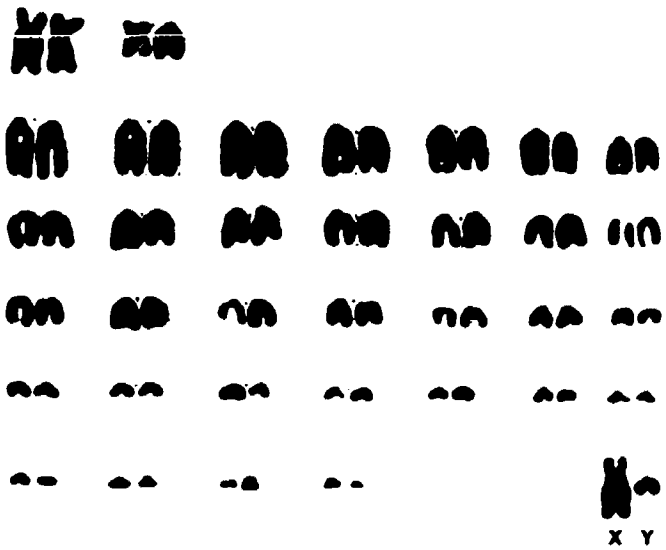


Fig. 8. Representative karyotype of Dipodomys gravipes. Baja California, Mexico.

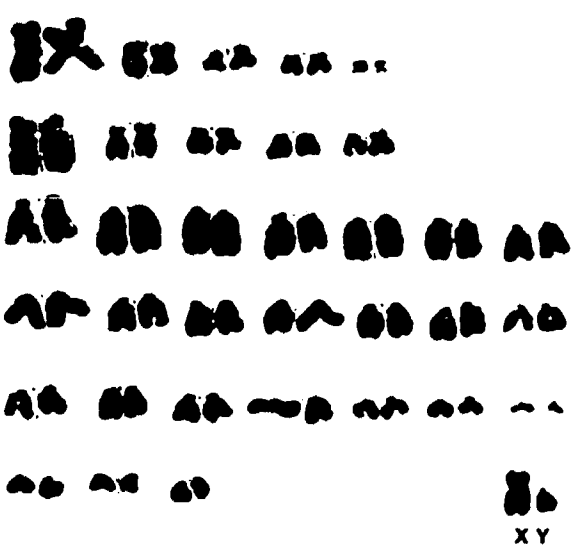


Fig. 9. Representative karyotype of Dipodomys stephensi. Riverside Co., California.

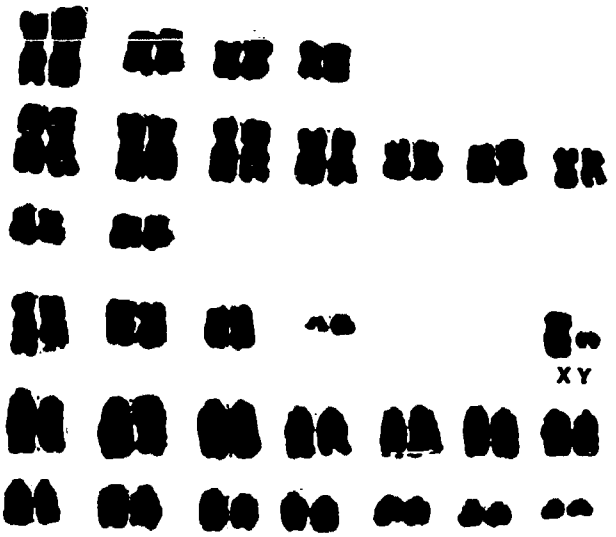


Fig. 10. Representative karyotype of Dipodomys panamintinus caudatus. Clark Co., Nevada.

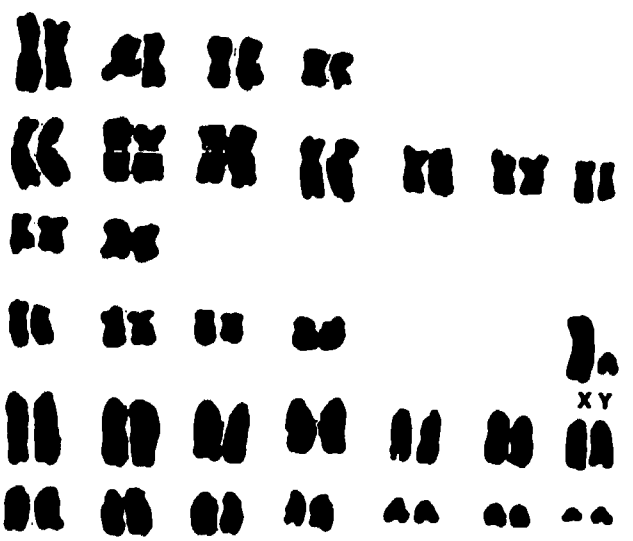


Fig. 11. Representative karyotype of Dipodomys panamintinus mohavensis. San Bernadino Co., California.

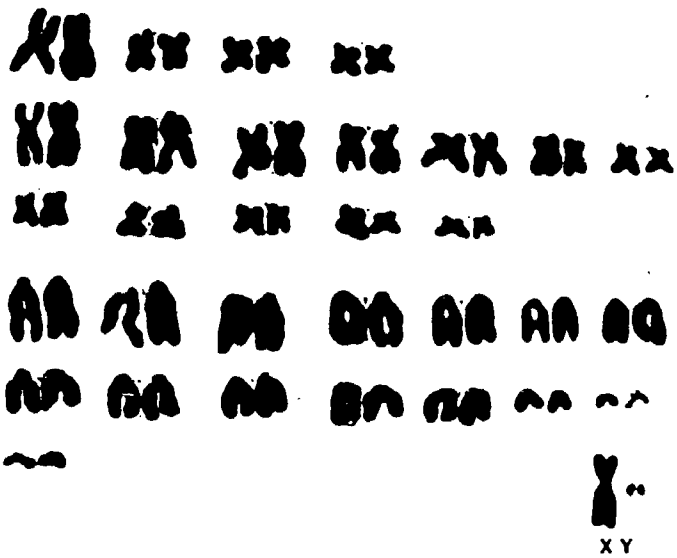


Fig. 12. Representative karyotype of Dipodomys heermanni. San Benito Co., California.

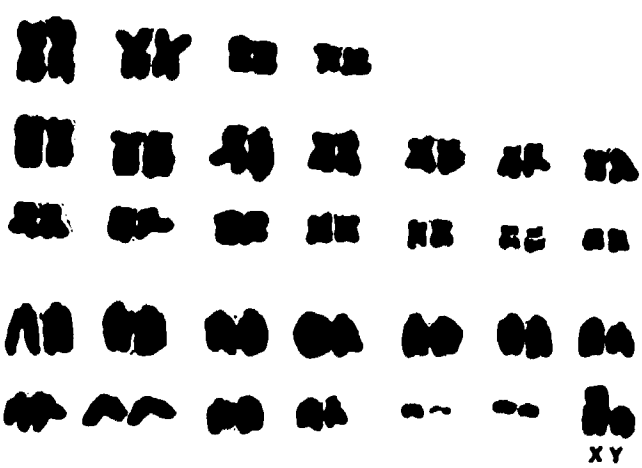


Fig. 13. Representative karyotype of Dipodomys ingens. Fresno Co., California.

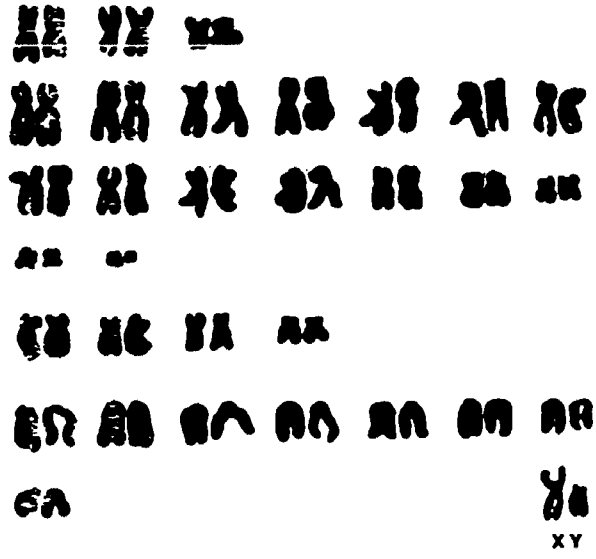


Fig. 14. Representative karyotype of Dipodomys deserti. Clark Co., Nevada.

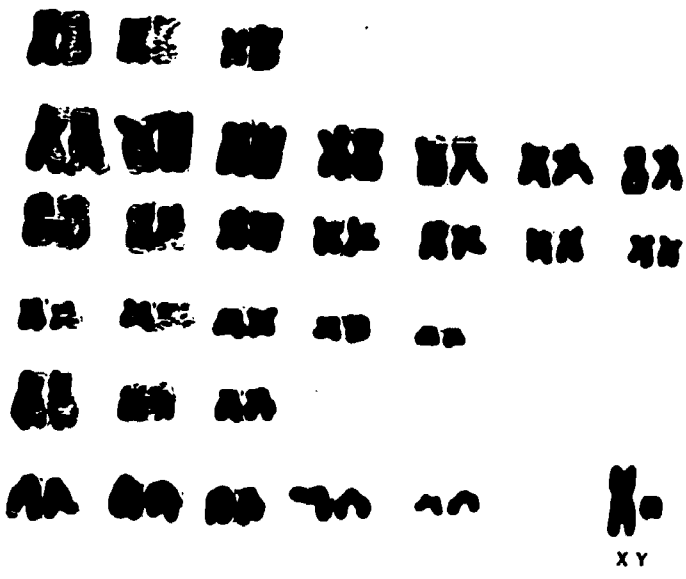


Fig. 15. Representative karyotype of Dipodomys agilis perplexus. San Bernadino Co., California.

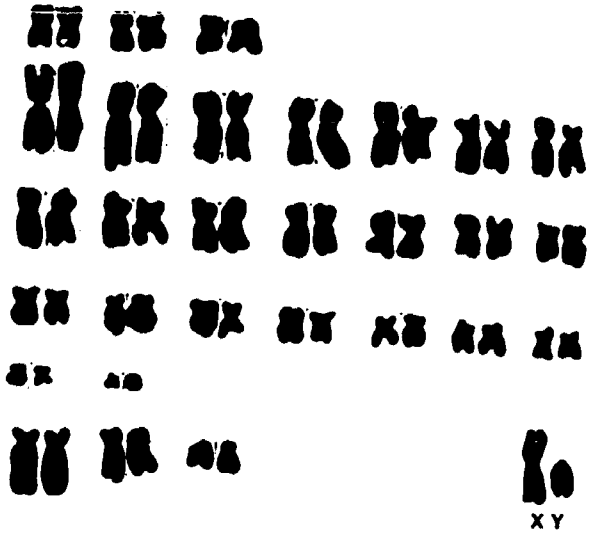


Fig. 16. Representative karyotype of Dipodomys agilis plectilis. Baja California.

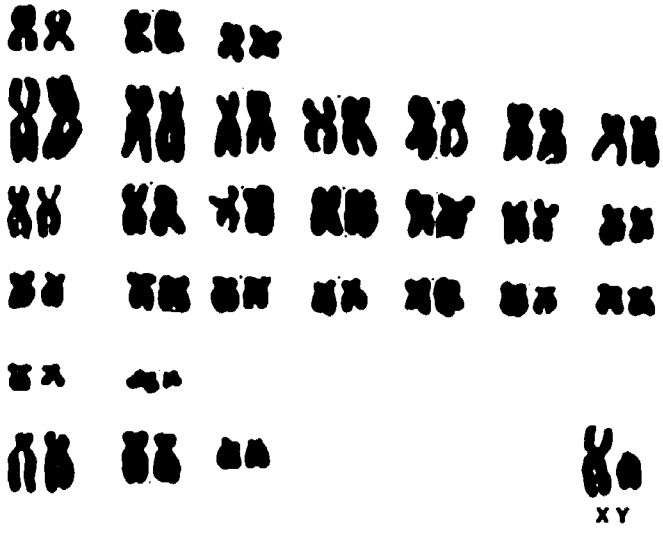
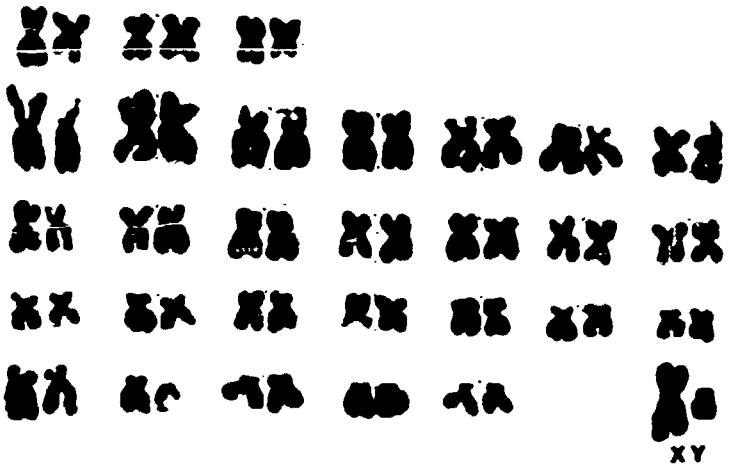


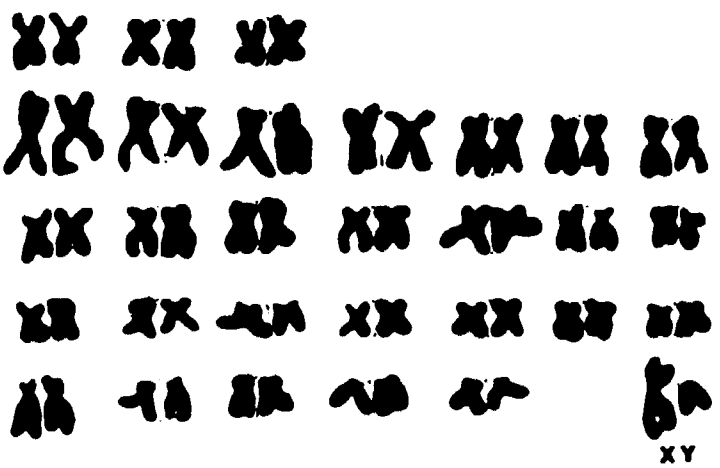
Fig. 17. Representative karyotype of Dipodomys peninsularis pedionomus. Baja California, Mexico.

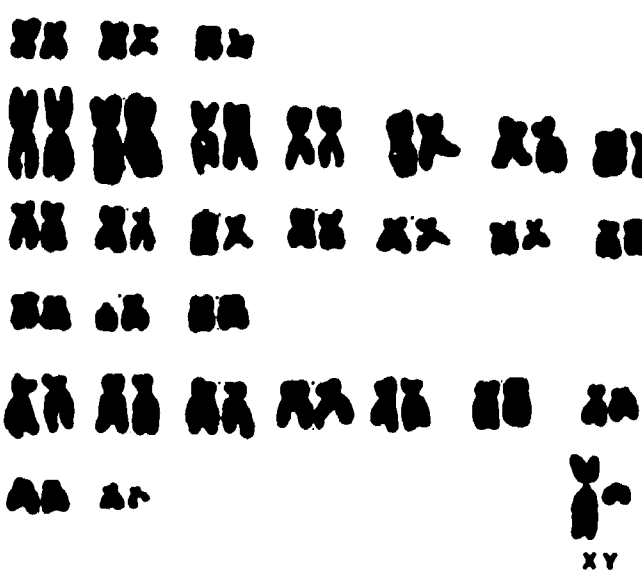
Fig. 18. Representative karyotype of *Dipodomys venustus*. Santa Clara Co., California.

Fig. 19. Representative karyotype of *Dipodomys elephantinus*. San Benito Co., California.

Fig. 20. Representative karyotype of *Dipodomys microps*. Washington Co., Utah.


XY


XY


XY

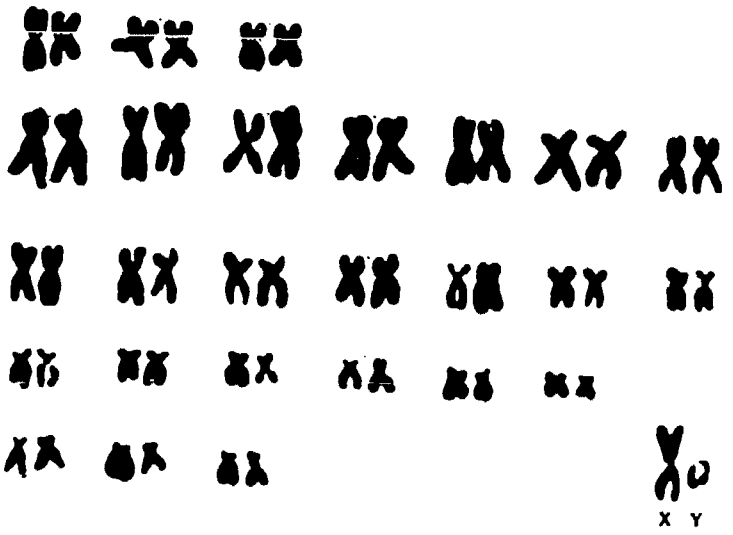


Fig. 21. Representative karyotype of Dipodomys nitratoides. Fresno Co., California

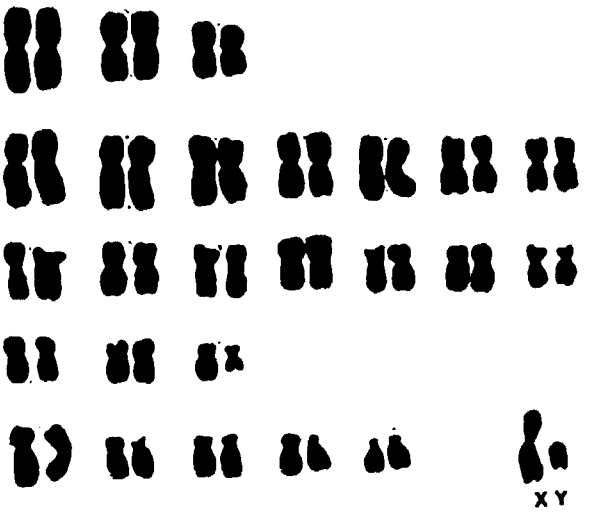
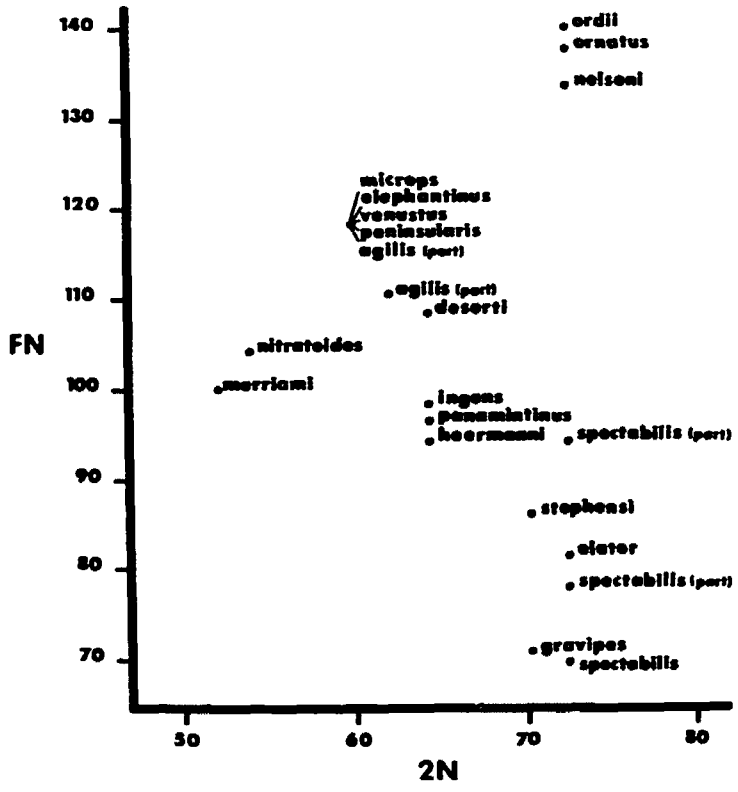


Fig. 22. Representative karyotype of Dipodomys merriami. Clark Co., Nevada.

Fig. 23. Fundamental number (FN) plotted against diploid number (2N) to illustrate the direction or directions of karyotypic change. See text for explanation.

Fig. 24. Meiotic chromosomes (metaphase I) of Dipodomys merriami. Clark Co., Nevada.


140
130
120
110
100
90
80
70
FN
50
60
70
80
2N
ordii
ornatus
nelsoni
microps
elephantinus
venustus
peninsularis
agilis (part)
agilis (part)
deserti
nitratoides
merriami
ingens
panamintinus
hoermanni
spectabilis (part)
stephensi
eleter
spectabilis (part)
gravipes
spectabilis


X
Y

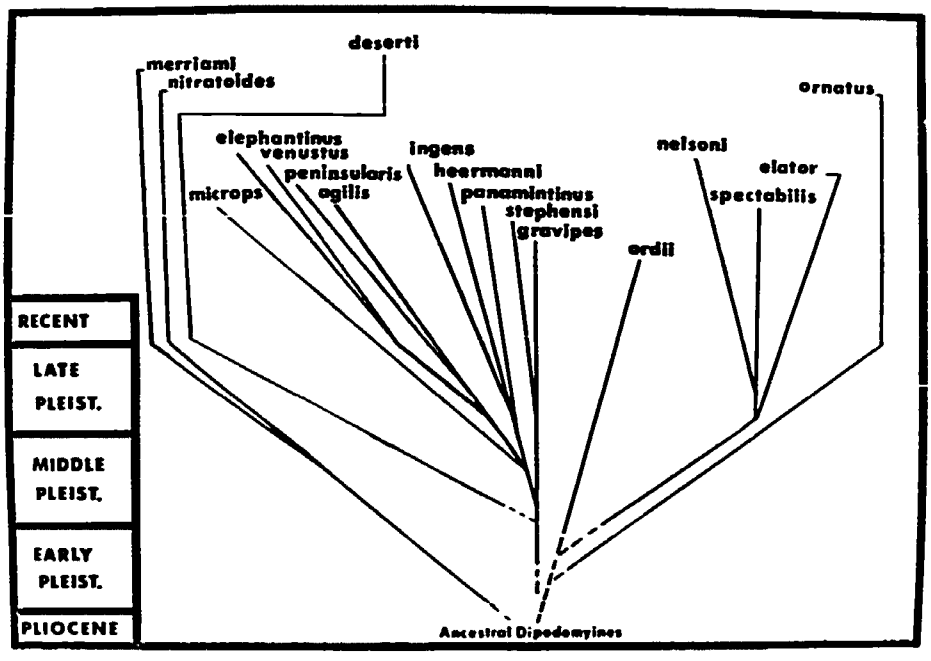


Fig. 25. Dendrogram illustrating the proposed phylogenetic relationships of the recent species of Dipodomys. See text for explanation.

CHAPTER IV

DISCUSSION

The kangaroo rats represent a compact genus of 22 currently recognized species that are remarkably similar in morphology. Their similarities of structure, resulting, in part, from parallelism in developing specializations for rapid bipedal locomotion in response to xeric environments with reduced plant cover, makes the task of establishing their phylogenetic relationships more difficult. Kangaroo rats have been grouped on the basis of tooth structure (Wood, 1935:155); by general external and cranial morphology (Grinnell, 1922:96); by use of the baculum (Burt, 1936: 152; 1960); by skeletal indices of specialization and compaction of the viscera (Setzer, 1949:496); and finally by combining several sources of information including field knowledge of most species (Lidicker, 1960:134). All of these groupings differ somewhat in the alignment of the various species, but the original groupings of Grinnell (1922) have not been greatly altered by later work. Davis (1942) modified the groupings of Grinnell by removing *D. elator* from the Phillipsi Group and placing that species in a separate group in alliance with *D. spectabilis*. He

also combined Grinnell's Ordii and Compactus groups and changed the position of the Phillipsi Group to follow the Heermanni Group. The plan presented by Lidicker (loc. cit.) probably best represents the actual relationships. Only the groupings arrived at by Lidicker (1960) and Setzer (1949) are included below as a reference point for the arrangement presented in this study:

Setzer (1949)

ORDII GROUP
- ordii
- microps

PANAMINTINUS GROUP
- panamintinus
- stephensi

HEERMANNI GROUP
- heermanni
- agilis
- ingens
- venustus
- elephantinus

SPECTABILIS GROUP
- spectabilis
- nelsoni

MERRIAMI GROUP
- merriami
- nitratoides
- insularis
- phillipsi
- ornatus
- elator

Deserti Group
- deserti

Lidicker (1960)

ORDII GROUP
- ordii

HEERMANNI GROUP
(Subgroup A)
- heermanni
- ingens
- panamintinus
- stephensi
- gravipes

(Subgroup B)
- agilis
- venustus
- elephantinus
- paralius
- peninsularis

MICROPS GROUP
- microps

SPECTABILIS GROUP
(Subgroup A)
- elator
- spectabilis
- nelsoni

(Subgroup B)
- deserti

PHILLIPSI GROUP
- phillipsi
- ornatus

MERRIAMI GROUP
- insularis
- merriami
- nitratoides

I agree with Lidicker (1960:133) that Setzer was misled to some extent by stressing total skeletal specializtion in deciding interspecific relationships. Setzer (1949:493) indicates that characters of the baculum between

subspecies of D. ordii are as great as the differences which Burt (1936:154-55) found between the full species, D. agilis and D. microps, and discounts the value of the baculum as an adequate basis for determining the natural relationships of the species groups. Lidicker (loc. cit.) holds the opposite view and believed that Burt (1936) presented data based on bacula that added evidence of relationship between D. deserti and D. spectabilis. My own studies of the bacula of kangaroo rats confirm Setzer's opinion, and I find that in most cases use of the baculum to decide relationships in kangaroo rats has resulted in error. An example of this error was the decision by Blair (1954) that the baculum of D. elator was similar to that of D. merriami and unlike that of D. spectabilis. Proper study of kangaroo rat bacula can yield worthwhile data, however, and it is the way that such data has been obtained and used that has tended to negate tha validity of observations based on that structure. Far too often only one or a few bacula of each species are compared and the lack of information concerning age effects and population variation has led to wrong conclusions. Burt (1960), though still using small samples, corrected his earlier work (1936). His groupings based on bacula (1960:45-7), while not listed as species groups, are similar to the arrangement presented in this study. Lidicker (1960:133) apparently used Burt's earlier description of the baculum

of D. spectabilis based upon immature specimens in stating that bacular evidence linked D. spectabilis and D. deserti.

These previous studies have offered some information concerning phylogeny within the genus, but even in the latest arrangement by Lidicker (1960), the affinities of certain species such as D. deserti, D. microps, and D. ornatus were not well understood. Use of indices of specialization without attention to different habitat preferences has obscured true relationships in many cases. Most kangaroo rats have not been able to adapt to the low, hot valleys of the Colorado and Mojave deserts, and the few that have, such as D. merriami and D. deserti, show specializations for saltation and other adaptations to such xeric conditions to a much greater degree than the majority in the genus. Under intense selective pressure, such specializations could have been rapidly achieved. Thus, adaptations to different habitats may have obscured true phylogenetic relationships. Another aspect to consider in deciding relationships and patterns of distribution is that most kangaroo rats tend to be highly territorial in behavior, both towards their own and other species. Seldom can more than two species be taken in the same trap line, and when two or more species do occur together, they are generally dissimilar in size and living requirements. The species groups in Dipodomys appear to be natural superspecies of closely related, largely allopatric and pre-

sumably reproductively isolated populations. While intergroup sympatry occurs, intragroup sympatry is rare. Certainly, the niche available to ricohetal rodents is a narrow one. Such narrowness of niche would require that a considerable degree of behavioral or ecological differentiation must occur before sympatry is possible. The situation seen in the kangaroo rats in this regard is similar to the even narrower niche available to subterranean rodents and the high degree of allopatry of such forms in most regions.

Paleontology.--the fossil record of the family heteromyidae is fragmentary, at best, but enough is known to present a general phylogeny of the group. Wood (1935) and Hibbard (1958, 1960) are two of the major contributors to the paleontology of this group. Setzer (1949) and Lidicker (1960) both present reviews of the evidence at hand and derive probable phylogenys based on that evidence. From these accounts it appears that all extant species of the genus Dipodomys have arisen since early Pleistocene times from an ancestral lineage which includes the middle Pliocene to early Pleistocene genus Prodipodomys and which probably diverged from other heteromyid subfamilies during early to middle Miocene times. The genus Prodipodomys shares many features with Dipodomys but is more primitive in dentition and limb structure. The appearance of Prodipodomys appears to coincide with the development of

a modern type of desert plains habitat in the area regarded by Lidicker (1960:131) as the center of dispersal for Dipodomys, namely, southeastern California and southwestern Nevada south to the northern deserts of Mexico. There is evidence that this area has been characterized by arid climates since Miocene times. Hibbard (1958) indicates that three living species of kangaroo rats have fossil representatives or very close relatives known from Wisconsin Age deposits. These are: D. ordii from Burnet Cave, New Mexico; D. agilis from Rancho La Brea, California; and D. ingens from the McKittrich tar seeps in California. Probably all known species of the genus were distinct before Wisconsin times. Hibbard (1960:13) lists specimens that are near D. ordii in both the Cragin Quarry and Jinglebob faunas of Sangamon Age. The presence of D. ordii in these interglacial deposits may indicate that this species and perhaps others of the genus had developed earlier, perhaps during Middle Pleistocene times. The occurrence of good fossil evidence for Dipodomys in the Colorado Desert region of California from early-middle Pleistocene (Late Aftonian) occurring sympatrically with Prodipodomys (Downs and White, 1968) lends support to the general time sequence presented in this study for the development of the Heermanni Group kangaroo rats in California.

Karyotype Evolution.--the kinds and degree of karyotype variation demonstrated by members of the genus Dipodomys

illustrates the futulity of labeling a particular type of chromosome complement as "primitive" or "specialized" and the terms genralized and derived better describe such variation. Low diploid numbers with many bi-armed chromosomes are often regarded as advanced or specialized while high diploid numbers with many acrocentric or telocentric chromosomes are regarded as primitive. That a generalized karyotype may be found in a species with a specialized or derived morphology is well demonstrated by a comparison of the morphologically generalized Dipodomys microps (Setzer, 1949) with 60 bi-armed autosomes with the morphologically more specialized D. deserti possessing 64 chromosomes including numerous uni-armed elements. Variation of this type is most often found in groups displaying what Tobias (1956) has termed Multiformity, wherein chromosome evolution has kept pace with and probably contributed to speciation within a group. Either centric fusion or pericentric inversions or both of these mechanisms may be responsible for the karyotypic changes observed in some groups. Genera and even families of mammals are known to utilize a single mechanism of karyotype re-arrangement to account for most observed changes. The genus Peromyscus with a diploid count of 48 and widely differing fundamental numbers in the various species or subspecies serves well to illustrate a group in which pericentric inversions are incorporated (Hsu and Arrighi, 1968). The family Bovidae

serves to ilustrate a group in which centric or Robertsonian fusions are most common (Wurster and Benirschke, 1968).

In the kangaroo rats a high diploid number (72) appears to be ancestral, and this number has been retained in some species and reduced in others. Indeed, there seems to be two types of evolution occurring in the genus. One is a slow accumulation of minor gene differences in semi-isolated populations while the other is more rapid and accompanied, if not dependent upon, chromosomal re-arrangements. The first type is seen in both D. ordii and D. merriami. Both possess karyotypes that appear to have reached the "end of the line" in chromosomal re-arrangement and possess chromosome complements in which all autosomes are bi-armed. The second type is seen in the many species of the Heermanni Group with their slightly differing karyotypes and possession, by most, of several to many chromosomes with terminal centromeres. The picture presented by members of this group appears to, though admittedly to a lesser degree, parallel that seen in the annual plants which Lewis (1966) presents as a case for Saltation or saltatory evolution. According to Lewis (1966:4) spatial isolation and inbreeding in small populations contributes to chromosomal re-organization in plants, and this may also apply to kangaroo rats and other mammals possessing the necessary type of chromosome complement. Pericentric inversions appear to be the most common type of karyotypic

change in the kangaroo rats, and the stepwise change in fundamental number without a change in diploid number as seen in the Spectabilis Group reflects this tendency. Change in diploid number without corresponding change in fundamental number was not found in the genus, and centric fusion appears to have been less common, though important, in the evolution of kangaroo rat karyotypes. Where chromosome reduction has occurred within the genus, it has probably been accomplished through centric fusions and possibly also by other types of translocations. The number of incorporated centric fusions distinguishes the Heermanni and Merriami groups from each other and from the other species groups, and both of these groups contain members with different diploid numbers. In D. panamintinus caudatus a small segment of one pair of autosomes apparently is missing when compared to the condition seen in D. panamintinus mohavensis. Possibly this difference results from an unequal translocation of the missing segment to another autosome.

Phylogenetic Implications.--on th basis of karyotypic analysis, five major groups of kangaroo rats are evident. These groups correspond quite closely to the groupings proposed by Setzer (1949) and Lidicker (1960) but differ in certain important aspects from either of those schemes. The groups are arranged below according to their degree of karyotypic re-organization. I believe that the groupings

andn their alignment based upon chromosome morphology more accurately reflect true relationships in the genus. Species groups based on morphological criteria involving specializations toward an extrmely arid, sandy desert habitat are not entirely valid since few kangaroo rat species occur in such habitats, and other specializations from the "ancestral" condition such as those possessed by brush dwelling forms are apparent and are as important.

Spectabilis Group
elator
spectabilis
nelsoni

Phillipsi Group
phillipsi
ornatus

Ordii Group
ordii

Heermanni Group
(Subgroup A)
gravipes
stephensi

(Subgroup B)
heermanni
panamintinus
ingens

(Subgroup C)
deserti

(Subgroup D)
agilis
peninsularis
venustus
elephantinus

(Subgroup E)
microps

Merriami Group
nitratoides
merriami

The above scheme differs importantly in the placement of D. deserti and D. microps in the Heermanni Group. Lidicker (1960) placed D. deserti with the Spectabilis Group and D. microps in its own group following the Heermanni Group. Setzer (1949) placed D. deserti in its own group at the end of the list and D. microps with the Ordii Group. The only other arrangement for these two species that is in keeping with the karyotypic data would be to place each in a separate group following the groups they are placed in above. Such an arrangement is not easily justified for D. microps but, considering morphological specialization

alone, is more easily accepted in the case of D. deserti. I agree with Lidicker in interpreting the Merriami Group as the most derived group. His division of the Heermanni Group into two subgroups also is supported to some extent by the karyotypic evidence though actually four or five distinct subgroups are apparent. I question whether D. venustus and D. elephantinus are both full species as did Setzer (1949:499). The karyotypes of these two appear to be identical. The karyotypes of D. agilis plectilis and D. peninsularis pedionomus are also identical to each other and even though possession of identical karyotypes does not necessarily mean conspecificity, it is doubtful that the two warrent separation as full species. More evidence to support this decision is given below. No other species examined, whether closely related morphologically or not, had identical karyotypes. I have no objections to the placement of the Ordii Group as the most generalized form in terms of morphological specialization. Although D. ordii possesses the most generalized morphology, it nevertheless has a more re-organized karyotype than do members of the Spectabilis or Phillipsi Groups. In view of the extreme specializations possessed by D. merriami and D. deserti inhabiting the more arid portions of the Mojave Desert, I believe that groupings and relationships of the species groups are more accurately approximated by the data derived from karyotypes. The inability of this

type of grouping system to illustrate both phylogenetic relationships and specialization is obvious in that the Heermanni Group contains the most highly specialized species, D. deserti, while the group itself stands well below the Merriami Group. The "phylogenetic tree", Fig. 25, although with its own inherent weaknesses, perhaps best reflects phylogeny and specialization. The degree of specialization shown in this dendrogram is only approximate, and while the degree of specialization assigned to some species by Setzer (1949:489) based on morphological indices serves as an underlying basis of comparison, I have also kept in mind specialization in habitat requirements and the overall effects of different habitat requirements on pedal and cranial specialization. Diploid numbers increase from left to right, and FN increases vertically in closely related groups. It will be noted that certain morphologically specialized species will be elevated despite low fundamental number.

ORIGINS OF THE SPECIES GROUPS

I have based the following theoretical account of the origins of Dipodomys species groups mainly on evidence derived from karyotypic analysis. The occurrence of only 2N=72 forms except for D. merriami throughout the major part of the range of the genus indicates to me that kangaroo rats may have first evolved in the semi-arid

grasslands of northern Mexico and central United States, and may first have developed their evolutionary trends toward bi-pedal locomotion in response to open, semi-arid grassland situations rather than in response to true desert conditions as is often held to be the case. Most of the species occur only in the limited western section of the range and only a few species occur in the major portion of the range of the genus. The wealth of species in the western section stems from the many isolate desert valleys and mountains in that region and the chromosomal characteristics of the early invaders into that region.

Heermanni Group.--during early to middle Pleistocene times, the ancestors of the Heermanni complex penetrated the Colorado-Mojave deserts and eventually spread throughout the Great Basin, California, and Baja California. This ancestral form is best represented cytologically and perhaps morphologically by D. stephensi and D. gravipes (2N=70). Possibly D. stephensi was isolated in the San Jacinto and San Bernardino valleys by the last major mountain building activity in the Middle Pleistocene as postulated by Lidicker (1960:207) for D. merriami parvus Rhoads. For some reason, the isolated stephensi-gravipes populations retained the ancestral karyotype with little modification while other related populations continued on a course of chromosomal reorganization. It is possible that these populations represent relicts of a once much larger

population that was maintained for a considerable time during middle to late Pleistocene in contrast to the population isolates that went on to become D. heermanni and its close relatives. Huey (1962:479) named D. cascus from the Bonsall region of San Diego County, California, describing it as a wide-faced form related to D. stephensi and D. gravipes and commented upon the broken chain of wide-faced forms ending in northwestern Baja California. These wide-faced populations would seem to point toward a derivation of the narrow-faced forms of the Agilis subgroup from a wide-faced ancestor such as D. stephensi or D. gravipes. Lackey (1967:328) arranged D. cascus as a subspecies of of D. stephensi and indicated that stephensi differed from D. gravipes about as much as D. stephensi differed from D. panamintinus. Lackey (1967:329) found, however, that the baculum of stephensi was more similar to that of gravipes than either D. heermanni or D. panamintinus. D. gravipes is karyotypically similar to D. stephensi and very distinct from all other members of the Heermanni Group, indicating that the stephensi-gravipes populations have not been isolated from each other for any great period of time and possibly D. stephensi represents a rather recent isolate from the main gravipes population as evidenced by the greater number of pericentric inversions in the karyotype of stephensi. D. stephensi and D. gravipes are true relicts, now existing only in small

isolated areas and as such readily fit the role of ancestral remnant populations accorded them in this study. The narrow-faced forms D. antiquarius, D. paralius, and D. peninsularis, ar arranged by Lackey (1967:332), are all closely related to D. agilis both morphologically and, at least in the case of D. peninsularis, karyotypically. D. peninsularis possesses 60 chromosomes and a karyotype that is identical to that of the 2N=60 populations of D. agilis. The karyotype of D. agilis is very distinct from that of the D. stephensi-D. gravipes line but can be derived from the karyotype of D. heermanni by incorporation of only one centric fusion and several pericentric inversions. Lackey (1967:333) interpreted the morphological and ecological evidence available to him as suggesting that the narrow-faced subgroup was derived from an arid-dwelling wide-faced form, probably in Baja California and that D. peninsularis an early derivative of the stock that eventually led to D. agilis and other narrow-faced species. The karyotypic evidence does not agree with such a scheme, and I interpret the evidence available, both morphological and cytological, as indicating that D. agilis was derived from the stock that produced D. heermanni, if not actually D. heermanni or D. panamintinus, and developed as an isolate along the the coast of southern California. This isolation may have been achieved by the onset of pluvial climates (Illinoian?), mountain building activity, or both. Once having adapted

to an encroaching chaparral habitat, D. agilis would have been able to spread throughout the coastal mountains and into Baja California without competition from other kangaroo rats. After development as a distinct species, D. agilis may have spread northward beyond its present range producing population isolates that underwent still further karyotypic re-organization to emerge as D. venustus and D. elephantinus. These two species are highly specialized members of the Agilis subgroup that are adapted to chaparral covered slopes and brushy areas at the edge of clearings as are some subspecies of D. agilis. These northern narrow-faced forms were apparently excluded from the valleys by the presence of the wide-faced populations already in residence and perhaps also by their own preference for brush habitats (Lackey, 1967:331). That members of the Agilis subgroup will occupy more typical kangaroo rat habitats in addition to brushy areas is evidenced by the occurrence of D. agilis among scattered sage in open stands of Yellow Pine on the Sierra San Pedro Martir (Huey, 1927:7), and in open areas among scattered low shrubs near the sea, along with D. gravipes, 12 miles northeast of El Rosario, Baja California. D. peninsularis prefers level areas between widely scattered desert shrubs. Arriving at a suitable explanation for the origin of the Baja California forms is more difficult, due largely to the present lack of knowledge about the distribution and

relationships of those populations. Morphologically, all are close to D. agilis, and the karyotype of D. peninsularis indicates that probably all were derived from D. agilis as were D. venustus and D. elephantinus. Possibly sea level changes and mountain building accompanied by climatic changes were responsible for cutting off D. peninsularis in the southern end of the Baja Peninsula and D. paralius and D. antiquarius in the northern section. The main population of D. agilis was perhaps forced northward along the California coast. Later D. peninsularis spread northward to eventually contact the range of D. agilis, which had in turn spread southward. The above sequence of events would explain the present distribution of the Baja members of the Agilis subgroup if D. paralius, D. antiquarius, and D. peninsularis are indeed distinct species apart from D. agilis. At least two lines of evidence come to mind that would seem to challenge such an arrangement: the occurrence of two distinctly different karyotypes within D. agilis corresponding to a northern segment in central California and a southern component in southern California and Baja California and the allopative distribution of D. agilis, D. peninsularis, and D. antiquarius. The data presented by Lackey (1967:325) and my own examination of many specimens of this series of related "species" leads me to believe that all of them may actually be subspecies of D. agilis. Some of the reasons for such a decision are

as follows: the cranial measurements of D. antiquarius fit well within the peninsularis series; the karyotype of D. agilis plectilis and D. peninsularis pedionomus are identical, and both differ from D. agilis perplexus; an even clinal distribution of cranial measurements and coloration characters extends through the southern California forms of D. agilis into the D. peninsularis populations in such a way that it is difficult to distinguish the two "species" where they meet; I question the reproductive isolation of D. agilis from D. peninsularis as captive D.p. pedionomus males have repeatedly shown a willingness to mate with females of D. agilis and have been accepted by such females when in estrous, whereas males of forms such as D. stephensi and D. gravipes were attacked. Possibly there are three species within the Agilis subgroup composed of venustus-elephantinus (2N=60), the northern populations of D. agilis (2N=62), and the southern populations of D. agilis (2N=60)beginning with D.a. simulans and including the Baja California populations paralius, peninsularis, and antiquarius. If the various populations of agilis-like forms in Baja California currently regarded as species are actually merely subspecies,at best, then the evolutionary history of the group becomes less complex and the karyotypic evidence would suggest that they were isolated from the parental, more northern populations of D. agilis possibly only since the beginning or

during Wisconsin Pluvial times and have achieved their karyotypic and morphological distinctness comparatively recently. Current views on the rate of subspeciation in kangaroo rats (Lidicker, 1960:209) indicate that the present subspecies of the D. agilis and D. peninsularis series are no older than late Pleistocene and post-Pleistocene.

Meanwhile, earlier climatic changes favoring aridity apparently eliminated most kangaroo rats from the Mohave Desert. The remaining populations of the Heermanni Group, under severe selective pressures, adapted to burrowing in deep sand as a means of escaping the desert heat and water loss, and D. deserti emerged as one of the most highly specialized of the kangaroo rats. Residence and population spreading within this area may have been possible by other species only during Pluvial periods cooler than the present, for only two species, D. merriami and D. deserti, have been able to successfully colonize the desert floor of that region with any degree of permanence. All other species occurring in the region do so, at present, only in isolated, cooler areas of scattered grass and tree yucca above the desert floor. The karyotype of D. deserti (2N=64) shows a general similarity to other members of the Heermanni Group and karyotype re-organization has proceeded to about the same degree as in D. heermanni or D. panamintinus, both of which also possess 64 chromosomes.

The karyotype of D. microps is similar in many respects to that of D. agilis perplexus and may have been derived from an early population of D. agilis that spread into the Great Basin regions of Nevada and Utah. Quite possibly, however, the similarity between the karyotypes of these two species results from both populations having been derived from a common ancestral population, probably D. heermanni or D. panamintinus, during or since Middle Pleistocene times. Isolation of D. microps may have resulted during a Pluvial period at which time the amount of inhabitable durface area would have bee greatly reduced, since many of the basins to the south and west of the major part of the present range of D. microps were filled with water. The present ranges of the subspecies of D. microps possibly result from population spreading and subsequent isolation only since Wisconsin or post-Wisconsin times as indicated by the amount of differentiation within the group and the presence of distinct subspecies in areas that would have been under the waters of Pleistocene lakes. Durrant (1952:501) presents evidence on the effects of late Pleistocene Lake Bonneville on the distribution of both D. ordii and D. microps and suggests that the latter species entered the Bonneville Basin from the west and arrived more recently than did D. ordii.

As I have already inferred, D. heermanni was apparently derived from an ancestral stock related to D. stephensi and

D. gravipes, as an isolate in the central valleys of California west of the Tehachapi Mountains, by mid-Pleistocene times. The two remaining species of the Heermanni Group, D. ingens and D. panamintinus, are probably derived from D. heermanni, and at least in the case of ingens, comparatively recently. D. ingens differs from D. heermanni by two pericentric inversions that reduce the number of telocentric pairs by two, while panamintinus differs from heermanni by one such arrangement plus four pairs of chromosomes that have short inversions not matched by heermanni. The relatively unspecialized morphology of panamintinus and the differences present in its karyotype suggests that isolation and subsequent differentiation from heermanni occurred while the parent population still possessed 19 or more pairs of telocentric chromosomes. The occurrence of distinct subspecies within both species would also seem to support an early separation. The large size of D. ingens would appear to contradict this supposition concerning its origin, however, the karyotypes of ingens and heermanni are so markedly similar that I have concluded that ingens was derived only after the karyotypic characteristics of heermanni were sijilar to that we now find. D. ingens was apparently isolated in the Carrizo Plains area whereas panamintinus represents an earlier derived population that may have been isolated from populations leading to heermanni by the rise of the Tehachepi Range and perhaps

also by climatic conditions. The wane of Wisconsin Pluvial conditions left "islands" of D. panamintinus above the desert floor, and a small amount of karyotypic difference already occurs between these populations reflecting their isolation. The effects of the extremely arid Mojave Desert can hardly be over-emphasized in the restriction of eastward movement by the developing Heermanni complex or in the development of the highly specialized sand-dwelling form, D. deserti. The Colorado River may have served as a barrier for some members of the Heermanni Group but did not prevent movement of deserti at its southern end.

In my opinion the high degree of allopatry shown by members of the Heermanni Group lends support to the above phylogenetic scheme.

Spectabilis Group.--members of this group apparently evolved on the high deserts of northern Mexico and southern New Mexico-southern Arizona and, while acquiring behavioral and morphological specializations, have retained a generalized karyotype (2N=72). D. elator appears to be a relict species that morphologically may best represent the ancestral population from which D. spectabilis was derived. The karyotype of D. elator is only slightly more derived than that of D. spectabilis spectabilis which possesses the most generalized and what is presumed to be the ancestral karyotype for the genus. Seemingly, D. elator developed as isolated form separated from the main population early in

the development of D. spectabilis or else is a true relict of a once more wide-ranging group ancestral to spectabilis that was eliminated by the spread of its highly competitive offspring, D. spectabilis. The karyotype of D. elator is similar to that of the cytologically more generalized subspecies of D. spectabilis, but the morphology of the bi-armed chromosomes and the short arms on some of the chromosomes representing pericentric inversions not indicated in the FN count may indicate a very long period of separation between these two species.

D. nelsoni was apparently derived directly from D. spectabilis as a population isolated in central Mexico and retains many of the morphological characteristics of D. spectabilis and a diploid count of 72. The great difference in the number of pericentric inversions incorporated into the karyotype as opposed to the number in the cytologically known subspecies of D. spectabilis indicates that nelsoni was isolated as a small population apart from the parental population for some time and then subsequently spread its range to again meet the range of D. spectabilis. Interspecific competition with D. spectabilis may prevent D. nelsoni from further extending its range. When the karyotype of D. nelsoni and the northern subspecies of D. spectabilis are compared, the degree of difference supports the species recognition here accorded D. nelsoni. However, the subspecies of D. spectabilis differ from each other in

the same way that they differ from nelsoni but to a lesser degree. In spectabilis the number of pericentric inversions may increase from north to south in the chain of subspecies reaching from Arizona south deep into Mexico. At least such an increase is present in the karyotypes of the subspecies north of Mexico. If the trend continues southward, then D.s. cratodon Merriam may possess a karyotype similar to that of D. nelsoni. The possiblity also exists that D.s. cratodon is actually a separate species; I know of no reliable evidence to determine if the karyotypically distinct populations of D. spectabilis or D. agilis are capable of interbreeding without significant reduction of fertility or viability in the offspring.

Phillipsi Group.--only D. ornatus has been examined thus far, but D. phillipsi is undoubtedly closely related and probably karyotypically similar. D. ornatus retains the ancestral diploid count of 72, but most chromosomes show small second arms derived through pericentric inversions. In the case of ornatus the arms are of sufficient length to be counted in the FN whereas in D. elator, they were fewer and smaller and not counted. The karyotype of ornatus does not closely resemble that of any other kangaroo rat studied and differs enough to indicate an earlier separation from the ancestral stock before any of the other 72 chromosome forms acquired their specific or group characteristics; the Phillipsi Group is at least as distinct from the

Heermanni and spectabilis groups as the latter are from each other. D. ornatus and D. merriami were taken in the same trap line near Bledos in San Luis Potosi, Mexico, and appeared to have very similar ecological requirements. Possibly members of the Phillipsi Group were prevented from extending their range northward due to competition with other species such as the ecologically flexible D. merriami. Baker and Greer (1962:103) indicate that in Durango ornate kangaroo rats occup elevated grasslands ehich might be expected to support D. spectabilis and that D. ornatus appears to have occupied these areas after the retreat of the larger species from the region. Farther to the north, habitat suitable for D. ornatus is held by yet another form, D. ordii, which apparently does not mix with ornatus where their ranges meet in Mexico.

Ordii Group.--the large series of interbreeding populations comprising the species D. ordii probably developed from a small ancestral population derived from a complex of generalized kangaroo rats inhabiting the plains region of Oklahoma-Kansas and adjoining regions of New Mexico and Texas. The species as now known retains a generalized morphology and possesses a karyotype with a high diploid number of 72 but has incorporated many pericentric inversions. Possibly this highly modified karyotype was established, presumably in a small population, before the species becaome so widespread because all subspecies

examined to date possess the same karyotype. D. ordii probably represents a separate line of development conserving the ancestral generalized morphology and giving rise to no other lines of descent. The populations from which D. ordii was derived, however, also could have been ancestral to one or all of the other species groups with the possible exception of the Merriami Group.

Merriami Group.--members of this group show the greatest degree of karyotype modification from the presumed ancestral condition; all autosomes are bi-armed, and the diploid count has been greatly reduced. Seemingly, this group has evolved separately from the other kangaroo rats for a considerable period of time and probably represents an old line of descent of forms that adapted early in their development to xeric conditions as indicated by their ability to conserve metabolic water, habitat preferences, and specializations toward saltation. That the ancestral diploid count for the group was higher, as theorized, is supported by the higher count of D. nitratoides (2N=54) as opposed to D. merriami (2N=52). D. nitratoides may have developed after isolation in the San Joaquin Valley of California by the rise of mountain barriers and before D. merriami developed its specific characters and its diploid count of 52. D. merriami probably developed in the Mojave and Colorado deserts (Lidicker, 1960:206); derived from the same general ancestral population that also

gave rise to D. nitratoides. At least it appears that merriami acquired its diploid count of 52 after the isolation of nitratoides and before becoming so widespread since all subspecies examined possess the same karyotype. It is perhaps significant that both of the two most "successful" species of kangaroo rats possess a chromosome complement of entirely bi-armed autosomes that was apparently set before the species became so widespread.

Johnson and Selander (1971), in their extensive analysis of protein variation in kangaroo rats, found that species of Dipodomys possessed low levels of genetic variability in comparison with other organisms which have been studied. D. merriami is relatively variable genically whereas D. ordii possesses a relatively low level of genic heterozygosity. In kangaroo rats the level of genetic variability possibly reflects past population size during speciation as much as any other factor.

The karyotype of D. insularis is not yet known but should contribute toward understanding the history of this group since, as indicated by Lidicker (1960:207), members of this species may have been isolated from D. merriami for a great period of time.

CHAPTER V

SUMMARY AND CONCLUSIONS

The mitotic chromosomes of 18 species of the kangaroo rats, genus Dipodomys, were evaluated as indicators of interspecific and species group relationships. The principle method of analysis involved pairing of putative homologues from photographic cut-outs of mitotic metaphase chromosomes to produce a representative karyotype for each population studied. Analysis of the karyotype included determination of the diploid number, number of major autosomal arms (FN), placement of each chromosome pair in a specific arm-ratio class, and determination of sex chromosomes. Meiotic metaphase material was used in some cases as an aid in determining general sex chromosome morphology. Diploid number was found to range from 52 to 72 while fundamental number ranged from 70 to 140.

Five major species groups of kangaroo rats were indicated by the results of karyotype analysis. These were similar to the species groups of other workers in general content, however, some important re-arrangements in the groups, both in content and phylogenetic order was necessary on the basis of karyotypic relationships.

Over most of the range of the genus all species of Dipodomys except D. merriami had 72 chromosomes; while in the Great Basin, California-Baja California forms and the wide-ranging D. merriami, lower counts occurred. The species of kangaroo rats with 72 chromosomes were few in number (5) and occurred over the greater portion of the geographic range of the genus; the ancestral count is thought to be 72. Of these 72 chromosome forms, D. ordii had been regarded by most other investigators as the most primitive member of the genus because of its generalized morphology. Karyotypic features of D. ordii indicated that its position as the basal member of the genus was not justified and the monotypic Ordii Group was placed after the other 72 chromosome species groups, the Spectabilis and Phillipsi groups, which were placed before the Ordii Group in that order. The Spectabilis Group contained D. elator, D. spectabilis, and D. nelsoni. Karyotypically, D. spectabilis was considered nearer the ancestral condition for the genus, and D. nelsoni was considered to be a direct line from D. spectabilis. D. elator was considered to be a specialized relict derived from the same stock that gave rise to D. spectabilis. The Phillipsi Group contained D. ornatus which also had 72 chromosomes but the group did not appear to be closely related karyotypically to any other kangaroo rat group.

The Heermanni Group contained the majority of the species and was characterized by diversity in diploid and fundamental numbers, as well as having a considerable degree of morphological variety. Five subgroups are indicated in this group, characterized, in general, by differences in diploid number. The stephensi subgroup (my subgroup A) was thought to be the most generalized of the Heermanni Group and perhaps best represents the ancestral populations that produced the Heermanni Group. The two species contained in this subgroup, D. gravipes and D. stephensi both possessed 70 chromosomes and had fundamental numbers of, respectively, 71 and 86. The 64 chromosome forms, D. heermanni, D. ingens, and D. panamintinus were considered to be less generalized than the preceeding subgroup and their karyotypes appeared to have been derived from 70 chromosome forms by centric fusions and pericentric inversions. The "wide-faced" D. heermanni subgroup (my subgroup B) is thought to have produced the "narrow-faced" or D. agilis subgroup (my subgroup C): this latter subgroup included D. agilis with populations in southern California and Baja California that possessed 60 chromosomes and populations in central California that pcssessed 62 chromosomes. D. agilis was considered to be ancestral to the 60 chromosome forms, D. venustus, D. elephantinus, and D. peninsularis (and also to the related forms not sampled, D. paralius and D.

antiquarius). Of these, all except D. venustus may eventually be considered to be only subspecifically distinct or less. D. microps is placed in the monotypic microps subgroup (my subgroup D) although it also possessed 60 chromosomes and is thought to have been derived from the same ancestral stock ad D. agilis. D. deserti was also considered to be a Heermanni Group species and possessed 64 chromosomes and a karyotype that was similar in general aspects to the other 64 chromosome forms such as D. panamintinus. The karyotype of deserti contained enough structural differences compared to that of the other 64 chromosome forms to indicate that it had been a distinct line for a long period of time. This difference in karyotype and the specialized morphology of D. deserti indicated, as with D. microps, that it belonged in a subgroup of its own, the deserti subgroup (my subgroup D).

The Merriami Group contained D. merriami and D. nitratoides. This group is characterized, karyotypically, by low diploid numbers of 52 and 54, respectively, and comparatively high fundamental numbers of 100 and 104. These values indicated that centric fusions have been more common in the evolution of this group, which has been distinct from other kangaroo rats for a considerable time.

Conclusions based upon the results of this study were as follows:

1. The genus Dipodomys probably developed during late Pliocene or early Pleistocene time in the arid grassland regions of northern Mexico and southcentral-southwestern United States.

2. Five major groups of kangaroo rats evolved after the initial radiation of the genus in early Pleistocene times.

3. While the primary radiation of the genus was thought to have been centered in the semi-arid grasslands, as indicated above, the most extensive speciation within the genus occurred within the Heermanni Group in the Great Basin and California-Baja California due to the varied topography and climates of that region and the chromosomal characteristics of the ancestral populations.

4. D. spectabilis of the Spectabilis Group karyotypically characterized the ancestral condition for the genus. The relict species, D. elator, is a specialized form probably derived from the same ancestral populations that gave rise to D. spectabilis. D. nelsoni was derived directly from D. spectabilis.

5. The Phillipsi Group was morphologically specialized but karyotypically generalized; this group was quite distinct karyotypically and not closely related to the other groups, and appeared to have been separated early in the radiation of the 72 chromosome forms.

6. The monotypic Ordii Group, although morphologically generalized (possibly due to grassland habitat preferences)

was karyotypically farther separated from the ancestral 72 chromosome forms than were the Spectabilis and Phillipsi groups. The karyotype of D. ordii indicated that this species developed as an isolated, presumably small population, until a high level of karyotypic stability was reached before becoming the most wide-spread member of the genus. While not directly ancestral to any other extant species or group, the populations ancestral to D. ordii possibly also produced the other kangaroo rat groups with possible exception of the Merriami Group

7. The Heermanni Group is comprised of five subgroups forming a series of populations differing in morphological and karyotypical specializations. These are listed in decending order, beginning with the subgroup best representing the ancestral populations: (A) stephensi subgroup; (B) heermanni subgroup; (C) deserti subgroup; (D) agilis subgroup; (E) microps subgroup.

8. D. stephensi and D. gravipes were closely related relict species that best represent the ancestral condition for the Heermanni Group.

9. D. heermanni developed in the interior valleys of central California from ancestors related to D. gravipes and D. stephensi; D. ingens was derived directly from D. heermanni while D. panamintinus may have been derived from D. heermanni earlier or a population that was ancestral to both.

10. D. deserti was not a member of the Spectabilis Group as had been suggested by Lidicker (1960) but was found to be a member of the Heermanni Group that developed in the Mojave and Colorado deserts and was highly specialized for occupying an extremely arid, deep sand habitat.

11. D. agilis was derived from D. heermanni or D. panamintinus or a population ancestral to these species, and developed as an isolate along the coastal slopes of California. Once having adapted to an encroaching chaparral habitat, D. agilis spread widely throughout the coastal hills of California and south into Baja California. Isolated populations of D. agilis produced the D. venustus-elephantinus complex and the D. peninsularis-antiquarius-paralius complex. Of these taxa, only D. venustus is of certain species distinction.

12. The "wide-faced" forms in southern California-Baja California were not directly ancestral to the "narrow-faced" Heermanni Group forms in Baja California as had previously been suggested by Lackey (1967).

13. D. microps was not a member of the Ordii Group as earlier arranged by Setzer (1949) but was a comparatively recently derived Heermanni Group species which developed in the Great Basin regions of Nevada and Utah.

14. The Merriami Group was more distinct from other kangaroo rats than the latter are from each other, possibly indicating a longer separation of the group from the ancest-

ral line. Karyotypically, members of this group had diverged farther from the putative ancestral condition (72 chromosomes) and possessed the lowest diploid numbers (52-54) in the genus. D. nitratoides developed in the central valleys of California as an isolate from the ancestral population that gave rise to D. merriami. I believe D. merriami to have developed in the Mojave-Colorada deserts region as a small population that, under severe selective pressures, achieved a highly stabalized karyotype before spreading widely. The higher diploid number of D. nitratoides supports this scheme. The spreading and subsequent subspeciation of D. merriami was remarkably similar to that of D. ordii in that both achieved highly modified karyotypes before becoming the most widespread and "successful" species of the genus.

15. Results from the study of bacula supported the species groups as arranged on the basis of karyotye data.

16. Study of meiotic chromosomes is an aid to the preparation of reliable karyotypes for some species of kangaroo rats.

17. The species groups within the genus Dipodomys actually represent superspecies groups of closely related, largely allopatric and presumably reproductively isolated populations.

18. Chromosome analysis is an effective tool for deter-

mination of both interspecific and species group relationships in the genus Dipodomys and is considered a more reliable measure of phyletic relationships than indices of skeletal and cranial specialization.

LITERATURE CITED

Baker, R. H. and J. K. Greer. 1962. Mammals of the Mexican State of Durango. Michigan State Univ., Publ. Museum, Biol. Ser., 2:25-154.

Blair, W. F. 1954. Mammals of the mesquite plains biotic district in Texas and Oklahoma and subspeciation in the central grasslands. Texas Jour., Sci., 3:248.

Burt, W. H. 1936. A study of the baculum in the genera Perognathus and Dipodomys. J. Mamm., 17:145-156.

Cross, J. C. 1931. A comparative study of the chromosomes of rodents. J. Morphol., 52:373-402.

Davis, W. B. 1942. The systematic status of four kangaroo rats. J. Mamm., 23:328-333.

Downs, T. and J. A. White. 1968. A vertebrate faunal succession in superposed sediments from late Pliocene to middle Pleistocene in California, 41-47. In Tejhol, Jiri (ed.). Tertiary/Quaternary Boundary. Academia, Prague, 88 pp., 1968 (Proc. Sec. 10, Internatl. Geol. Congr.).

Durrant, S. D. 1952. Mammals of Utah. Taxonomy and distribution. Univ. Kansas Publ., Mus. Nat. Hist., 6: 1-549.

Ford, C. E. and J. L. Hamerton. 1956. A colchicine, hypotonic-citrate, squash sequence for mammalian chromosomes. Stain Tech., 31:247-254.

Grinnell, J. 1921. A revised list of the species in the genus Dipodomys. J. Mamm., 2:94-97.

_____ 1922. A geographical study of the kangaroo rats of California. Univ. Calif. Publ. Zool., 40: 71-234.

Hall, E. R. and K. E. Kelson. 1959. Mammals of North America. The Ronald Press, New York, 1:xxx + 1-546 + 79.

Hibbard, C. W. 1958. Summary of North American Pleistocene Mammalian Local Faunas. Papers Mich. Acad. Sci., Arts and Letters, 43:1-32.

_____ 1960. Two Late Pleistocene Faunas from southwestern Kansas. Contrib. Mus. Paleontol. Univ. Mich., 12:47-96.

Hsu, T. C. and F. E. Arrighi. 1968. Chromosomal evolution in the genus Peromyscus (Cricetidae, Rodentia). Cytogenetics, 5:355-359.

Huey, L. M. 1951. The kangaroo rats (Dipodomys) of Baja California Mexico. Trans. San Diego Soc. Nat. Hist., 11:205-256.

_____ 1962. Two new species of broad-faced, five-toed kangaroo rats (genus Dipodomys). Trans. San Diego Soc. Nat. Hist., 12: 477-480.

Johnson, Walter D. and R. K. Selander. 1971. Protein variation and systematics in kangaroo rats (genus Dipodomys). Syst. Zool., 20: in press.

Lackey, J. A. 1967. Biosystematics of Heermanni group kangaroo rats in Southern California. Trans San Diego Soc. Nat. Hist., 14:315-342.

Lee, M. R. 1969. A widely applicable technique for direct processing of bone marrow for chromosomes of vertebrates. Stain Tech., 44: 155-157.

Lewis, H. 1966. Speciation in Flowering Plants. Science, 152:167-172.

Lidicker, W. Z., Jr. 1960. An analysis of intraspecific variation in the kangaroo rat Dipodomys merriami. Univ. Calif. Publ. Zool., 67: 125-218.

Matthey, R. 1951. The chromosomes of the Vertebrates. Pp. 159-180, in M. Demerec (ed.), Advances in Genetics, IV.

_____ 1952. Chromosomes de Muridae (II.), Experentia, 8:389-390.

_____ 1956. Nouveau apports a la cytologie comparée des Rongeurs. Chromosoma (Berl.), 7:670-692.

Meredith, R. 1969. A simple method for preparing Meiotic Chromosomes from mammalian testis. Chromosoma (Berl.), 26:254-258.

Patton, J. L. 1967. Chromosome studies of certain pocket mice, genus Perognathus (Rodentia: Heteromyidae). J. Mamm., 48:27-37.

Scherz, R. G. 1962. Blaze-drying by igniting the fixitive for improved spreads of chromosomes in leucocytes. Stain Tech., 37:386.

Setzer, H. W. 1949. Subspeciation in the kangaroo rat Dipodomys ordii. Univ. of Kansas Publs. Mus. Nat. Hist., 1:473-573.

Tobias, P. V. 1956. Chromosomes, sex-cells, and evolution of a mammal. London: Percy Lund, Hemphries and Co., Ltd. 1956. 420 pp.

Wood, A. E. 1935. Evolution and relationships of the heteromyid rodents. Ann. Carnegie Mus. XXIV:73-262.