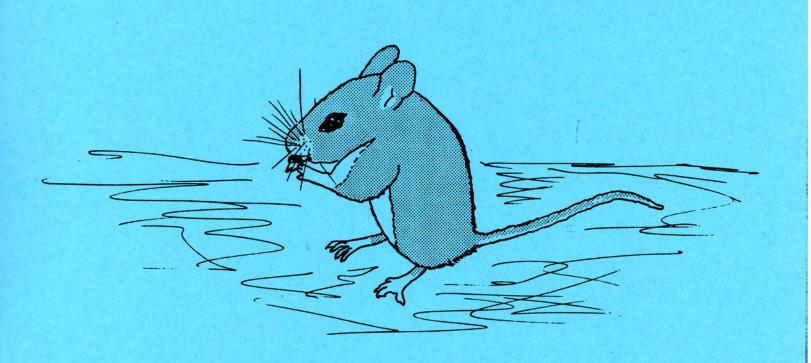
# PEROMYSCUS NEWSLETTER

# NUMBER SEVENTEEN



**MARCH 1994** 

Cover: Deer mouse. Based on photo by
Breck P. Kent in "Mouse" Calendar.
(See News and Comment, page 5)

# IN ISSUE NUMBER 17:

In this issue we feature species hybridization in *Peromyscus*. Historically one of the attributes which attracted evolutionary biologists to the genus was the potential for hybridization within and among some of the 55 odd existing species. Dice and his colleagues during the 1930s demonstrated that viable hybrids could be produced in certain crosses between *Peromyscus* species which are members of the same species group. Subsequently, others utilized this ability to examine a wide range of genetic and reproductive phenomena. The capacity of species to form hybrids also is relevant information for systematics, as Carleton (1989) has pointed out. The Table on pages 14 - 21 presents a synopsis of interspecific hybridization studies accomplished to date, including those attempted by artifical insemination and *in vitro* fertilization.

Also, we have an update on the Hantavirus-*Peromyscus* connection, with some additional information on the spread of hantaviral pulmonary syndrome within the United States. See page 6.

Offprints of the hantavirus summary, which appeared in PN #16 (September '93) are available from the Stock Center on request and without charge.

And concerning hantavirus, in CDC sampling of numerous Stock Center animals of all species and stocks we maintain, all *Peromyscus* consistently test negative for the virus. We are continuing the moratorium on accepting additional animals into the Center unless they are clearly hantavirus-free. We also screen routinely for 14 murine viruses and *Borrelia* (Lyme disease).

We appreciate the contributed entries for this issue. We are certain that most of you will find items of interest. Please continue to send news of your *Peromyscus* research.

Deadline for entries for the next issue is 15 September 1994.

# PEROMYSCUS NEWSLETTER is produced by the

Peromyscus Genetic Stock Center
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Institute of Biological Research and Technology
University of South Carolina
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# **NEWS, COMMENT and ANNOUNCEMENTS**

Our thanks to Harold Egoscue who gave the Stock Center a collection of skins representing mutant *P. maniculatus*. Among the specimens is the original wild-caught white-bellied non-agouti (J. Hered. 62:372). The Center has a color slide of this animal received in a previous donation from Harold.

# XXXXXXX

Kelly Hogan, Ira Greenbaum and their associates have formalized the species *Peromyscus keeni* to include many Pacific Northwest representatives of the *P. maniculatus* species group (J. Mamm. 74:819ff). Included in *P. keeni* are *P. oreas* and *P. sitkensis* as well as the subspecies *P. m. algidus*, *P. m. hylaeus*, *P. m. keeni*, *P. m. macrorhinus* and *P. m. prevostensis:* This assemblage has long been suspected of being specifically distinct from *P. maniculatus* based on cytogenetic and other information, often being referred to informally as "the *oreas* complex". The epithet "*keeni*" has priority in the literature, however.

Ramone Baccus writes that she has moved to the Bureau of Land Management at Malta, Montana. She says it will be "nice to be back in Peromyscus country" after a few years in Alaska.

**Glenn Haas** of Boulder City NV reports some interesting history of his experiences collecting rodent ectoparasites. See his entry under "Contributions".

<><><><><>

Several volumes of back issues of *The Journal of Heredity* were donated to the Stock Center by Mike Felder. Mike with his, former students, Yao-wu Zheng and Monica Bey, recently published cDNA sequences from *P. maniculatus* for *Adh-1* and that of a second class representing the *Adh-2* gene (J. Biol. Chem. 268:24933ff). The *Adh-1* gene is deleted in ADH-negative deer mice.

++++++++

A significant paper by **Jerry Wolff** recently was published in Nature (359:409ff). He reports data that support the view that juvenile dispersal mechanisms in P. leucopus are important in inbreeding avoidance. In another recent paper (J. Mamm. 74:846ff), Jerry examines whether the "Chitty effect" (larger body size correlated with high population density) occurs in Peromyscus. His initial results indicate that it might.

~~~~~~~~~~~~~~~~

Those of us who work with laboratory-reared deer mice are frequently asked: "How closely related to *Peromyscus* is *Mus*?" or "when did the ancestry of *Mus* and *Peromyscus* diverge?" These questions have been difficult to answer. Fossil (Jacobs and Pilbeam, *J. Human Evol.* 9:551ff) indicate separate lineages by 15 mybp. DNA/DNA hybridization data suggests ~16 mybp separation (Catzeflis *et al. MAMMAL PHYLOGENY* II pp 159ff). At the other extreme Vincent Sarich (pers. comm.), based on an immunological clock, proposed ~35 mybp. A 1992 paper by O'hUigin and Li (*J. Mol. Evol.* 35:377ff) may provide the best estimate. Considering divergence of hamsters (*Mesocricetus, Cricetulus*) from *Mus* and *Rattus*, based on nucleic acid sequences of 28 genes of these and outgroup representatives, the cricetine and murine lines diverged 32 to 46 mybp. Assuming the *Peromyscus* derives from a cricetine (*sensu lato*) ancestry, the *Peromyscus - Mus* divergence is likely no later than the *Mus - Mesocricetus* split.

The very attractive MOUSE calendar for 1994 features 12 excellent full color 9" X 13" photographs of *Peromyscus maniculatus* and *P. leucopus* in natural settings. No *Mus* or other critters. Although 1994 is well underway, peromyscologists may still want to obtain one. It is definitely a "look-see", not a "write on" calendar. The photos could be framed for display long after the year is done. It is available from Camden House Publishing, 7 Victoria Road, Camden East, Ontario K0K 1J0, Canada. The price is US\$ 9.95.

\* \* \* \* \*

1993 was the most active year to date for the Peromyscus Genetic Stock Center. We supplied more than 1000 specimens to external users for research and educational purposes. About 25% of the external utilization was hantavirus research-related. In addition to external use, more than 225 specimens were utilized by investigators at the University of South Carolina.

# XXXXXXXXXXXX

We learned, very belatedly, that Prof. Ardzroony Packchanian has passed away. Packchanian was on the Microbiology faculty of the University of Texas Medical School at Galveston. He conducted some collaborations on deer mouse research with the late R.R. Huestis of the University of Oregon as early as 1938. His final contribution to the Peromyscus literature was a decription of a new hairless mutant in P. californicus (1984. J. Hered. 75:229ff). For many years Packchanian maintained a modest deer mouse colony which included albino and other mutations derived from those kept by Huestis.

# \/\/\/\/\/

We received a letter very complimentary of the Newsletter from Nancy Shefferly of the University of Michigan Museum of Zoology. She refers to our readership as a "Peromyscus club of sorts". We appreciate the encouragement!

#####

IT'S OUT!! The newly revised *Peromyscus* Standardized Karyotype has just been published in *Cytogenetics and Cell Genetics* (1994.181ff). Several years of dedicated effort by **Ira Greenbaum** and his collaborators are represented by this review and revision. There will now be a sound frame of reference for those involved in gene mapping and cytogenetics of *Peromyscus* and allied genera. It will also be an important reference for phylogenetic and systematic investigations of myomorphic rodents. We urge attention to this document by anyone interested in *Peromyscus* evolution and genetics.

•••••

THE YOUNGEST PUBLISHED PEROMYSCOLOGIST ??? Daniel Compton, Age 10, is coauthor of the paper "Transport of a Juvenile Pinyon Mouse (*Peromyscus truei*)" which appeared recently in *Great Basin Naturalist* (1993. 53:316-317).

# HANTAVIRUS UPDATE

The hantaviral disease which emerged recently in the United States, initially referred to as "Four Corners disease" or "Navaho flu", has now been officially designated Hantaviral Pulmonary Syndrome or HPS. The virus itself has been named "Four Corners virus" (FCV) and another isolate from California (appropriately!) "Muerto Canyon virus". The deer mouse, *Peromyscus maniculatus*, has been implicated as the major host for this virus. Basic information about the disease and the role that deer mice play in its transmission are contained in *PN* #16.

53 cases had been confirmed by CDC as of the end of 1993, and several additional cases have been reported since. The peak number of cases appearing during May, June and July of 1993 centered in New Mexico, Arizona and Colorado. Until recently all cases were recorded from west of the Mississippi. By early this year cases were known in Indiana, Florida and New York. The virus in the Florida case appears to be a new strain carried by Sigmodon hispidus.

Amino acid sequence alignment of Four Corners virus with those of other hantaviruses indicates that it is related more closely to the Puumala and Prospect Hill viruses than to the Hantaan, Belgrade and Soule hantaviruses. The former are associated with microtine and cricetine hosts, while the latter are murine-associated (Hjelle *et al.* 1993). FCV has the highest mortality rate of any known hantavirus disease and affects primarily the pulmonary system, whereas the Hantaan group results in renal complications. Some evidence suggests that the severe symptomology associated with human hantavirus infection is the result of an exaggerated immune response.

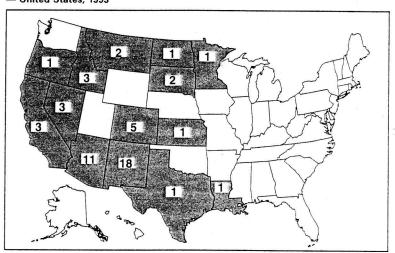


FIGURE . Number of reported confirmed cases of hantavirus pulmonary syndrome — United States, 1993

# Sources for more information:

Discover. December 1993 issue. (Cover story) Denise Grady, "Death at the Corners" 14:82-91.

Hjelle et al. 1994. J. Virol. 68:592-596.

MMWR 1993. (Oct. 29) 42:816-820.

MMWR 1994. (Jan. 28) 43:45-48.

Science 5 November 1993 issue (v.262): News & Comment 832-836; Hughes et al. 850-851; Nichol et al. 914-917.

# PEROMYSCUS STOCK CENTER

What is the Stock Center? The deer mouse colony at the University of South Carolina has been designated a genetic stock center under a grant from the Special Projects Program of the National Science Foundation. The major function of the Stock Center is to provide genetically characterized types of *Peromyscus* in limited quantities to scientific investigators. Continuation of the center is dependent upon significant external utilization, therefore potential users are encouraged to take advantage of this resource. Sufficient animals of the mutant types generally can be provided to initiate a breeding stock. Somewhat larger numbers, up to about 50 animals, can be provided from the wild-type stocks.

A user fee of \$10 per animal is charged and the user assumes the cost of air shipment. Animals lost in transit are replaced without charge. Tissues, blood, skins, etc. can also be supplied at a modest fee. Arrangements for special orders will be negotiated. Write or call for details.

# Stocks Available in the Peromyscus Stock Center:

|                                                       | ***************************************                                                                                          |
|-------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------|
| WILD TYPES                                            | ORIGIN                                                                                                                           |
| P. maniculatus bairdii<br>(BW Stock)                  | Closed colony bred in captivity since 1948.  Descended from 40 ancestors wild-caught near Ann Arbor MI                           |
| P. polionotus subgriseus<br>(PO Stock)                | Closed colony since 1952.  Derived from 21 ancestors wild-caught in  Ocala Nat'l. Forest FL. High inbreeding coefficient.        |
| P. polionotus leucocephalus<br>(LS Stock)             | Derived from beachmice wild-caught on Santa Rosa I., FL. and bred by R. Lacy. Fourth to seventh generation in captivity.         |
| P. leucopus<br>(LL Stock)                             | Derived from 38 wild ancestors captured between 1982 and 85 near Linville NC. Eighth to tenth generations in captivity.          |
| P. californicus insignis<br>(IS Stock)                | Derived from about 60 ancestors collected between 1979 and 87 in Santa Monica Mts. CA. Seventh to tenth generation in captivity. |
| P. aztecus                                            | Derived from animals collected on Sierra Chincua,<br>Michoacan, Mexico in 1986<br>Fourth to seventh generation in captivity.     |
| P. maniculatus X P. polionotus F <sub>1</sub> Hybrids | Sometimes available.                                                                                                             |

# MUTATIONS AVAILABLE FROM THE STOCK CENTER<sup>1</sup>

| Cost | Colors |
|------|--------|
| Coat | Colors |

# **ORIGINAL SOURCE**

Albino c/c

Sumner's albino deer mice

(Sumner, 1922)

Ashy ahy/ahy

Wild-caught in Oregon ~ 1960

(Teed et al., 1990)

Black (Non-agouti) a/a

Horner's black mutant

(Horner et al., 1980)

Blonde bl/bl

Mich. State U. colony

(Pratt and Robbins, 1982)

<sup>2</sup>Brown b/b

Huestis stocks

(Huestis and Barto, 1934)

Dominant spotting S/+

Wild caught in Illinois

(Feldman, 1936)

Golden nugget bgn/bgn [in P. leucopus]

Wild caught in Massachusetts

(Horner and Dawson, 1993)

Gray g/g

Natural polymorphism

From Dice stocks (Dice, 1933)

Ivory i/i

Wild caught in Oregon

(Huestis, 1938)

<sup>3</sup>Pink-eyed dilution p/p

Sumner's "pallid" deer mice

(Sumner, 1917)

Platinum pt/pt

Barto stock at U. Mich.

(Dodson et al., 1987)

<sup>2</sup>Silver si/si

Huestis stock

(Huestis and Barto, 1934)

Tan streak tns/tns

Clemson U. stock from N.C.

(Wang et al. 1993)

Variable white Vw/+

Michigan State U. colony

(Cowling et al. 1994)

White-belly non-agouti aw/aw

Egoscue's "non-agouti"

(Egoscue, 1971)

Wide-band agouti AND/a

Natural polymorphism. U. Michigan stock

(McIntosh, 1954)

Yellow y/y

Sumner's original mutant

(Sumner, 1917)

# MUTATIONS AVAILABLE FROM THE STOCK CENTER<sup>1</sup> (continued)

| Other Mutations and Variants                                         | ORIGIN                                                                    |
|----------------------------------------------------------------------|---------------------------------------------------------------------------|
| Alcohol dehydrogenase negative<br>Adh°/Adh°                          | South Carolina BW stock<br>(Felder, 1975)                                 |
| Alcohol dehydrogenase positive<br>Adh <sup>f</sup> /Adh <sup>f</sup> | South Carolina BW stock<br>(Felder, 1975)                                 |
| <sup>4</sup> Boggler <i>bg/bg</i>                                    | Blair's P. m. blandus stock<br>(Barto, 1955)                              |
| Cataract-webbed cwb/cwb                                              | From Huestis stocks.<br>(Anderson and Burns, 1979)                        |
| <sup>4</sup> Epilepsy <i>ep/ep</i>                                   | U. Michigan artemisiae stock<br>(Dice, 1935)                              |
| <sup>3</sup> Flexed-tail <i>f/f</i>                                  | Probably derived from Huestis<br>flexed-tail (Huestis and<br>Barto, 1936) |
| Hairless-1 hr-1/hr-1                                                 | Sumner's hairless mutant<br>Sumner (1924)                                 |
| Hairless-2 hr-2/hr-2                                                 | Egoscue's hairless mutant (Egoscue, 1962)                                 |
| ⁴Juvenile ataxia <i>ja/ja</i>                                        | U. Michigan stock<br>(Van Ooteghem, 1983)                                 |

Enzyme variants. Wild type stocks given above provide a reservoir for several enzyme and other protein variants. See Dawson et al. (1983).

**Note:** Some of the mutations are immediately available only in combination with others. For example, silver and brown are maintained as a single "silver-brown" double recessive stock. Write the Stock Center or call (803) 777-3107 for details.

<sup>&</sup>lt;sup>1</sup>Unless otherwise noted, mutations are in *P. maniculatus*.

<sup>&</sup>lt;sup>2</sup>Available only as silver/brown double recessive.

<sup>&</sup>lt;sup>3</sup>Available only as pink-eye dilution/flexed-tail double recessive.

<sup>&</sup>lt;sup>4</sup>Available from Behavior Mutant Center

# OTHER RESOURCES OF THE PEROMYSCUS GENETIC STOCK CENTER:

Limited numbers of other stocks, species, mutants and variants are on hand, or under development, but are not currently available for distribution. For additional information or details about any of these mutants or stocks contact: Janet Crossland, Colony Manager, Peromyscus Stock Center, (803) 777-3107.

Small numbers (c. 5) of deer mice from either of the two distinct inbred lines (H1 and H8) are available from the Stock Center on a limited basis.

Preserved or frozen specimens of types given above.

Tissues, whole blood or serum of types given above.

PLEASE CALL WITH INCLUDIES

Flat skins of mutant coat colors or wild-type any of the species above.

Reference library of more than 1900 reprints of research articles and reports on *Peromyscus*. Copies can be xeroxed and mailed.

Materials are now available through the *Peromyscus* Molecular Bank of the Stock Center. Allow two weeks for delivery. Included is purified DNA or frozen tissues from any of the stocks listed above. Several genomic and cDNA libraries and a variety of molecular probes are available. (See next page)

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|                |                      |  |  |

Peromyscus Genetic Stock Center University of South Carolina Columbia SC 29208 (803) 777-3107

# PEROMYSCUS BEHAVIOR MUTANT CENTER

A Special Stock Center for behavior mutants of deer mice currently is housed at the University of South Carolina-Aiken. The following variants are available from this center.

# **CONVULSIVE MUTANTS:**

Four different convulsive mutants are maintained. Of these four, only two, Chemogenic Convulsive (CNV) and Epilepsy (ep), have been formally described in the literature.

Alamogordo Convulsive (ALG). Affected animals are convulsive after about three months of age and throughout life, with convulsions gradually increasing in severity. In severe seizures, these animals are likely to arch the head and back, to the point of falling over backwards in spasm. This latter behavior is more common in older animals.

Chemogenic Convulsive (CNV). Affected animals are convulsive from about one month of age and throughout life, with convulsions gradually increasing inseverity. CNV/- mutants tend to display convulsive behavior more readily than ALG/- mutants, however the episode is likely to be much less severe.

**Epilepsy** (ep). Convulsions can be elicited in these animals from about twenty-one days of age. These animals usually grow deaf however by about three months of age, and thereafter can no longer be made to convulse. A "waltzing" behavior is often seen in these animals. Differences in the Organ of Corti and the central auditory pathway are associated with this mutation.

**Thompson Falls Convulsive** (*tf*). Homozygotes convulse throughout life and do not grow deaf. "Waltzing" is not commonly seen. The seizure pattern has a slightly later onset (about three months) and tends to be more severe, sometimes resulting in death.

# AGE-DEPENDENT ATAXIAS:

**Boggler** (*bg*). This is an autosomal recessive mutation characterized by increasing ataxia, tremor, and loss of fine motor coordination. Additional finding suggest that diminished tactile responsiveness also occurs with advancing age. These deficits are correlated with axonal dystrophy and neuronal loss in the CNS.

**Juvenile Ataxia** (*ja*). This is an autosomal recessive mutation which exhibits a marked ataxia from the time locomotor activity first begins until about forty-five days of age. The phenotype appears to be exaggerated or ameliorated by changes in dietary carbohydrates. Neuronal changes and loss is evident by 120 days of age.

For information about any of these variants, please contact:

Dr. Suellen A. VanOoteghem
Department of Anatomy
School of Medicine
University of West Virginia
Morgantown WV 26506
(304) 284-5443

# Materials on Deposit in the Peromyscus Molecular Bank

| Accession<br>Number | Item            | Description               | Species         | Donor          | Location <sup>1</sup> |
|---------------------|-----------------|---------------------------|-----------------|----------------|-----------------------|
| Probes and (        | Clones:         |                           | 3               |                | -                     |
| Pr-01               | LINE1           | pDK62                     | P. maniculatus  | D. Kass        | c                     |
| Pr-02               | LINE1           | pDK55                     | P. maniculatus  | D. Kass        | C                     |
| Pr-03               | ADH1            | pADH F72                  | P. maniculatus  | M. Felder      | В                     |
| Pr-04 <sup>2</sup>  | Mys             |                           | P. leucopus     | (Requested)    |                       |
| Pr-05 <sup>2</sup>  | SAT             |                           | P. leucopus     | (Requested)    |                       |
| Pr-06               | 6PGD            | pB5 clones                | P. californicus | S. Hoffman     | Α                     |
| Pr-07               | MHC PeleI       | 38dp2                     | P. leucopus     | M. Crew        | Α                     |
| Pr-08               | MHC PeleI       | 52ap6                     | P. leucopus     | M. Crew        | Α                     |
| Pr-09               | MHC PeleI       | 40Bgl                     | P. leucopus     | M. Crew        | Α                     |
| Pr-10               | MHC PeleI       | 53Pv1                     | P. leucopus     | M. Crew        | Α                     |
| Pr-11               | MHC PeleI       | 37B2                      | P. leucopus     | M. Crew        | Α                     |
| Pr-12               | MHC PeleI       | 37B4                      | P. leucopus     | M. Crew        | Α                     |
| Pr-13               | MHC PeleII      | α3E23                     | P. leucopus     | M. Crew        | Α                     |
| Pr-14               | MHC PeleIII     | 17E2                      | P. leucopus     | M. Crew        | Α                     |
| Pr-15               | MHC PemaI       | pr44                      | P. maniculatus  | M. Crew        | Α                     |
| Libraries:          |                 |                           |                 |                |                       |
| Lb-01               | lambda genomic  | : liver (ADH+)            | P. maniculatus  | M. Felder      | В                     |
| Lb-02               | lambda cDNA     | liver                     | P. maniculatus  | M. Felder      | В                     |
| Lb-03               | lambda genomic  | testis                    | P. leucopus     | M. Crew        | Α                     |
| Lb-04               | cosmid genomic  | testis                    | P. leucopus     | R. Baker       | Α                     |
| Lb-05               | lambda genomic  |                           | P. californicus | S. Hoffman     | Α                     |
| Frozen Tissu        | ie for DNA:     |                           |                 | ,              |                       |
| S-01                | bairdii (BW)    | liver, other <sup>3</sup> | P. maniculatus  | Stk. Ctr.      | Α                     |
| S-02                | subgriseus (PO) |                           | P. polionotus   | Stk. Ctr.      | Α                     |
| S-03                | leucopus (LL)   | liver, other              | P. leucopus     | Stk. Ctr.      | A                     |
| S-04                | wild-caught SC  | liver, other              | P. gossypinus   |                | A                     |
| S-05                | aztecus         | liver, other              | P. aztecus      | J. Glendinning | A                     |
| S-06                | insignis (IS)   | liver, other              | P. californicus | S. Hoffman     | A                     |
| S-07                | inbred PmH1A    |                           | P. maniculatus  | Jackson Lab    | A                     |
| S-08                | inbred PmH8     | liver, other              | P. maniculatus  | Jackson Lab    | A                     |

<sup>1</sup>Location code: A = USoCar SAI 01; B = USoCar CLS 603; C = USoCar CLS 707

<sup>&</sup>lt;sup>2</sup>Not currently available.

<sup>&</sup>lt;sup>3</sup>kidney, spleen, testis, carcass.

# EXPERIMENTAL SPECIES HYBRIDIZATION IN PEROMYSCUS

The following pages summarize published reports of inter-specific hybridization experiments involving *Peromyscus*. Some of the hybridization attempts were conducted in connection with research done for other purposes, *e.g.* gene mapping or embryological studies. In addition to the interspecific crosses given here, there are many reports of hybridization between subspecies within species that we have not addressed in this issue of *PEROMYSCUS NEWSLETTER*.

Most of the references cited in the tables and given on page 22 are cited in **Bibliography of** *Peromyscus* (**Rodentia**): **Genetics** by Bruce Buttler of Canadian Union College. We are aware of several unpublished interspecific hybrid experiments with *Peromyscus*, but have omitted these from the listing in the tables.

Most reported attempts of *Peromyscus* species hybridization have not produced liveborn offspring. Some attempts have involved early embryonic development or *in vitro* fertilization, from which no attempt was made to secure mature hybrid animals. Viable (*i.e.* weaned, but not necessarily fertile) progeny have been obtained from the following *Peromyscus* species crosses:

P. eremicus X P. interparietalis

P. guardia X P. interparietalis

P. guardia X P. collatus

P. truei X P. difficilis (nasutus)

P. leucopus X P. gossypinus

P. maniculatus X P. polionotus

P. maniculatus X P. melanotis

P. maniculatus X P. keeni (oreas)

See tables for more details and literature citations.

# SUMMARY OF EXPERIMENTAL INTERSPECIFIC HYBRIDIZATION ATTEMPTS IN PEROMYSCUS.

| CROSS (FEMALE X MALE)                      | COMMENTS                                                | REFERENCES                |
|--------------------------------------------|---------------------------------------------------------|---------------------------|
| INTERGENERIC CROSS ATTEMPTS                |                                                         |                           |
| P. maniculatus X Mus (musculus) domesticus | Artificial insemination. Five mice. No results.         | Maddock and Dawson (1974) |
| Mus (musculus) domesticus X P. maniculatus | in vitro fertilization. 100 ova. No results.            | Fukuda et al. (1979)      |
| Mus (musculus) domesticus X P. maniculatus | in vitro fertilization. 167 ova. No results.            | Hanada and Chang (1978)   |
| Mus (musculus) domesticus X P. polionotus  | in vitro fertilization. 261 ova. No results.            | Fukuda et al. (1979)      |
| Mus (musculus) domesticus X P. leucopus    | in vitro fertilization. 167 ova. No results.            | Hanada and Chang (1978)   |
| P. maniculatus X. Rattus norvegicus        | Artificial insemination. Five mice. No results.         | Maddock and Dawson (1974) |
| Rattus norvegicus X P. maniculatus         | in vitro fertilization. 108 ova. No results.            | Hanada and Chang (1978)   |
| Rattus norvegicus X P. maniculatus         | in vitro sperm injection. 17/67 ova normal pronuclei.   | Thadana (1980)            |
| Rattus norvegicus X P. leucopus            | in vitro fertilization. Sperm penetration. 18/164 ova.  | Hanada and Chang (1978)   |
| P. maniculatus X Meriones unguiculatus     | Artificial insemination. Six mice. No results.          | Maddock and Dawson (1974) |
| P. maniculatus X Mesocricetus auratus      | Artificial insemination. First cleavage. 2/18 mice.     | Maddock and Dawson (1974) |
| Mesocricetus auratus X P. maniculatus      | in vitro fertilization. Sperm penetration. 271/867 ova. | Hanada and Chang (1978)   |
| Mesocricetus auratus X P. leucopus         | in vitro fertilization. Sperm penetration. 2/200 ova.   | Hanada and Chang (1978)   |
| P. maniculatus X Oryzomys palustris        | Artificial insemination. First cleavage. 2/6 mice.      | Maddock and Dawson (1974) |
| P. maniculatus X. Sigmodon hispidus        | Artificial insemination. First cleavage. 1/12 mice.     | Maddock and Dawson (1974) |
| P. maniculatus X Ochrotomys nuttalli       | Artificial insemination. Seven mice. No results.        | Maddock and Dawson (1974) |
| P. maniculatus X Reithrodontomys humilus   | Artificial insemination. Sperm penetration. 1/6 mice.   | Maddock and Dawson (1974) |

| CROSS (FEMALE X MALE)                        | COMMENTS                                                                          | REFERENCES                                      |
|----------------------------------------------|-----------------------------------------------------------------------------------|-------------------------------------------------|
| INTERSUBGENERIC CROSS ATTEMPTS               |                                                                                   |                                                 |
| P. maniculatus X P. (Podomys) floridanus     | Artificial insemination. 2nd - 3rd cleavage. 1/6 mice.                            | Maddock and Dawson (1974)                       |
| P. maniculatus X P. (Haplomylomys) eremicus  | Caged pairs. No progeny. 29 attempts.                                             | Dice (1933)                                     |
| P. leucopus X P. (Haplomylomys) eremicus     | Caged pairs. No progeny. 24 attempts.                                             | Dice (1933)                                     |
| INTERSPECIES-GROUP CROSS ATTEMPTS            |                                                                                   |                                                 |
| P. boylii <sup>1</sup> X P. (comanche) truei | Caged pairs. No progeny. 9 attempts.                                              | Blair (1943)                                    |
| P. maniculatus X P. truei                    | Artificial insemination. Early cleavage 9/30 attempts.                            | Maddock and Dawson (1974)                       |
| P. leucopus <sup>1</sup> X P. maniculatus    | Caged pairs. No progeny. 69 attempts.                                             | Dice (1933)                                     |
| P. maniculatus X P. leucopus                 | Artificial insemination. 44 attempts. I to term, not viable. 13 to mid-gestation. | Dawson et al. (1972), Maddock and Dawson (1974) |
| P. maniculatus X P. gossypinus               | Artificial insemination. 24 attempts. 13 early cleavage. 1 implantation.          | Maddock and Dawson (1974)                       |

| CROSS (FEMALE X MALE)                                     | COMMENTS                                                       | REFERENCES               |
|-----------------------------------------------------------|----------------------------------------------------------------|--------------------------|
|                                                           |                                                                |                          |
| INTERSPECIFIC CROSS ATTEMPTS WITHIN SUBGENUS HAPLOMYLOMYS | N SUBGENUS HAPLOMYLOMYS                                        |                          |
| P. eremicus X P. californicus                             | Six caged pairs. No progeny.                                   | Dice (1933)              |
| P. eremicus X P. interparietalis                          | Five caged pairs. Nine progeny. F <sub>1</sub> fertile.        | Brand and Ryckman (1969) |
| P. eremicus X P. interparietalis                          | Two caged pairs. No progeny.                                   | Lawlor (1971)            |
| P. interparietalis X P. eremicus                          | Five caged pairs. 17 progeny. $F_1$ fertile.                   | Brand and Ryckman (1969) |
| P. interparietalis X P. eremicus                          | Two caged pairs. No progeny.                                   | Lawlor (1971)            |
| P. eremicus X P. guardia                                  | Five caged pairs. No progeny.                                  | Brand and Ryckman (1969) |
| P. guardia X P. eremicus                                  | Five caged pairs. No progeny.                                  | Brand and Ryckman (1969) |
| P. eremicus X P. collatus                                 | Four caged pairs. No progeny.                                  | Lawior (1971)            |
| P. collatus X P. eremicus                                 | Three caged pairs. No progeny                                  | Lawlor (1971)            |
| P. interparietalis X P. guardia                           | Five caged pairs. No progeny.                                  | Brand and Ryckman (1969) |
| P. guardia X P. interparietalis                           | Five caged pairs. No progeny.<br>One mass mating. Two litters. | Brand and Ryckman (1969) |
| P. guardia X P. interparietalis                           | Caged pair. No progeny.                                        | Lawlor (1971)            |
| P. interparietalis X P. collatus                          | Caged pair. Progeny viable.                                    | Lawlor (1971)            |
| P. collatus X P. interparietalis                          | Caged pair. No progeny.                                        | Lawlor (1971)            |

| CROSS (FEMALE X MALE)                                             | COMMENTS                                                                                                       | REFERENCES                                               |
|-------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------|----------------------------------------------------------|
| INTERSPECIFIC CROSS ATTEMPTS WITHIN THE P. TRUE! SPECIES GROUP    | IN THE P. TRUE! SPECIES GROUP                                                                                  |                                                          |
| P. truei <sup>1</sup> X P. (nasutus) difficilis                   | Caged pairs. 3/17 fertile.<br>F <sub>1</sub> females fertile, males sterile.                                   | Dice and Liebe (1937)                                    |
| $P$ . truei $^1 	imes P$ . (nasutus) difficilis                   | Two pairs in social choice cages. No hybrids detected/67 progeny.                                              | Tamsitt (1961)                                           |
| P. truei <sup>1</sup> X P. (nasutus) difficilis                   | F, females fertile, males sterile, with impaired. spermatogenesis. $90\%$ reduction in F <sub>1</sub> progeny. | Moree (1946, 1948)                                       |
| INTERSPECIFIC CROSS ATTEMPTS WITHIN THE P. BOYLII SPECIES GROUP   | IN THE P. BOYLII SPECIES GROUP                                                                                 |                                                          |
| P. boylii X P. stephani                                           | Two caged pairs. No progeny.                                                                                   | Lawlor (1971)                                            |
| P. stephani X P. boylii                                           | One caged pair. No progeny.                                                                                    | Lawlor (1971)                                            |
| INTERSPECIFIC CROSS ATTEMPTS WITHIN THE P. LEUCOPUS SPECIES GROUP | IN THE P. LEUCOPUS SPECIES GROUP                                                                               |                                                          |
| P. leucopus <sup>1</sup> X P. gossypinus                          | 70 $F_1$ hybrids produced. $F_2$ produced.                                                                     | Dice (1937)                                              |
| P. leucopus <sup>1</sup> X P. gossypinus                          | Three F <sub>1</sub> hybrids produced.                                                                         | Foreman (1966)                                           |
| P. leucopus <sup>1</sup> X P. gossypinus                          | Thirty-nine mating choice cages.                                                                               | Bradshaw (1968)<br>No hybrids detected among 25 progeny. |

| CROSS (FEMALE X MALE)                       | COMMENTS                                                                                                                                                                               | REFERENCES                               |
|---------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------|
| INTERSPECIFIC CROSS ATTEMPTS WITHI          | INTERSPECIFIC CROSS ATTEMPTS WITHIN THE P. MANICULATUS SPECIES GROUP                                                                                                                   |                                          |
| P. maniculatus <sup>1</sup> X P. polionotus | Viable, fertile F, from 1 of "a number" of caged pairs.                                                                                                                                | Sumner (1930, 1932)                      |
| P. maniculatus <sup>1</sup> X P. polionotus | Progeny from one of five caged pairs.                                                                                                                                                  | Dice (1933)                              |
| P. maniculatus X. P. polionotus             | Progeny from seven of 23 caged pairs. $\mathbf{F_1}$ fertile.                                                                                                                          | Watson (1942)                            |
| P. maniculatus X P. polionotus              | Viable progeny from 20 of an unknown number Some $\boldsymbol{F}_1$ males sterile.                                                                                                     | Blair and Howard (1944),<br>Blair (1944) |
| P. maniculatus X P. polionotus              | Viable progeny from 10 of 31 caged pairs.                                                                                                                                              | Liu (1953a)                              |
| P. maniculatus X P. polionotus              | Viable progeny from 20 of 44 caged pairs. Some F, pre-weaning mortality. F, average smaller than either species. Successful backcrosses to either species and F <sub>2</sub> produced. | Dawson (1965, 1966, 1971)                |
| P. maniculatus X P. polionotus              | Viable, fertile progeny from 1 of 20 caged pairs homozygous for the brown mutation.                                                                                                    | Dawson et al. (1969)                     |
| P. maniculatus X P. polionotus              | Viable F, progeny from 17 of 30 caged pairs. Near-term fetuses and placentas collected from six matings. F, fetal and placental weights less than in either species. Placenta compact. | Rogers and Dawson (1970)                 |
| P. maniculatus X P. polionotus              | Karyotype of $\mathbf{F_1}$ female given.                                                                                                                                              | Te and Dawson (1971)                     |
| P. maniculatus X P. polionotus              | Nine F <sub>1</sub> hybrids produced in test of transferrin inheritance.                                                                                                               | Griswold and Dawson (1971)               |

|                                            |                                                                                                                       | 2 220                                       |
|--------------------------------------------|-----------------------------------------------------------------------------------------------------------------------|---------------------------------------------|
| CROSS (FEMALE X MALE)                      | COMMENTS                                                                                                              | REFERENCES                                  |
| INTERSPECIFIC CROSS ATTEMPTS WITH          | INTERSPECIFIC CROSS ATTEMPTS WITHIN THE P. MANICULATUS SPECIES GROUP (Continued):                                     | ontinued):                                  |
| P. maniculatus X. P. polionotus            | Artificial insemination. 10 of 12 fertile.                                                                            | Maddock and Dawson (1974)                   |
| P. maniculatus X P. polionotus             | 41 F, hybrids produced in test of alcohol dehydogenase inheritance.                                                   | Felder (1975), Burnett and<br>Felder (1978) |
| P. maniculatus X P. polionotus             | in vitro fertilization. 89 of 115 ova fertilized, 79 normal, 8 abnormal.                                              | Fukuda et al. (1979)                        |
| P. maniculatus X P. polionotus             | Seven of 24 caged pairs pregnant. 37 of 38 embryos survived to term.                                                  | Maddock and Chang (1979)                    |
| P. maniculatus X P. polionotus             | 87 F, neonates and 47 fetuses measured in test of transspecific immunity.                                             | Dawson et al. (1982)                        |
| P. maniculatus X P. polionotus             | $\mathbf{F}_1$ hybrids and progeny from backcrosses to $P^1$ maniculatus produced in genetic linkage tests.           | Dawson (1982), Dawson et al. (1983)         |
| P. maniculatus X P. polionotus             | 15 pregnant females assayed for progesterone.                                                                         | Bell and Dawson (1983)                      |
| P. maniculatus X P. polionotus             | 10 F <sub>1</sub> hybrids used in test of burrowing behavior. F <sub>1</sub> ahimals burrow same as $P$ , polionotus. | Dawson et al. (1988)                        |
| P. maniculatus X P. polionotus             | $50\mathrm{F}_1$ weaned from 3 of 5 matings attempted.                                                                | Dawson et al. (1993)                        |
| P. maniculatus X P. polionotus             | Viable F <sub>1</sub> backcrossed in linkage test.                                                                    | Cowling et al. (1994)                       |
| P. maniculatus X P. melanotis              | Viable progeny from one of two caged pairs. Male $\mathbf{F}_1$ fertile in backcross to $P$ . maniculatus.            | Clark (1966)                                |
| P. maniculatus <sup>1</sup> X P. melanotis | No progeny from 137 pairs.                                                                                            | Bowers et al. (1973), Bowers (1974)         |

| CROSS (FEMALE X MALE)                              | COMMENTS                                                                                                                                                    | REFERENCES                                        |
|----------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------|
| INTERSPECIFIC CROSS ATTEMPTS WITHIN                | INTERSPECIFIC CROSS ATTEMPTS WITHIN THE P. MANICULATUS SPECIES GROUP (Continued):                                                                           | ntinued):                                         |
| P. maniculatus $^1$ X P. oreas = keeni             | No progeny from 12 pairs.                                                                                                                                   | Dice (1949)                                       |
| P. maniculatus X P. oreas = keeni                  | No progeny from ten caged pairs.                                                                                                                            | Liu (1954)                                        |
| P. maniculatus <sup>1</sup> X P. oreas = keeni     | No results.                                                                                                                                                 | Sheppe (1961)                                     |
| P. maniculatus <sup>1</sup> X P. sitkensis = keeni | No laboratory progeny.                                                                                                                                      | Thomas (1972)                                     |
| P. polionous X P. maniculatus                      | Progeny from 1 of 23 caged pairs. $\mathbf{F}_1$ large sized. Died soon after birth.                                                                        | Watson (1942)                                     |
| P. polionotus X P. maniculatus                     | Viable F, from 22 of an unknown number of caged pairs. Some F <sub>1</sub> fertile in backcrosses.                                                          | Blair and Howard (1944),<br>Blair (1944)          |
| P. polionotus X P. maniculatus                     | $F_1$ progeny from 10 of 64 caged pairs. Placentas large, prenatal mortality of $F_1$ common.                                                               | Liu (1953a, 1953b)                                |
| P. polionotus X P. maniculatus                     | Ten of 99 caged pairs produced liveborn. F average larger than either species birth to adult. Maternal and F fetal mortality high. F fertile in both sexes. | Dawson (1965, 1966, 1971)<br>Dawson et al. (1982) |
| P. polionotus X P. maniculatus                     | Live born progeny four of 35 pairs. 16 near term conceptuses collected from six matings. $F_1$ placentas large (6X reciprocal) and vesiculate.              | Rogers and Dawson (1970)                          |
| P. polionotus X P. maniculatus                     | Eight of 12 caged pairs pregnant. 14 living near-term embryos collected. Fetal and placental weights average more than either species.                      | Maddock and Chang (1979)                          |

<sup>1</sup>Female in cross not given.

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# Chimeric (Allophenic) and Transgenic Peromyscus

There have been very few attempts to produce allophenic chimeras in *Peromyscus*. Only one report has been published to date. Klein and Markert (1981) produced intraspecific embryonic aggregate chimeras of wild-type *P. maniculatus bairdi* and homozygous blonde (bl/bl) mutant deer mice. They reported one male obvious coat color chimera delivered by caesarian section which survived to maturity. It eventually sired 34 progeny in nine litters, all of the blonde phenotype when mated to a blonde female. Another male was produced by caesarian section and survived to maturity. It showed chimerism of the coat only in a patch of blonde around one eye. This animal produced 11 progeny in 3 litters, all wild-type. Chimerism was also demonstrated in the retina of the eye. A third chimaeric fetus was recovered from a dead host female.

No transgenic *Peromyscus* have been reported. There is one report of *Peromyscus* trangenes inserted into *Mus*. Pine *et al.* (1988) microinjected four *P. leucopus mys* element (retroposon) clones into *M. domesticus* male pronuclei. Three transgenic mice were recorded with Mys transcripts produced. The consensus target sequence recognized by the *mys* element 3' was deduced to be > ATCC......TT/GAAGTT.

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# ----- PEROMYSCUS MOLECULAR GENETICS: Nucleic Acid Sequences Published:

A. INDIVIDUAL COPY NUCLEAR GENES.

Adh-1, Adh-2. Alcohol dehydrogenase-1 and 2. (Zheng et al. 1993)

ADH-1. Essentially full length sequence of nucleotides 1-1304 obtained from two overlapping cDNA clones. 374 amino acid sequence startsite begins at base 58 and with a stop codon at 1247. Deduced amino acid sequence exhibits 94% identity with *Mus Adh-1*. The *Adh-1* negative allele is due to deletion of most or all of the *Adh-1* sequence.

ADH-2. Full length sequence of nucleotide 1 - 1898 obtained from cDNA clones. 374 amino acid sequence startsite begins at base 136 and terminates at base 1261. *Peromyscus Adh-2* amino acid sequence exihibits 67% identity with human ADH-6 and 51-58% with other human ADHs.

Hbb. Beta globin complex. Partial sequences of structural adult beta globin genes in *P. maniculatus* (Padgett *et al.*, 1987):

Features: Twelve lambda clones represent a total of 80 kb in three sections with gaps of undetermined length. Clones isolated using three *Mus Hbb* probes. Sequences given for three regions ([a]110, [b]110 and [c]219 bp, respectively) from each *Hbb-b1* and *Hbb-b2*, and for two regions ([a]110 and [c]219 bp) from *Hbb-b3* adult beta globin genes. The second of the three beta globin coding blocks is located, except for the initial two codons, in the third sequenced region for each of the three genes. No termination codons are present in the coding sequences. *Hbb-b1* and *b2* have identical coding sequences and match for all but two non-coding bases in regions sequenced. *Hbb-b3* varies from *b1* and *b2* at ten sites in the third region, which contains the second coding block, and at numerous sites in the non-coding first region. Region two was not sequenced for *Hbb-b3*. Homologies with *Mus* and other mammals are discussed together with molecular evolution of the beta globin gene.

Mhc. Major histocompatibility complex. Exon 5 sequences of three distinct MHC Class I subtypes representing genes Pele-A, B and C of P. leucopus.

Features: Sequences reported for 121, 115 and 114 bases of three clones (52a, 40b and 53) representing the three MHC Class I genes. Homology with equivalent human, mouse and hamster genes indicated. (Crew et al., 1990)

Approximate 110 base sequences reported for 10 MHC Class I *Pele-A* exon 5 (transmembrane-coding) genes. Hypervariability from consensus base 48 to 82, with deletion and repeats within that region differing among genes. (Crew *et al.*, 1991)

RFLP characterization of MHC Class II polymorphism reported. (Crew et al., 1989)

rRNA (Ribosomal RNA gene complex) of *P. boylii* and *P. eremicus*. Also *Onychomys* species *O. leucogaster*, *O. torridus* and *O. arenocola*. [Restriction maps]

Features: Includes 18S, 5.8S and 28S coding regions and two internal transcribed spacers and the 5' external transcribed spacer. Homology among rodent species shown. 28S gene restriction mapped also shown for *P. (Megadontomys) thomasi.* (Allard and Honeycutt, 1991)

Tnf. Tumor necrosis factor/cachectin and Lt (lymphotoxin). Occurs within the major histocompatibility complex (Mhc) of P. leucopus.

Features: 2304 base sequence, from 400 bases upstream to 160 beyond stop codon, includes complete *Tnf* sequence with putative TATA box and four exons. Homology with rabbit, human, mouse and hamster shown. (Crew *et al.* 1992)

# Molecular Genetics of Peromyscus Nucleic Acid Sequences Reported (Continued)

# B. REPEAT ELEMENTS.

L1 (=LINE-1) long interspersed repeat family in P. maniculatus:

Features: Sequences of two clones (pDK55 and pDK62) from *P. maniculatus* of 1.5 and 1.8 kb, respectively, representing diverged DNA families. Homology between *Peromyscus* DNA families on same order as either is to L-1 elements representing *Rattus*, *Mus* or human. (Kass *et al.*, 1992)

Homology with *Mus* and other mammalian *L1* elements shown by Southern blotting and sequence matching. Concerted restriction site variation among seven *Peromyscus* species frequently corresponds to species-group taxonomic level (Kass *et al.*, 1992).

Mys-1 element in P. leucopus (Wichman et al., 1985; Pine et al., 1988):

Features: 2843 bp. 343 bp terminal repeats (1-343) and (2501-2843). Open reading frame [1] 489 bp (595-1083) and ORF [2] 642 bp (1552-2193) with a single interrupt codon at 1795. ORF [1] translated reveals homologies with other known reverse transcriptase proteins. 20 bp pyrimidine tract (344-364); internal direct repeats 1243-1280, 1281-1318; T A sequences beginning at 1516 and at 2240. Lys tRNA binding site at 2487-2498. Mys elements 2 - 8 share common restriction sites. Mys probe hybridizes with P. gossypinus and other cricetid, but not murid, genomic digests. Mys elements probably occur in 500 to 1000 copies per haploid genome in both P. leucopus and P. gossypinus.

Mys elements hybridize to all chromosomes of four species of Peromyscus leucopus and P. maniculatus complex tested by in situ hybridization, but excluded from C-bands, and preferentially hybridizing to G-bands. Also preferential hybridization to X and Y chromosomes. (Baker and Wichman (1990)

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# NONRADIOACTIVE DNA FINGERPRINTING OF THE WHITE-FOOTED DEER MOUSE PEROMYSCUS MANICULATUS

The potential utility of DNA fingerprinting with the SNAP probe (manufactured by Syngene, a Molecular Biosystems, Inc. company) for distinguishing degrees of relatedness within populations of *Peromyscus maniculatus* was tested. This probe is the Jeffreys 33.6 human myoglobin minisatellite sequence as a 22mer oligodeoxyribonucleotide, with alkaline phosphatase covalently attached to the central thymidine nucleotide (Edman et al., 1988). I believe Syngene is no longer in business, but similar probes can be obtained from a number of other suppliers. Using the SNAP probe Cummings and Hallett (1991) detected polymorphic fragments in *HaellI*-digested DNAs from *Peromyscus maniculatus* deer mice and *Perognathus parvus* pocket mice, each individual caught at a different widely-spaced location in Washington State. Mice with familial relationships were not analyzed in their study.

We analyzed the DNAs of *P. maniculatus* individuals from one limited area of the Univ. of Nebraska field station near Mead, Nebraska as well as the DNAs of *P. maniculatus* individuals bred in the laboratory. Using the restriction endonuclease *Hinf1* and the SNAP probe, at least 15 bands could be detected in each sample. Band sharing between DNAs from sibling or parent/offspring pairs was 53±13%, similar in both mean and standard deviation to that observed by Cummings and Hallett between DNAs from sibling or mother/offspring pairs of montane voles, *Microtus montanus*. Band sharing between DNAs from *P. maniculatus* individuals without known relationships in our study, however, were higher than we expected and averaged 40±16%, which was more similar to that reported by Cummings and Hallett for *Perognathus parvus* (38±7%) than for *Peromyscus maniculatus* (19±5%).

Our preliminary results therefore suggest that a different probe, one that detected a more variable DNA sequence, might be required for measuring relatedness among individuals from a single *Peromyscus maniculatus* population. We would appreciate hearing from others that may have had success in distinguishing different degrees of relatedness in single *Peromyscus maniculatus* populations using either a multilocus hybridization probe other than the Jeffreys 33.6 minisatellite, or a single-locus probe, or PCR primers. We would like to use nuclear DNA, but would be willing to use mitochondrial DNA if that works better. Thanks!

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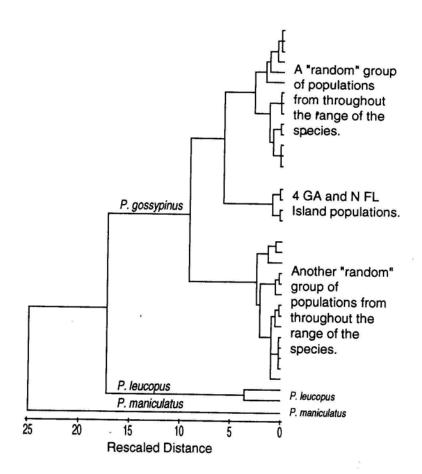
# Studies of the Cotton Mouse (Peromyscus gossypinus), II

My revisionary studies concerning the subspecific taxonomy of the cotton mouse (*Peromyscus gossypinus*) continue, and I am very interested in discussing these finding and ideas with other interested researchers. In a paper recently submitted for review to the Journal of Mammalogy, I have proposed synonymizing the Federally endangered Key Largo cotton mouse, presently referred to *P. g. allapaticola*, with the more widespread mainland subspecies, *P. g. palmarius*. In this study, I examined cotton mice, white-footed mice (*P. leucopus*), and deer mouse (*P. maniculatus*) to assess the taxonomic validity and affinities of the mice on Key Largo, Florida, presently referred to *P. g. allapaticola*. I performed genetic analysis on 40 loci in 658 mice from 35 populations (6 Island, 29 mainland) and morphometric analysis on 18 characters on 1167 mice from 36 populations. Every population was genetically and/or morphologically different from every other population for at least one character. However, when I examined all populations simultaneously, the differences between the Key Largo and other populations were not significant. While each population was statistically distinct, none was unusually distinct. Therefore, the Key Largo population does not warrant recognition as a separate subspecies and should be referred to the widespread mainland subspecies, *P. g. palmarius*.

These results are quite similar to those I found for the mouse population on Cumberland Island, Georgia, formerly referred to *P. g. anastasae* (Boone et al., 1993). As above, this island population appeared to be morphologically distinct, but in fact was not any more distinct than would be expected among randomly chosen populations. This is a species where we must be particularly careful to ensure that subspecies of *P. gossypinus* are not simply monuments to "the persistence and patience of the systematist" (Lidicker, 1962:168).

Since Boone et al. (1993) was submitted for publication, I have greatly expanded the data sets (allozymes and morphology), and I am tempted to revise my earlier position concerning all the Georgia and north Florida islands (Amelia Island, FL north to Skidaway Island, GA). Populations on these islands do appear as a distinct cluster in both genetic and morphometric spaces (Fig. 1). However, I find it difficult to conclude that they should each be a different subspecies.

We have also been investigating a possible case of reticulate evolution on Cumberland Island, Georgia where it appears that a subspecies level of distinction may have been achieved by hybridization between *P. gossypinus* and *P. leucopus*, and by "hybridization" between *P. g. gossypinus* and *P. g. megacephalus*. The inadvertent movement of animals as a result of trade between island residents and people around Baton Rouge, Louisiana, and trade with "Northerners" can account for the distribution of alleles at two loci (Transferrin and GPI). Of course, common ancestry can also account for these observations. We have found a few hybrid *P. gossypinus* X *P. leucopus* individuals from areas where these species are sympatric, so hybridization on Cumberland Island is a possibility.



# Figure Legend

Figure 1. Phenogram of three *Peromyscus* species produced from UPGMA cluster analysis (17 morphometric characters, correlation matrix extraction, varimax rotation) of population centroids.

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# UNUSUAL LENTICULAR HYDRATION IN CATARACT-WEBBED DEER MICE

Cataract-webbed (cw/cw) is an interesting autosomal recessive trait in Peromyscus that results in cataracts and syndactyly (see PN#13, p 15). The webbed-toe phenotype is identifiable before weaning and is 98% penetrant, whereas cataract development occurs over a wide age range beginning as early as 3 months, but more typically developing between 6 and 12 months with about 88% penetrance. With its later onset the cataract phenotype of cw/cw mice might provide an analogy to senile cataract in humans.

In other murine cataract models, such as Philly and Nakano, the development of lens opacity is often associated with perturbed ion and water metabolism (Kador et al., 1971; Kinoshita, 1980). Lenses in new born laboratory mice contain about 65% water and during the next few weeks the lenses experience a marked dehydration to values approaching 50%. This lower value is normally maintained throughout the life of the individual. Mutants that develop cataract within the first months, however, have lenses whose water content returns to 65%. In one model, the Nakano cataract, a faulty Na-K-ATPase has been hypothesized to be directly responsible for the development of lens opacity.

To determine whether similar phenomena may be evident in cataract-webbed mice, the effect of cw and cataract development on lenticular water content was examined. Two experimental and one control group were used. One experimental group contained animals with syndactyly and clearly observable cataract (cw/cw-cat), the other group consisted of animals that had webbed toes but had not yet developed cataract (cw/cw-clear). The control group (Cw/cw) consisted of F1 mice resulting from a cross between homozygous (cw/cw) and wild-type (Cw/cw) mice. The ages of the mice in each of these groups ranged between 5 and 13 months.

The results we have obtained thus far suggest that cw may well influence lenticular water content and in ways that differ from other cataract models. Lenses from the control group gave water values in the expected 50-55% range. On the other hand most of the (cw/cw) clear lenses were even more dehydrated with the majority falling in the 37-50% range. The mutant lenses with frank cataract showed the greatest range of values from severely dehydrated (35-40%), normal (50-55%), and overhydrated 55-65%. Thus, many of the clear cw/cw lenses are physiologically distinguishable from wild-type lenses in a way suggestive that altered water metabolism may be a contributing factor to the later development of frank cataract. Since the degree of cell hydration is a function of ion metabolism it would be most worthwhile to study the effect of cw on lenticular ion content and transport.

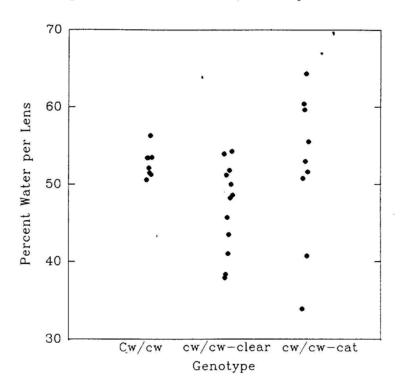


Figure 1: The effect of cw/cw on percent water

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# Pleiotropic effects of the agouti coat-color Peromyscus

Pleiotropic effects of the agouti coat-color locus on the behavior of 75 deer mice (*Peromyscus maniculatus*) homozygous for either the agouti or the nonagouti allele were assessed using standardized behavioral tests (handling, open field, and platform jump) originally developed for use with rats. Nonagouti deer mice and rats are less aggressive in handling tests. They exhibit fewer attacks and permit more physical contact. In open-field and platform-jump tests they are less active overall, but groom more than animals with the agouti allele. In conjunction with previously described regional differences in the distribution of neural catecholamines between allelomorphs, these data suggest that at least one major coat-color locus exhibits consistent behavioral patterns across species. Thus pleiotrophic effects of structural genetic loci may influence complex behaviors.

A second investigation of the pleiotropic effects of the agouti coat-color locus examined tyrosine hydroxylase immunoreactivity in the two allelomorphs. Scattered tyrosine hydroxylase cells were observed around the suprachiasmatic and supraoptic nuclei of both color morphs. Immunoreactivity was not uniform across the hypothalamus but was partitioned into dorsal and ventral regions. The dorsal group included cells in the zona incerta and paraventricular nucleus as well as cells near the dorsal caudal fornix, whereas the ventral group included cells in the arcuate nucleus as well as cells near the suprachiasmatic and supraoptic nuclei. The numbers of immunopositive cells were similar between the color morphs in the median preoptic area, paraventricular area, zona incerta, substantia nigra, ventral tegmental area, parabrachialis pigmentosa, retrorubral field, dorsal raphe, and locus coeruleus, but significantly different in the arcuate nucleus. Nonagouti deer mice have more tyrosine hydroxylase immunoreactive cells in the arcuate nucleus, denser immunoreactive fibers in the ventral hypothalamus, and also have much larger ventricles than agouti animals. I believe, these are the first neuroanatomical differences recorded for color morphs of *Peromyscus* and they suggest that the agouti coat-color locus may have pleiotropic effects on the neurobiology of deer mice. The conservative nature of the agouti locus across mammals suggests that such pleiotropic effects should be present in other species.

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A colleague and I are preparing a report on fleas found in nests of *Peromyscus maniculatus* collected mostly in Arizona and New Mexico. Since 1969 I have been handling live and dead deer mice and other species of *Peromyscus* without gloves or mask. I must have examined well over 1,000 mice and never became infected with any rodent disease.

Field workers who took part in a survey in north-central New Mexico in 1969-1971 were vaccinated against plague as an epizootic of sylvatic plague was occurring in the study area then (Haas et al. 1973). The collateral epidemic of bubonic plague resulted in at least 13 human cases in the state.

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\* \* \*

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The nature of our research involves an evolutionary analysis of DNA repetitive sequences. This work includes the ID (identifier) sequences, characterized as short interspersed elements (SINEs) that are found throughout the rodent genome as a result of retroposition events. ID copy numbers vary by orders of magnitude between guinea pig (~100-200), hamster (~1000-2000), Mus (~10,000-20,000), and rat (~130,000). We now show a more sporadic array of copy number within the order Rodentia, utilizing additional species, including *Peromyscus*, which approximates the number in Mus.

The BC1 RNA gene, regarded as the source gene for ID copies, is transcribed primarily in the brain, but observed in other tissues, such as the spleen and the testes of *Peromyscus*. We observed a slight size variation between the transcripts of *P. californicus* and *P. maniculatus*. By cloning the BC1 gene in these species, we observed a concordance between DNA sequence and transcript size. Additionally, we have observed variants in the highly conserved ID core sequence of the BC1 gene which is maintained in species of the *Peromyscus* subgenus. This has provided us with an excellent opportunity to investigate the evolutionary history of this repeat within the genus which we are currently pursuing. We are also sequencing and analyzing the BC1 genes from several rodent species in an attempt to resolve the copy number enigma.

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# KSU RESEARCHERS FIND HANTAVIRUS IN SOME KANSAS DEER MICE

A team of researchers at Kansas State University has been working with the Centers for Disease Control and Prevention in Atlanta to test for the presence of hantavirus in natural populations of deer mice in Kansas. The deer mouse, *Peromyscus maniculatus*, is the animal suspected of transmitting hantavirus, which causes hantavirus pulmonary syndrome in humans.

K-State researchers trapped deer mice in two areas of Kansas where no cases of hantavirus pulmonary syndrome have been reported.

The first locality trapped was Konza Prairie Research Natural Area, south of Manhattan. None of more than 50 deer mice trapped had antibodies to hantavirus in their blood.

The second region of study was in north central Kansas in the area of <u>Lucas</u>, in Russell, Lincoln and Osborne counties.

Deer mice were trapped in the habitat in which they are normally found - in grazed pastures, fencerows and agricultural fields. Deer mice are not found commonly in urban centers or around farm houses and buildings. In contrast to research findings from Konza Prairie, 17 of 201 deer mice showed the presence of hantavirus.

Researchers at K-State and CDC were not surprised by the presence of hantavirus in north central Kansas due to the distribution of known cases of hantavirus pulmonary syndrome in the United States. For example, cases of this disease have been diagnosed from California to Florida and New Mexico to North Dakota. The hantavirus likely will be found in rodents in the Manhattan area and statewide as more research is done.

The presence of hantavirus should not cause undue alarm. In contrast to initial reports, the hantavirus is not a new virus. Recently, CDC found that a man had died of hantavirus pulmonary syndrome in San Francisco in 1980. This was found by analyzing preserved tissue samples from the victim.

Other information that comes from the virus itself suggests that hantavirus has occurred in deer mice for hundreds to thousands of years. CDC has discovered that several genetic strains occur in different regions of the United States. For example, the strain found in North Dakota, Montana and Idaho is different from the strain that occurs in New Mexico, Nevada and California.

What is new about the hantavirus is that its presence is known in the United Stated and that the CDC has developed tests to detect it. Other hantaviruses are also known from Europe and Asia.

Knowing the hantavirus occurs in Kansas should not cause extreme concern; however, the public should not put themselves at risk.

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## Spatial Memory in Peromyscus

The purpose of the present exploratory study was to test the spatial memory abilities of deermice (Peromyscus) in a radial maze. The radial maze has a number of arms radiating from a central choice area (like spokes around a wheel hub). The maze was constructed of clear plexiglass with 5 equally spaced arms (24 cm long by 5.25 cm wide and 15 cm high) radiating from an octagonal starting platform (perimeter 5 x 8.5 cm). Each arm contained a 4 cm high clear plexiglass barrier installed 8 cm from the central arena to slow the running speeds of the deermice. A food cup (thimble), 1.5 cm deep and 2 cm in diameter, was located at the distal end of each arm. An octagonally shaped clear plexiglass cylinder (20 cm high) was used to restrain the mice to the central platform at the start of each trial. The maze was placed on a table top in a small room where several extramaze stimuli, such as a wall poster and electromechanical programming equipment, were available. Deermice were given one trial per day for 26 days after meeting a criterion of consuming all 5 pieces of food on 2 consecutive days. On each trial all the paths were baited with a small portion of food (approximately half a Cocoa Pebble) and deermice were allowed to run the maze until they consumed the five pieces of food, or until 5 min, had elapsed. The sequence of arms chosen and time to complete each trial were recorded. Entering an arm was defined as crossing over the 4 cm barrier. Once a food pellet was consumed that arm of the maze remains without food for the balance of the trial. Animals learn to enter only those arms of the maze from which food has not yet been obtained on that trial. Numerous studies have been conducted to rule out the use of food smells or other odor cues in the selection of the appropriate maze arms. Between trials, animals were provided with access to Purina Lab chow and water ad lib. The animal were food-deprived for six hours prior to each training session.

Ten comparatively young (approximately 1 year old) male deermice were used in this study (5 wild-type, +/+ and 5 boggler, bg/bg). Wild-type designates that these animals were demonstrated to be behaviorally normal. Boggler deermice were specifically chosen because they have been demonstrated to show an increasing ataxia with age, and an increasing axonal dystrophy with advancing age. It would be interesting to determine if any indication of memory impaired also occurred in these animals.

#### RESULTS AND DISCUSSION

The results demonstrate that both wild type and boggler *Peromyscus* rapidly acquire the radial arm maze task, as do other rodents. Figure 1 shows the mean number of correct choices out of the first five choices made, averaged over two-trial blocks. As early as the third block of training, the mice were choosing an average of more than four different arms within the first five choices. This value reliably exceeded that expected by chance alone. As with other rodents, the sequence of arms chosen appeared random. Moreover, the failure of probe tests (in which the maze was randomly rotated between trials, to disrupt performance) suggests that the deermice relied on extramaze cues to solve the maze. Examination of choice protocols for each individual animal revealed that none of the deermice solved the radial maze problem by adopting a kinesthetic strategy involving response chains or algorithms, that is they did not go sequentially from one arm to the next in clockwise or counterclockwise direction.

It is interesting to note that the performance of young boggler deermice is indistinguishable from that of young wild-type animals. This suggests that the ability of young boggler deermice to remember for short periods of time where they have been last, based on extramaze clues is not affected.

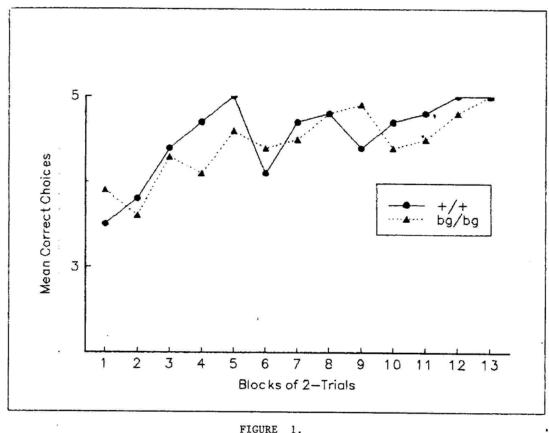


FIGURE 1.

Olton and Samuelson (1976) found that also rats possess a remarkable spatial memory capacity, remembering the location of food in a radial arm maze. Available evidence indicated that rats perform efficiently without using response chains or entering maze arms in a fixed order from one trial to the next. Several recent investigations (Pico and Davis, 1984; Goldowitz and Koch, 1986) have reported that mice (Mus musculus) also demonstrate a working memory capacity in the radial maze.

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My research interests in *Peromyscus* ecology continue with new projects I have started since arriving at Trinity last year. The first project includes describing the mating and social organization of two coexisting *Peromyscus* species in the mountains of northern New Mexico. I am interested in how microhabitat differences between these species (*P. truei* and *P. boylii*) may or may not influence home ranges and social organization. I am using a combination of radiotelemetry and intensive live-trapping during the summer months. I will also be using molecular techniques, specifically PCR amplification of "microsatellite" loci, to monitor male reproductive success. I would be interested in hearing from anyone who has had any luck with PCR primers for amplification of microsatellite loci. To date I have tested some *Mus* primers with mixed results. At this point I am convinced we need to develop *Peromyscus* specific primers.

Of course as soon as I began my field work this past summer in New Mexico the Hantavirus phenomenon was upon us. We proceeded cautiously with our field work and in the process provided around 40 tissue samples to the CDC for analysis. Fortunately for us all samples tested negative for this virus. I found the CDC most cooperative and helpful and I encourage anyone in a similar situation to contact them.

Besides working during the summer in New Mexico, I am also supervising undergraduates working on small mammal field projects around San Antonio. One of these students is investigating the behavioral interactions between red imported fire ants and rodents, including *P. leucopus*.

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# BILATERAL ADRENALECTOMY ACCELERATES THE ONSET OF SPRING FUR GROWTH IN PEROMYSCUS

A study was conducted to determine the affects of bilateral adrenalectomy on the initiation of spring fur growth in the deer mouse (Peromyscus maniculatus). On January 30, 1993 seven adult male mice were assigned randomly to one of two groups. On February 4, mice in group one (N=4) were sham adrenalectomized under sodium pentobarbital anesthesia and each animal received an empty Silastic implant representing controls. Mice in group two (N=3) were bilaterally adrenalectomized and in addition each mouse received an implant containing 100 mg deoxycorticosterone, as a mineralocorticoid supplement. Starting February 11, dorsal guard hair measurements were taken to the nearest millimeter approximately every other day until March 20. In control mice, hair growth began on February 22 and was complete by March 4, revealing a hair growth cycle of approximately 10 days. In none of the control mice did hair growth completely fill in the dorsal area. Hair growth was observed in all three adrenalectomized mice, on February 18, four days earlier than for control mice. In addition, all adrenalectomized mice exhibited active hair growth over the entire dorsal surface and by March 2, had replaced the entire coat. These findings support the hypothesis that adrenal hormones may play a role in regulating the onset of hair growth cycles, and possibly influence hair replacement patterns as well. (This work will be presented at the 1994 Annual Meeting of The Idaho Academy of Science, Idaho State University, Pocatello, ID).

<u>COMMENT</u>: If you have an interest in the regulation of hair growth cycles of *Peromyscus* or have information regarding the phenomenon, we would appreciate your input. We would like to establish this animal as a model for future hair growth research projects (if the danger of Hantavirus can be controlled). This has been somewhat challenging, because unlike other mammals (such as mink) the literature base is small with regard to its fur growth cycles, especially factors regulating the onset of fur growth. Furthermore, unlike mink that grow a distinct winter or summer pelage, *Peromyscus* appears to exhibit hair growth cycles during all months of the year, with a peak in October. Does it truly exhibit a summer or winter pelage? In addition, based on our findings and those in the literature, *Peromyscus* does not appear to replace its entire pelage following a single hair growth cycle. Any help on this perplexing little critter would be greatly appreciated. Thanks, JR.

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Evaluation of effects of fluorescent powder on Peromyscus maniculatus

Fluorescent powder tracking is a popular technique for tracking nocturnal rodents, and we have been using powder tracking to study the movements and microhabitat use of deer mice in north-central Colorado. Our Animal Care and Use Committee expressed concerns over the possible short-term and chronic pathological effects of fluorescent powder on mice. In collaboration with Drs. Jamie Young and Sue VandeWoude from the Department of Pathology, we undertook a study to determine whether the application of powder has deleterious effects on deer mice.

We applied fluorescent powder to wild-caught mice housed in field enclosures on the Central Plains Experimental Range in June 1993. We sacrificed individuals 3, 6, 16, and 27 days later, and examined the carcasses for the presence and amount of powder. We also performed gross and microscopic examinations of body tissues for evidence of pathological lesions and other adverse reactions to powder. Our examinations were focused on the respiratory, gastrointestinal, and integumentary systems because we expected the greatest potential for irritation in these tissues. We compared our results to those from necropsies of captive untreated mice that had been collected from enclosures at the same time.

As expected, the amount of powder in the tissues decreased over time, and the powder particles appeared to pass harmlessly through the digestive tract and from the skin. The greatest irritation was in the respiratory system, and we found microscopic powder in the lungs of mice as late as day 27. We detected a mild to moderate histiocytic pneumonia associated with powder particles in one mouse from each collection group (27% of treated mice), and the most severe reaction was observed in a mouse from the day 3 group. However, two-thirds of the treated mice had powder in their lungs without any histiocytic reaction, and we found little evidence of inflammation or lesions in other tissues. Further, all treated mice survived the duration of the study, which supports field observations that suggest little direct mortality from tracking. Although we recommend that researchers take steps to minimize exposure of respiratory tissues to powder during application, we concluded that the powder tracking technique has no significant pathological effects that would preclude its continued use.

We hope that our results will demonstrate to other researchers and their institutions that the technique is relatively safe for ecological studies of small rodents, and will reduce the need for other research into this aspect of the powder tracking method. A manuscript describing our results currently in "in press" in the Journal of Mammalogy, with publication expected in 1994.

We also collected blood samples from most of the mice we sacrificed. Blood samples from 20 mice were sent to Dr. Tom Ksaizek at the Center for Disease Control to test for presence of hantavirus antibodies. All twenty mice were negative for hantavirus.

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Abstract (From Wolff, J. O. 1993. Does the "Chitty Effect" occur in *Peromyscus?* J. Mammal. 74:846-851): I tested for the "Chitty Effect: in white-footed mice, *Peromyscus leucopus*, by looking for the presence of a large-bodied, dominant, genetic morph in peak populations. Large-sized animals were present primarily during high but not low densities. However, large-sized males nested with females less often than did small males and they were not dominant to small males. Body mass of offspring was not correlated with that of their mothers, and thus, large size was not a unique genetic morph. Large-sized animals were older than small animals having survived through two breeding seasons under conditions of high food availability. The occurrence of large-sized animals in this fluctuating *Peromyscus* population was similar to that proposed by Chitty (1958) for microtines, but whether it is analogous or not may be conjectural until comparable studies are conducted on *Microtus* species.

Abstract (From Wolff, J. O. 1994. Reproductive success of solitarily and communally nesting white-footed mice. Behav. Ecol. [in press]): To determine the fitness consequences of communal nesting in white-footed mice, *Peromyscus leucopus*, and deermice, *Peromyscus maniculatus*, I compared the reproductive success of field populations of females nesting solitarily, in communal groups of more than one female, in extended families of successive litters, and communal groups with extended families. Mean first litter size of weanlings and juveniles ≥6 wks old did not differ significantly for pups raised under the four nesting situations. Similarly, for pups born into extended families, litter sizes of pups from second litters did not differ significantly from those of first litters or from pups born to solitarily nesting females. Delayed dispersal of juvenile females did not result in resource competition or inhibition of reproduction. Thus, reproductive success of females was not significantly affected by additional members in the nest. At least 26 of 28 communally nesting females were close relatives. Solitary nesting is the common breeding pattern in *Peromyscus* and extended families and communal nesting are alternative reproductive tactics in response to limited space, delayed dispersal, and local grouping among related females.

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