PEROMYSCUS NEWSLETTER

NUMBER FIFTEEN

MARCH 1993

Cover: Non-enhanced *Peromyscus maniculatus* chromosome spread, electronically imaged using Dage-MTI camera system and Sony MultiScan printer.
(Courtesy of Z. Wang)

IN **PN** ISSUE NUMBER FIFTEEN .....

We have contributed entries from Alaska, California, Connecticut, Georgia, Illinois, Massachusetts and Mexico. Please continue to send your informal reports for our "Contributions" section. Each entry should be limited to two single-spaced pages. Entries will be published verbatim, with editing only for format. We limit photos, drawings and tables to one per entry max. Since PEROMYSCUS NEWSLETTER is not a formal publication and reports often contain tentative or preliminary results, entries in the "Contributions" section should not be cited in other publications without prior consent of the contributor.

John J. Christian, Professor *Emeritus*, State University of New York at Binghamton, is our "Peromyscus Pioneer". We want to express our thanks to Stuart Landry of SUNY-Binghamton for preparing an excellent biographical sketch of Dr. Christian. We also want to thank Sandra Michael for her assistance. While Jack Christian is well known for his work with microtine rodents, he is also a major contributor to our knowledge of *Peromyscus* endocrinology and population dynamics. Of more than 100 publications he authored, his papers "Adrenal weight in prairie deer mice (*Peromyscus maniculatus bairdii*) and white-footed mice (*Peromyscus leucopus noveboracensis*)" (1967. *J. Mamm.* 48:598-605), "Social subordination, population density and mammalian evolution" (1970. *Science* 168:84-90) and "Population density and reproductive efficiency" (1971. *Biol. Reprod.* 4:248-294) were particularly significant in contrasting *Peromyscus* with microtine and murine rodents with regard to the role of adrenal corticoids in regulating population density.

It has now been three years since we last updated our lists of presumptive protein loci reported from surveys of natural *Peromyscus* populations. Information from papers by Janecek (*J. Mamm.* 71:301ff), Rogers and Engstrom (*J. Mamm.* 73:55ff), Schnake-Greene *et al.* (Southwest. Nat. 35:54ff), Sugg *et al.* (*J. Mamm.* 71:309), Sullivan and Kilpatrick (*J. Mamm.* 72:681ff), and Sullivan *et al.* (*J. Mamm.* 72:669ff) is incorporated into updated tables on the *P. aztecus, P. boylii, P. mexicanus* and *P. truei* species groups in this issue. Other species groups will be revised in the September 1993 issue.

In the current issue we are also updating "*Peromyscus* Molecular Genetics" - See page 13.

Again, please remember to send your entries and news.....
..... Deadline for *PN* #16 is **15 September 1993.**

PEROMYSCUS NEWSLETTER is produced by the

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University of South Carolina
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with support in part from
National Science Foundation Grant # DIR-9000352.

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NEWS AND COMMENT

We want to thank ROBERT BAKER of Texas Tech University and SUSAN HOFFMAN of Livermore National Lab for providing materials to the *Peromyscus* Molecular Bank. Also thanks to BOB YOUNG of the OB-Gyn Department of the University of South Carolina for allowing us access to his cytogenetics lab and assistance with our efforts at *in situ* gene mapping in deer mice..

****

We are happy to report that the *Peromyscus* Genetic Stock Center finished 1992 with the highest degree of utilization since its beginning in 1985. We have furnished more than 3000 live animals and hundreds of preserved specimens, tissues and other materials to researchers and educators at 69 institutions located in 27 states the District of Columbia and two foreign countries.

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BRUCE BUTTLER has a preliminary version of his Bibliography of *Peromyscus* Reproduction. Those interested in having a copy should contact Bruce at Canadian Union College, College Heights, Alberta, Canada T0C 0Z0.

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We learned recently of the death of **EMMET T. HOOPER in July 1992. Dr. Hooper exercised a major influence on the taxonomy and systematics of** *Peromyscus*. He was the subject of one of our "Peromyscus Pioneer" biographical sketches (See *PN* # 6, Sept. 1988). His presence among peromyscologists will be missed.

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Two other prominent mammalogists, J. KNOX JONES of Texas Tech and MARIE LAWRENCE of the American Museum, also passed away during the year.

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STEVE UPTON and co-workers at Kansas State University report cross transmission of the coccidian *Eimeria arizonensis* **between** *Peromyscus* **and** *Reithrodontomys* (1992. J. Parasit. 78:406ff)), **which may be of interest to some of our readers.**

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ALLERGIES to Peromyscus.... Several cases of severe allergy to *Peromyscus* have come to our attention over the years. Most recently, an animal care supervisor at the Stock Center brought a worker's compensation case involving allergic reactions to deer mice. Individuals who intend to work with these animals, especially in a laboratory environment, should be aware of this potential hazard....

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 (BW Stock)	Closed colony bred in captivity since 1948. Descended from 40 ancestors wild-caught near Ann Arbor MI
P. polionotus subgriseus (PO Stock)	Closed colony since 1952. Derived from 21 ancestors wild-caught in Ocala Nat'l. Forest FL. High inbreeding coefficient.
P. polionotus leucocephalus (LS Stock)	Derived from beachmice wild-caught on Santa Rosa I., FL. and bred by R. Lacy. Third to sixth generation in captivity.
P. leucopus (LL Stock)	Derived from 38 wild ancestors captured between 1982 and 85 near Linville NC. Seventh to ninth generations in captivity.
P. californicus insignis (IS Stock)	Derived from about 60 ancestors collected between 1979 and 87 in Santa Monica Mts. CA. Fifth to ninth generation in captivity.
P. maniculatus X *P. polionotus* F_1 Hybrids	Sometimes available.

MUTATIONS AVAILABLE FROM THE STOCK CENTER

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 colony (Pratt and Robbins, 1982)
Brown *b/b*	Huestis stocks (Huestis and Barto, 1934)
Dominant spotting *S/-*	Wild caught in Illinois (Feldman, 1936)
Gray *g/g*	Natural polymorphism From Dice stocks (Dice, 1933)
Ivory *i/i*	Wild caught in Oregon (Huestis, 1938)
Pink-eyed dilution *p/p*	Sumner's "pallid" deer mice (Sumner, 1917)
Platinum *pt/pt*	Barto stock at U. Mich. (Dodson *et al.*, 1987)
Silver *si/si*	Huestis stock (Huestis and Barto, 1934)
White-belly non-agouti a^W/a^W	Egoscue's "non-agouti" (Egoscue, 1971)
Wide-band agouti $A^{Nb/-}$	Natural polymorphism Univ. Michigan stock (McIntosh, 1954)
Yellow *y/y*	Sumner's original mutant (Sumner, 1917)

Note: Some of the coat color 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.

MUTATIONS AVAILABLE FROM THE STOCK CENTER (continued)

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

*Available only on pink-eye dilution background.

**Available from Behavior Mutant Center

Other Resouces of the *Peromyscus* Genetic Stock Center:

Preserved or frozen specimens of types given above.

Tissues, whole blood or serum of types given above.

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

Reference library of more than 1700 reprints of research articles and reports on *Peromyscus*. Copies can be xeroxed and mailed.

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.

INBRED PEROMYSCUS

The Stock Center has acquired the inbred lines of *P. maniculatus bairdii* developed by Muriel Davisson and others at Jackson Laboratory. These lines are closely related and each is derived from more than twenty sib-mated generations, hence they are "highly" inbred. Two of the lines were separated at the 15th sib-mated generation and represent closely related, but separate lines, and are currently designated PmH1 and PmH8. Lines of H8 were split subsequent to generation I_{20} and, hence, constitute sublines of H8. The Stock Center is in the process of further development of these lines. Small numbers (*c.* 5) of either of the two distinct lines (H1 and H8) are available from the stock center on a limited basis. We suggest that these animals may be useful in molecular or other genetic investigations where a uniform genome is desirable. It is anticipated that greater numbers will be available in the future as production stocks are established. Please contact the Stock Center for more information.

PEROMYSCUS MOLECULAR BANK

Materials are now available through the *Peromyscus* Molecular Bank of the Stock Center. Allow two weeks for delivery.

Purified DNA from fresh and/or frozen tissues of following species:

P. maniculatus	*P. leucopus*	*P. aztecus*	*P. californicus*
P. polionotus	*P. gossypinus*		
P. melanotis			

Genomic DNA libraries:

P. maniculatus (Source: M. Felder)
P. leucopus (Source: M. Crew)

cDNA libraries:

P. maniculatus liver (Source: M. Felder)

DNA Probes:

LINE1 element probes pDK55 and pDK62
(Source: D. Kass from *P. maniculatus* genomic library)

Adh-1 (cADHF72) and Adh-3 (cADHF65) probes
(Source: M. Felder from *P. maniculatus* cDNA library)

Additional materials soon to be acquired. Please call with inquiries.

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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:

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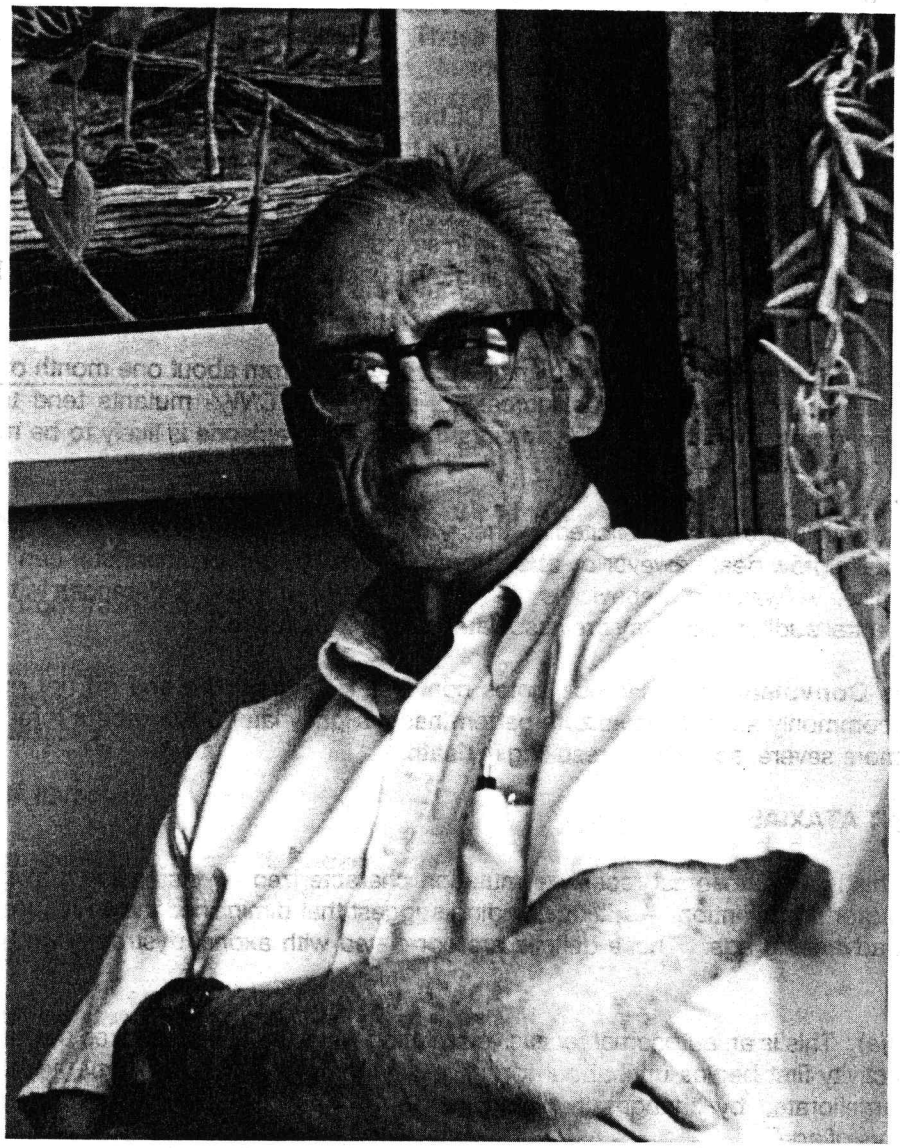

JOHN J. CHRISTIAN

John J. Christian

Although John Christian's work is too far ranging to qualify him strictly as a "Peromyscologist", he deserves inclusion as one of the pioneer workers in the field of endocrinology of small mammals, including *Peromyscus*.

John J. Christian was born on April 12, 1917 at Scranton, Pennsylvania and graduated from Princeton University *cum laude* in 1939. He next entered the College of Physicians and Surgeons of Columbia University where he spent three years, but left before obtaining the M.D., having decided that the actual practice of medicine was not for him. After a stint working as an aircraft engineer, Christian entered the service during World War II, serving as the commander of a PT boat and seeing action in the Pacific. At the end of the war he went to work collecting for Kenneth Doutt on the Pennsylvania Mammal Survey conducted by the Carnegie Museum, finally returning to school first at the University of Pennsylvania, and then at Johns Hopkins School of Hygiene and Public Health, where he received his Ec.D degree in 1954. From 1954 through 1959, he worked as a research associate at the Division of Vertebrate Ecology at Johns Hopkins, going from there to Penrose Research Laboratory at the Philadelphia Zoo, and on to the Albert Einstein Medical Center where he worked from 1962 to 1970. In that year he went to the State University of New York at Binghamton as Professor of Biology where he has been ever since.

This bald and truncated account of Christian's career does not do justice to the breadth and variety of his interests. While he was serving in the Navy, for instance, he took advantage of his situation by recording the seabirds that he encountered off the coast of Halmahera. There were no manuals or field guides at the time (for an area that had not been extensively collected), but Christian used his skills as an artist to record the appearance of the birds so that his identification of them could be confirmed by museum ornithologists later. His wartime notebook is still one of the best sources of information on the distribution of these birds. He remains a bird watcher and photographer to this day as well as a skillful wildlife artist. Indeed, he is an amateur, in the best sense of the word, of oriental art as well as maintaining a lively appreciation of renaissance art and impressionism. He is, moreover, a devotee of the music of the big band, swing era, when he is not listening to the symphonies of Gustav Mahler.

Christian's interest in animal demography was grounded in his participation in that seminal investigation of the population of Norway rats in Baltimore conducted by John Emlen, David Davis and John Calhoun in the late forties and fifties. It was Christian who first saw the possibility of applying the findings of Hans Selye of the effects of stress due to crowding on adrenal function to the problem of small mammal population fluctuations. His paper, "Adrenal and Reproductive responses to population size in mice in freely growing populations" which appeared in *Ecology* (37:258ff) received the Mercer award for outstanding achievement in ecology in 1957. The influence of this paper and those that followed has been far-reaching. Whatever may be the arguments on the cause, or causes, of "cycles", Christian's work has firmly established the reality of the effect of crowding on the reproductive capacity of small mammals both in the laboratory and in the wild.

All the while Christian's laboratory work was in progress he was continually engaged in collecting mammals and recording, for each specimen, a broad array of anatomical measurements including adrenal weights, size and condition of reproductive organs, numbers and sizes of embryos, placental scars (in each horn of the uterus) and of course, weights. There are multiyear series from the Letterkenny Army Base (near Chambersburg, Pennsylvania), from his own 144 acre tract at Starlight (Wayne County), Pennsylvania, and from his family place near Fairhaven, Vermont. These collections form the largest series of mammal specimens documented in this detail of which I have any knowledge. Both the specimens and the notebooks are now housed in the Department of Mammalogy at the American Museum of Natural History. It is interesting to note that Christian found *Peromyscus maniculatus bairdii* to be abundant at Letterkenny, and collected numerous specimens, whereas subsequent investigators have been unable to find it there at all.

An interesting detail of peromyscology discovered by Christian's laboratory is the fact that the typical keratinization of the vagina of rodents occurring at estrus is, in *Peromyscus leucopus*, so excessive that the vagina is packed with extravasated keratinized cells so as to form a "cast" which is extruded from the vagina as a solid plug. Indeed the hyperkeratinosis is so extreme in this form that it is difficult to recognize the classic stages of estrus from smears, since keratinized cells are present most of the time.

Although formally retired, Christian continues to actively pursue research projects. One of his ongoing interests is the pathology of wild animals. He was able to document and publish the pathological effects of the effluvium of the Love Canal on small mammals in that area (a project that elicited no enthusiasm from the New York State Department of Health) and of late he has been investigating the effects on the reproductive physiology of female mice kept in exclusively female colonies except for one male, a condition that results in bizarre social interreactions and ovarian pathology.

John Christian has been my colleague for better than twenty years, and I have the impression that his scientific adversaries read his character the wrong way round. From the manner and order in which his publications have appeared they are apt to conclude that he is a laboratory physiologist-endcrinologist applying his expertise to field situations, whereas the truth is much more nearly the opposite. Jack is at heart a field naturalist in the mold of Ernest Thompson Seton, John Muir, or Joseph Grinnell who has on occasion gone into the laboratory to seek answers to problems of mammalian biology raised by observations in the field. I know of very few bench endocrinologists who have followed the footprints of a *Peromyscus leucopus* in the snow to see if it would climb a tree. (It did.)

Stuart O. Landry
SUNY-Binghamton

I. Sequences reported:

A. INDIVIDUAL COPY NUCLEAR GENES.

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)

B. REPEAT ELEMENTS.

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)

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 (Burton *et al.*, 1986). Concerted restriction site variation among seven *Peromyscus* species frequently corresponds to species-group taxonomic level (Kass *et al.*, 1992).

Sat (Satellite DNA) in *Peromyscus*.

Features: Four cross-hybridizing inserts isolated from *P. leucopus* genomic library clones were characterized as satellite DNA by ladder-like southern patterns. All hybridized *in situ* to centromeric regions of *Peromyscus* chromosomes, but not to chromosomes of non-peromyscine rodents. (Hamilton *et al.*, 1992)

II. DNA Libraries:

A. GENOMIC LIBRARIES.

P. leucopus. Lambda library constructed from *P. l. leucopus* from Georgia. Dr. H. A. Wichman, University of Idaho (Wichman *et al.*, 1985).

P. maniculatus. Constructed from *P. m. sonoriensis* from California, using lambda phage Charon 4A vector. (Dr. M. Edgell and associates, Dept. of Bacteriology and Immunology, Univ. North Carolina, Chapel Hill NC 27514) (Padgett *et al.*, 1987).

P. leucopus. EMBL3 library constructed from *P. l. noveboracensis* descended from Argonne National Laboratory colony, originally collected on the site.

P. leucopus. Cosmid library constructed by R. Baker and associates. (Janecek *et al.*, in press)

B. cDNA LIBRARIES.

P. maniculatus. Two constructed from liver mRNA by M.R. Felder, University of South Carolina (Zheng and Felder, in press).

III. Mitochondrial DNA:

RESTRICTION ENZYME ANALYSIS.

P. polionotus, P. maniculatus and *P. leucopus.* Digest with EcoRI, HindIII, BstEI, BstEII, HaeIII and PstI. 25 combinational types (haplotypes) from 23 populations indicated. (Avise *et al.*, 1979)

P. maniculatus, P. polionotus, and *P. leucopus.* Digest with HincII, BglII, HindIII, BstEII, EcoRI, BamHI, Xba and HpaII. 61 combinational types in *P. maniculatus*, 22 combinational types in *P.polionotus*, and 12 combinational types in *P. leucopus.* (Lansman *et al.*, 1983; Avise *et al.*, 1983)

P. maniculatus. Digest with EcoRI, HindIII, BstII, PstI, BglII, AvaI, AvaII, MboI and HinfI. 26 combinational types from 26 populations from California Channel Islands and southern California mainland. (Ashley and Wills, 1987, 1989)

P. leucopus. Digest with BamHI, BglII, BstEII, EcoRI, HincII, HindIII, HpaII and XbaI. 7 combinational types from six populations, representing two cytotypes and a hybrid zone in Oklahoma. (Nelson *et al.*, 1987)

IV. DNA Fingerprinting

P. californicus. Fingerprints from 23 to 4 kb *Hin*fI digested DNA fragments probed with RNA minisatellite human probe. Eighty-two progeny from 22 complete families scored from wild population in Hastings Natural History Reservation, CA. (Ribble, 1991).

P. maniculatus. Fingerprints from 23 to 2 kb *Hae*III digested DNA fragments probed with SNAP (Molecular Systems) 22 base repeat probe. Five individuals from Whitman County, WA, scored. Unsatisfactory results with *Mus* minisatellite repeat and cytosine-adenine-cytosine 5-repeat probes. (Cummings and Hallett, 1991)

V. Transgenics:

Four *P. leucopus mys* repeat element (retroposon) clones (1, 2, 4, 7) microinjected into *Mus domesticus* male pronuclei. Three transgenic *Mus* recorded. *Mys* transcripts produced. Consensus target sequence recognized by *mys* element 3' deduced: > **ATCC . . . . . T(T/G)AAGTT.** (Pine *et al.*, 1988)

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VARIANT GENETIC LOCI IN NATURAL POPULATIONS OF PEROMYSCUS

Numerous electrophoretic studies of allozymes and other proteins in natural populations of *Peromyscus* have been conducted beginning in the late 1960's. These studies revealed numerous polymorphisms within populations and species, as well as variation among potentially interbreeding species, e.g. *P. maniculatus* and *P. polionotus*. Variants of a protein are generally presumed to identify a genetic "locus", although formal mendelian analysis might not have been accomplished.

PEROMYSCUS NEWSLETTER periodically lists in tabular form the known genetic loci in *Peromyscus* species or species groups. We distinguish between loci which have been formally **demonstrated** and **presumptive** loci. The latter are usually protein variants from natural populations identified by electrophoresis. Separate listings for the two categories are published in PN.

In this issue Tables 1. through 4. summarize presumptive variant loci identified in four species groups: *Peromyscus aztecus*, *P. boylii*, *P. mexicanus*, and *P. truei* species groups. Similar tables in PN #8 and #9 lists variant presumptive loci reported in other *Peromyscus* species and species groups. These tables are updated at three year intervals. Those not in the current issue will be published in the September 1993 issue of PEROMYSCUS NEWSLETTER.

Since limited interbreeding in captivity is frequently possible among different species within a species group, we treat a species group as a single gene pool. Thus while two species may each be monomorphic for alternate alleles, by hybridization heterozygotes can be produced and genetic analysis conducted. Linkage analysis and gene regulation potentially can be investigated using species hybrids. Such systems are currently used in both *Mus* and *Peromyscus*. Thus, the tables serve as a reference to identify reported variants at given loci. Completely monomeric loci, *i.e.* loci for which no variation within the species or species group has been reported, are not listed.

Only variants reported in research publications, abstracts excluded, are listed in the tables. References are listed at the foot of each table.

Table 1. VARIANT PROTEIN LOCI REPORTED FROM NATURAL POPULATIONS OF THE *PEROMYSCUS AZTECUS* SPECIES GROUP

Protein	Locus	Species	References
Amylase	*Amy-1*	*P. aztecus* *P. spicilegus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)
Carbonic anhydrase	*Car-3*	*P. aztecus* *P. spicilegus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)
Esterase	*Es-1* *Es-2* *Es-5*	*P. aztecus* *P. spicilegus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)
Glutamate oxaloacetate transaminase	*Got-1*	*P. aztecus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)
Hemoglobin	*Hba-1*	*P. aztecus*	Sullivan and Kilpatrick (1991)
Isocitrate dehydrogenase	*Idh-1* (*Icd-1*) *Icd-2*	*P. aztecus* *P. spicilegus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)
Lactate dehydrogenase	*Ldh-2*	*P. spicilegus*	Sullivan and Kilpatrick (1991)
Malate dehydrogenase	*Mdh-1*	*P. winkelmanni*	Sullivan and Kilpatrick (1991)
Malic enzyme	*Me-1* (*Mod-1*)	*P. winkelmanni*	Sullivan and Kilpatrick (1991)
Peptidase	*Pep-1* (A) *Pep-4* (D)	*P. aztecus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)
Phosphogluconate dehydrogenase	*Pgd-1*	*P. spicilegus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)

(Continued)

Table 1. Protein variants in *P. aztecus* group natural populations (Continued)

Protein	Locus	Species	References
Phosphoglucomutase	*Pgm-2* *Pgm-3*	*P. aztecus* *P. spicilegus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)
Sorbitol dehydrogenase	*Sdh-1*	*P. aztecus* *P. winkelmanni*	Sullivan ande Kilpatrick (1991)
Transferrin	*Trf*	*P. aztecus* *P. winkelmanni*	Sullivan and Kilpatrick (1991)

Reference

Sullivan, J.M. and C. W. Kilpatrick. 1991. J. Mamm. 72:681-696.

Table 2. VARIANT PROTEIN LOCI REPORTED FROM NATURAL POPULATIONS OF THE *PEROMYSCUS BOYLII* SPECIES GROUP

Protein	Locus	Species	References
Alcohol dehydrogenase	*Adh*	*P. attwateri*	Sugg *et al.* (1990)
Albumin	*Alb*	*P. boylii* *P. pectoralis*	Jensen and Rasmussen (1971) Avise *et al.* (1974) Kilpatrick and Zimmerman (1975) Zimmerman *et al.* (1975) Kilpatrick and Zimmerman (1976a) Kilpatrick (1984) Rennert and Kilpatrick (1986) Werbitsky and Kilpatrick (1987)
Amylase	*Amy-1*	*P. boylii*	Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987)
Carbonic anhydrase	*Car-1* *Car-3*	*P. boylii* *P. beatae*	Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Sullivan and Kilpatrick (1991)
Catalase	*Cas-1*	*P. attwateri*	Sugg *et al.* (1990)
Creatine kinase	*Ck-1*	*P. attwateri*	Schnake-Greene *et al.* (1990)
Esterase	*Es-1* *Es-3* *Es-4* *Es-5* *Es-6* *Es-7*	*P. boylii* *P. attwateri* *P. pectoralis* *P. polius* *P. beatae*	Rasmussen and Jensen (1971) Avise *et al.* (1974) Kilpatrick and Zimmerman (1975) Zimmerman *et al.* (1975) Kilpatrick and Zimmerman (1976a) Kilpatrick (1984) Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Sugg *et al.* (1990) Sullivan and Kilpatrick (1991)
Glucose dehydrogenase	*Gdh-1*	*P. attwateri*	Sullivan *et al.* (1991)

(Continued)

Table 2. Protein variants in *P. boylii* group natural populations (Continued)

Protein	Locus	Species	References
Glutamate dehydrogenase	*Gtdh-1*	*P. attwateri*	Sugg *et al.* (1990)
Glutamate oxaloacetate transaminase	*Got-1*	*P. boylii* *P. pectoralis* *P. attwateri* *P. simulans*	Avise *et al.* (1974) Kilpatrick and Zimmerman (1975) Zimmerman *et al.* (1975) Kilpatrick and Zimmerman (1976a) Kilpatrick (1984) Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Sullivan *et al.* (1991)
α-glycerophosphate dehydrogenase	*Gpd-1* *Gpd-2*	*P. boylii* *P. pectoralis*	Mascarello and Shaw (1973) Avise *et al.* (1974) Janecek (1990)
Glucose-6-phosphate dehydrogenase	*G6pd-1* (*H6pd-1*)	*P. pectoralis* *P. boylii*	Avise *et al.* (1974) Kilpatrick (1984) Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Sullivan *et al.* (1991) Rogers and Engstrom (1992)
Hemoglobin	*Hb-1* *Hb-2*	*P. boylii* *P. pectoralis* *P. attwateri* *P. simulus*	Rasmussen *et al.* (1968) Avise *et al.* (1974) Kilpatrick and Zimmerman (1975) Zimmerman *et al.* (1975) Kilpatrick and Zimmerman (1976a) Kilpatrick and Zimmerman (1976b) Kilpatrick (1984) Sullivan *et al.* (1991)
Hexose-6-Phosphate dehydrogenase	*H6pd-1*	*P. boylii*	Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987)

(Continued)

Table 2. Protein variants in *P. boylii* group natural populations (Continued)

Protein	Locus	Species	References
Isocitrate dehydrogenase	*Idh-1* (*Icd-1*) *Icd-2*	*P. boylii* *P. pectoralis* *P. attwateri* *P. simulus*	Mascarello and Shaw (1973) Avise *et al.* (1974) Kilpatrick and Zimmerman (1976a) Kilpatrick (1984) Remert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Schnake-Greene *et al.* (1990) Sugg *et al.* (1990) Janecek (1990) Sullivan *et al.* (1991)
Lactate dehydrogenase	*Ldh-1* *Ldh-2* *Ldh-3*	*P. boylii* *P. pectoralis* *P. polius* *P. attwateri*	Mascarello and Shaw (1973) Avise *et al.* (1974) Kilpatrick and Zimmerman (1975) Kilpatrick and Zimmerman (1976a) Kilpatrick (1984) Schnake-Greene *et al.* (1990) Sugg *et al.* (1990) Janecek (1990)
Leucine aminopeptidase	*Lap-1*	*P. boylii* *P. attwateri*	Kilpatrick (1984) Janecek (1990)
Malate dehydrogenase	*Mdh-1* *Mdh-2*	*P. boylii* *P. pectoralis* *P. attwateri*	Avise *et al.* (1974) Kilpatrick and Zimmerman (1976a) Schnake-Greene *et al.* (1990) Sugg *et al.* (1990) Janecek (1990)
Mannose phosphate isomerase	*Mpi-1*	*P. attwateri*	Sugg *et al.* (1990)
Nucleoside phosphorylase	*Np*	*P. attwateri*	Schnake-Greene *et al.* (1990) Sugg *et al.* (1990) Rogers and Engstrom (1992)
Peptidase	*Pep-1* *Pep-2*	*P. attwateri*	Schnake-Greene *et al.* (1990) Sugg *et al.* (1990) Janecek (1990)

(Continued)

Table 2. Protein variants in *P. boylii* group natural populations (Continued)

Protein	Locus	Species	References
Phosphogluconate dehydrogenase	*Pgd-1*	*P. boylii* *P. pectoralis* *P. attwateri*	Avise *et al.* (1974) Kilpatrick and Zimmerman (1975) Zimmerman *et al.* (1975) Kilpatrick and Zimmerman (1976a) Sugg *et al.* (1990) Janecek (1990)
Phosphoglucose isomerase	*Pgi-1*	*P. boylii* *P. pectoralis* *P. attwateri* *P. simulus*	Avise *et al.* (1974) Kilpatrick (1984) Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Sullivan *et al.* (1991) Rogers and Engstrom (1992)
Phophoglucomutase	*Pgm-1* *Pgm-2* *Pgm-3*	*P. boylii* *P. pectoralis* *P. attwateri*	Mascarello and Shaw (1973) Avise *et al.* (1974) Kilpatrick and Zimmerman (1976a) Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Sugg *et al.* (1990) Janecek (1990)
Sorbitol dehydrogenase	*Sdh-1*	*P. boylii*	Janecek (1990)
Superoxide dismutase	*Sod-2*	*P. boylii*	Janecek (1990)
Transferrin	*Trf*	*P. boylii* *P. pectoralis* *P. attwateri* *P. polius*	Rasmussen and Koehn (1966) Avise *et al.* (1974) Kilpatrick and Zimmerman (1975) Zimmerman *et al.* (1975) Kilpatrick and Zimmerman (1976a) Kilpatrick (1984) Rennert and Kilpatrick (1986) Rennert and Kilpatrick (1987) Werbitsky and Kilpatrick (1987) Sullivan *et al.* (1991)

(Continued)

Table 2. Protein variants in *P. boylii* group natural populations (Continued)

Protein	Locus	Species	References
Xanthine dehydrogenase	*Xdh-1*	*P. boylii* *P. attwateri*	Kilpatrick (1984)
Unspecified protein	*"Gp"*	*P. boylii*	Janecek (1990)

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Sullivan, J.M., C.W. Kilpatrick and P.D. Rennert. 1991. J. Mamm. 72:669-680.

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Table 3. VARIANT PROTEIN LOCI REPORTED FROM NATURAL POPULATIONS OF THE *PEROMYSCUS MEXICANUS* SPECIES GROUP (*Sensu* Carleton, 1989)

Protein	Locus	Species	References
Esterase (NADA)	*Es-3*	*P. mexicanus* *P. gymnotis*	Rogers and Engstrom (1992)
Glutamate oxaloacetate transaminase	*Got-1*	*P. mexicanus*	Rogers and Engstrom (1992)
6-Glycerophosphate dehydrogenase	*Gpd-1*	*P. mexicanus*	Rogers and Engstrom (1992)
Glucose phosphate isomerase	*Gpi-1*	*P. mexicanus* *P. gymnotis*	Rogers and Engstrom (1992)
Isocitrate dehydrogenase	*Icd-2* (*Idh-2*)	*P. mexicanus*	Rogers and Engstrom (1992)
Malate dehydrogenase	*Mdh-1* *Mdh-2*	*P. yucatanicus* *P. mexicanus*	Rogers and Engstrom (1992)
Malic enzyme	*Me*	*P. mexicanus* *P. gymnotis*	Rogers and Engstrom (1992)
Mannose phosphoisomerase	*Mpi-1*	*P. mexicanus* *P. zarhynchus*	Rogers and Engstrom (1992)
Nucleoside phosphorylase	*Np-1*	*P. zarhynchus*	Rogers and Engstrom (1992)
Peptidase	*Pep-1* (B) *Pep-2* (D)	*P. mexicanus*	Rogers and Engstrom (1992)
Phosphoglucomutase	*Pgm-1* *Pgm-2*	*P. mexicanus* *P. zarhynchus*	Rogers and Engstrom (1992)

Reference:

Rogers, D.S. and M.D. Engstrom. 1992. J. Mamm. 73:55-69.

Table 4. VARIANT PROTEIN LOCI REPORTED FROM NATURAL POPULATIONS OF THE *PEROMYSCUS TRUEI* SPECIES GROUP

Protein	Locus	Species	References
Albumin	*Alb*	*P. truei* *P. difficilis*	Brown and Welser (1968) Jensen and Rasmussen (1971) Johnson and Packard (1974) Zimmerman *et al.* (1975) Avise *et al.* (1979)
Esterase	*Es-1* *Es-2* *Es-3* *Es-4* *Es-5* *Es-6*	*P. truei* *P. difficilis*	Rasmussen and Jensen (1971) Johnson and Packard (1974) Zimmerman *et al.* (1975)
Glutamate oxaloactate transaminase	*Got-1* (*Aat-1*) *Got-2*	*P. truei* *P. difficilis* *P. gratus*	Zimmerman *et al.* (1975) Avise *et al.* (1979) Janecek (1990) Sullivan *et. al.* (1991)
α-glycerophosphate dehydrogenase	*Gpd-1* *Gpd-2*	*P. truei* *P. difficilis* *P. gratus*	Mascarello and Shaw (1973) Johnson and Packard (1974) Avise *et al.* (1979) Janecek (1990)
Isocitrate dehydrogenase	*Icd-1* (*Idh-1*) *Ich-2*	*P. truei* *P. difficilis* *P. gratus*	Mascarello and Shaw (1973) Johnson and Packard (1974) Avise *et al.* (1979) Rogers and Engstrom (1992) Janecek (1990)
Lactate dehydrogenase	*Ldh-1* *Ldh-2*	*P. truei* *P. gratus*	Mascarello and Shaw (1973) Janecek (1990)
Malate dehydrogenase	*Mdh-2*	*P. diffilis* *P. gratus*	Janecek (1990)

(Continued)

Table 4. Variant protein loci in *P. truei* group natural populations (Continued)

Protein	Locus	Species	References
Nucleoside phosphorylase	*Np-1*	*P. truei* *P. difficilis* *P. gratus*	Janecek (1990)
Peptidase	*Pep-1* *Pep-2* *Pep-3*	*P. difficilis* *P. gratus* *P. truei*	Janecek (1990)
6-Phosphogluconate dehydrogenase	*Pgd-1*	*P. truei* *P. difficilis* *P. gratus*	Mascarello and Shaw (1973) Johnson and Packard (1974) Zimmerman *et al.* (1975) Avise *et al.* (1979) Janecek (1990) Sullivan *et al.* (1991)
Phosphoglucose isomerase	*Pgi-1*	*P. truei* *P. difficilis* *P. gratus*	Avise *et al.* (1979) Sullivan *et al.* (1991)
Phosphoglucomutase	*Pgm-1* *Pgm-2* *Pgm-3*	*P. truei* *P. difficilis* *P. gratus*	Mascarello and Shaw (1973) Johnson and Packard (1974) Janecek (1990)
Sorbitol dehydrogenase	*Sdh-1*	*P. difficilis*	Janecek (1990)
Transferrin	*Trf*	*P. truei* *P. difficilis*	Avise *et al.* (1979) Johnson and Packard (1974) Sullivan *et al.* (1991)

References:

Avise, J.C., M.H. Smith and R.K. Selander. 1979. J. Mamm. 60:177-192.

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Janecek, L.L. 1990. J. Mamm. 71:301-308.

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Sullivan, J.M., C.W. Kilpatrick and P.D. Rennert. 1991. J. Mamm. 72:669-680.

Zimmerman, E.G., B.J. Hart and C.W. Kilpatrick. 1975. Comp. Biochem. Physiol. 52B:541-545.

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REPRODUCTIVE ASPECTS OF *PEROMYSCUS EREMICUS* IN BAJA CALIFORNIA SUR, MEXICO.

One of the projects that we are currently studying at the Centro de Investigaciones Biologicas de Baja California Sur is reproduction of the cactus mouse *Peromyscus eremicus*, in agricultural areas in the north of La Paz, Baja California Sur, Mexico.

This work attempts to establish a relationship between the reproduction of *Peromyscus eremicus* and the agricultural activity that takes places in the region since this is the most problematic species in the lower peninsula.

Rodents collected from agricultural and wild areas were compared to study differences in relative abundance and reproductive cycles.

Sherman traps were used for the collection of representative numbers of females and males in each sample. The specimens were transported live to the laboratory, and sacrificed. The testicles of the males and the reproductive apparatus of the females, were removed.

For males took morphometrical data of the testicles, and then the sperm was prepared to study for microscopic description and morphology. The sperm count was in related to testicle size and month of collection for statistical analysis.

Females were dissected to determine the number, size, and implant location of the embryos, for correlated with development. The measurements of the body and skull were taken, for statistical analyses between sexes.

* * *

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DISCOVERY OF BLACK MORPH *PEROMYSCUS* IN SOUTHEAST ALASKA

This summer, as part of a plan to inventory the small mammals of southeast Alaska, we trapped on 15 islands of the Alexander Archipelago. We would like to report the finding of three abnormal *Peromyscus maniculatus*. After examining them, we concluded that they are expressing the "black" or "extreme non-agouti" mutation (Horner, et al., 1980).

Two are from Shrubby Island (56° 14'N, 132° 58'W). They were caught approximately 50 meters from each other, one in a Sherman live trap and one in a museum special snap trap in a 17-year old clear-cut. Both are entirely black except for a white throat and mouth, white feet, and some white spotting on the belly and ventral side of the tail. With two exceptions, the specimens match Horner et al.'s (1980) description of the coat color mutant "black" or "extreme non-agouti". The first exception is that the hind feet are white beyond the toes to about the middle of the tarsals. Another difference is that some of the vibrissae are completely white, but most vibrissae follow Horner et al.'s description as being "black for at least their basal third, many becoming gradually lighter, even silvery, terminally."

A third black *Peromyscus maniculatus* was live trapped at Anita Bay, Etolin Island (56° 11'N, 132° 27'W). It was not apparent at first that it was expressing this mutation, since it is a juvenile. However, after comparing it to other juvenile *P. maniculatus* in the UA collection, we concluded it to be a juvenile black morph. It lacks the distinct bicoloration of body and tail of normal individuals. The dorsum is slightly darker than the sides and ventrum, which are of the normal gray of the sides of young *P. maniculatus*. This differs slightly from other black *Peromyscus* which are "slightly reddish" as juveniles (V. Hayssen, pers.comm.). All feet are white and there is a restricted amount of white about the lips and nose, less than on the adults from Shrubby Island.

We cryogenically preserved the heart, liver, and kidneys from the three specimens. Also, we made chromosome preparations from the live specimens from Shrubby and Etolin Islands; all materials are in the UA Museum. Kirstin Bagne first opened the Sherman live trap on Shrubby and noted that the live animal was unusually docile. There is some evidence that "black" mutants show a more depressed behavioral profile (V. Hayssen, pers. comm.). All other *Peromyscus* captured (n=107 for Shrubby, n=11 for Etolin) appear to be of normal coloration.

Horner et al. (1980) originally reported this mutation in *Peromyscus maniculatus gracilis* in the Hubbard Brook Experimental Forest near West Thornton, New Hampshire (BEH reported two black mice trapped out of approx. 1000 total). No other occurrences of this mutant have been found in the scientific literature. The wide geographic disjunction of this mutation may have implications for the estimate of the occurrence of this mutation. Either we are witnessing independent mutation events at this locus, or this recessive allele has been maintained in populations since divergence of the ancestors of these distinct subspecies. The latter explanation seems improbable, given the number of *Peromyscus* that have been collected and the lack of additional reports.

[Conroy Continued]

The significance of this discovery to theories of *Peromyscus* dispersal and systematics in southeast Alaska may be important. Of over 1000 islands in southeast Alaska, only 24 have been trapped for small mammals. There may be other populations with this mutation which could give us insight into island biogeographic relationships and additional sampling of these islands will give us a better idea of the extent of this mutation. We would be very interested if any readers know of other incidences of black or extreme non-agouti coat color.

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Virginia Hayssen, Department of Biology, Smith College, MA.

Specimens:

UAM# 20878: Adult female, K. Bagne prep #59, Alaska Frozen Tissue Collection, AF2874 (Karyotyped and frozen cell suspension), 173-83-21-16=22.8g

UAM# 20875: Adult female, S. MacDonald prep #1585, Alaska Frozen Tissue Collection AF3026, placental scars 3 left, 2 right, 191-92-22-17=22.0g

UAM# 20883: Juvenile female, S. MacDonald prep #1377, Alaska Frozen Tissue Collection AF2598 (Karyotyped and frozen cell suspension), 186-99-23-18=16.7g

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I am one year into my doctoral research on the foraging behavior of *Peromyscus*. Although *Peromyscus* are very generalist foragers, according to optimal foraging theory constraints still govern their decisions. Elements of foraging considered in my study include handling time and search time constraints affecting *P. leucopus* when nutrition and palatability of test foods are held constant. I hypothesize that increases in handling time and search time of food items will constrain the foraging decisions of *Peromyscus*, influencing their patch choice and patch use patterns. I further hypothesize that early experience with seeds requiring differing handling times will influence their foraging behavior. A final hypothesis is that different levels of perceived predation risk will affect patch use patterns.

Nine enclosures are arranged inside a 10 X 10 X 3 m high shed open only on the east side. Each enclosure is a 1.8 m cube of pine and hardware cloth where mice can choose to forage in artificial food patches. Two enclosures have video cameras and time lapse videotape recorders. In each, a camera is positioned so that the field of view encompasses the food patch. Feeding patches of two different sizes will allow me to test foraging between and within patches when handling time of food within is varied. Large wooden food patches measure 60 X 60 X 16 cm deep and are covered with a clear Plexiglas lid. Thirty-six plastic Petri dishes are glued to the bottom of each patch about 3 cm apart in a 6 X 6 grid. Small wooden food patches 30 X 30 X 16 cm deep are designed to hang at different heights inside each enclosure. None Petri dishes are glued to the bottom of each small patch in a 3 X 3 grid, providing one-quarter the area and one-quarter the number of food sites of the large patch.

Hulled and unhulled sunflower seeds are provided in the dishes and numbers eaten are recorded after a night of foraging. Handling and search times for the seeds are increased by covering seeds with sand, covering Petri dishes with wire mesh covers, or encasing seeds in empty gelatin capsules. Videotaping and direct observation are used to record search and handling times of seeds and search patterns of individuals. With the videotape, I can determine when the mouse enters and leaves a patch, how many Petri dishes it searches and in what order, and how many seeds of each type are eaten. Perceived predation risk experiments will include open, elevated patches and visual or olfactory stimuli.

I expect results indicating that *Peromyscus* are constrained by search and handling time requirements, but only when the cost is very high. In one completed experiment, there was a significant preference for unencapsulated seeds when half were in the open, and half were encased in gelatin capsules. The cost of procuring seeds with a high handling time was evidently sufficient to show an effect. Both adult and juvenile mice lacking experience with handling hulled seeds are expected to choose seeds without hulls. Mice are expected to avoid patches that have visual or olfactory cues suggestive of a predator. Search paths of mice within patches are expected to be efficient, as measured by number of dishes searched divided by path length.

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Peromyscus Isozyme Systems Interpretable Using Blood Samples

Population genetic studies of small mammals often rely on destructive sampling to obtain tissues for electrophoretic analysis. However, interpreting isozyme systems from red blood cells and plasma may encourage nondestructive techniques, which may be desirable if the focus of the study is related to evolutionary ecology or conservation biology.

Populations of two small mammal species, *Peromyscus leucopus* and *P. maniculatus*, are being studied at Mountain Lake Biological Station in the Allegheny Mountains of Virginia. The objective is to determine the dynamics of gene and morphological variation over space and time as the populations decline. Animals were captured with Sherman live traps in areas treated as either capture-recapture sites or removal sites. Under either sampling regime, blood was collected from every individual that weighed 10 grams or more. Typically, 0.04 ml of blood (0.02 ml/hematocrit tube) was taken from each individual using the suborbital sinus puncture technique with heparinized hematocrit tubes.

Starch-gel electrophoresis is being conducted at Savannah River Ecology Laboratory. Since blood was collected from all individuals (i.e. those captured on either capture-recapture sites or removal sites), it was critical to determine the maximum number of interpretable loci using either red blood cells or plasma. Buffers systems used for electrophoresis include: (1) AC=N-(3-Amino Propyl) Morphine-Citrate pH 6.0; (2) LIOH=Lithium Hydroxide pH 8.1; and (3) TC6.7=Tris Citrate pH 6.7/6.3. Most of the buffers and stains are described in either Selander *et al.* (1971) or Harris and Hopkinson (1976); however, some may have been revised in our lab.

Table 1 lists the isozymes and the appropriate buffer systems that are interpretable using either red blood cells or plasma. Also listed are the number of variable loci found in the *Peromyscus* populations that are being studied in southwestern Virginia. Of the 17 loci interpretable with plasma or red blood cells, eleven are polymorphic. The primary reason for determining the maximum number of polymorphic loci interpretable with blood is consistency in data collection and interpretation. It is necessary that the allozymes are scored using the same tissue type since different tissues can result in varying interpretations. This consistency in data collection will enable comparisons of populations from removal sites and capture-recapture sites.

To address the question of temporal and spatial morphological variation, it was necessary to employ destructive sampling to obtain cranial and post-cranial skeletons. Tissues were obtained from these individuals, which accounts for nine additional polymorphic loci. However, in cases where destructive sampling is not necessary, I encourage the use of blood for electrophoretic analysis. Non-destructive techniques are especially valuable in studies that involve life history traits such as reproductive success and survival.

Table 1. INTERPRETABLE PROTEIN SYSTEMS

ISOZYME	BUFFER	NUMBER OF LOCI *Interpreted*	*Variable*
Adenosine Deaminase (ADA)*	TC6.7	1	1
Carbonic Anhydrase (CA)*	AC	3	2
Glucose Phosphate Isomerase (GPI)*	AC	1	1
Isocitrate Dehydrogenase (ICD)*	TC6.7	1	0
Lactate Dehydrogenase (LDH)*	TC6.7	1	1
Malate Dehydrogenase (MDH)*	TC6.7	1	1
Peptidase (PEP)*	LiOH	4	2
Plasma Protein B (PPB)**	LiOH	1	0
Post-Albumin (P-Alb)**	LiOH	1	1
Purine Nucleoside Phosphorylase (NSP)*	AC	1	1
Sorbitol Dehydrogenase (SDH)*	AC	1	0
Transferrin (Trf)**	LiOH	1	1
Total		17	11

* red blood cells

** plasma

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Yawning is characterized by an involuntary, wide opening of the mouth, sometimes accompanied by tongue protrusion, closing of eyelids, and stretching of forelimbs. Although yawning occurs in a broad array of taxa, the proximate (neuronal mechanism) and ultimate (adaptive advantage) causes of yawning are unknown. Dopamine agonists (e.g. apomorphine, bromocriptine) reliably induce yawning and/or stretching in laboratory murids and rhesus monkeys but not in domestic cats (Code & Tang 1991; Marini 1981; Protais, et al. 1983).

Yawning is a rare behavior in deermice. To see if dopamine agonists could induce yawning behavior in *Peromyscus*, we injected bromocriptine (i.p. 10 mg/kg in canola oil) or vehicle into 12 male *Peromyscus maniculatus gracilis* of two color morphs (six agouti and six non-agouti [the black Horner mutant]) and counted the number of stretches and yawns in the subsequent 90 minutes. Vehicle injected animals exhibited no stretching or yawning, whereas 25% of the bromocriptine injected animals (i.e. three animals, one black and two agoutis) both yawned and stretched. All stretches and yawns were observed in the last 60 minutes of the observation period. Yawning and stretching were still rare behaviors (only 1-2 yawns and 12-13 stretches per animal). Differences between the color morphs were not apparent.

The incidence and frequency of induced yawning is much less than that observed for rats with similar dosages (Ushijima et al. 1988), however the deermice we used were all old animals (three years of age or more). The pharmacological effects of dopamine and dopamine agonists may change with age, thus different data might be obtained with younger deermice.

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EFFECTS OF PARITY ON RECRUITMENT IN A NATURAL POPULATION OF *PEROMYSCUS LEUCOPUS*

One of the goals of our research was to determine what factors were most important in determining whether an individual was recruited. Recruitment is the addition of new individuals to a population by reproduction or immigration. By markings litters of white-footed mice at an early age it was possible to identify recruits from the reproductive efforts of residents. We defined recruitment as the attainment of 18 grams. Mice weighing 16 grams are sexually mature (Jackson, 1952), therefore mice identified as recruits in our study should have the potential to reproduce. However, there are seasonal differences in age at maturity, which may influence the weight at maturity, and separation from the opposite-sex parent may be necessary to complete sexual maturation (Wolff, 1992).

We conducted a two year study of *P. leucopus* in a 2 hectare oak/hickory woodlot in Northwestern Ohio. The goal was to determine how season and mother's parity (which litter it is) influence recruitment. We employed a 15 X 26 livetrapping grid and 90 nestboxes to monitor this population. We marked litters of mice in nestboxes at as young an age as possible, usually within a week of birth. Mother's parity was estimated by examination of previous capture data. Lactation, obvious abdominal swelling, or a sudden weight loss of 5 g were considered as evidence of partuition (Goundie and Vessey, 1986).

During 1991 and 1992, 75 litters were contacted. Litter size averaged 4.93 (SE = 0.14, Range = 2-8, mode = 5). *P. leucopus* have a midsummer lull in breeding (Rintamaa et al. 1976), therefore litters were divided into spring or fall born. No seasonal differences in litter size were found (ANOVA d.f. = 73, $F = 0.185$, $p > 0.5$). In 1991, 27 of 69 males (39.1%) and 24 of 77 females (31.2%) were recruited into the population.

A two-way ANOVA was used with season of birth (spring or fall) and mother's parity (early or late litters) as independent variables and the number of recruits per litter as the dependent variable. The interaction between season and parity was significant (d.f. = 33, $F = 8.3$, $p = 0.0072$). Recruitment from the first and second litters was higher in both seasons than from third or later litters. This difference was more pronounced in the spring than in the fall. Parity significantly affected recruitment (d.f. = 33, $F = 13.51$, $p < 0.001$), but season did not (d.f. = 33, $F = 1.58$, $p > 0.2$).

Mother's parity and the interaction of season and parity significantly affected recruitment in 1991, results from 1992 have not yet been analyzed. Previous reports of seasonal differences in recruitment may need to be re-evaluated given our finding of a strong effect of parity.

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The mutant mice that included the mouse in the photograph in my last report now have a variety of colors in their coats. The dorsal coats vary from a light reddish brown to a darker grayish brown. The ventral coat varies; white, gray mottled, creamy yellow or an orangish yellow in different animals. In some, the color continues into the pectoral area, usually a bit lighter there, except in the animals with the gray mottled color. That often is even darker in the pectoral section.

The other group that produced the near-hairless offspring about a year ago also has one member with the orangish yellow underside. This one has almost a solid color the whole length of its body. I have been taking photographs of them to document this color variation. I have not developed the film yet so do not know how successful the photographs will be. It is not easy to catch them upside down.

The behaviors of these two groups are very different. The latter group are fairly docile. Any battles are not very violent and usually end before any obvious wounds are inflicted. Usually two or three of them are squeezed on top of the water bottle (of course two of them are more off than on, hanging from the hardware cloth top) taking a nap. When one of the ones hanging from the top gets tired it just pushes in to get its back on the bottle, and the one that had been resting on the bottle then hangs from the top. This trade-off sometimes goes on for many minutes or even hours.

The other group are violent and often kill the young at about the time that their color changes to brown. I have rescued a number of badly injured young ones that have survived injuries that I was sure would kill them. I thought they were killing the young males, but now I am not so sure. The last one I removed was a female with a back that looked "peeled". I put all except the old mother and her male into another tank. These included a young mother and a couple of pups that were just opening their eyes, one juvenile that was still small, but brown, and two other grays that were probably littermates of the "peeled" put (about five or six weeks old I would guess). For several days all seemed to go well. The "peeled" one seemed to be healing nicely. Then one night the "peeled" one was hanging to the cagetop trying to get out. I removed her and discovered that she had severe bites on her back. One was a deep crater, her ears were torn, and she was obviously relieved to be in a cage by herself. I used to wonder if they killed because there was something wrong with the animal they were killing. This has proved to be untrue because I have removed too many very badly injured animals that have survived and are still living. So far this little one is still alive and seems to be in good health.

Do any of you have any ideas about the reason a certain animal is sought out to be killed? I am still looking for some reason.

I find using my camcorder excellent for documentation of *Peromyscus* behavior. I have filmed a female carrying her pups that did not yet have their eyes open. She took them out of the nest, ran the length of the tank, going over jars, the exercise wheel, even doing a sort of run up the tank side (almost a flip),over and over again. Then she would return the pup to the nest jar and bring out another pup doing the same routine with it. This would be done for long periods of time a couple times a day and into the night.

An interesting result is that she and some of the young which are now several months old are all making the same trip, around the tank, up and over the wheel and jars. She taught them well! The only difference is that now none of them seem to come out of the jars during the day. It is a very quiet cage until late at night.

I read with interest the report in the last Newsletter written by Virginia Hayssen at Smith College. All my mice are of the wild variety and I have observed the same variations in grooming that she has reported. I am recording these grooming sessions too, because they are not equal. I have noted that some of the shorter grooming seems to be the result of nervousness. The animals do this when they are aware of a strange person nearby or a new situation (object in the tank moved, a new object introduced, another mouse moving in a threatening way) or before eating. They do a quick wash. The longer grooming seems to take place when they are feeling secure, have eaten and played, are generally comfortable with their surroundings. Those are the times when they concentrate on cleaning the whole body and work on the feet too.

Now I will watch which mice are doing the prolonged grooming to see if it is just certain individuals or any of them. It may be that some individuals are more relaxed than others. This will be something to watch in the future. It should be interesting to see if the mice at Smith will settle down to longer grooming if there are no distracting elements in their surroundings. Since I do not know the disposition of the black deermice, I think Virginia will have to tell us if they are more relaxed in general or if it is just that she has families that are more docile and relaxed than her agouti deer mice family. Certainly there are differences in my mice from family to family as inferred in the material I have written about my two families that produce the mutants.

I shall certainly watch the grooming activity of these different groups closely between now and the next issue.

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The effect of multiple hits on phylogenetic analysis of DNA sequence data: Congruence analysis using *Peromyscus*.

Nucleotide sequence data increasingly are being used to infer phylogenies from almost every group of organisms. The processes that effect the distribution of multiple substitutions at a single site differ among types of genes; therefore genes vary in phylogenetic usefulness. The well corroborated relationships among taxa of the *P. maniculatus* and *P. leucopus* groups are being used to assess the effects of multiple hits on phylogenetic reconstruction from three mitochondrial genes: Cytochrome b (Cyt b), Cytochrome oxidase subunit II (COII), and small ribosomal subunit (12s rRNA). To date, 350 bp from domain 3 of the 12s gene, 950 bp from the Cyt b gene, and 680 bp from the COII gene have been collected for *P. melanotis*, *P. polionotus*, *P. leucopus* (both southern and northern cytotypes), and *P. gossypinus*. Variation (inter- and intraspecific) is greatest is the Cyt b gene, followed by COII, then 12s. Phylogenetic analysis of the 3' 350 bp fragment of Cyt b using *Onychomys* (sequence from Brett Riddle) as an outgroup produced three equally parsimonious trees, all of which were congruent with the well corroborated relationships among these taxa. Use of distant outgroups *Rattus* or *Mus* resulted in poor resolution. The effect of outgroup saturation on ingroup topology is being addressed by examination of progressively distant outgroup taxa (*Onychomys*, *Neotoma*, and *Sigmodon*). Phylogenetic analysis of the 12s data yields topologies that are incongruent with the well corroborated relationships of these taxa. Character analysis of the changes in domain 3 of the 12s gene among these taxa suggests that the few sites that do vary are saturated.

This project is a Ph. D. dissertation being conducted in the laboratory of **Chris Simon**.

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Survey of Chaparral Mammals and Video Microscopy of Mammal Hair

Our lab will be surveying the vertebrates of the Santa Ana Mountains near Tucker Wildlife Sanctuary in Orange County, California. As part of this survey, we will be taking a population census of various small mammal species, and we expect *Peromyscus* to have a major contribution to that survey.

For his Master's thesis, Christopher Kodani is investigating the diet and range size of Gray Fox. Diet will be determined by using scat analysis. Since rodents make up a significant portion of Gray Fox diet, we will spend much of our time tackling the problem of identifying prey species by their hair. We are interested in identifying characteristics unique to each species' hair, as well as investigating differences between individuals of each species and variation of hair structure on each individual. In order to facilitate this work, we are developing a video microscopy and digitizing system, which has the ability to store images on diskette. Thus, we will be able to build a video library of mammal hairs.

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DEADLINE FOR NEXT ISSUE IS SEPTEMBER 15th

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