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Sue M. Abraham

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Abstract

SPECIES RECOGNITION IN WILD-CAUGHT PEROMYSCUS CALIFORNICUS AND PEROMYSCUS BOYLII (RODENTIA, CRICETIDAE)

by

Sue M. Abraham

Experiments were performed to compare homospecific and heterospecific species choice in sympatric populations of Peromyscus californicus and P. boylii. Mate selection performance of males and females were also compared. For both species there was no significant difference between males and females in mate selection performance, and selection Female P. boylii showed the highest appeared to be random. preference for its own species as indicated by the amount of time spent in the homospecific chamber, but this choice was not statistically significant. When data for the two species are combined, the percent of time in the homospecific chamber is significantly different for males and females with females spending more time in the homospecific chamber. This pair of species exhibits less tendency to show preference for its own species than several other species of Peromyscus.

LOMA LINDA UNIVERSITY

Graduate School

SPECIES RECOGNITION IN WILD-CAUGHT PEROMYSCUS CALIFORNICUS AND PEROMYSCUS BOYLII (RODENTIA, CRICETIDAE)

by

Sue M. Abraham

A Thesis in Partial Fulfillment

of the Requirements for the Degree

Master of Arts in Biology

June 1991

Each person whose signature appears below certifies that this thesis in his opinion is adequate, in scope and quality, as a thesis for the degree Master of Arts.

, Chairman

Leonard R. Brand, Professor of Biology

David L. Cowles, Asst. Prof. of Biology

James Gibson, Assistant Research Scientist

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INTRODUCTION

Several experiments dealing with species recognition between different species of *Peromyscus*, and the mechanisms involved in this process, have been conducted. Experiments by Blair (1953) indicated that the sympatric species, *Peromyscus truei* and *P. nasutus* choose homospecific mice in species discrimination studies. Furthermore, studies conducted by Smith (1965) indicated that allopatric and sympatric males of *Peromyscus eremicus* and *P. californicus* showed a preference for females of their own species.

More recently Carter and Brand (1986) compared homospecific and heterospecific choices of two closely related wild-caught species of *Peromyscus*, *P. californicus* and *P. eremicus*. Their investigations revealed that both species significantly chose the homospecific animal and the most significant homospecific choice was exhibited by mice from sympatric populations. Other studies of special recognition in *Peromyscus* have been done, and in most lab experiments they chose the homospecific stimulus animal. There were exceptions, however, for example in studies by Moore (1965) and by Reese (unpublished) some populations of *Peromyscus* did not exhibit homospecific choice.

Experiments dealing with species recognition in other rodents have also been performed. Murphy (1977) observed the sexual preference of female hamsters (genus *Mesocricetus*). Individual females were studied by placing them in an arena with a pair of stimulus males from two different species. When one male of the pair was a conspecific, estrous females of all three species significantly preferred the conspecific male.

Other experiments dealing with kin recognition in Rodentia, and more specifically kin recognition in Peromyscus also contribute to our understanding of social choice in these animals. Holmes and Sherman (1982) studied the ontogeny of kin recognition in two species of ground squirrels (Spermophilus parryii and S. beldingi). Avoidance of incestuous breeding was exhibited between siblings of Peromyscus maniculatus and P. eremicus (Dewsbury, 1982). The effect was stronger in P. eremicus. Studies on kin recognition in Peromyscus leucopus (Grau, 1982) revealed that both sexes investigate related strangers of opposite sex more than unrelated strangers. Kin recognition may benefit individuals in establishing subpopulations and inbreeding avoidance.

Aldhous (1989) studied intraspecies cross-fostering and its effects on the development of intrasexual kin discrimination in male laboratory mice, *Mus musculus L*. The results suggested that cues of self may be learned and influence

discrimination by juveniles, and discrimination is much influenced by cues of familiar littermates. Evidence for discrimination was not exhibited by subject animals when adult. These studies indicate an individual recognition process in rodents that is evident in laboratory choice experiments and that can be altered by early experience.

The cross-fostering process has also been used in interspecies studies of recognition to investigate the role of learning in the development of a species identity. Carter and Brand (1986) studied cross-fostering in Peromyscus californicus and P. eremicus and indicated that species choice was altered by cross-fostering and the results varied with species. Cross-fostered P. californicus had random species choice. However, cross-fostering of P. eremicus resulted in significant choice for the heterospecific species. Controls, which were laboratory raised, chose significantly for the homospecific chamber.

Earlier experiments by McCarty and Southwick (1977) indicated that *Peromyscus leucopus* males actually switched from homospecific to heterospecific preference after crossfostering with *Onychomys torridus*. Females also showed decreased preference for homospecific odors. These studies indicate that at least in some species the species identity is learned and can be altered by early experience.

Other studies have investigated the sensory mechanisms involved in species recognition. Moore (1965) suggested

olfactory discrimination as an isolating mechanism for Peromyscus maniculatus. However, P. polionotus exhibited a much lower level of discrimination. The difference between these species may be because P. polionotus leucocephalus is an insular form and has no midventral odor gland. Bowers and Alexander (1967) also indicated in Mus musculus that individual recognition and species recognition was based on olfactory clues.

Doty (1972) studied female Peromyscus maniculatus bairdii in an olfactorium and noted that reactions are influenced by gonadal state and that olfaction plays a role in sexual isolation between this species and *P. leucopus* noveboracensis. Significant species preference was exhibited by estrous females, but not by diestrous females.

More recently, Drickamer (1984) studied captures of two species of *Peromyscus* at live traps baited with male and female odors, and concluded that: 1) *Peromyscus leucopus* and *P. maniculatus* exhibit an attraction to homospecific odor/or avoidance of heterospecific odor. 2) Within each species there is a strong heterosexual odor preference. 3) There are no seasonal differences in these responses.

The research cited above indicates that odor is one important cue involved in species recognition. Experiments dealing with the early environment and its effect on odor recognition in Rodentia have been conducted by several investigators. Echandia et al. (1982) showed that

laboratory rats reared on lemon scent from birth to weaning acquire a permanent preference for lemon-scented bedding and that females have less of a preference than males. The role of preputial gland odors in female *Mus musculus* was studied by Hayashi (1979). Females reared by mothers with preputial glands preferred males with preputial glands to males without such glands. They also preferred females with preputial glands to females with preputial glands removed. Apparently the odors from these glands are important in individual preference.

Earlier Parkes and Bruce (1961) reviewed the relevant evidence regarding odor, which is one important factor in neurohormonal responses "affecting estrous, pseudopregnancy, and pregnancy in the mouse." Geyer (1981) indicated that in choice tests young rodents (*Peromyscus leucopus, Microtus pinetorum* and *Mus musculus*) prefer familiar odors and that rodents discriminate odors early in life. Furthermore, vocalizations of young rodents are affected by nest odors and the relationship between odors and vocalization may be different in different species.

Dagg and Windsor (1971) studied olfactory discrimination limits in gerbils and their results indicated that the gerbil possesses a keen sense of smell. In their conclusion they pointed out that since vision is limited or nonexistent in burrows where small mammals live, an acute sense of smell

is important, enabling an individual to immediately identify other individuals as part of their home group or as aliens.

In summary, previous research has indicated that most species of Peromyscus, but not all, exhibit homospecific choice in laboratory studies of species recognition. This choice can in some species be altered by early experience. It is also clear that odor is an important cue in species recognition, but the role of other sensory cues is not as well understood, probably as a result of other cues being more difficult to study than odor, for strictly mechanical These studies are often difficult to compare reasons. because they have not used the same methods. The research described in this paper is part of a study which attempts to standardize the approach to studying species recognition and the effects of cross-fostering on species identity.

The animals used in the experiments described in this paper, *Peromyscus californicus* and *P. boylii*, occur sympatrically in some areas in California. No natural hybrids of these two species are known to exist. The purpose of this research is to investigate species recognition in two closely related species, *Peromyscus californicus* and *P. boylii*, and attempt to quantify the species recognition of males as well as females from both species.

MATERIALS AND METHODS

Collection and Maintenance of

Experimental Subjects

The animals used in these experiments were wild-caught Peromyscus californicus and P. boylii. Most subjects were captured in aluminum Sherman live traps in Southwestern San Bernardino County, elevation 5500 ft., in sympatric areas along the border between yellow pine forest and chaparral from spring through fall. A few were caught in Western Riverside County, elevation 678 ft. in chaparral.

Trapped mice were identified and housed individually in laboratory cages. Animals were given at least five days to acclimate to their surroundings before they were used in an experiment. Each cage was provided with a mixture of rolled oats, cracked corn, and bird seed as well as water *ad libitum*. Fresh lettuce was provided periodically. Fresh pine shavings were regularly provided for bedding. The animals' rooms were maintained at 23°C, with the lights turned on from 0230 to 1430 hours (Pacific daylight time, PDT).

<u>Apparatus</u>

Testing was performed in four testing units, each consisting of five linearly arranged acrylic chambers with 0.64 cm. wire mesh tops or acrylic tops (Figure 1). Design of

the units was similar to that used by Carter and Brand (1986). The two outer chambers of each unit housed the stimulus mice (heterospecific and homospecific mice of the opposite sex from the test mouse). End chambers were separated from the center chamber by 0.64 cm. wire mesh. The three center chambers were connected by two tunnels (Figure The movement and location of the test mouse in the cen-1). ter chambers and of the stimulus mice in the outer chambers was detected by photodetector cells (Figure 2). This arrangement allowed the test mouse to move freely through the three center chambers and to choose to spend time next to one or both of the stimulus mice, or to avoid them both by staying in the center chamber. The amount of time spent next to each of the stimulus mice was used as an indication of preference for that species.

Commodore 64 computers provided continuous analysis of the photodetector signals, producing a record of time spent by the test mouse in each of the center chambers, and location of the test and stimulus mice in each of the four outer chambers, throughout the entire experimental period. The computer surveyed the photodetector signals at 0.10 second increments, and if any mouse had moved, the data were recorded. A video record of all mouse activity in one of the chambers was also made throughout each experiment.

The computer also continuously calculated a value called the index of association. Each time any mouse moved,

the computer recorded the time since the last mouse movement. This time was multiplied for each stimulus mouse by a number representing the inverse of the distance between the stimulus and test mice. The number was determined by counting the number of detector-to-detector intervals separating the two mice. At the end of each time block, these values were added and averaged to yield the index of association, which is a number between 0 and 11. The larger the number, the closer the average distance between the test and stimulus mouse. A value of 0 indicates that the test mouse was never in the chamber next to that stimulus mouse.

Experimental Procedure

Preparation for testing began at 1430 hours by placing a previously untested male or female mouse into the center chamber of the experimental unit. Entry to the adjacent chambers was blocked by sliding acrylic barriers for a period of 24 hours. During this acclimation period and throughout the entire testing time, food and water were provided only in the center chamber. This acclimation period in the center chamber allowed for the test animal to become familiar with the novel environment, and provided, through exposure, a preference for the center chamber, in case the first chamber encountered should influence preference. At the end of the acclimation period, stimulus mice of the opposite sex were placed in the detachable end chambers, and these were placed in the test position at the ends of the

chambers housing the test animals. One end chamber had a homospecific stimulus mouse while the other was heterospecific. Acrylic barriers were then removed and the behaviorrecording devices were activated. Each test lasted two hours and ten minutes. Experiments by Carter and Brand (1986) indicated this approximate test duration to be the most effective. All mice were used as test animals only once. The light regime and temperature were the same in the experimental rooms as in the animal room.

Forty trials were conducted, using P. californicus males (N = 9); females (N = 10); P. boylii males (N = 12); and females ((N = 11).

At the end of each test the animals were returned to the animal care rooms and the experimental units were thoroughly washed in hot water and detergent with a sponge. The units were rinsed in hot water and allowed to air dry, or were dried with paper towels.

The two hour 10-minute trial was divided into 30 minute (or less) time blocks and analyzed by time block and for the entire trial. The pooled data were analyzed statistically with paired-t comparisons for time in the homospecific chamber versus time in the heterospecific chamber. Additional analysis was done using a fixed effects model 2-way analysis of variance.

RESULTS

Female P. boylii showed the highest preference for its own species as indicated by amount of time spent in the homospecific chamber (68.6%), but this choice was not significant (t = 1.61; p = .141) (Table I). Female P. californicus exhibited essentially random choice with 46.8 percent of the time next to the homospecific chamber and 53.2 percent next to the heterospecific. The two male groups showed a preference for the heterospecific chamber, but in neither case was the result statistically significant. Male P. boylii spent 70.8 percent of the time next to the heterospecific (t = 1.68; p = .1213), and male P. californicus spent 82.2 percent of the time next to the heterospecific (t = 2.21; p = .058). Male P. californicus had the strongest preference for heterospecific choice (82.2%), which was close to being statistically significant (p =.058). When the results are measured in amount of time (in minutes) in each chamber, rather than percent time, the results are essentially the same.

The analysis of variance was accomplished using SPSSPC multiple linear regression with sex and species coded as dummy variables (Afifi & Clark, 1990). Using this dummy variable technique, females were chosen as a reference group

for sex and P. boylii as the reference group for species. Α sex*species interaction term was also included in the model. None of the three terms, sex, species, or sex*species interaction, proved to be statistically significant. The t statistics and p-values for the beta's describing the terms sex, t = -1.7, p = 0.09 indicating males spent more were: time in the heterospecific chamber but not significantly more; species, t = -1.3, p = 0.21 indicating *P. californicus* spent more time in the heterospecific chamber but not significantly more; and for the interaction term, sex*species, t = 0.03, p = 0.97 indicating no significant interaction. For the constant term in the model t = 1.6, p = .12 showing no overall sex by species time difference in the two chambers.

The index of association adds information to the analysis (Table III). It measures not only time spent in a given chamber, but also the position of the test mouse and stimulus mouse, how close they are to each other, and how much time was spent at various distances from each other. The index of association yields a number between 0 and 11, and higher numbers correlate to more time spent closer to the indicated stimulus mouse. Female *P. boylii* exhibited the highest mean for homospecificity with a value of 2.52. Female *P. californicus* indicated the next highest value of 2.02. Male *P. californicus* had a value of .92 for homospecificity. Male *P. boylii* exhibited the lowest value of

0.71 for homospecificity when comparing the mean of the index of association. The index of association does not indicate a significant homospecific preference for any of these groups, and indicates essentially the same species preference relationships as seen in Table I.

Table IV compares the amount of time and percent of time spent by the test mouse in the middle chamber and in the end chambers. All test mice spent over 60 percent of the time in the middle chamber. As previously stated, female *P. boylii* showed the greatest percentage of time (27.1%) in the homospecific end chamber of the four experimental groups, and male *P. boylii* showed the least percentage of time in the homospecific chamber (5.2%). Female *P.* californicus spent 17.9 percent of the time, and male *P.* californicus spent 6.6 percent of the time in the homospecific end chamber. Male *P. californicus* spent the greatest percentage of time in the heterospecific end chamber, 30.6 percent, and female *P. boylii* spent the least time (12.4%). Male *P. boylii* spent the greatest percentage of time in the middle chamber, 82.3 percent.

DISCUSSION

In my experiments, P. boylii and P. californicus do not demonstrate a clear preference for their own species. My data indicate that there is no significant difference between males and females in mate selection performance, and selection appears to be random.

This lack of significant preference is apparently not the result of the type of experimental apparatus or procedure, since recent experiments by Reese (unpublished) with *P. boylii* and *P. maniculatus*, using the same apparatus and procedure, did indicate a consistent significant homospecific choice by *P. boylii*. The data from Reese's experiments show that both male and female *P. boylii* selected conspecifics significantly more than the heterospecific stimuli mice, but that male and female *P. maniculatus* selected randomly.

Other investigators of *Peromyscus* have also observed several other subspecies that do not indicate homospecific choice in species preference experiments. Studies on *Peromyscus californicus* and *P. eremicus* indicated that "significant homospecific choice was made by mice from sympatric but not from allopatric populations" (Carter & Brand, 1986). Earlier, however, in a study by Smith (1965), *P.*

californicus from allopatric populations made a significant homospecific choice, but allopatric *P. eremicus* did not make a significant choice. Smith used mice from a different locality than Carter and Brand (1986) and the difference in results may be due to utilizing different populations.

Hill (1970) found that female, but not male, *P.* maniculatus bairdii consistently moved into an adjacent area of a laboratory test situation and nested with an oppositesexed conspecific, suggesting possible female mate selection in this species. However, neither male nor female *P. leucopus noveboracensis* did so.

Moore (1965) found in studies of *P. maniculatus* and *P. polionotus leucocephalus*, that male and female *P. maniculatus* exhibited significant homospecific choice, but in *P. polionotus* choice for the homospecific animal was not significant. The species choice behavior of *P. maniculatus* was quite different in the experiments of Reese (unpublished) and Moore (1965). The two sets of experiments are not very comparable. They used different subspecies of *P. maniculatus*, and Reese used *P. boylii* as the second species in his study, while Moore used *P. polionotus*. Another difference is that Reese used sympatric populations while the two populations used by Moore were from New Mexico and Florida. Furthermore, in the study by Reese (1990) the mice had access to olfactory, sound and visual stimuli,

whereas, Moore's (1965) design utilized olfactory stimuli alone.

In our experiments the estrous state of the females was not monitored, since similar experiments by Carter and Brand (1986) indicated no significant difference between choice behavior associated with estrous and non-estrous females. In future research it may be beneficial to determine whether this is equally true for other species.

Although none of the four groups in these experiments could be demonstrated to be making a significant choice (Table I), there was a considerable difference between some of the groups. Males of both species spent over 70 percent of their time in the heterospecific chamber (not counting time in the center chamber), and for male P. californicus this heterospecific choice was nearly significant. In contrast, female P. californicus showed random choice, and P. boylii spent 68.6 percent of their time in the homospecific chamber. When data for the two species are combined, the percent of time in the homospecific chamber is significantly different for males and females (N = 21; t = 2.8833; p = .009; t-test) with females spending more time in the homospecific chamber. (However, the fixed effects model indicated that this factor was not quite significant, p = .09.) This result, if correct, could be predicted from theoretical considerations, since females have more to lose from making mistakes in mate selection (Clutton-Brock & Harvey, 1978).

In this study, as in the research of Carter and Brand (1986), female mice show a stronger homospecific choice than males.

In nature, these species apparently do not interbreed. The absence of homospecific choice in these experiments does not necessarily mean that they have no behavioral reproductive barriers, but in the laboratory situation any behavioral homospecific preference that they may have does not show up as it does in some other species. Table I. A comparison of the mean percent of time spent in the homospecific chamber and in the heterospecific chamber by the four experimental groups. These values represent only the time spent in the two choice chambers, and exclude time spent in the center neutral chamber (see Table IV). Data from test periods of two hours and ten minutes. (Paired t-test)

Test Mouse (Species) % Homospecific % Heterospecific

P. californicus

Male	17.8%	82.2%	9	2.21	0.058	ns
Female	46.8%	53.2%	10	0.184	0.858	Ns
<u>P. boylii</u>						
Male	29.2%	70.8%	12	1.68	0.121	NS
Female	68.6%	31.4%	11	1.61	0.141	NS

NS = Not significant (p < .05)

Factor	t	p	
Sex	-1.7	0.09	
Species	13	0.21	
 Interaction	0.03	0.97	

Table II. A comparison of the results of the fixed effects model 2-way analysis of variance. Data from test periods of two hours and ten minutes. Table III. A comparison of the mean of the Index of Association. Data from test periods of two hours and ten minutes. (The Index of Association measures not only time spent in a given chamber, but also the position of the test mouse and stimulus mouse. Numbers can range from 0 to 11. Larger numbers indicate more time spent closer to the indicated stimulus mouse.) (Paired t-test)

Test Mouse (Species)	Homospecific	Heterospecific	<u>N</u>	<u>t</u>	P	
<u>P. californicus</u>						
Male Female	0.92 2.02	2.54 1.32	9 10	2.175 0.619	0.061 0.551	NS NS
<u>P. boylii</u>						
Male Female	.71 2.52	1.16 1.12	12 11	1.30 1.67	0.22 0.126	NS NS

NS = Not significant (p < .05)

Table IV. Amount of time in minutes and as percentage of time spent in the end chambers and in the center chambers expressed as means of the four experimental groups.

Test Mouse (Species)	Time in homospecific		Time in	n middle	Time in heterospecific		
	end chamber		cha	mber	end chamber		
,	Min.	8	Min.	8	Min.	*	
P. californicus							
Male	8.43	6.6%	80.0	62.8%	39.0	30.6%	9
Female	22.8	17.9%	79.4	61.9%	25.9	20.2%	10
<u>P. boylii</u>							
Male	6.6	5.2%	105.3	82.3%	16.0	12.5%	12
Female	34.9	27.1%	78.1	60.6%	15.97	12.4%	11

Figure 1. Diagram of mouse monitor arena showing the five acrylic chambers for mice. Letters A-E designate the five chambers.



Figure 2. Diagram of mouse monitor arena showing the five chambers, the locations of photodetectors (D), and connection to the computer. An emitter is located across from each detector. From I.E. Rouse, et al, 1990.



Frame containing detectors, emitters and part of the interface (cage cards).

APPENDICES

Appendix I. A comparison of homospecific and heterospecific choice by female *Peromyscus boylii*. Data from two hours and ten minutes test period.

Test Mouse Female Peromyscus boylii

Individuals showing preference for homospecific

Time associate with <i>boylii</i>	d Index of Association	Time associated with californicus	Index of Association
minutes		minutes	
22.21	1.77	9.70	0.93
75.84	5.18	0.50	0.09
4.23	0.53	1.96	0.22
35.28	2.94	11.55	1.04
79.50	6.98	4.02	0.35
76.42	4.68	0.87	0.20
$\bar{\mathbf{x}}$ = Mean time	associated with	P. boylii \pm SEM =	34.87 <u>+</u> 8.97

Individuals showing preference for heterospecific

4.15	0.62	6.06	0.44
0.22	0.02	0.27	0.16
36.25	2.11	52.68	3.19
26.32	1.02	61.36	3.75
23.16	1.86	26.73	1.91

 \overline{x} = Mean time associated with *P. californicus* <u>+</u> SEM = 15.97 <u>+</u> 6.56

т	=	1.600
N	=	11
\mathbf{DF}	=	20
Ρ	=	*
x	=	Mean time \pm SEM
Ν	=	Number of experiments
\mathbf{DF}	=	Degrees of freedom
Р	=	Probability *Not significant

Appendix II. A comparison of homospecific and heterospecific choice by male *Peromyscus boylii*. Data from two hours and ten minutes test period.

Test Mouse Male Peromyscus boylii

Individuals showing preference for homospecific

Time associated with <i>boylii</i>	Index of Association	Time associated with <i>californicus</i>	Index of Association
<u>minutes</u>		minutes	
21.44	2.06	15.32	1.55
4.89	1.17	2.78	0.39
1.25	0.12	0.56	0.05
2.04	0.30	1.90	0.25
0.49	0.09	0.04	0.02
0.73	0.46	0.58	0.04
0.22	0.28	0.05	0.00

 \overline{x} = Mean time associated with P. boylii + SEM = 6.61 + 2.12

Individuals showing preference for heterospecific

16.74	1.51	60.44	4.17
3.77	0.21	19.24	1.43
2.34	0.14	4.54	0.42
10.90	0.78	17.88	1.35
14.54	1.41	68.74	4.22

x = Mean time associated with *P. californicus* \pm SEM = 16.01 \pm 6.89

T = 1.681 N = 12 DF = 22 P = * $x = Mean time \pm SEM$ N = Number of experiments DF = Degrees of freedom P = Probability *Not significant

Appendix III. A comparison of homospecific and heterospecific choice by female *Peromyscus californicus*. Data from two hours and ten minutes test period.

Test Mouse Female P. californicus

Individuals showing preference for homospecific

Time associated with <i>boylii</i>	Index of Association	Time associated with <i>californicus</i>	Index of Association
minutes		minutes	
5.75	0.45	112.09	9.81
2.83	0.18	4.66	0.50
1.94	0.18	13.14	2.42
3.40	0.18	4.85	0.31
12.97	0.88	32.95	2.89
6.27	0.61	8.55	0.58
15.68	1.61	16.26	1.02
-			

x = Mean time associated with P. californicus \pm SEM = 22.84 \pm 10.34

Individuals showing preference for heterospecific

34.80	1.44*	23.77	1.67*
85.75	4.95	7.84	0.39
89.89	2.67	4.29	0.62

 \overline{x} = Mean time associated with *P. boylii* ± SEM = 25.93 ± 10.76

Т	=	0.184
N	=	10
\mathbf{DF}	=	18
Ρ	=	0.858 (*)
x	=	Mean time <u>+</u> SEM
N	=	Number of experiments
DF	=	Degrees of freedom
Ρ	=	Probability *Not significant

Appendix IV. A comparison of homospecific and heterospecific choice by male *Peromyscus californicus*. Data from two hours and ten minutes test period.

Test Mouse Male P. californicus

Individuals showing preference for homospecific

Fime associated with <i>boylii</i>	Index of Association	Time associated with <i>californicus</i>	Index of Association
<u>minutes</u>		minutes	
2.15	0.18	7.79	0.59
10.10	0.77	15.22	1.59
0.96	0.09	1.95	0.19

 \overline{x} = Mean time associated with *P. californicus* <u>+</u> SEM = 8.44 <u>+</u> 1.40

Individuals showing preference for heterospecific

5.77	0.35*	2.89	0.39*
119.01	3.99	7.07	1.16
19.12	2.38	13.04	1.21
48.52	4.06	6.27	0.54
81.37	7.05	10.15	1.19
64.24	4.02	11.57	1.45

 \overline{x} = Mean time associated with *P. boylii* + SEM = 39.14 + 13.18

T	=	2.206
N	=	9
\mathbf{DF}	=	16
Ρ	=	0.058 (*)
x	=	Mean time <u>+</u> SEM
Ν	=	Number of experiments
DF		Dograad of froodom
DI		Degrees of freedom
P	=	Probability *Not significant

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