THE RELATIONSHIP BETWEEN AGE OF GROUPING AND WEIGHT OF THE REPRODUCTIVE ORGANS OF MALE PRAIRIE DEERMICE

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Of the Requirements for the Degree of

Master of Arts

By

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APPROVAL SHEET

This thesis is submitted in partial fulfillment of

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ABSTRACT

Groups of five female and five male *Peromyscus maniculatus bairdii* were assembled at 21 days of age and killed at 40, 60, 80 and 100 days of age, or grouped at 40, 60, and 80 days and killed at 100 days of age. Controls were bisexual pairs mated at 21 days and killed at similar age levels as the experimentals.

Male deermice grouped at or before 40 days of age had significantly lighter testes and vesicular glands than controls ($.05 > P$). The weights of the vesicular glands and testes of mice grouped at 60 days of age or older were not significantly different than controls. The weights of the adrenal glands, both relative and absolute, were not correlated with these changes. It is believed that sexual maturity was reached in controls between this interval of 40 to 60 days of age.

The data suggest that the reproductive organ weights of male deermice may be inhibited if they are grouped before sexual maturity occurs. Similar results occurring in natural populations might have a profound effect on population dynamics.

THE RELATIONSHIP BETWEEN AGE OF GROUPING AND WEIGHT OF THE REPRODUCTIVE ORGANS OF MALE PRAIRIE DEERMICE

INTRODUCTION

As long ago as Plato's time it was recognized that population increases are produced by the forces of immigration and birth, while reductions are the result of emigration and death (Cole, 1957). Changes in these forces have been noted with changes in the physical environment, such as temperature, humidity, storms, etc. (Allee, et. al., 1949). Information gathered from laboratory studies of mammalian populations, however, show that when the physical environment is controlled or held constant, populations do not increase indefinitely, but control their growth as a result of factors thought to develop intrinsically (Brown, 1953, Calhoun, 1950, 1952, Chitty, 1960, Christian, 1956, Southwick, 1955, Terman, 1965).

Increase in the relative density of populations of small mammals have been correlated with a number of changes within the population. Among these are behavioral changes such as increases in aggression, disturbance of maternal care, dispersion and changes in the social structure. In addition, physiological changes occur which increase mortality, decrease growth, and reduce reproductive function (see Thiessen and Rodgers, 1961 and Christian 1963, 1964 for reviews). Although some of these changes may be species specific, others may be related to general mechanisms of population control.

Laboratory studies have shown that freely growing populations of _Peromyscus maniculatus bairdii_ are able to control their own numbers (Terman, 1965). Males born into these populations had significantly lower testes and vesicular gland weights then bisexually paired controls of similar age (Terman, 1963). It is not known whether these lower weights were due to an inhibition of growth or a regression in weight. Nor is it clear whether these results would be duplicated by grouping during some important stage of development or whether it is necessary for the mice to be part of the populations from birth.

The purpose of this study was to investigate, by randomly grouping deermice for prescribed periods of time, whether (1) the reproductive organ weights of deermice from these freely growing, asymptotic populations would be duplicated and (2) to study the relationship between the age of grouping and the reproductive maturation of deermice.

MATERIALS AND METHODS

Materials:

Prairie deermice (Peromyscus maniculatus bairdii) were obtained from a colony in which non-sibling matings were the practice. All mice were housed in 5" X 11" X 6" cages with wood shavings as bedding and food and water supplied in excess. Animals were placed in clean cages with fresh bedding every two weeks. Further, the temperature in the room was kept within the approximate limits of 70° to 80° F. and the lights were on each day from 7:00 a.m. to 7:00 p.m.

The Experimental Design:

The experimental design (Figure 1) consisted of randomly assigning 21 day old mice to one of three treatments: Pairing as bisexual pairs until death, grouping five males and five females until death, pairing as bisexual pairs and subsequently grouping as five males and five females.

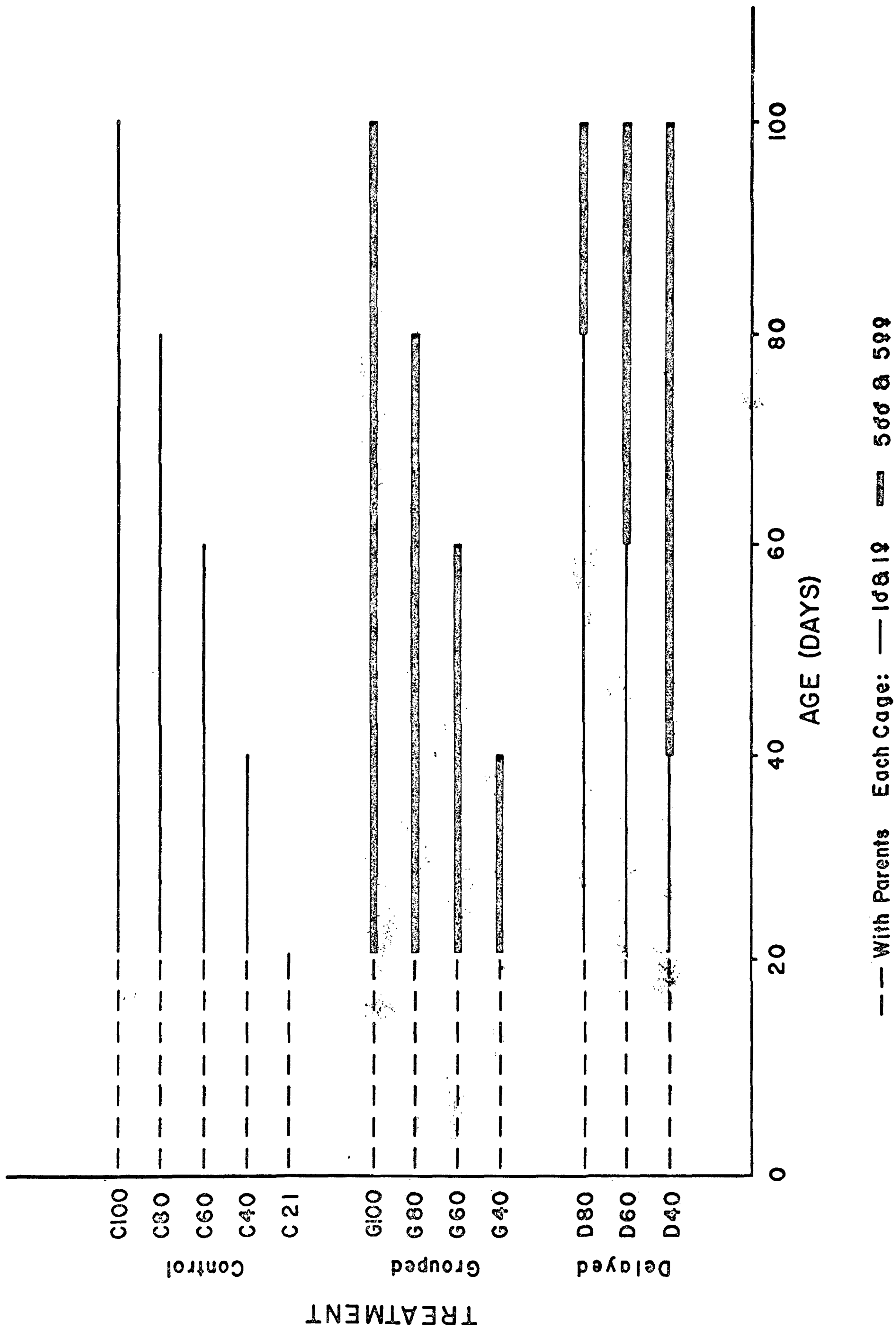
The bisexual pairs served as controls. These mice were weaned when 21 days old, toe clipped, weighed and placed with a mouse of the opposite sex born to other parents. The age difference of the mice of each pair did not exceed 10 days. Four replications of five pairs were killed with chloroform at either 40, 60, 80, or 100 days of age. The notation of Figure 1 refers to these as C40, C60, C80, and C100 respectively.

The "Grouped" treatment consisted of assembling 21 day old mice into groups of five males and five females. A maximum of one male and one female per litter was placed in each group, and the range of ages within a group did not exceed 10 days. Four replications (groups) were

Figure 1

The Experimental Design

Figure 1


TREATMENT
Control
C100
C80
C60
C40
C21
Grouped
G100
G80
G60
G40
Delayed
D80
D60
D40
0
20
40
60
80
100
AGE (DAYS)
With Parents
Each Cage: 1♂&1♀
5♂♂ & 5♀♀

killed at each of 40, 60, 80, and 100 days of age, these being listed in Figure 1 as G40, G60, G80 and G100 respectively. The date of death was determined by the mean age of the group.

The "Delayed Grouping" treatment consisted of maintaining mice as isolated bisexual pairs from 21 days of age until 40, 60, or 80 days of age and then assembling as groups of five males and five females. Four replications (groups) were assembled at each age and are noted in Figure 1 as D40, D60 and D80. All "Delayed Groups" were killed at 100 days of age regardless of when they were assembled. The date of death was determined in the same manner as the "Grouped" treatment.

Dissection and Weighing of the Organs:

All mice were killed with chloroform, weighed, examined for the amount of fat, size and condition of the reproductive organs, and fixed in 10% formalin. The testes, vesicular glands, and adrenals of the males were later cleaned and weighed to the nearest 1/100 of a milligram on a Sartorius balance.

RESULTS

Presented here are the data pertaining only to the males (testes, vesicular glands, absolute and relative adrenals, and body weights). Table 1 lists the mean of the means for the four replicates of each treatment and the standard errors for each.

Testes Weights:

The student "t" test (Snedecor, 1956) was used to make all comparisons between the testes weights. The mean testes weight of the four replicates of one treatment (five males per replicate, 20 males per treatment) were compared with the mean of four replicates of another treatment (n = 4). Tables II through IV list the "P" value of the "t's" calculated in this manner. "F" tests of the variances indicated that there were no significant differences between treatment variances.

Grouping mice at weaning resulted in markedly lower testes weights at all ages when compaired to controls (Table II). These differences were highly significant at 40 and 60 days of age, and significant at 100 days of age (.025 > P). However, the difference between C80 and G80 was significant only at .10 > P. These comparisons are graphically illustrated in Figure 2.

Table III compares the testes weights of mice killed at 100 days of age. These data indicate that mice grouped at weaning (G100) and at 40 days of age (D40) had lighter testes weights compared with animals maintained as isolated pairs and killed at 100 days (.025 > P and .10 > P, respectively). Grouping at 60 and 80 days of age (D60 and D80) resulted

TABLE I

The Means of the Testes, Vesicular Glands, Absolute and Relative Adrenal Weights, and the Body Weights With Their Standard Errors (in milligrams)

Replicates		Testes	Vesicular Glands	Absol. Ad.	Rel. Ad.	Body Wt.
C21	4	18.09 ± 0.94	0.93 ± 0.03	1.34 ± 0.03	15.1 ± 0.26	8.9 ± 0.41
C40	4	107.36 ± 8.86	17.03 ± 5.65	2.04 ± 0.14	14.0 ± 0.84	14.5 ± 0.26
C60	4	195.23 ± 16.87	61.95 ± 8.32	2.44 ± 0.22	16.8 ± 1.88	14.8 ± 0.47
C80	4	233.55 ± 31.47	81.97 ± 3.44	2.53 ± 0.22	16.7 ± 1.46	15.1 ± 0.39
C100	4	181.57 ± 20.47	93.24 ± 8.45	2.28 ± 0.22	16.0 ± 1.7	14.5 ± 0.59
G40	4	56.32 ± 7.37	4.67 ± 1.57	2.09 ± 0.05	15.8 ± 0.53	13.3 ± 0.20
G60	4	130.75 ± 14.93	29.07 ± 7.94	2.30 ± 0.05	15.4 ± 1.17	15.3 ± 0.77
G80	4	160.48 ± 35.89	48.78 ± 18.11	2.52 ± 0.24	17.1 ± 1.50	14.8 ± 0.78
G100	4	125.93 ± 16.10	57.25 ± 25.53	2.19 ± 0.26	14.9 ± 0.80	14.8 ± 0.80
D40	4	127.87 ± 26.94	32.39 ± 8.98	2.10 ± 0.20	14.6 ± 1.17	14.6 ± 1.17
D60	4	226.43 ± 37.33	133.30 ± 32.37	2.58 ± 0.20	16.4 ± 0.62	16.0 ± 0.62
D80	4	226.47 ± 26.44	112.39 ± 25.10	2.36 ± 0.17	14.2 ± 0.62	16.7 ± 0.30

TABLE II

Comparisons of the Mean Testes Weights of Control and Grouped Males

Age at Death	Comparison and Relative Weight Control Grouped	Level of Significance Student "t" test
40	C40 > G40	.001
60	C60 > G60	.01
80	C80 > G80	.10
100	C100 > G100	.025

TABLE III

Comparisons of the Mean Testes Weights of Control, Grouped or Delayed Grouping Males Killed at 100 Days of Age

Treatment	C100	G100	D40	D60
G100	C100 > .025			
D40	C100 > .10	G100 = N.S.		
D60	C100 < N.S.	G100 < .025	D40 < .025	
D80	C100 < N.S.	G100 < .005	D40 < .025	D60 = N.S.

N. S. = P > .10

Figure 2

A Comparison of the Mean Paired Testes Weight of Controls and Grouped

Figure 2

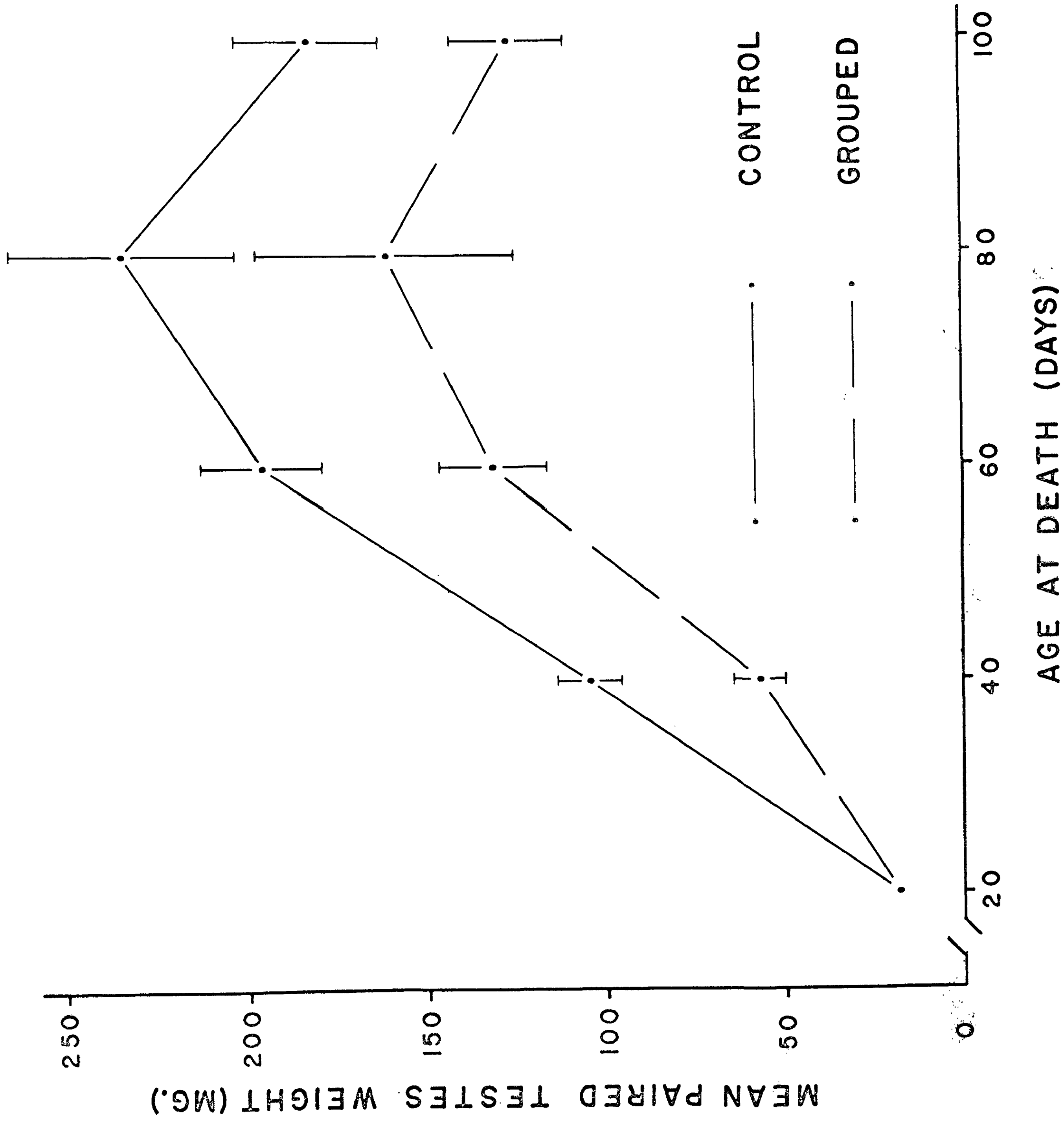
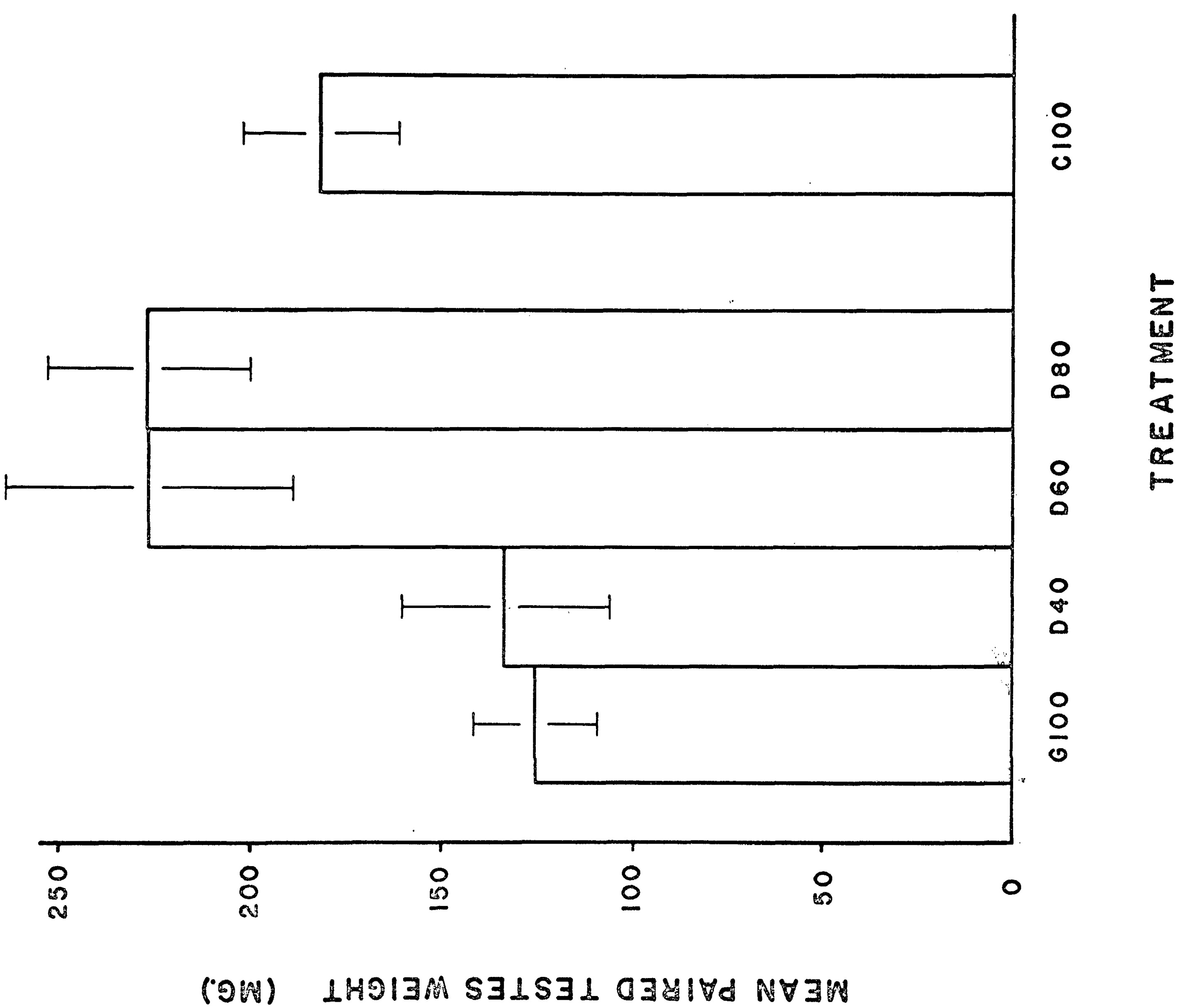

MEAN PAIRED TESTES WEIGHT (MG.)
0
50
100
150
200
250
AGE AT DEATH (DAYS)
20
40
60
80
100
CONTROL
GROUPED

Figure 3.

A Comparison of the Mean Paired Testes Weights of the Delayed Grouping and Controls Killed at 100 Days of Age

Figure 3


MEAN PAIRED TESTES WEIGHT (MG.)
250
200
150
100
50
0
G100
D40
D60
D80
C100
TREATMENT

in heavier testes weights than controls killed at 100 days, although not significantly so (Figure 3).

Further comparisons show that the average testes weights of the G100 and D40 mice were nearly identical as were the means of the D60 and D80 mice (Table I and III). A comparison of those grouped before 40 days (G100 and D40) with those grouped after 40 days (D60 and D80) shows that the testes weights of G100 and D40 were each significantly lighter than those of D60 and D80 in all four tests (Table III).

Table IV compares the testes weights of mice killed at 100 days of age with those of controls killed at 40, 60, 80, and 100 days of age. The testes weights of C40 mice were significantly lighter ($.005 > P$), while the testes weights of C60 mice were not significantly different than mice of the same treatment killed at 100 days of age (C100). The testes weights of C80 mice were heavier than those of the C100's but not significant at $P < .05$. Further comparisons tabulated in Table IV indicate that the testes weights of C40 mice (40 days old) were not significantly different than those of mice grouped at 21 (G100) and 40 (D40) days of age but killed at 100 days; but were significantly lighter than those grouped at 60 and 80 days (D60 and D80) and killed at 100 days ($.025 > P$ and $.001 > P$, respectively). Further the testes weights of controls killed at 60 and 80 days of age were significantly heavier than those grouped at 21 and 40 days of age (G100 and D40) and killed at 100 days, but not significantly different than those grouped later.

Vesicular Gland Weights:

Tables V through VII present comparisons of the vesicular gland weights in the same order as the testes weights are presented in Tables III through IV. The Mann-Whitney "U" test was used to compare the ves-

TABLE IV

Selected Comparisons of the Mean Testes Weight Between Treatment Groups Killed at 100 Days of Age and Controls Killed at Various Ages

Treatment	C40	C60	C80	C100
C100	C40 < .005	C60 > N.S.	C80 > .10	—
G100	C40 < N.S.	C60 > .01	C80 > .025	C100 > .025
D40	C40 < N.S.	C60 > .025	C80 > .025	C100 > .10
D60	C40 < .025	C60 < N.S.	C80 < N.S.	C100 < N.S.
D80	C40 < .001	C60 < N.S.	C80 < N.S.	C100 < N.S.

N. S. = P > .10

icular gland weights rather than the "t" test because in a large proportion of comparisons the variances were significantly different when tested with the "F" test. The Mann-Whitney "U" test (Seigel, 1956) is a non-parametric test which may be used in this situation.

Grouping mice at weaning resulted in significantly lighter vesicular weights at all ages when compared with controls (Table V). Vesicular gland weight differences of the grouped and control mice were highly significant at 40 and 60 days of age (.002 > P and .002 > P) and significant at 80 and 100 days of age (.02 > P and .05 > P, respectively). These results were generally similar to the testes comparisons and are graphically illustrated in Figure 4.

Table VI compares only treatment groups which were killed at 100 days of age. Those mice grouped at 21 (G100) and 40 (D40) days of age had significantly lighter vesicular glands than those kept as isolated pairs until killed at 100 days (C100). Those grouped after 40 days (D60 and D80) are heavier than controls killed at 100 days (C100), but not significantly so.

Contrary to the results reported for the testes (Table I and III), the mean vesicular gland weights of mice grouped at 21 (G100) and 40 (D40) days of age were not similar (.05 > P). The means of those grouped at 60 (D60) and 80 (D80) days of age while not similar, were not significantly different from one another. A comparison of those mice grouped at 21 days and killed at 100 days with all other groups killed at 100 days of age (Table VI) shows that mice grouped at 21 days of age had significantly heavier vesicular gland weights than those grouped at 40 (.05 > P) days, but significantly lighter vesicular gland weights than controls killed at 100 days (.05 > P), and mice grouped at 60 (D60) and 80 (D80) days of age (.05 > P, and .002 > P, respectively). These relationships are illustrated

TABLE V

Comparisons of the Mean Vesicular Gland Weights of Control and Grouped Males

Age At Death	Comparisons and Relative Weight Control		Grouped	Level of Significance (Mann-Whitney "U" Test)
40	C40	>	G40	.002
60	C60	>	G60	.002
80	C80	>	G80	.02 *
100	C100	>	G100	.05

asterisk signifies a significant difference in the variance

TABLE VI

Comparisons of the Mean Vesicular Gland Weights of Controls, Grouped or Delayed Grouping Males Killed at 100 Days of Age (Mann-Whitney "U" Test)

Treatment	C100	G100	D40	D60
G100	C100 > .05	—		
D40	C100 > .002	G100 > .05	—	
D60	C100* < N.S.	G100 < .05	D40* < .02	—
D80	C100 < .10	G100 < .002	D40 < .002	D60 < N.S.

N. S. = P .10

asterisk signifies a significant difference in the variance

Figure 4

A Comparison of the Mean Paired Vesicular Gland Weight of Controls and Grouped

Figure 4

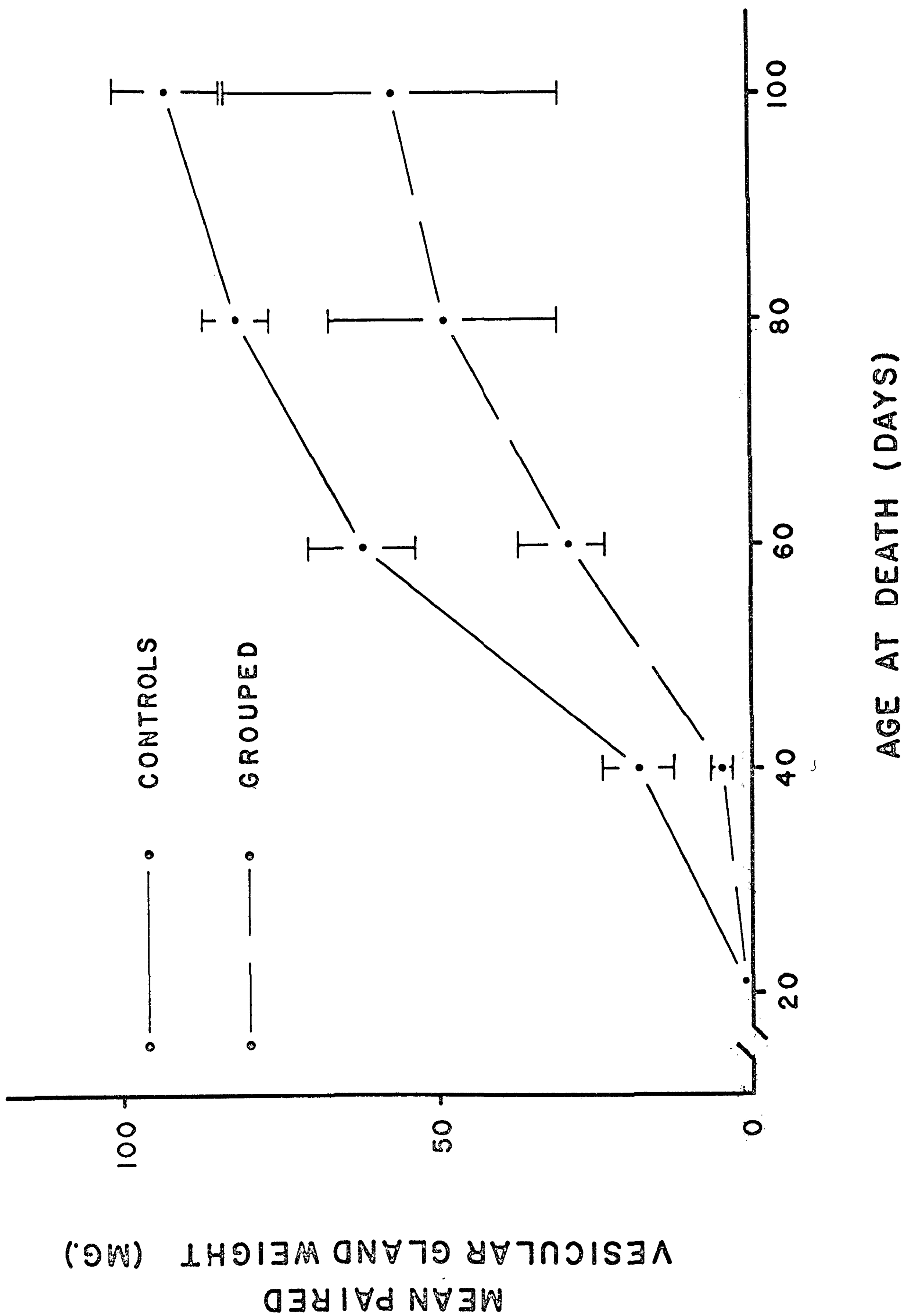
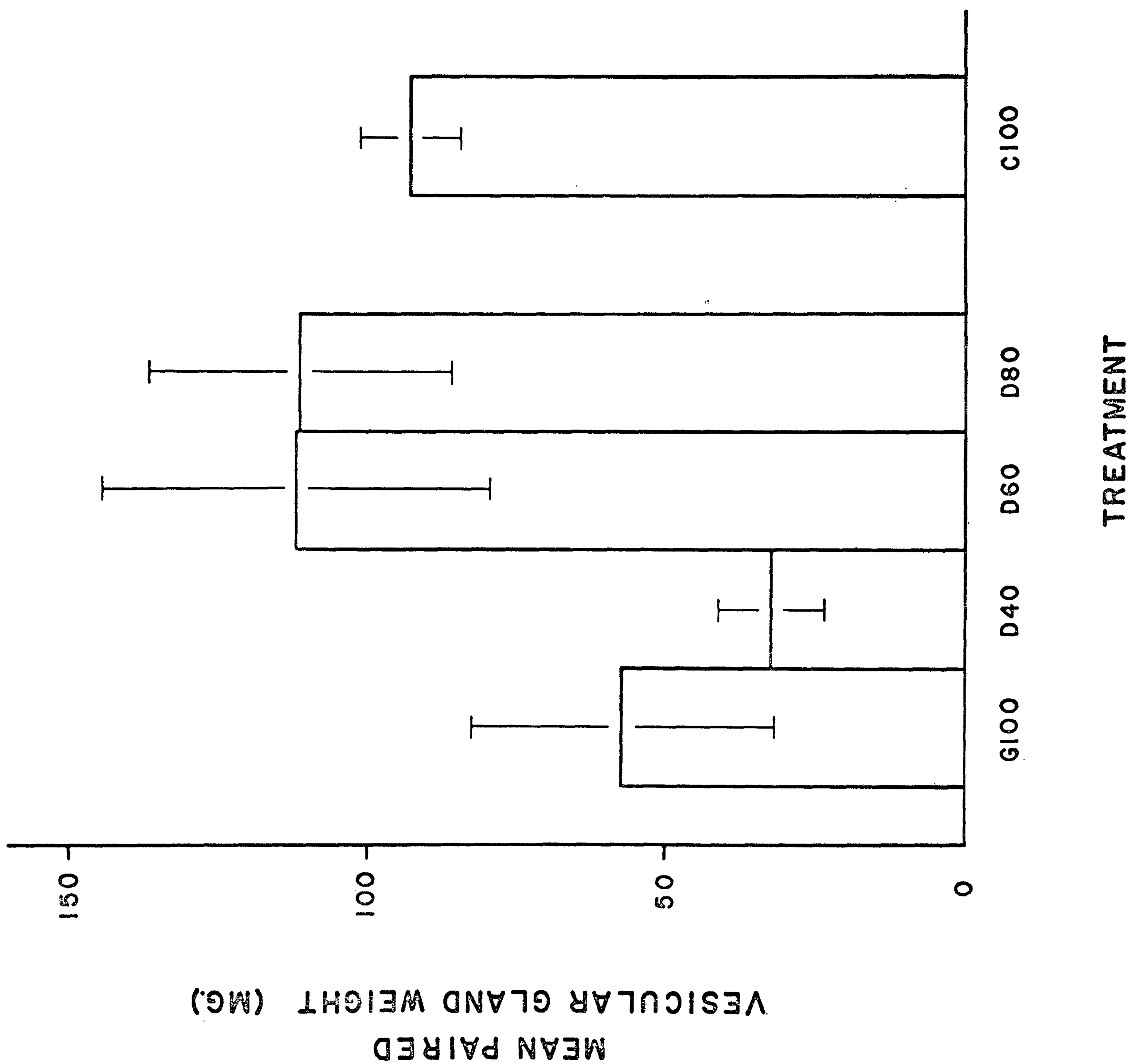

CONTROLS
GROUPED
AGE AT DEATH (DAYS)
20
40
60
80
100
MEAN PAIRED
VESICULAR GLAND WEIGHT (MG.)
0
50
100

Figure 5

A Comparison of the Mean Paired Vesicular Gland Weight
of the Delayed Grouping and Controls
killed at 100 Days of Age

Figure 5


MEAN PAIRED
VESICULAR GLAND WEIGHT (MG.)
150
100
50
0
G100
D40
D60
D80
C100
TREATMENT

in Figure 5.

Table VII compares the vesicular gland weights of mice killed at 100 days of age and controls killed at 40, 60, 80 and 100 days of age. The vesicular glands of controls killed at 40 and 60 days were significantly smaller than those of controls killed at 100 days of age ($.002 > P$ and $.05 > P$, respectively), while that of controls killed at 80 days were smaller but not significantly so. The vesicular glands of controls killed at 40 days (C40) were significantly lighter ($.002 > P$) than those of mice grouped at 21 days of age and killed at 100 (G100), but were not significantly different than those grouped at 40 days and killed at 100 days (D40). The vesicular glands of controls killed at 60 days (C60) were not significantly different than the G100's, but were significantly heavier than the D40's, even though these controls were 40 days younger!

Absolute Adrenal Weights:

Tables VIII through X present the comparisons for the absolute adrenal weights in the same sequence as the previous weights have been presented. The "t" test was used to test the significance of the differences in weight because only one comparison indicated a difference in variance, and when this test was repeated using the Mann-Whitney "U" test there was no change in the "P" value.

It is evident from the data of Table VIII that grouping mice at weaning resulted in a significantly heavier absolute adrenal weights than controls only at 40 days of age ($.05 > P$). After 40 days of age the weights of the controls were heavier than the Grouped, but not significantly so.

Table IX presents a comparison of all treatment groups which were killed at 100 days of age. Of the ten comparisons listed none were sig-

TABLE VII

Selected Comparisons of the Mean Vesicular Gland Weight
Between Treatments Killed at 100 Days of Age
and Controls Killed at Various Ages
(Mann-Whitney "U" Test)

Treatment	C40	C60	C80	C100
C100	C40 * < .002	C60 < .05	C80 < N.S.	———
G100	C40 * < .002	C60 < N.S.	C80 < .02	C100 < .05
D40	C40 < N.S.	C60 < .002	C80 < .002	C100 < .002
D60	C40 * < .002	C60 < N.S.	C80 < N.S.	C100 < N.S.
D80	C40 * < .02	C60 < .02	C80 < N.S.	C100 < .10

N. S. = P .10

asterisk signifies a significant difference in the variance

TABLE VIII

Comparisons of the Mean Absolute Adrenal Weights of Control and Grouped Males

Comparison and Relative Weight Control		Grouped	Level of Significance (Student "t" Test)
C40	<	G40	.05
C60	<	G60	N.S.*
C80	<	G80	N.S.
C100	<	G100	N.S.

N. S. = P .10

asterisk signifies a significant difference in the variance

TABLE IX

Comparisons of the Mean Absolute Adrenal Weights of Control, Grouped, or delayed Grouping Males Killed at 100 Days of Age

Treatment	C100	G100	D40	D60
G100	C100 > N.S.	———		
D40	C100 > N.S.	G100 > N.S.	———	
D60	C100 < N.S.	G100 < N.S.	D40 < .10	———
D80	C100 < N.S.	G100 < N.S.	D40 < N.S.	D60 < N.S.

N. S. = P .10

nificantly different at $.05 > P$, and only one was significantly different at $.10 > P$ - the absolute adrenal weights of mice grouped at 40 days (D40) were lighter than those of mice grouped at 60 days (D60).

Table X compares the absolute adrenal weights of mice killed at 100 days of age and controls killed at 40, 60, 80, and 100 days of age. Again there were very few differences which were significant. Controls killed at 40 days of age (C40) had lighter adrenal weights than mice grouped at 60 (D60) and 80 (D80) days and killed at 100 days ($.05 > P$ and $.025 > P$, respectively) and isolated pair controls killed at 80 days of age (C80) exhibited a trend toward significantly heavier adrenals than those grouped 21 days (N.S.) and at 40 days ($.1 > P$) and killed at 100 days.

The Relative Adrenal Weights:

Tables XI through XIII present comparisons of the relative adrenal weights in the same order as previously followed. The "t" test was employed here because the "F" test of the variances indicated no significant differences between the variances of the treatments. All relative adrenal weights are given in terms of adrenal weight/body weight X 100.

Grouping mice at weaning resulted in significantly heavier relative adrenal weights when compared to isolated pair controls at 40 days of age ($.05 > P$). At 60, 80, and 100 days of age there were no significant differences between grouped and control animals.

Table XII contains the results of comparisons of treatments killed at 100 days of age. Of the 10 comparisons listed only one is significant at $.05 > P$; the C100 mice had significantly heavier relative adrenal weights than those grouped at 80 days and killed at 100 days (C80). A trend toward significantly heavier relative adrenal weights ($.10 > P$) was found in those mice grouped at 80 days and killed at 100 days (D80) when com-

TABLE X

Selected Comparisons of the Mean Absolute Adrenal Weights Between Treatments Killed at 100 Days of Age and Controls at Various Ages

Treatment	C40	C60	C80	C100
C100	C40 < N.S.	C60 > N.S.	C80 > N.S.	——
G100	C40 < N.S.	C60 > N.S.	C80 > N.S.	C100 > N.S.
D40	C40 < N.S.	C60 > N.S.	C80 > .10	C100 > N.S.
D60	C40 < .05	C60 < N.S.	C80 < N.S.	C100 < N.S.
D80	C40 < .025	C60 > N.S.	C80 > N.S.	C100 < N.S.

N. S. = P .10

TABLE XI

Comparisons of the Mean Relative Adrenal Weights of Control and Grouped Males

Comparison and Relative Weight Control		Grouped	Level of Significance (Student "t" Test)
C40	<	G40	.05
C60	>	G60	N.S.
C80	<	G80	N.S.
C100	>	G100	N.S.

N. S. = P .10

TABLE XII

Comparisons of the Mean Relative Adrenal Weights of Control, Grouped or Delayed Grouping Males Killed at 100 Days of Age

Level of Significance (Student "t" test)

Treatment	C100	G100	D40	D60
G100	C100 > N.S.	———	———	———
D40	C100 > N.S.	G100 > N.S.	———	———
D60	C100 < N.S.	G100 < N.S.	D40 < N.S.	———
D80	C100 > .05	G100 < N.S.	D40 < .10	D60 > N.S.

N. S. = P .10

pared to those grouped at 40 days and killed at 100 days (D40).

Table XIII compares the relative adrenal weights of mice killed at 100 days of age and controls killed at 40, 60, 80 and 100 days of age. Only three of the 19 comparisons listed were significantly different. Isolated pair controls killed at 60 days (C60) had significantly heavier relative adrenal weights than the controls killed at 100 days ($.01 > P$); the controls killed at 40 days (C40) had significantly lighter relative adrenal weights than those grouped at 60 days (D60) and killed at 100 ($.025 > P$); and the controls killed at 100 days (C100) had significantly heavier relative adrenal weights than those grouped at 80 days (D80) and killed at 100 days ($.05 > P$). These differences do not seem to fall into a discerniable pattern.

Body Weights:

Tables XIV through XVI contain comparisons of the body weights in the same sequence as the previous weights have been presented. The "t" test was again used because there were only two comparisons exhibiting significant differences between the variances of the treatments. These two comparisons were corrected to obtain the proper "P" value (Snedecor, 1956) and were also tested by the Mann-Whitney "U" test.

Grouping mice at weaning resulted in significantly lighter body weights at 40 days of age (Table XIV) than controls killed at 40 days. Mice grouped at 21 days and killed at 60, 80, and 100 days of age were not significantly different than controls of the same age.

Table XV presents a comparison of all treatment groups which were killed at 100 days of age. These data indicate that grouping mice at 21 (G100) and 40 (D40) days and killing at 100 days did not result in significantly different body weights when compared to Controls killed at

TABLE XIII

Selected Comparisons of the Mean Relative Adrenal Weights
Between Treatments Killed at 100 Days of Age
and Controls Killed at Various Ages

Level of Significance (Student "t" Test)

Treatment	C40	C60	C80	C100
C100	C40 <	C60 >	C80 >	———
	N.S.	.01	N.S.	
G100	C40 <	C60 >	C80 >	C100 >
	N.S.	N.S.	N.S.	N.S.
D40	C40 <	C60 >	C80 >	C100 >
	N.S.	N.S.	N.S.	N.S.
D60	C40 <	C60 >	C80 >	C100 <
	.025	N.S.	N.S.	N.S.
D80	C40 <	C60 >	C80 >	C100 >
	N.S.	N.S.	N.S.	.05

N. S. = P .10

TABLE XIV

Comparisons of the Mean Body Weights of Control and Grouped Males

Comparison and Relative Weight			Level of Significance
Control		Grouped	(Student "t" Test)
C40	>	G40	.005
C60	<	G60	N.S.
C80	>	G80	N.S.
C100	<	G100	N.S.

N. S. = P .10

TABLE XV

Comparisons of the Mean Body Weights of Control, Grouped or Delayed Grouping Males Killed at 100 Days of Age

Level of Significance (Student "t" Test)

Treatment	C100	G100	D40	D60
G100	C100 < N.S.	—	—	—
D40	C100 < N.S.	G100 > N.S.	—	—
D60	C100 < .05	G100 < N.S.	D40 < N.S.	—
D80	C100 < .005	G100 < .025	D40 < .10	D60 < N.S.

N. S. = P .10

100 days of age. Grouping at 60 (D60) and 80 (D80) days of age, however, produced significantly heavier body weights when compared to the C100's (.05 > P and .005 > P, respectively). The D80's are also significantly heavier than those grouped at 21 (G100) and 40 (D40) days and killed at 100 days (.025 > P and .10 > P, respectively). The comparison of the D80's with the D40's indicated a significant difference in variance between the groups. When this comparison was tested with the Mann-Whitney "U" test the body weights of the D80 mice were significantly heavier than those of the D40's (.002 > P). All other comparisons of mice killed at 100 days of age showed no significant differences in body weights.

Table XVI compares the body weights of mice killed at 100 days of age with controls killed at 40, 60, 80 and 100 days. The 40, 60, and 80 day controls were not significantly different in body weight from the controls killed at 100 days of age (C100). The C40's, C60's and C80's were also not significantly different from mice grouped at 21 (G100) and 40 (D40) days and killed at 100 days. However, the controls killed at 40 and 60 days had significantly lighter body weights than those mice grouped at 60 (D60) and 80 (D80) days and killed at 100. The controls killed at 80 days of age (C80) were not significantly different in body weights than those mice grouped at 60 days and killed at 100 (D60), but were significantly lighter than those grouped at 80 (D80) and killed at 100 (.005 > P). These comparisons further emphasize the heavier body weights of mice which were grouped at 60 and 80 days and killed at 100 days of age.

TABLE XVI

Selected Comparisons of the Mean Body Weights Between Treatments Killed at 100 Days of Age and Controls Killed At Various Ages

Level of Significance (Student "t" Test)

Treatment	C40	C60	C80	C100
C100	C40 =	C60 >	C80 >	——
	N.S.	N.S.	N.S.	
G100	C40 <	C60 =	C80 >	C100 <
	N.S.	N.S.	N.S.	N.S.
D40	C40 <	C60 >	C80 >	C100 <
	N.S.*	N.S.	N.S.	N.S.
D60	C40 <	C60 <	C80 <	C100 <
	.025	.10	N.S.	.05
D80	C40 <	C60 <	C80 <	C100 <
	.001	.005	.005	.005

N. S. = P .10

asterisk signifies a significant difference in the variance

DISCUSSION

The importance of the social environment to the regulation of population numbers is becoming apparent (Brown, 1953, Christian and Davis, 1964, Louch, 1956, 1958, Southwick, 1955, Terman, 1961, 1962, 1965, Thiessen and Rodgers, 1961). In any such system of population regulation the immature members play an important role because any condition which adversely affects their development (especially reproductive development) may directly result in a decline of the reproductive rate of the next generation. The results of the present study appear to have relevance to these considerations.

In this experiment deermice which were grouped at weaning (21 days of age) and killed at 20 day intervals (40, 60, 80 and 100 days) averaged lighter testes weights than controls maintained as bisexual pairs and killed at these same ages. With the exception of animals killed at 80 days of age these differences were significant at the .05 level of probability. In other experiments in which male house mice were grouped for varying lengths of time the grouped animals had lower testes weights than isolated controls (Christian, 1955a, 1956, 1963, Christian and Davis, 1964, Theissen, 1963).

Grouping deermice at 40 days and killing at 100 days (D40) also produced lower testes weights than controls killed at the same age (C100). Further, the testes weights of the D40's were not statistically different than those grouped at 21 days and killed at 100 (G100) even though the latter were grouped for a longer period of time.

The extent of the effect of grouping is shown by the fact that the testes weights of both the G100's and the D40's were not significantly different than controls killed at 40 days of age (C40), although these controls were 60 days younger! Further, controls killed at 60 days of age (C60) and 40 days younger than the G100 and D40 mice had significantly heavier testes weights. In addition the testes weights of the G100's and the D40's were significantly lighter than those of mice grouped at or after 60 days of age (D60 and D80) while the latter two groups (D60 and D80) did not differ significantly from the testes weights of controls killed at 100 days. These findings indicate that the weights of the testes of the G100 and the D40 mice have changed little from the time they were grouped (see Figure 6).

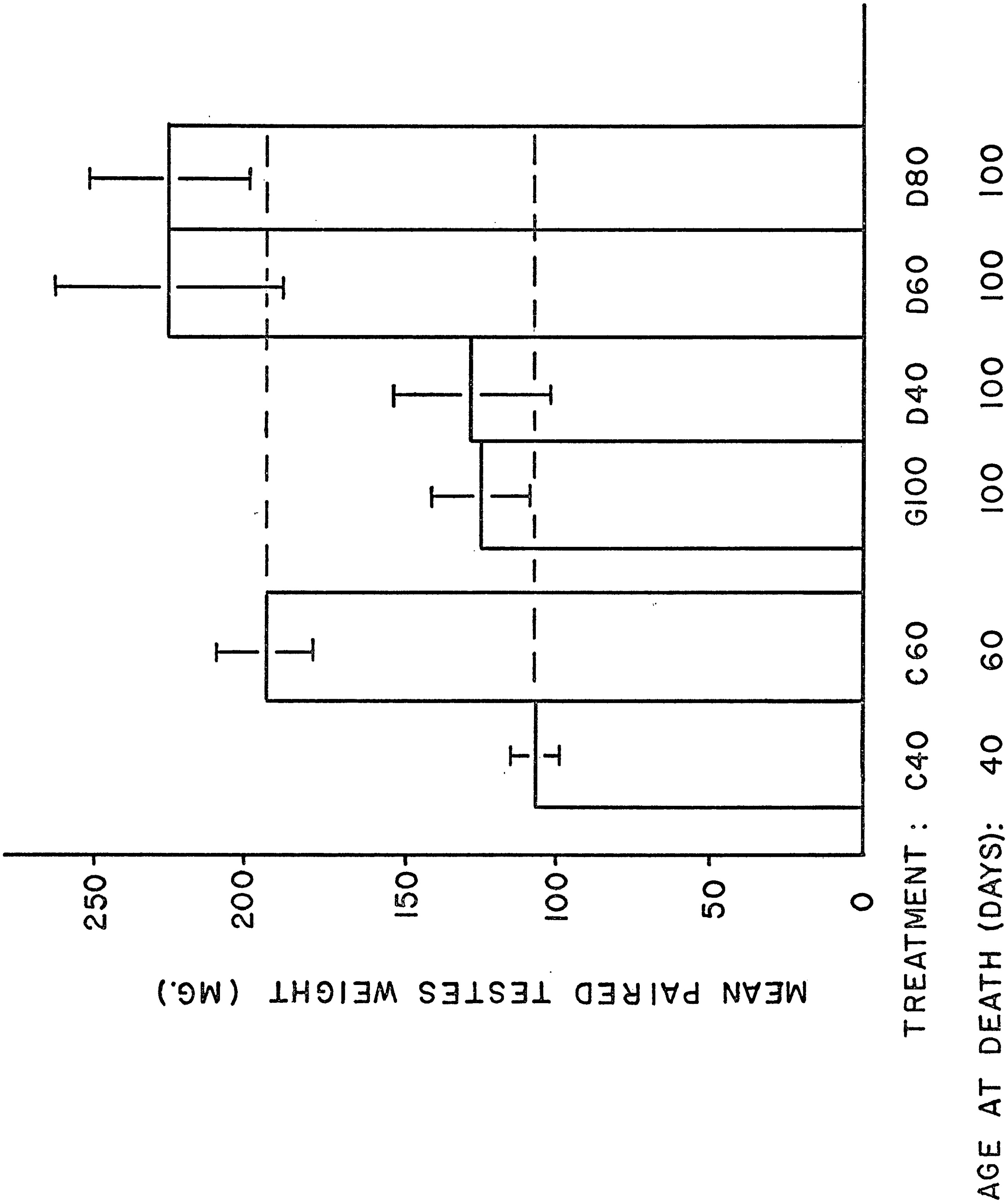
The above data suggest that grouping deermice before a particular age (at or before 40 days) inhibits testes growth. An examination of the testes weights of the controls indicates that sexual maturation occurs after this age (40 days), if it is assumed that testis weight is a direct measure of testicular function (Christian, Lloyd and Davis, 1965). Controls killed at 40 days of age (C40) had significantly lighter testes than controls killed at 100 days (C100), while those controls killed at 60 and 80 days of age (C60 and C80) had testes weights which were not significantly different from those of the C100's. Further the testes of the C40's were significantly lighter than the C60's. If the testes weights of the C100's are used as a criterian for sexual maturity it seems evident that male deermice become sexually mature after 40 days, but before 60 days of age. Evidence to support this contention is given by Clark (1938) who measured the number of motile sperm in the epididymus of P. maniculatus and estimated that the average age of sexual maturity is 58 days of age.

Figure 6

A Comparison of the Mean Paired Testes Weights of Experimentals Killed at 100 Days of Age and Controls Killed at 40 and 60 Days of Age

note: The dotted lines represent the testes weights before (C40) and after (C60) the estimated age of sexual maturity.

Figure 6



MEAN PAIRED TESTES WEIGHT (MG.)
250
200
150
100
50
0
TREATMENT : C40 C60 G100 D40 D60 D80
AGE AT DEATH (DAYS): 40 60 100 100 100 100

The age at which pregnancies first occur in the controls gives another estimate of the age of sexual maturity for the males since the females mature earlier (Clark, 1938). In the present experiment no control females killed at 40 days of age (C40) were pregnant, but of the controls killed at 60 days of age (C60) three were pregnant.

The above evidence strongly suggests that the average age of sexual maturity in male P. m. bairdii is after 40 days of age but before 60, and probably within the range of 50 to 55 days of age.

The D40's and the "Grouped" treatment (G40, G60, G80 and G100) are the experimentals which were grouped before the suggested time of sexual maturation, and their testes weights are the only ones which are significantly altered by the experimental treatment when compared to appropriate controls.

The causes of the lower testes weights of the grouped males is not known. Several studies suggest that intraspecific competition within groups may alter endocrine mechanisms and result in lower reproductive organ weights (Christian, 1959, 1964, Christian, Lloyd and Davis, 1965).

Grouping also produced changes in the vesicular gland weights which were in most respects similar to those of the testes. The vesicular glands of the "Grouped" treatment were all significantly lighter than controls and the "Delayed Grouping" treatment resulted in lighter vesicular glands in those mice grouped before sexual maturity - at 21 and 40 days of age. Those grouped after this time were actually heavier than controls killed at 100 days of age, but this difference was not significant at the $P = .05$ level.

Not all comparisons of vesicular gland weights are similar to those of the testes. One difference is that the vesicular glands did not increase in weight as rapidly as the testes did. This is easily seen by comparing Figures 2 and 4. Jameson (1950) reported that the vesicular glands of P. maniculatus do not begin to grow until the testes have almost reached their mature size. This time lag is probably due to a dependance of the vesicular glands on androgens produced by the testes (Turner, 1966).

A second difference with respect to the vesicular gland weights is that they are more variable than the weights of the testes. Due to this fact a non-parametric test was used for comparison of these organs. Others have reported that grouping house mice resulted in significantly lower vesicular gland weights than controls, but the weights of the grouped house mice were also variable (Christian, 1955a, Southwick and Bland, 1959).

A third difference in the response to grouping of the vesicular glands compared to the testes response is that mice grouped at 40 days of age (D40) had significantly lighter vesicular glands than those grouped at 21 days of age (G100), even though the G100's were grouped for a longer time. The testes weights of the same animals were not statistically different. The reason for this difference is not clear.

The weights of the adrenal glands of animals in this experiment were not significantly different from controls nor were they correlated with changes in the reproductive organs, with the exception of the G40's. These mice, which were grouped at 21 days and killed at 40 days of age (G40), had significantly lighter testes and vesicular glands, significantly heavier absolute and relative adrenal weights, and significantly lighter body weights than controls killed at the same age. This was the only one of seven experimental treatments ("Grouped" and "Delayed Grouping") that showed all these differences, and their meaning is not clear. The failure of grouping to produce significant adrenal hypertrophy in deermice is in general agreement with the studies of Bronson (1963) and Bronson and Eleftheriou (1963).

The relationship of specific behavioral responses to the results found in this experiment is not known. Behavioral data taken before these mice were sacrificed might provide some answers to this question, but these have not yet been analyzed.

It is interesting to note that Christian's papers show that grouping albino house mice at approximately 40 to 45 days of age (the exact age is not given) for a period of one week did not always result in lower testes weights of the grouped mice (Christian, 1955a, 1955b, 1955c, 1959). A similar experiment by Southwick and Bland (1959) resulted in no significant changes in testes weights. The results of the present study suggest that the inconsistency of testes response to grouping reported by these authors may have been due to grouping at variable intervals with respect to the time of sexual maturation.

The results of the present study may have some relevance to the dynamics of natural populations of deermice. The growth phases of populations are characterized by having a high proportion of young animals while declining phases have fewer young. The higher relative densities of growing populations may result in similar inhibition of the reproductive systems of immature males as found in the present experiment and perhaps result in lower reproductive function for these animals. If this happens reproduction may drop, and the population may cease growing or decline dependant upon factors producing mortality. Similar results have occurred in freely growing laboratory populations of P. m. bairdii. Significantly fewer females (6.1%) born into these populations became pregnant compared with controls (83.3%) (Terman, 1965). Further, males born into these populations had significantly lighter testes and vesicular glands when compared to controls (Terman, 1963), even though these mice were over 90 days of age.

It is recognized that the above mechanism may not be the only one which would curb or control population numbers in nature. The physical and biotic environment make a complex matrix of factors, most of which may act under specific circumstances to influence population dynamics. The affect of grouping on the reproductive development of males as demonstrated in the present experiment reflect the actions of social factors intrinsic to the population environment. In nature, such factors may contribute directly to population control through reproductive effects or indirectly by triggering dispersal and/or increasing vulnerability to factors producing mortality. The importance of social behavior or intraspecific competition as related to the physical environment and population has been suggested by several workers (Anderson, 1961, Christian, 1959, Nicholson, 1957, Terman, 1965).

The data obtained in the present experiment focus attention on the importance of the age of the animals at the time of exposure to grouping or crowding influence. The studies of several workers (Anderson, 1961, Howard, 1960, Nicholson, 1941, Sheppe, 1966) suggest that the age interval between weaning and sexual maturity is the time of movement and home establishment of young Peromyscus. Environmental pressures during this age interval may have similar affects under natural conditions as in the present experiment and may produce reproductive inhibitiom of indefinite duration. Such inhibition irrespective of the cause would have profound effects on the dynamics of natural populations.

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