# Weights of Testes, Thymi, and Accessory Reproductive Glands in Relation to Rank in Paired and Grouped House Mice (*Mus musculus*)[1] (35503)

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Previous work indicates that the weights of testes and of accessory reproductive organs may be sensitive indicators of alterations in androgen activity associated with changes in social pressure that occur in increasing populations, as well as with differences in rank of individuals (1, 2). Considerable work has been reviewed implicating androgens as a major factor in inter-male aggression (3). However, previous studies have been principally concerned with effects of injected androgens in castrates or the effects of encounters between paired males on androgen levels as measured by changes in weights of accessory reproductive organs. In examining mechanisms regulating population growth it has been hypothesized that long-term increases in social pressure lead to chronic changes in androgen activity that are reflected in weights of accessory reproductive organs. These changes vary among individuals within a given population and it has been further hypothesized that these individual responses of population social pressure also reflect the relative positions of individual animals in the social structure (2). Operating from the basis of these hypotheses the present study was designed to compare the effects of short-term exposure of paired animals with the effects of long-term exposure of animals in groups on testicular size and reproductive accessory organs in animals of different rank.

*Materials and Methods. I. Paired encounters.* One hundred and twenty male house mice (*Mus musculus*) were isolated at 21 days of age. When the mice had attained the age of 70 days, each mouse was placed for 10 min in a strange cage with one other mouse, making 60 paired encounters. Observations of behavior were made to determine "winners," "losers," and "draws." A mouse was designated a "winner" if it attacked and bit the other mouse and restricted the movement of the other mouse by threats and attacks. Mice that were recipients of repeated attacks and that fled without attempting to bite their attackers were designated as "losers." Mice in paired encounters in which neither mouse attacked the other, in which both mice totally withdrew from each other or in which each mouse equally threatened the other or equally attacked the other were designated as "draws." After 10 min had elapsed from the placing of the mice together, both mice were withdrawn and immediately sacrificed, weighed, eviscerated, and placed in 10% formalin. After fixation, testes, seminal vesicles, preputials, and thymi were dissected and weighed on a Roller-Smith torsion balance. Comparisons of the weights of the various organs were made between the winners, losers, and draws. Fisher's $t$ test of significance was used to analyze for differences among the organ weights. Differences having probability values of 5% or less were considered significant.

*II. Groups.* One hundred and fifty six male house mice (*Mus musculus*) were isolated at 21 days of age. At 70 days of age the mice were placed in groups of six mice each in cages 43.18 × 58.42 cm for a period of 3 weeks. At the end of the 3-week period the groups were deprived of food for 12 hr. The mice from each group were then placed in a strange observation cage of the same dimensions as the one in which they had been

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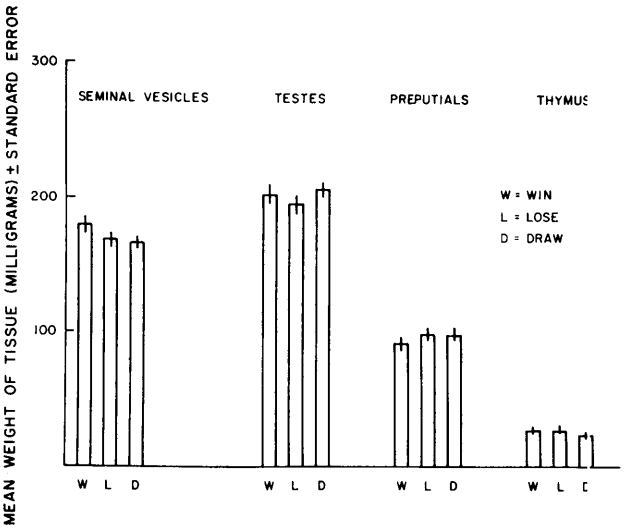


FIG. 1. Mean weights of seminal vesicles, testes, preputials, and thymi from mice in brief paired encounters.

grouped. A single food pellet was placed in the center of the cage. Observations of the groups were made to determine the rank of individuals. The "A" rank was assigned to the mouse that attacked and chased all other mice in the group and who successfully took the food pellet from all other mice. After the "A" mouse was determined, he was removed from the cage and observations continued to determine the "B" mouse. The "B" rank was assigned to the mouse that dominated by attacking and chasing the members of the remaining group. After the "B" mouse was designated he was removed from the group and subsequent ranks of C, D, and E were assigned to individual mice. The mice were sacrificed immediately after their removal from the groups and the same procedures of fixation, dissection, weighing, and statistical use of the Fisher *t* test were employed as outlined for the mice from the paired encounters above. The Spearman rank correlation coefficient (4) was also calculated for the organ weights.

*Results. I. Paired encounters.* There were no significant differences in the weights of any of the organs among the winners, losers, and draws (Fig. 1).

*II. Groups.* In a number of the groups of six males, one, or in a few instances, as many as three mice were dead at the end of the 3-week period. In groups in which all of the original six mice remained, it was not possible to separate the last two mice into different ranks. Hence, the remaining two mice were both assigned to rank of E. Thus, for purposes of comparing weights of organs only five ranks (A, B, C, D, and E) were employed. Although it was not initially intended as a measure of rank, it happened that the "A" mice in each group were the only ones in their respective groups that had tails that were unscarred and unstunted from being bitten.

In using the Fisher *t* test for analysis of the data, rank A mice differed significantly from mice of all other ranks for all of the measurements made with three exceptions, *i.e.*, rank A mice did not differ significantly from rank B mice with respect to weights of seminal vesicles or thymi and rank A mice did not differ significantly from rank D mice with respect to seminal vesicle weight (Fig. 2). Mice of ranks B through E did not differ from each other with respect to any of the measurements made. Spearman rank correlation coefficients for the organ weights and accessory glands were as follows: seminal vesicles, 0.90; preputials, 0.90; testes, 0.30; and thymi, 0.95.

*Discussion.* A number of studies have well established the role of androgens in aggressive behavior (5–8). In these studies, initiation of spontaneous aggression, frequency

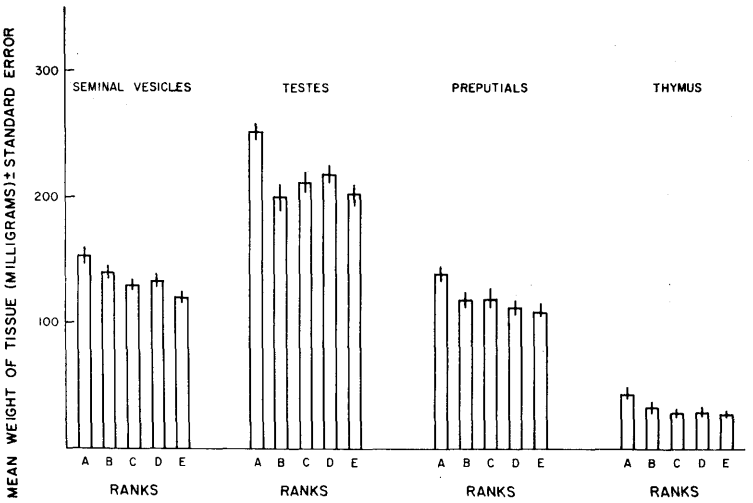


FIG. 2. Mean weights of seminal vesicles, testes, preputials, and thymi from grouped mice of different ranks.

and intensity of aggression have been shown to be directly influenced by levels of androgens. In the present study, using morphological criteria as indices of androgen activity, a direct relationship between rank and physiological response was observed, in that dominant animals in each group were clearly distinguishable from other members of the group, and in that there were also high levels of correlation with rank and weights of seminal vesicles, preputials and thymuses; the weights of each of these descending with decreasing rank. These results support earlier proposed concepts that endocrine feedback mechanisms operating in grouped animals and in populations at high density have differential effects on individual animals depending on the relative position of individuals in the hierarchy (1, 2). The results of the present study also support concepts of the presence of long-term chronic physiological effects on animals in groups (2). The contrasting absence of differences among organ weights of winners, losers, and draws in brief single-paired encounters in the present study suggests that care should be taken in dismissing the role of behavior in physiological processes on the basis of simple dominance–subordinate relationships that are not sustained over a period of time. In mice subjected to repeated brief encounters to fighter mice, decreased seminal vesicle weights were observed in the exposed mice as the number of encounters increased (9). The preputial gland has also been implicated as having a role in aggression; however, the relationship of preputial weight to rank was not clear (10). In the present study the preputial weights also served to separate dominant animals from others in their group. It is concluded from the results of this study that rank plays an important role in determining androgen secretion in groups of mice. Further work is necessary to assess the manner in which rank influences androgen levels and to determine how androgen activity operates to reenforce social dominance.

*Summary.* Indices of androgen activity in the form of reproductive organ and sexual accessory weights were correlated with winners, losers, and draws in brief paired encounters between male house mice (*Mus musculus*) and in mice from groups of six mice each that were housed together for 3 weeks. There were no significant differences in any of the parameters measured from the mice in brief paired encounters. Among the grouped mice the dominant mouse differed from almost all of the mice from other ranks with respect to testes, seminal vesicle, preputial, and thymus gland weights. Using an ordinal correlation coefficient, weights of sem-

inal vesicles, preputials, and thymus glands correlated highly with rank as organ weights decreased in mice of descending rank.

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