THE EFFECTS OF CROWDING UPON SOME ADRENAL RESPONSES IN MALE WHITE-FOOTED MICE (PEROMYSCUS LEUCOPUS)

By

JAMES STEVEN WALKER II

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Oklahoma State University

Stillwater, Oklahoma

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Thesis Approved:

Bryan P. Glass
Thesis Adviser

W. S. Newcomer

Milton R. Curd

N N Durham
Dean of the Graduate College

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CHAPTER I

INTRODUCTION

It is generally assumed that mammalian population growth is regulated directly by the availability of essential environmental necessities, such as food and shelter. If this assumption is correct, a mammalian population would not stop growing until its food supply and shelter were exhausted. Critical studies, however, show that many populations of rodents cease growing or decline in spite of adequate supplies of food, shelter, and other necessities (Davis, 1953; Clarke, 1955; Southwick, 1955a, 1955b; Christian, 1955b, 1956, 1959b; Louch, 1956; Lidicker, 1965; Terman, 1965a). Usually the subordinate animals are forced competitively into disadvantageous situations with regard to obtaining food or to being more subject to predation (Errington, 1943, 1954; Calhoun, 1948, 1949, 1950; Davis, 1953; Frank, 1953; Strecker and Emlen, 1953; Strecker, 1954; Christian and LeMunyan, 1958; Les, 1968; Glickman and Morrison, 1969). Christian (1959a) asserted that disease may be a factor in increasing mortality, but only because it is usually secondary to other factors, such as diminished resistance (Retzlaff, 1938; Frank, 1953; Chitty, 1954).

Populations of rats can be reduced by decreasing the capacity of the habitat in terms of food and cover, but starvation does not occur (Davis, 1953). These declines are affected by competitive situations which decrease infant survival and increase mortality of the subordinate

rats (Christian, 1961; Lidicker, 1965; Terman, 1965). Furthermore, most declines in numbers, once begun, continue to well below the point to which any lowering of the environmental capacity would have reduced them, and the rate of decrease is greater than would be expected (Christian, 1956, 1959b; Crowcraft and Rowe, 1957; Louch, 1958). These facts point to the existence of some underlying mechanism which regulates the size and growth of mammalian populations.

It seems that any explanation for the regulation of growth in natural populations would include an intrinsic adaptive system through which environmental factors would act. Social pressure in the form of behavioral competition is always present and is believed to be the key factor in regulating mammalian population growth, even though species vary considerably in their social organizations (Wynne-Edwards, 1962, 1965). It is thought that social interaction would regulate population development by eliciting physiological adaptive responses in some proportion to increased population density (Christian, 1950, 1955a, 1961, 1963a, 1963b, 1963c). Increased competition could stimulate pituitary-adrenocortical activity and decrease reproduction and resistance to disease in some proportion to density. These responses to social competition would be density-dependent and not geographically or specifically limted (Christian, 1959b). This feedback theory implies that environmental needs would act by increasing social competition.

Christian's (1950, 1963a) specific thesis was that this mechanism of endemic population growth limitation is intimately related to Selye's (1946) "General Adaptation Syndrome." This is illustrated in Figure 1. Christian (1950) suggested that wild animals in high populations experienced stress due mainly to the lack of food and cover, greater predation

pressure, and intraspecific strife. The alleged adaptive responses by individuals to these stresses were believed to bring about an overall lowering of resistance to disease as well as a lowering of the reproductive potential due to the exhaustion of the adreno-pituitary system.

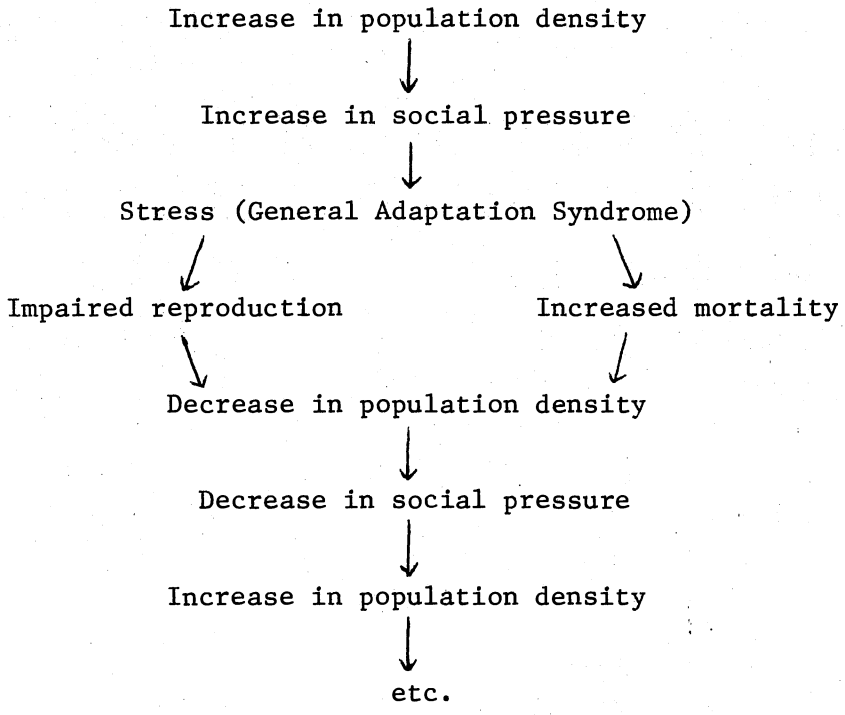

Figure 1. Christian's Stress Hypothesis (Christian, 1950)

Despite the large amount of material to support this hypothesis, there is still considerable uncertainty about its role and relative importance in the regulation of mammalian population growth, especially with regard to generalizing to a large number of species from the few species for which data are available. Therefore, the objective of this

investigation was to measure a variety of adrenal responses previously associated with stress as diagnostic and prognostic indicators of the effects of crowding upon male Peromyscus leucopus.

The responses which were measured were the percentages of: 1) eosinophils, 2) basophils, 3) lymphocytes, 4) monocytes, and 5) neutrophils; and 6) induced granuloma formation, 7) dried adrenal weights, and 8) plasma corticosterone levels.

The common white-footed mouse is abundant in woodlands or brushlands east of the Rockies. It has several characteristics not fully shown by the traditionally employed house mice (Mus musculus), Norway rats (Rattus norvegicus), deer mice (Peromyscus maniculatus), or meadow voles (Microtus spp.).

Namely, the degree of adrenocortical response to changes in population size depends upon the behavioral aggressiveness of the strain or species. The house mouse is very aggressive, whereas the deer mouse is much less so. Population increases have much more effect on the adrenal of the former species than on the latter. However, the adrenal indices of the two species respond in a similar manner when subjected to trained fighters of their own species (Bronson and Eleftheriou, 1964; Terman, 1965a, 1965b).

This makes P. leucopus an interesting species for further comparative study. The most pertinent qualities of P. leucopus for studies of this type are as follows:

1) It adapts quickly to captivity and is easily handled in the laboratory (Southwick, 1964).

2) It maintains excellent health and breeds well under standard husbandry procedures (Southwick, 1964).

3) Individuals tolerate very high population densities if they have been caged together since weaning (Southwick, 1964).

4) Conversely, adults that have been caged separately and as social strangers are highly intolerant of each other; they fight severely and show marked disruption of normal behavior and health (Southwick, 1964).

CHAPTER II

REVIEW OF LITERATURE

Introduction

It is appropriate first to discuss the stimuli which are known to elicit endocrine adaptive responses. These environmental changes, whether physical, chemical, thermal, or sociopsychological in nature, have typical dose-time-response relationships: the longer the stimulation, the greater the response (Sayers and Sayers, 1949). However, it should not be assumed that all adaptive responses are qualitatively similar. Some stimuli may elicit quite similar responses in kind and degree, whereas others may be manifested in different ways. All have in common that, if uncompensated, they will produce widespread physiologic changes which come under the heading of shock (Selye, 1950). However, whether or not the symptoms commonly associated with shock are elicited depends on the severity of the stimulus (Selye, 1950). It is not until these stimuli reach rather serious proportions and evoke marked responses that one considers the animal to be subject to "stress."

The variety of stimuli which have been found to elicit adaptive reactions is great (Selye, 1950). Nonetheless, it should be noted that emotional stimuli of a chronic nature can produce profound long-term physiologic reactions which may have widespread effects on the subject (Elmadjian *et al.*, 1958; Ratcliffe and Cronin, 1959; Christian, 1959b;

Mason, 1959; Hepworth, 1966; Bronson and Eleftheriou, 1964; Friedman et al., 1969; Plaut et al., 1969; Friedman and Glasglow, 1973). Social competition, social pressure, and chronic anxiety have been shown to produce marked physiologic responses (Clarke, 1955; Christian, 1959a, 1959b, 1963a; Mason, 1959; Louch and Higginbotham, 1967; Les, 1968; Plaut et al., 1969; Riegle, 1973).

Role of the Adrenal Cortex in Stress

The adrenalectomized animal or adrenal-insufficient individual has little ability to tolerate changes in the internal and external environment, such as cold, heat, infection, or prolonged exercise. The resistance to environmental change in such subjects is very low, and as a result of the stimulus they will die, whereas the normal individual can tolerate a greater stimulus. Selye (1956) has shown that when an animal is subjected to a severe but sublethal injury it reacts in a sterotyped manner. This reaction is triphasic and is referred to as the general adaptation syndrome (Figure 2). In Selye's (1956) own words, the general adaptation syndrome is the "sum of all non-specific systematic reactions of the body which ensue upon long-continued exposure to stress" (p. 64).

The first stage, which occurs immediately upon exposure to an injurious agent, is referred to as the alarm reaction. In Selye's (1956) words, this is the "sum of all non-specific systematic reactions elicited by sudden exposure to stimuli to which the organism is not adapted" (p. 64). The alarm reaction stage includes a phenomenon referred to as "shock." Shock is characterized by changes which are similar to those seen in acute adrenal insufficiency. A countershock

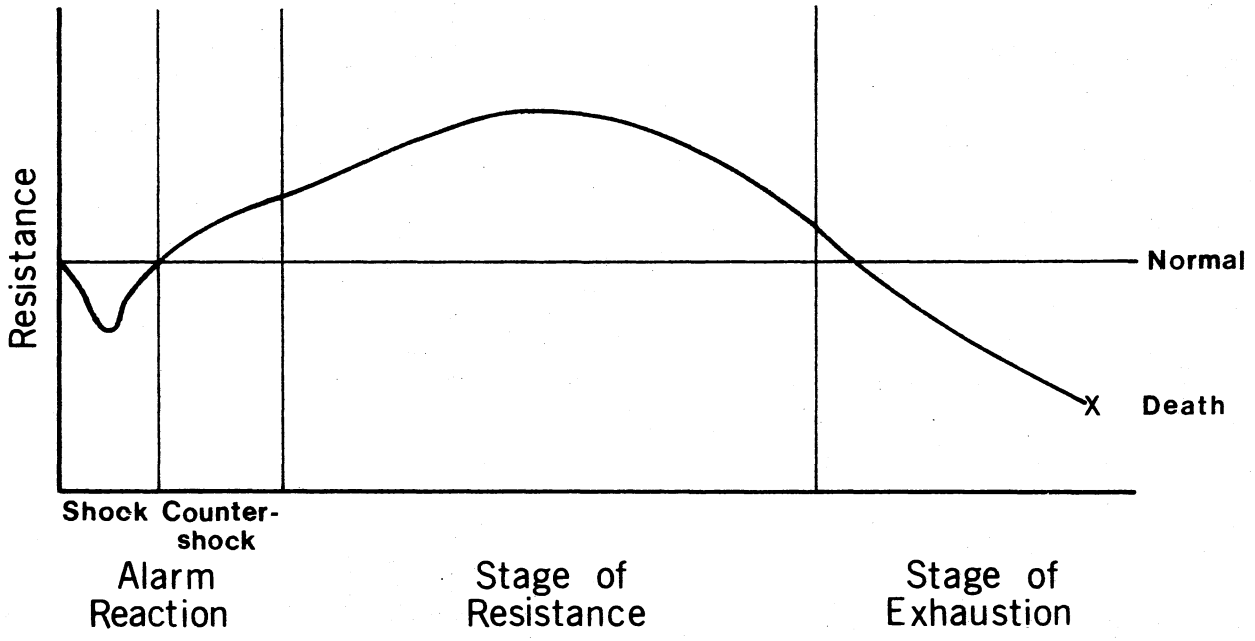

Figure 2. Changes in Resistance to a Specific Stressor in Different Phases of the General Adaptation Syndrome, According to Selye (Gorbman and Bern, 1962:331)

phase follows during which changes similar to those following corticoid administration occur. If the same stimulus is given to an adrenalectomized animal, the shock phase is extremely severe and the animals show no evidence of the countershock phase prior to death. However, survival can be attained if adrenal cortical extracts are given to the adrenalectomized animals. Thus, protection is afforded by adrenal steroids, and activation of the pituitary-adrenal axis must ensue.

If the organism continues to be exposed to this injurious influence, one notes an adaptation of the organism to the "stressor." This "stage of resistance" may continue for a long period of time. When the sustained injurious stimulus begins to have its full effect upon the organism, there is an apparent loss of adaptation, and the "stage of exhaustion" occurs. According to Selye's hypothesis, the prolonged excessive outpouring of critical hormones would play an etiological role in certain diseases. Yet, even though cortical hormones are essential for the full-blown manifestation of certain responses to stimuli, they need not be the causative agents of the responses. Thus, cortical hormones are required to support a normal response to a nonspecific stimulus and provide "permissive activity" (Ingle, 1952, 1954).

Indices of Adrenocortical Function

Lymphocyte Counts

After an alarm stimulus there is an immediate rise in the number of circulating lymphocytes, which is then followed by a characteristic lymphopenia and eventually a return to normal levels. The initial rise in circulating lymphocytes is at least in part due to the release of

epinephrine, but this release by no means accounts for the entire rise (Gordon, 1955). The prolonged release of lymphopenia which begins in the early stages of the increased pituitary-adrenocortical activity probably results from the destruction of medium and small-sized lymphocytes within the lymphatic organs by the corticoids (Gordon, 1955). However, there is also some reason to believe that lymphocytes may migrate to depot locations (Colfer, DeGroot and Harris 1950). Over a prolonged period of stress, the main cause for lymphopenia appears to be the carbohydrate-active corticoids involuting lymphoid tissue by producing degeneration and actual fragmentation of the lymphoid cells, inhibition of differentiation, and depression of lymphocytopoiesis (Selye, 1950; Dougherty, 1953; Santiesteban and Dougherty, 1954; Gordon, 1955; Weaver, 1955; Emel'yantsev and Sveshnikova, 1969). In addition, lymphocytolysis evidently serves to release a readily available store of amino acids and may serve to provide a flood of antibodies which are normally produced in the lymphoid tissues (Keuning et al., 1950; Dougherty, 1953; Kass et al., 1953; Sundberg, 1955).

Lymphocyte counts as criteria of stress in wild mammals are subject to one main criticism: the response is rapid enough and the counts labile enough so that there is real danger that counts may reflect alarming stimuli induced by handling, thus masking any other effects which may be the main point of the study.

Eosinophil Counts

The number of circulating eosinophils are beyond doubt diminished by adrenal factors and eosinophil counts are commonly used to assess the response of the adrenal cortex to ACTH or to indicate increased

pituitary-adrenocortical activity (Speiss and Meyer, 1949, 1951; Louch, Meyer and Emlen, 1953; Speirs, 1955; Hepworth, 1966; Tepperman, 1971). The reduction in eosinophils appears to result mainly from their increased destruction under the influences of cortical hormones (Essellier et al., 1954). A reduction in eosinophils can be affected by epinephrine as well as by cortical hormones, and there is evidence that the presence of cortical hormone is required for epinephrine to produce eosinopenia (Gordon, 1955). It is mainly with respect to whether or not cortical hormones are necessary for the eosinopenic action of epinephrine that the specificity of eosinopenia as an indicator of increased adrenocortical activity has been questioned. Nevertheless, if epinephrine produces a marked eosinopenia to the intact animal, eosinophil counts in mammals, especially wild, appear to have limited value. Fear resulting from handling, trapping, etc., not only would produce eosinopenia, but could produce it without necessarily having an increase in adrenocortical activity.

In a series of experiments, Southwick (1959) demonstrated that moving laboratory mice into a new environment daily could induce a marked increase in adrenocortical activity as determined by eosinophil counts. That the eosinopenia was not a result of handling was shown by the fact that mice handled in the same way, but not placed in a strange situation, responded with only a slight fall in circulating eosinophils. The mice transferred to new cages for a period of time every day adapted to the situation, as the eosinophil count returned to normal levels by the end of the eight-day experimental period. A third series of mice were placed in groups of four each day and these animals exhibited an 80 percent mean decline in their eosinophil counts and these counts

remained low as long as the mice were placed in groups. Presumably the mice responded to grouping with a marked increase in adrenocortical activity and did not adapt to the situation. It is clear from these results that merely placing laboratory mice in a stange situation is an emotional stimulus sufficient to result in a decline in circulating eosinophils and presumably in adrenocortical activity and that grouping constitutes a more profound stimulus to which these mammals failed to adjust.

Nevertheless, Louch (1958) used eosinophil counts to assess adrenocortical function in relation to changes in populations of voles and with adequate precautions the use of eosinophil counts for evaluating adrenocortical activity in natural populations seemed to have considerable value in these experiments. It was not possible to state definitely whether or not the declines in eosinophils were due to cortical or medullary hormones. However, in long-term studies with repeated counts, he concluded that there can be little doubt that eosinophil counts reflect adrenocortical rather than medullary functions (Louch, 1956). In studying three populations of voles, Louch (1956) found that there was a significant negative correlation between eosinophil counts and population density. But, the correlation between eosinophils and population density was not significant in the third population. The third population, however, never reached 30 animals in size. The published figures indicate a striking parallel between the rates of population growth and eosinophil counts for all three populations, and in all three populations there were significantly fewer circulating eosinophils when the populations of these voles were greater than 30 (Louch, 1956). The variability in Louch's data reflects to some

extent the difficulties in obtaining precise eosinophil counts, even though he was fully aware of these problems and took every step possible to avoid pitfalls (Louch et al., 1953; Louch, 1956). The problems involved in using eosinophil counts as indices of adrenocortical function have been discussed fully elsewhere (Thorn et al., 1953; Louch et al., 1953; Rosemberg et al., 1954; Speirs, 1955; Visscher and Halberg, 1955; Louch, 1956). Handling will cause an adrenal medullary and cortical discharge due to fear, excitement, and possibly rage in wild mammals, and therefore affect eosinophil counts (Southwick, 1959). Consequently, it is possible for variability to result from handling as well as by individual and unknown factors, unless appropriate precautions are taken (Louch, 1958).

Neutrophil Counts

A rise in circulating neutrophils accompanies increased pituitary-adrenocortical activity due to the inhibition of diapedesis. However, similar changes can be induced by so many factors that neutrophil counts are not very useful indices of such activity (Christian, 1963a).

Adrenal Weights

For depiction of chronic stress, adrenal weights are much more useful and much less subject to pitfalls than plasma corticosteroids, or circulating eosinophils or lymphocytes. All these measurements with the exception of adrenal weights are labile and reflect rapid changes in adrenal function, thus they may often reflect nothing more than the handling of an animal and may completely obscure the desired observations. Since adrenal weights vary from individual to individual in the

same species, it is necessary to obtain large enough samples to have a reliable criterion for evaluating adrenal changes in a population (Christian, 1963a).

Glucocorticoid Levels

The preceding indices are indirect assessments of adrenocortical function whereas glucocorticoid concentration is a direct measurement. In fact, the concentrations of glucocorticoids generate the preceding indirect indices, with the exception of adrenal weight. Glucocorticoids are secreted into the blood stream upon stimulation of the adrenal cortex by ACTH. As soon as a glucocorticoid, such as corticosterone, is synthesized it is released into the blood stream and transported both to the tissues on which it acts and to the brain and pituitary which regulate the rate of release of ACTH (Lissak and Endoczi, 1965). Physiological stress can enhance ACTH secretion as much as 20-fold within minutes (Eisenstein, 1967). The perception of stress is first transmitted to the perifornical area of the hypothalamus (Guillemin, 1974). This in turn transmits impulses into the median eminence where CRH is secreted into the primary capillary plexus of the hypophyseal portal system and then carried by this portal system to the sinuses of the adenohypophysis (Guillemin, 1974). CRH then excites the beta cells of the adenohypophysis to secrete ACTH, which in turn causes adrenal secretion of corticosterone (Branson, 1968).

Glucocorticoid effects on the blood and lymphoid tissues of the body have been indirectly described previously. The injection of a corticoadrenal hormone of the corticosterone type causes a very prompt decrease in the number of circulating eosinophils and lymphocytes.

Moreover, there is a marked decrease in the size of the lymphoid tissue mass of the body (Gordon, 1955; Dougherty, 1953).

An important group of glucocorticoid effects modify drastically the sequence of events involved in the inflammatory response and tissue repair. When a tissue is injured--mechanically, chemically, or by parasitic invasion--there is (1) an extravasation of fluid from the intravascular compartment into the tissue spaces, (2) an infiltration of the area with leukocytes, and (3) the beginning of the healing process which is characterized by the synthesis of connective tissues. Pharmacological doses of glucocorticoids inhibit all phases of this sequence (Sorensen, 1966).

Possible explanations for the preceding are as follows. First, it has been recently discovered that cortisol stabilizes the membranes of cellular lysosomes so that they rupture only with great difficulty (Weissman and Thomas, 1964). This stabilizing effect on the lysosome membrane could alone account for a major share of the anti-inflammatory effect of cortisol, because it would prevent the usual destruction of tissues that occurs in inflammation because of liberation of lysosomal enzymes. Second, cortisol decreases the formation of the powerful vasodilator bradykinin (Schachter, 1969). Bradykinin is split from an alpha globulin by the proteolytic enzyme kallikrein. It is possible that stabilization of the lysosome membrane prevents release of kallikrein into the cell from the lysosome. Third, cortisol decreases the permeability of the capillary membrane, and this is a significant factor in preventing the usual protein leakage into inflamed tissues (Eisenstein, 1967). Part of this decrease in capillary permeability might result from inhibition of bradykinin formation, because bradykinin

vasodilation tends to overstretch the capillary membrane (Luft, 1965).

Generalized Effects of the Endocrine Adaptive Responses

Inflammation and Granulation

The adrenocorticoids, cortisol, cortisone, hydrocortisone, and to a lesser extent corticosterone, exert powerful anti-inflammatory effects which stem from the suppression of growth of connective tissue, the depression of lymphocyte activity, and interference with the phagocytic process (Dougherty, 1953; Robinson and Smith, 1953; Thomas, 1953; Emel'yantsev and Sveshnikova, 1969; Riegle, 1973), but in addition they prevent the mobilization of all the usual elements of an inflammatory response around the site of an injury (Taubenhaus and Amromin, 1950; Dougherty, 1953; Robinson and Smith, 1953; Gordon, 1955; Dougherty and Schneebeli, 1955). Androgens and estrogens activate these anti-inflammatory responses. They decrease the destruction of fibroblasts and the invasion of polymorphonuclear leukocytes and macrophages (Zweifach, Grant and McCluskey, 1965). The appearance of epithelioid macrophages, giant cells, and formation of new fibroblasts and macrophages are suppressed by cortisone, either injected or implanted as pellets (Baker, 1954; Kemper et al., 1969). Dougherty and Schneebeli (1955) explained the inhibition of the inflammatory response around the site of injury in the following ways: when there is cellular injury, polypeptides are liberated locally which attract white blood cells, cause white cells to stick to the blood vessel walls in the area, produce local vasodilation, and increase capillary permeability (Menkin, 1955; Dougherty and Schneebeli, 1955; Macleod, 1970). Cortisone or cortisol suppress the

release of these polypeptides from the tissue (Macleod, 1970). Growth hormone, deoxycorticosterone, and aldosterone appear to exert a stimulating effect on inflammation and the development of granulation tissue (Selye, 1955; Dougherty and Schneebeli, 1955). These hormones, however, also inhibit the anti-inflammatory action of cortisone, hydrocortisone, and corticosterone and may enhance the local inflammatory response by increasing the local liberation of these polypeptides (Dougherty and Schneebeli, 1955; Macleod, 1970). In addition, there is some evidence that by stabilizing the lysosomal membranes glucocorticoids inhibit the breakdown of lysosomes that takes place in inflamed tissue (Macleod, 1970). Therefore, if there is a reduction in the secretion of growth hormone with a simultaneous increase in the production of ACTH and the adrenocortical steroids, there will not only be a direct suppression of the inflammatory response to infection or injury but also a withdrawal of the factors which ordinarily would stimulate such activity. The effects of various alarm stimuli or hormones on inflammation and granulation in the intact animal have been studied and measured by using experimental granulomas (Weier et al., 1950; Selye and Bois, 1954; Robert and Mezamis, 1957; Christian and Williamson, 1958). It has been shown that in addition to suppressing inflammation the carbohydrate-active corticoids also suppress the formation of granulation tissue and healing by preventing connective tissue growth.

Antibody Formation

Antibodies are formed mainly in the lymphatic tissues (Keuning and Van der Slikke, 1950; Kass et al., 1953a; Kelsal and Crabb, 1958; Zweifach, Grant and McCluskey, 1965). One school of thought maintains

that this function resides primarily in the plasma cells while another school of thought maintains that lymphatic cells in general are capable of manufacturing antibodies (Keuning and Van der Slikke, 1950; Dougherty, 1953; Kelsall and Crabb, 1958; Zweifach, Grant and McCluskey, 1965). It is not the intention of this literature review to enter this controversy, but it seems relatively certain that the lymphoid tissues are primarily responsible for the production of antibodies. A variety of experiments have shown that injected corticoids or the increased secretion of endogenous corticoids, brought about either by injected ACTH or in response to alarming stimuli, markedly suppress the formation of antibodies (Kass et al., 1953a; Zweifach, Grant and McCluskey, 1965). Protein manufacture, and therefore the formation of antibodies, requires the presence of nucleic acids in the cells, especially in the cytoplasm, and antibody formation is normally associated with an increase in nucleic acid content of the lymph organs (Kass et al., 1953a; Kelsall and Crabb, 1958). It has been shown that in addition to actually destroying lymphoid tissue, the glucocorticoids reduce the PNA content of the remaining lymphatic cells (Kelsall and Crabb, 1958). The ability of the reticuloendothelial system to dispose of phagocytized particulate material is also impaired (Thomas, 1953) even though phagocytosis may be stimulated (Gordon and Katsh, 1949; Thomas, 1953). By these several mechanisms the production of antibodies may be seriously impaired following the activation of the adrenal cortex although there appears to be a gradient response relationship (Dougherty, 1953; Dougherty and Schneebeli, 1955).

Resistance to Infection

The two immediately preceding topics deal with factors involved in the resistance to infection. It stands to reason that reducing the inflammatory response and depressing the formation of antibodies will inevitably impair the ability of an animal to resist infection. Cortisone, hydrocortisone, and ACTH have been shown to decrease resistance to a variety of experimental infections caused by numerous infectious agents including streptococcal, pneumococcal, tuberculosis infections in mice, rats and guinea pigs, brucellosis, malaria in monkeys, and others (Selye, 1951; Kligman et al., 1951; Schmidt and Squires, 1951; Schwartzman, 1952; Kass et al., 1953b; Le Maistre et al., 1953; Weinstein, 1953; Robinson and Smith, 1953; Cross, 1960; Parker, 1961; Ogilvie, 1965; Beisel, 1969; Miller and Lowell, 1969; Plaut et al., 1969; Bamgbose and James, 1969; Harley and Gallicchio, 1970). The pathogenicity of various agents has been increased by cortisone activity. For example, the virulence of Coxsackie infections in mice was greatly enhanced by cortisone (Boring et al., 1955). Also, poliomyelitis can be made a paralytic disease in the normally resistant hamster by cortisone or hydrocortisone (Schwartzman and Aronson, 1953). Viremias may likewise be prolonged appreciably by the adrenal corticoids (Whitney and Anigstein, 1953; Pollard and Wilson, 1955).

The influence of cortisone on helminthis infections has been studied by many investigators. Stoner and Godwin (1953) found that cortisone increases susceptibility of mice to trichina infections. Coker (1955) reported that cortisone suppressed the cellular response in the intestinal wall of mice infected with Trichinella spiralis, and that hormone treatment resulted in a longer persistance of more adult

worms in the intestine and the establishment of more larvae in the musculature. Weinstein (1955) found that cortisone administered to rats during or after immunization to Nippostrongylus muris caused a marked reduction in the cellular response in the skin to a challenging infection of infective larvae. Roman (1956) has also shown that adult mice usually resistant to Strongylus ratti could become infected if they were treated with cortisone. Olson (1957) and Briggs (1958), in their studies with Litomosoides carinii in an abnormal host, have also demonstrated that cortisone treatment resulted in a decrease in the number of parasites encapsulated in the pleural cavity. Davis and Read (1958) revealed that infection by Trichinella spiralis increased 20 percent in grouped mice compared to individual mice. Ogilvie (1965) found that daily treatment of rats with prednisolone during infection suppressed the initiation of acquired resistance to Nippostrongylus brasiliensis completely.

Whenever experiments with injected hormones are considered, the question arises whether or not the same events may occur as a result of increased endogenous secretion of the same or similar hormones. A criticism frequently made of experiments with exogenous hormones, especially with large doses, is that the results are pharmacologic rather than physiologic. However, it is by using isolated hormones in using isolated hormones in highly controlled situations that an understanding of the mechanisms is gained. Nevertheless, before one can extrapolate from these data to natural events, comparable changes must be shown to occur in natural or semi-natural conditions. Changes in host resistance may result from adverse environmental stimuli, probably as a result of adrenocortical activity. It has long been common knowledge that excess

fatigue, chilling, and a variety of comparable stimuli increase the susceptibility of humans to colds and other infections. It should be apparent that most of these same stimuli also increase the secretion of adrenocortical steroids. Commonly accepted truisms still do not constitute experimental evidence and proof of such conclusions, yet several experiments have shown that host resistance is decreased by exposing the animals to stimuli which are known to increase adrenocortical activity. When mice are exposed to 4° C. for a period of time, Coxsackie infections become much more pathogenic (Boring et al., 1956). Diminished resistance of mice to trichinosis and to tuberculosis have been demonstrated by procedures which also produce increased pituitary-adrenocortical activity and depress inflammation and granulation (Tobach and Block, 1956; Davis and Reed, 1958; Christian and Williamson, 1958).

CHAPTER III

MATERIALS AND METHODS

Materials

Standard Solutions

Corticosterone (11β, 21-hydroxypregn-4-ene-3,20,dione) obtained from Sigma Chemicals, was dissolved in ethanol at a concentration of 1 mg/l and refrigerated at 5° C until use. Standards contained 0.2, 0.4, 0.6, 0.8, and 1.0 µg corticosterone/ml distilled water along with a distilled water blank.

Reagents

The 2,2,4-trimethyl pentane, dichloromethane, and methanol which were used were of "nanograde" quality (Mallinckrodt Chemical Works). Ethanol was 100 percent analytical grade (U. S. Industrial Chemical Company). Water used in the assay was distilled and deionized. Concentrated hydrochloric and sulfuric acids were analytical grade (Baker Chemical Company).

Glassware

Glassware was rinsed with tap water before use, soaked in detergent (Sparkleen, Fisher Chemical Company), rinsed, and placed in a sulfuric acid-potassium dichromate cleaning solution overnight. Following seven

to eight rinses in tap water the glassware was resoaked in detergent, rinsed again with tap water and placed in concentrated hydrochloric acid overnight. After being rinsed seven or eight times each with tap water, deionized water, and distilled water, the glassware was finally rinsed two or three times with nanograde methanol and air dried. The microcuvettes used in the fluorometric determination were soaked in concentrated hydrochloric acid overnight, rinsed with tap water, deionized water, distilled water, methanol, and air dried.

Methods

General Procedures

The mice utilized throughout the study were male white-footed mice (Peromyscus leucopus) live-trapped in Payne County, Oklahoma from 3 June 1974 to 22 January 1975. In the laboratory, the mice were isolated in individual cages for a minimum of 15 days. During the isolation period there was no physical or visual contact between mice. Olfactory and auditory contact were not controlled. At the end of the isolation period, a cotton pellet wet with turpentine was implanted subcutaneously in the mice. Then the mice were placed into experimental cages (27.9 x 22.6 x 30.5 cm) in groups of 1, 2, 4, 8 and 16 animals per cage for a period of 20 days. These groups will be referred to in the rest of the study as groups 1, 2, 4, 8, and 16; thus, the group's designation is also the number of animals in the group. The mice were subjected to a 14-hour light and 10-hour dark cycle (lights on from 0700 to 2100 hours) and the temperature was maintained at 22°C$\pm$2°. Food and water were available ad libitum.

On the 20th day social rank of either A, B, or W was subjectively assigned to each mouse. The assessment of social rank was based upon appearance (scars, torn ears, bobbed tails, lost toes) relative to other mice. Social rank A was designated the dominant individuals and was characterized by very few scars, if any, and a generally immaculate pelage. Social rank B denoted the subordinate mice and was typified by a few scars in the rump region, knicks on the tail, and a pelage that was not well-groomed. Individuals indicated by social rank W were super-subordinate and were delineated by bobbed tails, torn ears, lost toes, scars in the rump region, and a coarse mottled pelage. Between 1900 and 2100 hours, the animals were weighed to the nearest 0.1 gm and then sacrificed. At that time, a blood smear was made, blood was collected for corticosterone determination, the cotton pellet was removed, dried, and weighed, and the dry weight of the adrenal glands was determined for each mouse. A more extensive description of the procedures for the measured indices is as follows:

Blood Smear

Differential white counts of the stained blood smears were conducted in accordance with standard hematological procedures (Cartwright, 1958; Wintrobe, 1961; Schalm, 1975). The neutrophils, eosinophils, basophils, lymphocytes, and monocytes were enumerated until the leukocytes totaled 200.

Corticosterone Determination

Blood Collection. Blood was collected in heparinized test tubes following decapitation. Ramaley (1972) reports that serum corticosterone

was elevated only at 3 minutes after initial handling in Sprague-Dawley rats. Accordingly, samples were completed within 1 minute after the mouse was first handled. The plasma was removed after centrifugation at 1500 rpm (500 x g) for 15 minutes and stored at -20°C until use.

Assay for Plasma Corticosterone. Plasma corticosterone was determined in 100 µl samples of each plasma aliquot using a modification of the Silber (1958) method, developed by Akin (1972). In this procedure plasma samples are mixed with 2 ml of iso-octane (2,2,4-trimethyl pentane) and mixed on a vortex mixer for 30 seconds. After centrifugation for 10 minutes at 1500 rpm (500 x g), the iso-octane was removed by aspiration. Iso-octane removed lipids less polar than C-21 glucocorticoids.

The plasma was extracted with 6 ml of methylene chloride (dichloromethane) and then discarded following vortexing for 1 minute and centrifugation for 10 minutes. The remaining acidic and phenolic impurities as well as estrogens were removed by 1 ml of 0.1 N NaOH. The alkaline layer was aspirated after mixing on a vortex mixer for exactly 5 seconds and centrifugation. This step was executed quickly due to the instability of steroids in alkali for longe periods of time. Then a 7 ml aliquot of the methylene chloride extract was transferred to a test tube containing 0.5 ml of 70 percent ethanolic sulfuric acid (3:7, v:v). The contents were vortexed for 1 minute. After centrifugation the supernatent was discarded by aspiration.

Fluorescence was measured on an Aminco-Bowan Spectrophotoflurometer exactly 95 minutes after the addition of the alcohol acid mixture. The following settings were used to read the resulting fluorescence: (1)

excitation wave length of 472 nm through a 3 mm slit; (2) emission wave length of 524 nm through a 3 mm slit; and (3) turret setting 2. The index of precision (lambda) for the assay was 0.26.

The fluorometric technique used in this study is unable to discriminate between cortisol and corticosterone. However, since it has been shown by several workers (Bush, 1953; Hofman, 1956, 1957; Wilson et al., 1958; Bloch and Cohen, 1960; Cortes et al., 1963; Eleftheriou, 1964; Treiman and Levine, 1969; Ogunsa et al., 1971) that corticosterone is the major glucocorticoid present in rodents, and especially Peromyscus, with very little if any hydrocortisone detectable, the nonspecificity of the technique does not matter. In the event that some hydrocortisone is present, it should be noted that hydrocortisone and corticosterone have different intensities at the wave lengths used, corticosterone being about four times that of hydrocortisone.

Recovery of Stable Corticosterone. This procedure was used to test for loss of corticosterone other than that associated with the extraction step. A 100 µl qliquot of 30 µg/100 ml of stable corticosterone was added to an aliquot of pooled plasma which was then assayed.

$$\% \text{ Recovery} = \frac{\text{Total corticosterone in } \mu g/100 \text{ ml} - \text{Corticosterone in pooled plasma in } \mu g/100 \text{ ml}}{\text{Corticosterone added in } \mu g/100 \text{ ml}}$$

The test for percentage recovery of stable corticosterone was 96 percent (Table XV in the appendix). No correction was necessary for corticosterone values obtained experimentally in this study, since the recoveries approached 100 percent.

Induced Granuloma Formation

The technique utilizing subcutaneous cotton pellets wet with turpentine to induce granuloma formation was used in the present study (Christian and Williamson, 1958). Using aseptic procedures, a cotton pellet (No. 4 from Richmond Dental Cotton Company, North Carolina) 5 mg in weight was wet with turpentine, blotted, and inserted beneath the skin of an ether-anesthetized mouse through a mid-dorsal longitudinal incision. The incision was closed with a wound clip after insuring that the pellet was sufficiently far from the incision so that it would be neither sloughed nor involved in the reaction to the solution. Upon sacrifice of the mouse, the cotton pellet together with the accumulated granuloma was removed and weighed.

Adrenal Gland Weights

At the termination of the experiment, the adrenal glands were removed, cleaned of connective tissue, dried at 100°C for 24 hours and weighed.

Statistical Analysis of Data

Data were subjected to analysis of variance (Barr and Goodnight, 1972). Multiple comparison procedures could not be applied because each density-group did not possess relatively equal variation. The significant differences between means of social ranks and/or groups were detected by a Student's t-Test derived by Welch (1947). This test takes into consideration different population variances.

CHAPTER IV

RESULTS

A total of 267 male white-footed mice (Peromyscus leucopus) were subjected to the experimental treatment of crowding. Twenty-seven mice died during the crowding period; 6 of which were in group 8 and 21 were in group 16. It is noteworthy that the mice that failed to live the duration of the crowding period were either immature or juvenile. Twelve of the 27 mice were cannibalized. Thus the results were based on a total of 240 surviving mice. Forty-eight mice were reported for each density.

Behavior

It was observed that the males of social rank A were the victors of all social conflicts encountered and moved about the cage at will. The members of social rank B were generally confined to corners of the cage and exhibited "huddling." Mice of social rank B displayed very little overt aggression at the densities of 2 and 4 mice per cage, whereas, at the densities of 8 and 16 intensive social interaction was evident. In contrast, social rank W was always characterized by a substantial restriction of movement and being found on the sides of the cage. If a member of social rank W entered the floor of the cage, he was promptly driven away. Because of this forced emigration, the body weight for the super-subordinate mice was less than for the two other social ranks.

The general lack of aggression did not become overt until the densities of 8 and 16 mice per cage were reached. Groups 1, 2, and 4 were "socially compatible" in a comparative sense. It is worthy to note that the mice that did not live the duration of the crowding period died within two days after initial subjection.

Differential Leukocyte Count

Table I presents the means for the various types of total leukocytes expressed as a percentage.

Eosinophils

The eosinophil percentages remained relatively constant for the five groups (Figure 3) with a comparison of the group means failing to detect any significance. The percentages of eosinophils of mice in the same social rank but of different groups did not follow any trend (Figure 3). However, a generalized eosinopenia was observed within each group as a function of decreasing social rank with the exception of group 8.

Basophils

No significant differences were present between the group means of the mice. In addition, analysis of the differences between social rank means did not reveal any significance or trends (Figure 4).

Lymphocytes

The lymphocytes decreased in a linear fashion as group size increased from 2 through 16 animals per group (Figure 5). The mean

TABLE I

MEAN DIFFERENTIAL LEUKOCYTE PERCENTAGES AND STANDARD DEVIATIONS OF MALE *PEROMYSCUS LEUCOPUS* IN RELATION TO CROWDING AND SOCIAL RANK

Group	Social Rank	No.	Eosinophils	Basophils	Lymphocytes	Monocytes	Neutrophils
1	A	48	15.73± 9.53	0.52±0.69	36.76±15.70	10.36±5.34	36.52±18.15
2	A	24	24.33± 8.04	0.33±0.48	48.00±11.66	11.21±5.61	15.92±14.07
2	B	24	14.94±10.89	0.58±0.74	37.17±13.17	9.71±5.04	37.60±16.85
		48	19.63±10.59	0.46±0.63	42.58±13.47	10.46±5.33	26.76±18.88
4	A	12	25.96± 7.53	0.25±0.62	40.83±10.11	9.17±5.48	23.79±16.14
4	B	23	14.69± 7.35	0.37±0.64	41.02±14.48	9.00±4.48	35.59±17.27
4	W	13	6.54± 6.08	0.73±0.85	29.11±10.26	10.27±5.10	53.35±11.84
		48	15.30±10.64	0.44±0.72	37.75±13.28	9.38±4.99	37.45±20.24
8	A	6	18.42± 9.91	0.50±0.77	41.00±18.73	9.25±6.17	30.83±24.68
8	B	35	19.69±10.58	0.43±0.70	33.53±15.17	9.93±6.03	36.60±19.17
8	W	7	8.21± 7.83	0.71±0.90	24.28±15.95	5.86±0.10	60.93±16.72
		48	17.85±11.10	0.48±0.81	33.11±19.25	9.25±4.88	39.43±25.54
16	A	3	27.00±11.53	0.17±0.28	36.67±20.21	1.00±0.50	35.17±10.20
16	B	38	17.89±10.80	0.43±0.63	28.64±11.47	10.03±4.86	43.00±14.94
16	W	7	16.86± 7.26	0.29±0.75	28.21± 7.27	11.28±3.98	43.36±13.16
		48	18.31± 9.23	0.39±0.45	29.08±12.28	9.64±6.53	42.56±11.23

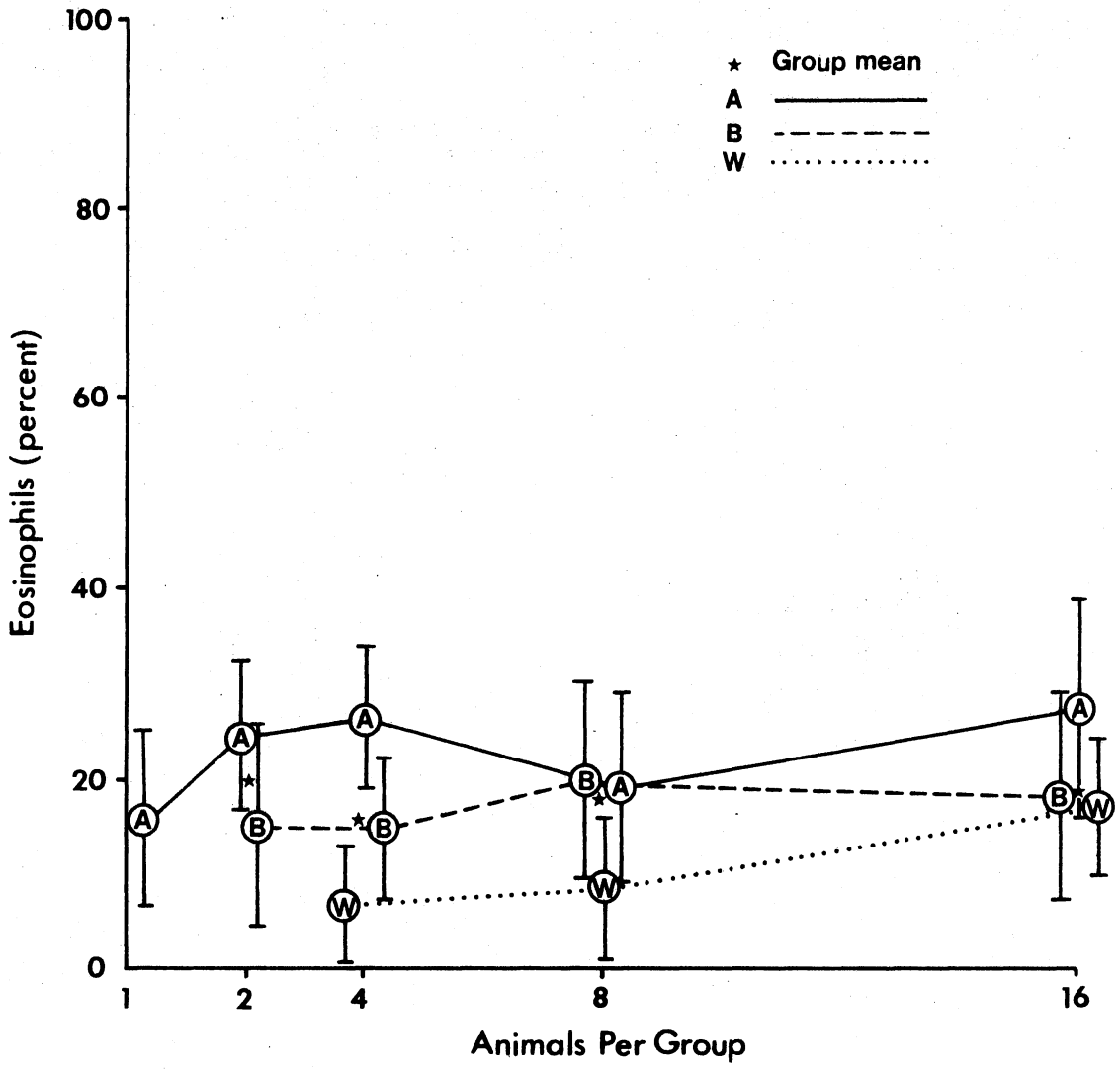

Figure 3. Mean Percentages of Eosinophils by Social Rank Plotted Against Group Size on a Logarithmic Scale

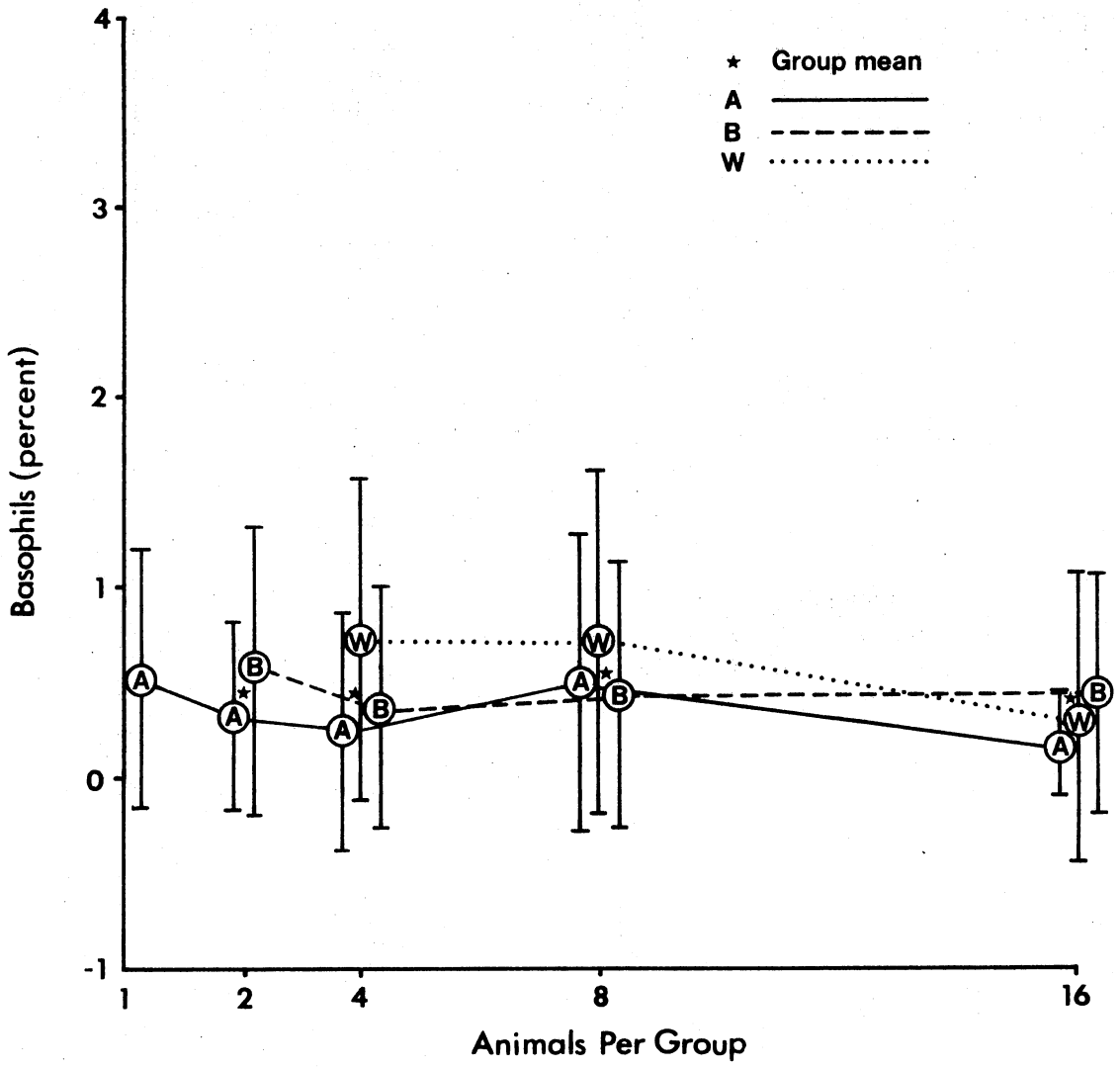

Figure 4. Mean Percentages of Basophils by Social Rank Plotted Against Group Size on a Logarithmic Scale

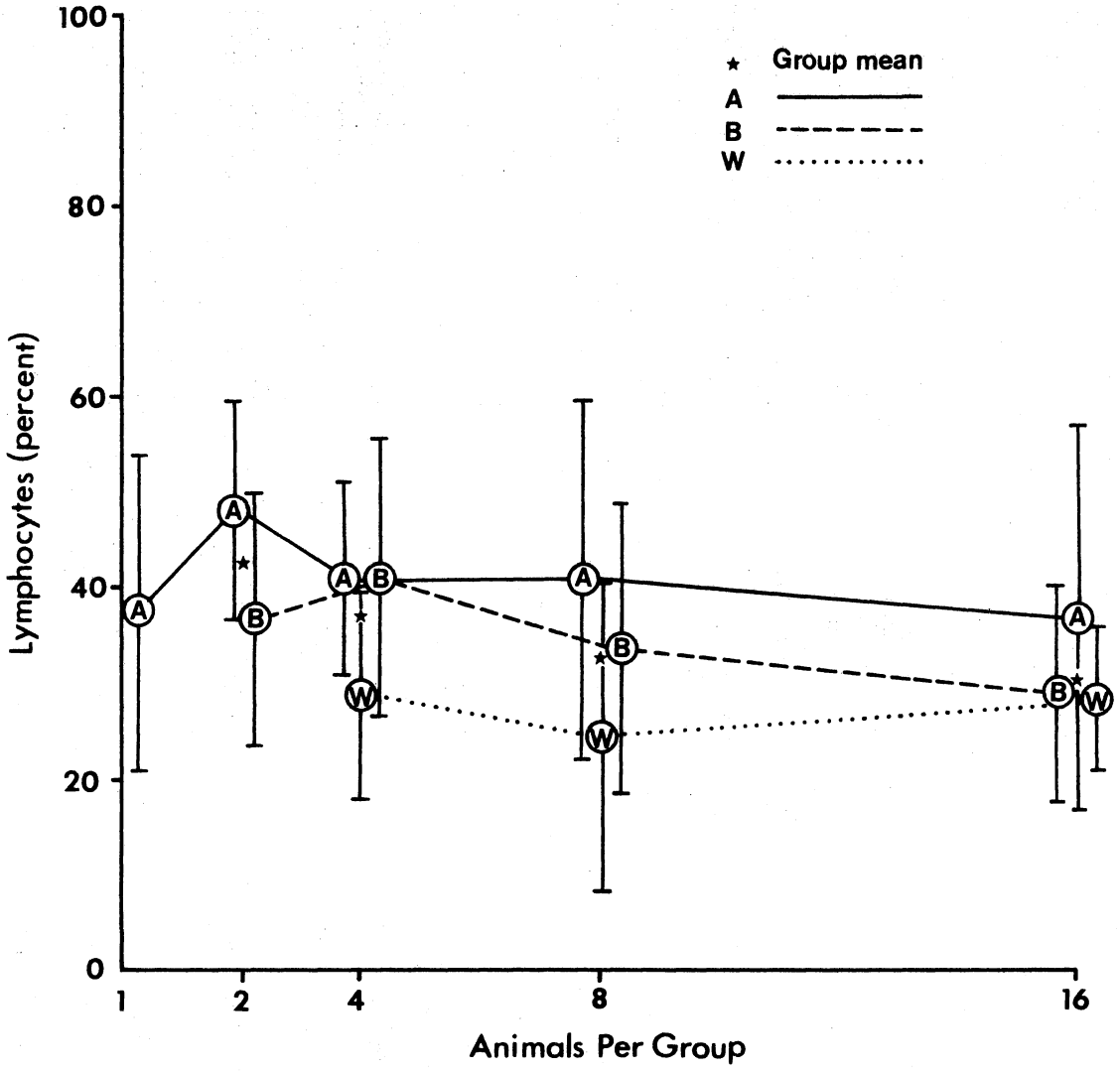

Figure 5. Mean Percentages of Lymphocytes by Social Rank Plotted Against Group Size on a Logarithmic Scale

lymphocytes percentages in groups 1, 2, 4, and 8 were comparatively the same (Table II, Figure 5). Significance at $P < .001$, $P < .01$, $P < .02$, and $P < .05$ occurred between groups 2 and 16, 4 and 16, and 2 and 8, respectively. The percentages between means within the same social rank but of differing groups remained constant except for the increase from 1_A to 2_A ($P < .01$). A generalized lymphopenia was evident within each group as a function of decreasing social rank exposing differences between 2_A and 2_B ($P < .01$) and 4_B vs. 4_W ($P < .01$).

TABLE II

COMPARISON OF DIFFERENCES IN MEAN LYMPHOCYTE PERCENTAGE AMONG ALL GROUPS (SIGNIFICANT P VALUES ARE INDICATED)

	Group 1 (36.76)	Group 2 (42.58)	Group 4 (37.75)	Group 8 (33.11)	Group 16 (29.08)
Group 16 (29.08)	7.68 P < .02	13.50 P < .001	8.67 P < .01	4.03 ns*	0
Group 8 (33.11)	3.65 ns*	9.47 P < .05	4.64 ns*		
Group 4 (37.75)	0.99 ns*	4.83 ns*	0		
Group 2 (42.58)	5.82 ns*	0			
Group 1 (36.76)	0				

*ns = non-significant P values.

Monocytes

Differences between the group monocyte means of mice were not observed. An examination of mice of the same social rank but of different groups did not disclose any trends, yet the mean for 16_A was exceptionally low. The presence of differences between social ranks within the same group was not characterized by any trends (Figure 6).

Neutrophils

The percentages of neutrophils for the various groups exhibited a marked decrease at group 2, followed by a gradual increase back to near basal levels. Groups 1, 4, 8, and 16 were comparatively equal (Table III, Figure 7). Significance at $P < .001$ occurred between groups 2 and 16. Significant differences at $P < .01$ existed between groups 2 and 8. Group 2 was significantly different from groups 1 and 4 at $P < .02$ (Table III). A generalized neutrophilia was observed as a function of decreasing social rank (Figure 7).

Induced Granuloma Formation

The effects of crowding and of relative social rank on the weights of experimentally induced granulomas in white-footed mice are summarized in Table IV. The mean granuloma weights of grups 2 and 4 were greater than those of groups 1, 8, and 16 (Figure 8). Group 1 was significantly different from groups 2 and 4 at $P < .001$ (Table V). Significance at $P < .05$ occurred between groups 4 and 8 (Table V). An examination of mice in the same social rank but in different groups revealed some interesting trends. Mice of social rank A tend to increase in almost a linear manner as group size increased from 1 through 16 animals per

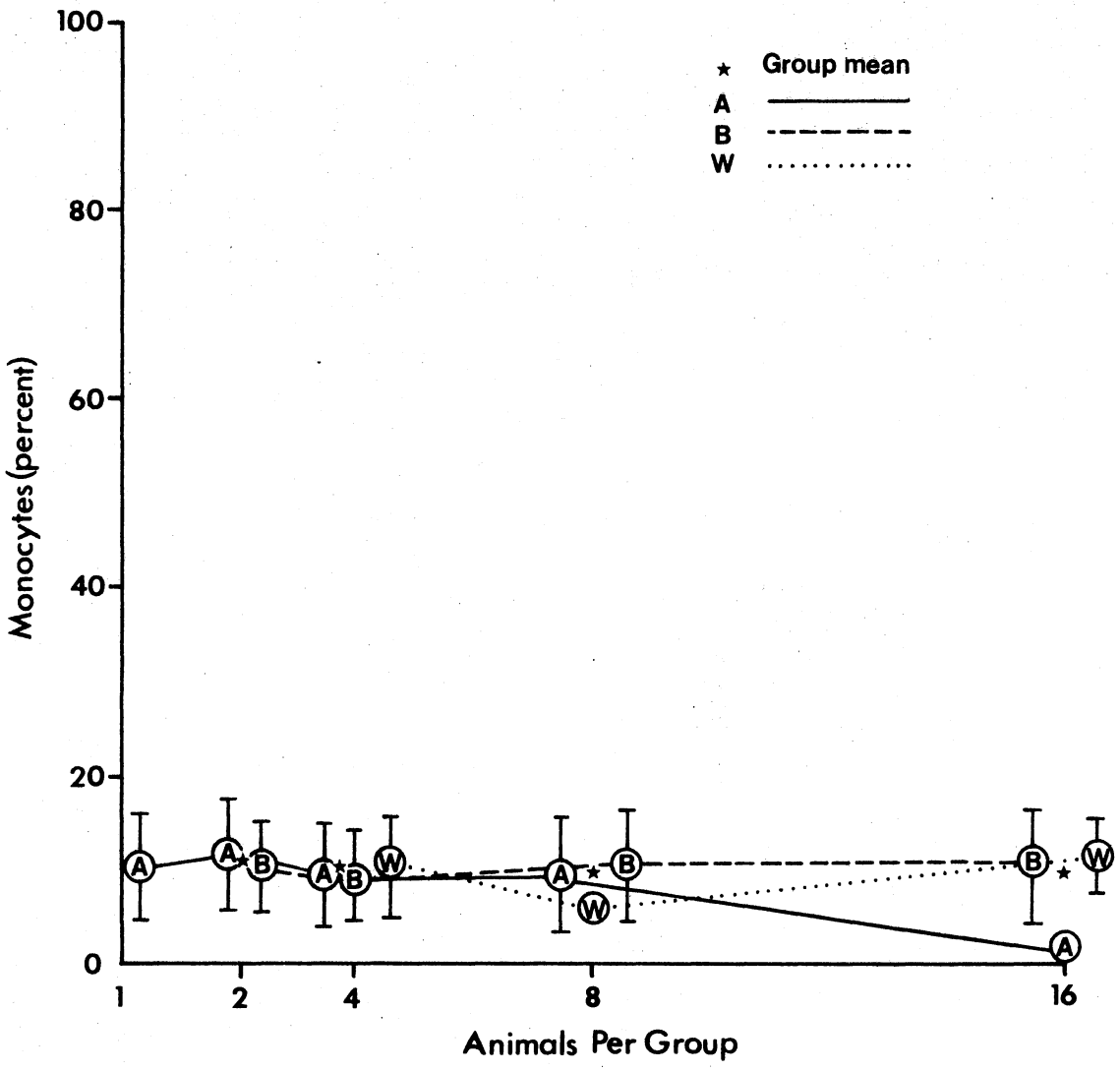

Figure 6. Mean Percentages of Monocytes by Social Rank Plotted Against Group Size on a Logarithmic Scale

TABLE III

COMPARISONS OF DIFFERENCES OF MEAN NEUTROPHIL PERCENTAGES AMONG ALL GROUPS (SIGNIFICANT P VALUES INDICATED)

	Group 1 (36.52)	Group 2 (26.76)	Group 4 (37.45)	Group 8 (39.43)	Group 16 (42.56)
Group 16 (42.56)	6.04 ns*	15.80 P < .001	5.11 ns*	3.13 ns*	0
Group 8 (39.43)	2.91 ns*	12.67 P < .01	1.98 ns*	0	
Group 4 (37.45)	0.93 ns*	10.69 P < .02	0		
Group 2 (26.76)	9.76 P < .02	0			
Group 1 (36.52)	0				

*ns = non-significant P values.

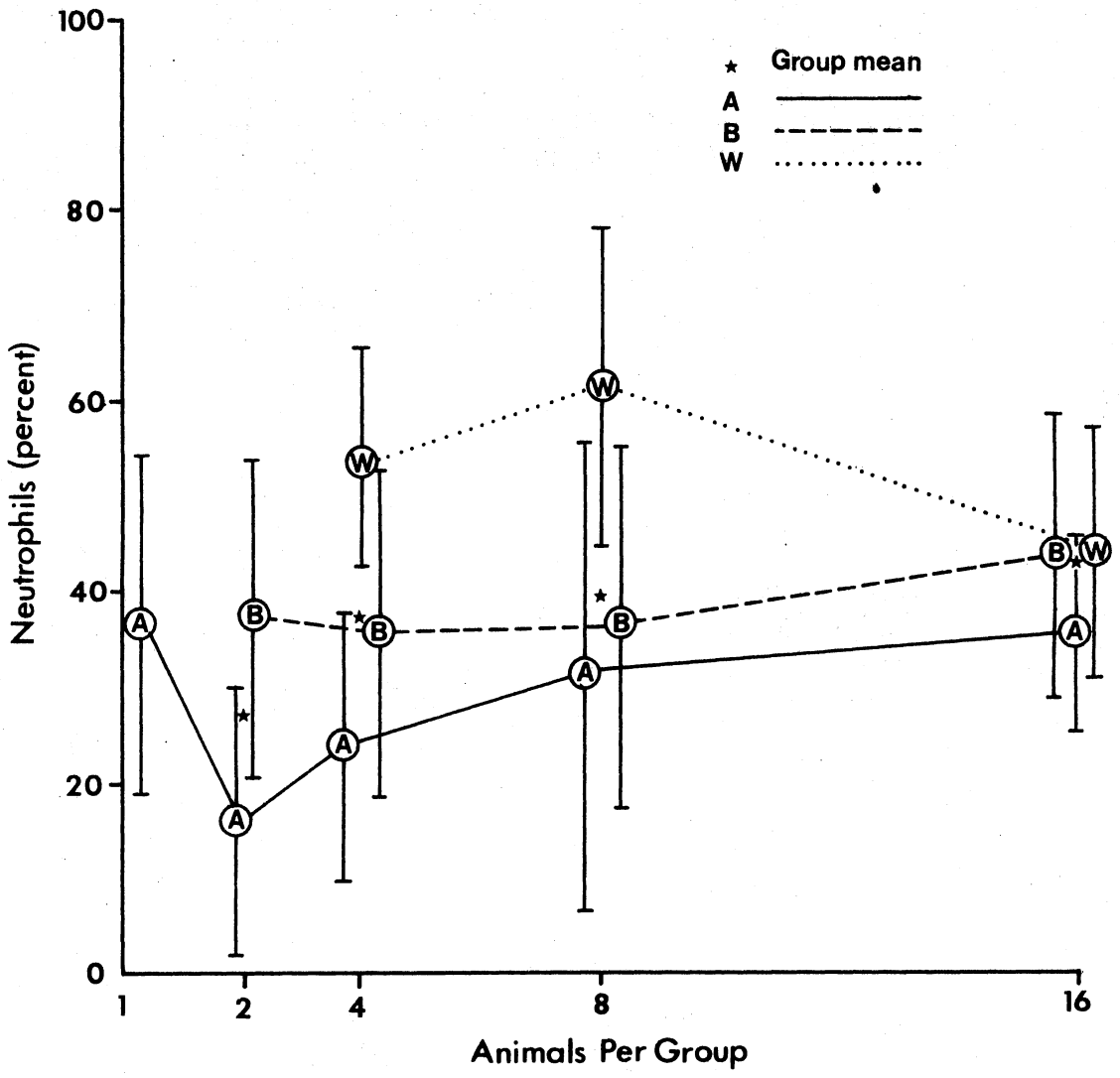

Figure 7. Mean Percentages of Neutrophils by Social Rank Plotted Against Group Size on a Logarithmic Scale

TABLE IV

MEAN WEIGHTS AND STANDARD DEVIATIONS OF GRANULOMAS INDUCED IN MALE *PEROMYSCUS LEUCOPUS* IN RELATION TO CROWDING AND SOCIAL RANK

Group	Social Rank	No.	Weight of Granulomas ± S.D. (mg)
1	A	48	633.36±47.11
2	A	24	681.23±40.44
2	B	24	655.17±30.60
		48	668.20±37.84
4	A	12	701.98±17.24
4	B	23	670.54±17.02
4	W	13	641.04±13.28
		48	670.41±27.26
8	A	6	758.23±68.24
8	B	36	640.28±40.41
8	W	7	607.61±17.16
		48	650.27±59.57
16	A	3	807.03±22.51
16	B	38	659.86±36.89
16	W	7	551.86±19.55
		48	653.31±64.94

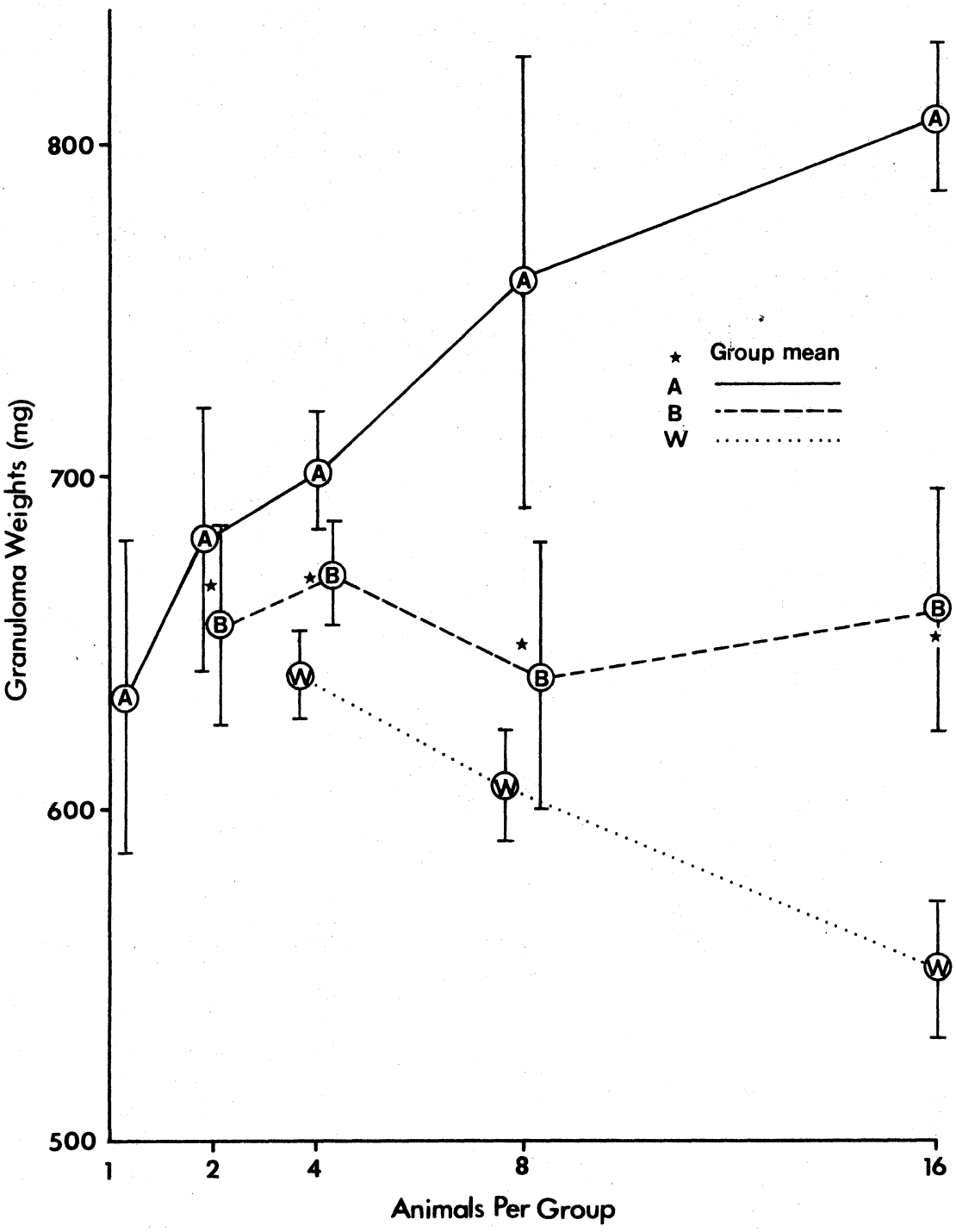

Figure 8. Mean Granuloma Weights by Social Rank Plotted Against Group Size on a Logarithmic Scale

group. Members of social rank B exhibited a marked variation around a central axis of 655 mg. It is interesting to note that with respect to this social rank group 4 was characterized by the largest value of 670.54 mg. The weights for social rank W decreased in a linear fashion as the group size increased. A generalized decrease in granuloma weight was observed as a function of decreasing social rank.

TABLE V

COMPARISON OF DIFFERENCES OF INDUCED GRANULOMA WEIGHTS AMONG ALL GROUPS (SIGNIFICANT P VALUES ARE INDICATED)

	Group 1 (633.36)	Group 2 (668.20)	Group 4 (670.41)	Group 8 (650.27)	Group 16 (653.31)
Group 16 (653.31)	19.95 ns*	14.89 ns*	17.10 ns*	3.08 ns*	0
Group 8 (650.27)	16.91 ns*	17.93 ns*	20.14 P < .05	0	
Group 4 (670.41)	37.05 P < .001	2.21 ns*	0		
Group 2 (668.20)	34.84 P < .001	0			
Group 1 (633.36)	0				

*ns = non-significant P values.

Adrenal Weights

Table VI presents the data obtained from the relative weights of dried adrenal glands for the mice. Adrenal weight of P. leucopus varied significantly in this study with crowding (Table VII). Mice from groups 2 and 4 revealed significantly smaller relative adrenal weights than isolated males ($P < .001$). This suggests that isolation may be a stressful situation for male P. leucopus. Increased group size did not produce enlarged adrenals between groups 2 and 4 and 8 and 16. There was considerable variation in adrenal weights for groups 8 and 16. The standard error of the means were approximately five times that of the control group and twice the variability of groups 2 and 4.

Following the observed decrease of relative adrenal weight from group 1 to group 2, members of social rank A exhibited only one significant change, that being between groups 4 and 8 ($P < .01$) (Figure 9). Furthermore, the same noticeable increase between groups 4 and 8 ($P < .001$) was also observed for social ranks B and W. These data support the hypothesis that relative adrenal weights increase as a function of decreasing social rank.

Plasma Corticosterone Levels

The mean levels of plasma corticosterone for the various groups of the study are summarized in Table VIII. Table IX shows that the mean corticosterone level for group 1 was significantly higher than for groups 2 and 4 at $P < .01$ and $P < .05$, respectively. Groups 2 and 4 were judged to be comparatively equal as well as groups 8 and 16. It is worthy to note that a substantial increase was seen in the comparison

TABLE VI

MEAN DRIED RELATIVE ADRENAL WEIGHTS AND STANDARD DEVIATIONS OF MALE *PEROMYSCUS LEUCOPUS* IN RELATION TO CROWDING AND SOCIAL RANK

Group	Social Rank	No.	Mean Dried Relative Adrenal Weights±S.D. (mg/100 g body wt.)
1	A	48	11.43±0.39
2	A	24	8.21±0.68
2	B	24	8.73±0.78
		48	8.47±0.77
4	A	12	8.28±0.47
4	B	23	9.30±0.26
4	W	13	9.82±0.40
		48	9.19±0.67
8	A	6	9.27±0.40
8	B	35	12.15±1.19
8	W	7	14.84±0.45
		48	12.18±1.79
16	A	3	9.40±0.36
16	B	38	12.05±1.40
16	W	7	15.29±0.57
		48	12.36±1.87

TABLE VII

COMPARISON OF DIFFERENCES OF RELATIVE DRIED ADRENAL WEIGHTS AMONG ALL GROUPS (SIGNIFICANT P VALUES INDICATED)

	Group 1 (11.43)	Group 2 (8.47)	Group 4 (9.19)	Group 8 (12.18)	Group 16 (12.36)
Group 16 (12.36)	0.93 P < .05	3.96 P < .001	3.17 P < .001	0.18 ns*	0
Group 8 (12.18)	0.75 P < .05	3.71 P < .001	2.99 P < .001	0	
Group 4 (9.19)	2.24 P < .001	0.72 ns*	0		
Group 2 (8.47)	2.96 P < .001	0			
Group 1 (11.43)	0				

*ns = non-significant P values.

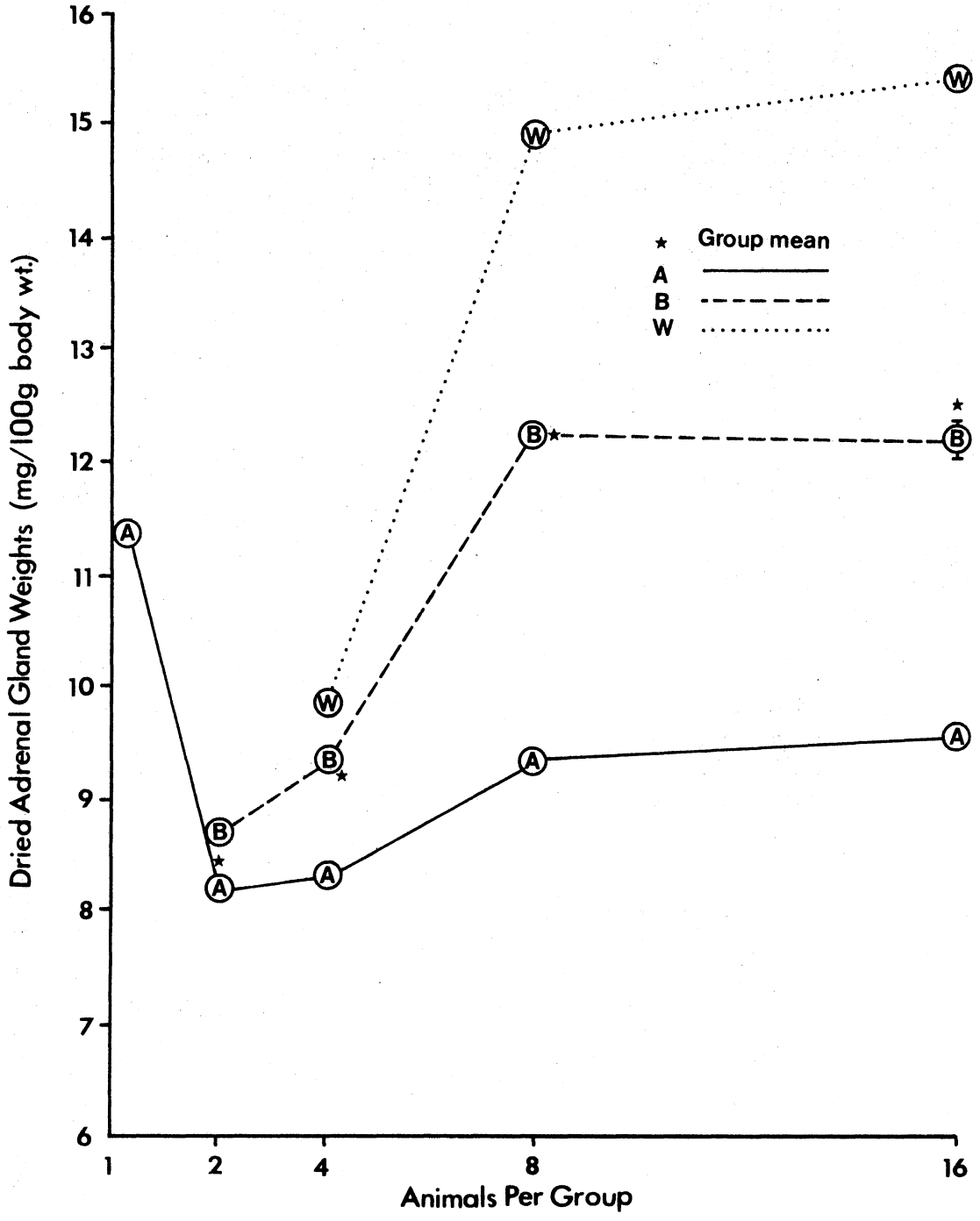

Figure 9. Mean Adrenal Gland Weights by Social Rank Plotted Against Group Size on a Logarithmic Scale

TABLE VIII

MEAN PLASMA CORTICOSTERONE LEVELS AND STANDARD DEVIATIONS OF MALE *PEROMYSCUS LEUCOPUS* IN RELATION TO CROWDING AND SOCIAL RANK

Group	Social Rank	No.	Plasma Corticosterone Levels±S.D. (µg/100 ml plasma)
1	A	48	45.45±11.31
2	A	24	30.92± 5.98
2	B	24	40.63± 5.22
		48	35.77± 7.41
4	A	12	27.33± 3.04
4	B	23	39.13± 4.61
4	W	13	49.77± 3.66
		48	39.06± 9.07
8	A	6	26.75± 2.42
8	B	35	51.71± 9.53
8	W	7	69.07± 2.58
		48	51.13±13.82
16	A	3	27.33± 2.51
16	B	38	51.09±12.33
16	W	7	76.57± 5.77
		48	53.32±15.87

of groups 4 and 8 (Figure 10) which is in agreement with this study's findings of adrenal weights. Again, an increase in the magnitude of the standard error was observed as group size increased.

TABLE IX

COMPARISON OF DIFFERENCES OF PLASMA CORTICOSTERONE LEVELS AMONG ALL GROUPS (SIGNIFICANT P VALUES INDICATED)

	Group 1 (45.45)	Group 2 (35.77)	Group 4 (39.06)	Group 8 (51.13)	Group 16 (53.32)
Group 16 (53.32)	7.87 P < .05	17.55 P < .001	14.26 P < .001	2.19 ns*	0
Group 8 (51.13)	5.68 ns*	15.36 P < .001	12.07 P < .001	0	
Group 4 (39.06)	6.39 P < .05	3.29 ns*	0		
Group 2 (35.77)	9.68 P < .01	0			
Group 1 (45.45)	0				

*ns = non-significant P values.

Examination of the groups with respect to social rank disclosed that social rank A decreased in a curvilinear fashion as a function of increasing group size, while social rank B exhibited a sigmoid-like trend with a marked increase betwen groups 4 and 8 (P < .001). Social rank W was observed to increase in a curvilinear fashion as the group

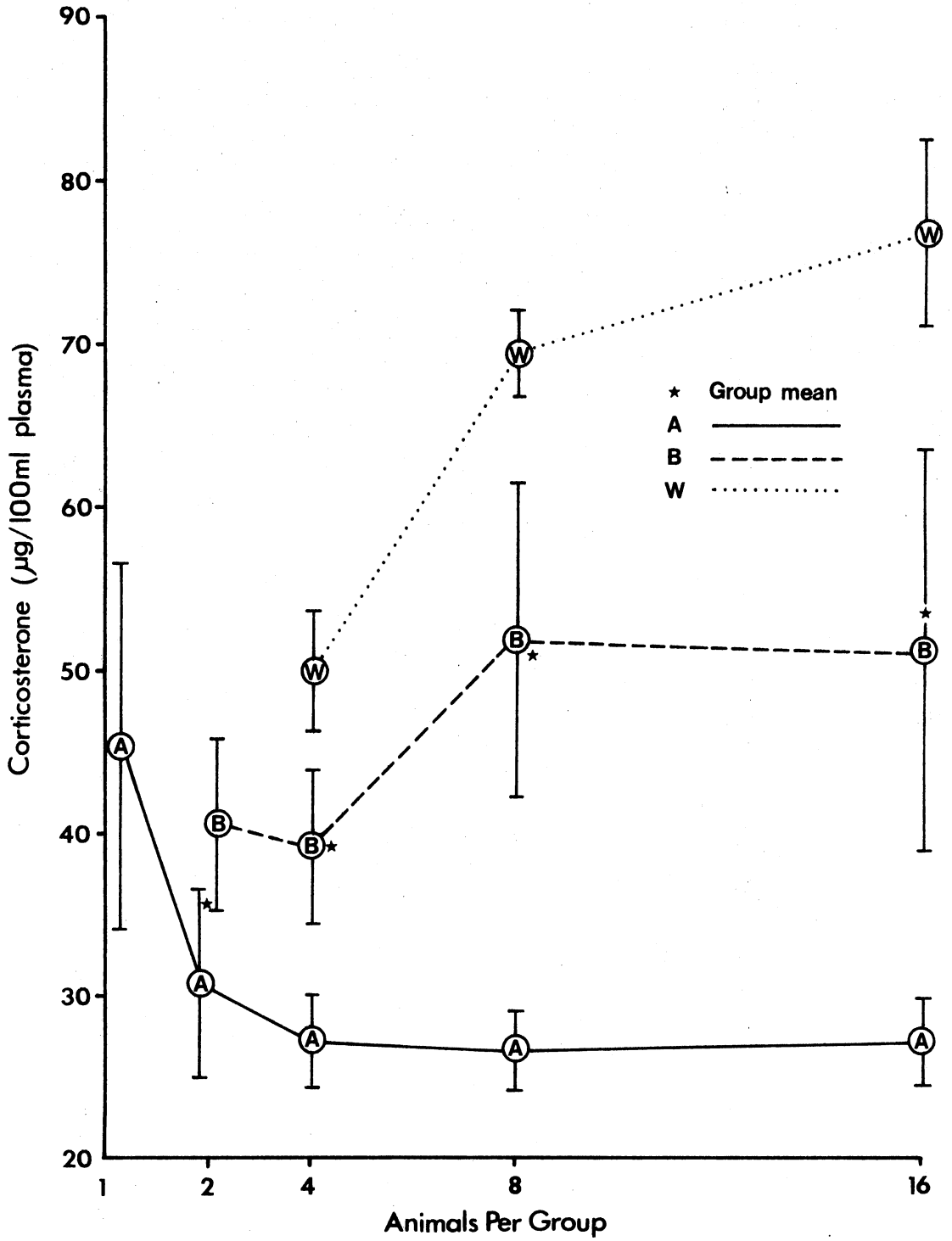

Figure 10. Mean Plasma Corticosterone Levels by Social Rank Plotted Against Group Size on a Logarithmic Scale

size increased. It was evident that plasma corticosterone levels increased as a function of decreasing social rank.

CHAPTER V

DISCUSSION

Introduction

Each group of mice can be considered as a population, though they did not possess all the properties of a population; the number was static and each population consisted only of males. However, taken collectively, the five groups can represent five instances in the development of a population. A population rarely saturates the area it occupies, but certain other aspects of the environment can become saturated. As a population increases, the number of animals with which any individual can interact increases proportionately even when the area per individual is constant. Increases in number of social interactions would presumably have effects on an individual similar to those in experiments where space per animal is varied.

Differential Leukocyte Count

The results of this study differ in a number of aspects from other reports on leukocyte changes in response to crowding. Southwick (1964) reported the eosinophil response of P. leucopus to behavioral and social disturbance followed the same pattern as in house mice (Southwick, 1959; Vanderbergh, 1960). Males placed in empty colony pens exhibited a 70 percent eosinophil decline within 4 hours, but by 72 hours counts

returned to normal. Males placed in colony pens containing a resident pair exhibited a 92 percent decline, and the eosinophil count remained significantly suppressed for the duration of the experiment (5 days). However, the changes in percentages of eosinophils, basophils, lymphocytes, monocytes, and neutrophils, observed in the present study, were neither great in extent in most instances nor consistent. Furthermore, those differences that were observed, were not beyond the ranges of variation. If any interpretation is warranted from these data, it is the negation of the validity of the utilized technique as an assessment of adrenocortical function.

Two methods are available for counting leukocytes, the differential count and the chamber count. The differential count has a large margin of error, and is, therefore, not suitable for enumeration of the respective types of leukocytes in the peripheral blood (Randolph and Stanton, 1954; Discombe, 1946; Best and Samter, 1951). These findings are in agreement with those of the investigator. Further examination of the blood smears by the current investigator revealed that the distribution of leukocytes was not symmetrical. In fact, it was found that the differential values for a given mouse were not reproduceable with any degree of consistency.

Nonetheless, in spite of the high coefficients of variation and apparent low precision of the differential leukocyte count as a quantitative index of adrenocortical function, the results of this study revealed a generalized eosinopenia, lymphopenia, and neutrophilia as a function of decreasing social rank within each group.

Granuloma Formation Rate

An examination of the data with respect to crowding should be conducted only after the omission of the isolated controls. The isolated mice possessed significantly lighter granulomas than groups 2 and 4. This suggests that isolation was stressful to these mice.

With this in mind, crowding clearly had a suppressive effect on granuloma formation in mice in this study. This, together with the known anti-inflammatory action of adrenal glucocorticoids (Dougherty, 1953; Taubenhaus and Amromin, 1950; Sorenson, 1966) and the positive correlations between plasma corticosterone levels and population density (Louch and Higginbotham, 1967; Pearson, 1962; Gartner et al., 1973) support the hypothesis that there is a sufficient increase in adrenocortical activity to depress inflammation as a result of crowding.

Of special interest is the observation that the granuloma weights of the respective social ranks become more separated as a function of group size, thereby suggesting a marked difference in corticosterone levels with respect to social rank.

Depression of inflammation in this study is believed to be due to increased levels of corticosterone. The fact that increased population density decreases resistance to infection by normal defenses, such as inflammation, has obvious importance for studies concerned with population dynamics.

Adrenal Weights

The adrenal glands of P. leucopus are exceptionally large, with the current investigator finding 1.71 to 3.51 mg (7.6 to 16.1 mg/100 g body weight) dry weight and Southwick (1964) reporting 10.5 to 16.5 mg (50.6

to 93.5 mg/100 g body weight) wet weight. This is approximately three times as large as the adrenal weights reported for P. maniculatus bairdi and P. m. gracilis (Bronson and Eleftheriou, 1963; Terman, 1965a, 1965b) and for house mice or meadow voles. Mice and voles previously studied had mean paired adrenal weights ranging from 3 to 5 mg (15.0 to 45.0 mg/100 g body weight) (Clarke, 1952; Christian and Davis, 1955; Christian, 1955; Louch, 1956, 1958). Male P. leucopus have larger adrenals than females, whereas the reverse is true for the rodent species examined except the nutria (Wilson and Dewees, 1962).

The results of the present study indicate that the adrenal weights of male P. leucopus change with population density. Similar results have been obtained with P. leucopus by Southwick (1964). The mean response of the mice in the various groups with respect to increasing density mainly reflected changes occurring in the mass of low-ranking individuals. This is because population increase occurred with a disproportionate increase in the number of subordinate animals. The number of dominant animals is relatively constant and hence the additional individuals must be subordinate.

It was observed that as group size increased, the mean adrenal weight increased with the exception of group 1. Christian (1965a) attributed the increase in adrenal weight to hypertrophy of the adrenal cortex. In addition, the increase in adrenal weight (commencing with group 2) was accompanied by an increase in variance and skewness of the frequency distribution curves.

Males from groups 2 and 4 revealed significantly smaller relative adrenal weights than isolated males ($P < .05$). It is suggested that isolation may be a "stressful" situation for male P. leucopus (the

postulated "isolation stress" of Weltman et al., 1962, 1966, 1967, 1968). These findings are in agreement with Southwick's (1964) reportings for the same species. The role of isolation in contrast to crowding needs to be considered. However, this topic will receive little attention here because isolation is not the normal situation and does not present the problems that crowding does. Furthermore, it is probable that an isolated rodent cannot be regarded as one at the lowest intensity of "grouping." Isolation was attained by simply separating the male mice for a matter of weeks. When these animals were put together again, they fought rather severely, but in a short period of time settled down to apparent rank organization. It is worthy to note that for these data isolation produced physiological stress reactions, as measured by relative adrenal weights, at almost the same level of those produced by crowding at the higher densities. As emphasized by Sigg et al. (1966), extreme isolation or density produces many homeostatic adjustments. These observed adrenocortical changes seem to obey the Yerkes-Dobson law in that minimal pituitary-adrenal activation occurs at an optimal population size. The interpretation for the present data suggests that the densities of 2 and 4 mice per cage meet such a criterion.

Further examination of the adrenal weights discloses that the most marked increase in weight, whether with respect to group means or to the various social ranks, occurred between groups 4 and 8. The adrenal weights of groups 8 and 16 were relatively equal. This implies that if Christian's postulated density-dependent population regulatory mechanism is, indeed, present for these data, its threshold lies somewhere between 4 and 8 mice per group. Hence, between these two levels of crowding,

the intensity of social interaction was of sufficient magnitude to increase the pituitary-adrenal system to deleterious levels.

It has been reported by a number of workers that there is a direct relationship between adrenal weights and population density for several species of mammals (Christian, 1950, 1963a, 1963b, 1963c). However, the unqualified statement that the mean adrenal weight of animals in a given population has increased can be interpreted in two ways. Either it could mean that the adrenal of every individual had increased approximately the same amount, or it could mean that the increase in adrenal weight had actually occurred in only a few individuals, with the majority of the animals being little affected. Observations were made for this set of data which indicate that the true situation lies somewhere between the two extremes.

When the dried adrenal weights from the various groups were compared, the over-all variation, as well as the mean adrenal weight, with the exception of group 1, was found to increase in group size. Virtually, all of the increase in variation around the group mean for groups 8 and 16 can be attributed to the individuals of social rank W. The mice which deviated from the main distribution were always in the direction of enlarged adrenals. Accordingly, the adrenals of members of social rank W were significantly larger than those of social rank B ($P < .001$). In Figure 11, the progressive separation of mean and median values, an indication of increasingly skewed distribution in the direction of the larger adrenals, it almost entirely ascribed to the individuals of social rank W. Even if these deviant individuals are censored, however, the mean relative adrenal weights of the remaining mice still followed the same trend. Welch and Klopfer (1961) have

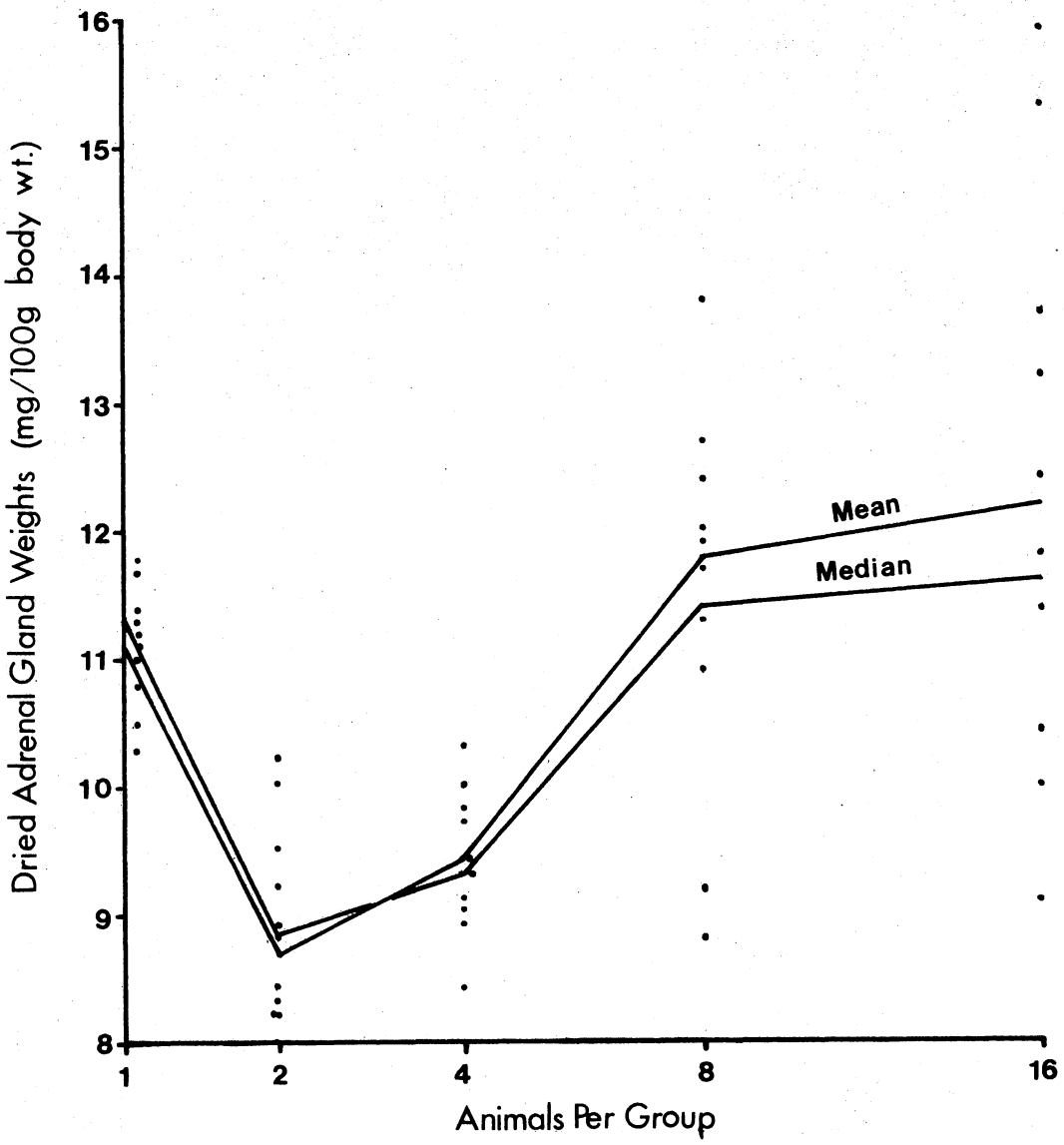

Figure 11. Milligrams of Adrenal Gland per 100 g Body Weight are Plotted for 10 Animals Drawn at Random from each of the Five Group Sizes. The Difference Between Median and Mean as well as the Dispersion of Points is Indicative of Increasing Skew in the Direction of Larger Adrenals

emphasized the significance of increased endocrine variability in the adrenal stress response of house mice. The data from this study support their concept.

In general, the effects of descending social rank upon weights of adrenal glands in mice parallel those of increasing density (Davis and Christian, 1957). The present study, as well, observed an increase in adrenal weight as a function of decreasing social rank. Adrenocortical hypertrophy as indicated by adrenal weight was greatest in the most subordinate animals and was slight or absent in dominant mice. The adrenals of those in between tended to fall in line in a reciprocal relationship to their dominance rank.

Welch and Klopfer (1961) also observed a situation similar to the one seen here. In addition to the men relative adrenal weights increasing with rising density, there was also a significant rise in variance. Their findings led them to propose that the probability of death from effects associated with adrenal hypertrophy is not shared equally by all members of the population.

> It appears that there are two predominant kinds of psychosocially influenced endocrine responses to crowding which oppose each other in the effect that they produce on the population. First, the true elevation in adrenal weight throughout the entire population together with the greater physiological homogeneity of the majority, favors the likelihood that a large portion of the population will experience the consequences of acute hyperfunction simultaneously. Second, this tendency is opposed by the increasing number of deviant individuals whose death will precede the others, and, reducing density, reduce the likelihood of a major population crash (Welch and Klopfer, 1961:259).

If this situation proves generally true, it provides a method for a selective reduction in density and thus the avoidance of a wholesale population crash.

Plasma Corticosterone Levels

For years adrenal function has been assayed by the weight of the adrenal glands. However, recent trends have questioned the validity of adrenal weights as an assessment of adrenocortical activity with the utilization of plasma corticosterone levels being a more direct criterion of this activity. A primary drawback has been the analysis of adrenal weight data. First, it should be brought to mind that adrenal weight includes the weight of the adrenal medulla as well as the cortex. Second, the relation between adrenal weight and body weight is most likely not linear (Krebs, 1964), and it is not valid to compare adrenal weights by using values which are given in mg adrenal wt/gm body weight. Third, adrenal weight may be altered by a number of different factors, not necessarily related to the production of glucocorticoids (Brain and Nowell, 1969a). Fourth, several attempts at finding relations between adrenal weights and microtine population densities have failed (Christian, 1961; H. Chitty, 1961; Krebs, 1964). Indeed, adrenocortical functions and adrenal weight are often not closely comparable parameters as Barthe and Desaulles (1970) have recently found that atrophied rat adrenals (produced by hypophysectomy) can be stimulated to give a normal response (in terms of plasma corticosterone levels) by 10-day treatment with synthetic ACTH, whereas 30-day treatment is required to restore adrenal weight to the pre-hypophysectomy level. In addition to this difficulty, it now appears likely that changes in sex steroids can influence adrenal weight (Brain and Nowell, 1969b, 1971a, 1971b; Brain et al., 1971).

An integration of the present data should be made by comparing only the groups with 2, 4, 8, and 16 mice. The solitary mice suffered

a change of endocrine responses caused by "isolation stress." This observation is supported by the significantly larger mean corticosterone level and higher standard error than reported for groups 2 and 4 ($P < .05$). These findings are in agreement with the results of the adrenal weights for the study. Isolation and high population density have, therefore, in common an activation of pituitary adrenal function, but they differ from each other in that the former causes pituitary-gland stimulation (indirect evidence from castration experiments), whereas the latter diminishes reproductive function (Christian, 1963). From this study of isolation and crowding, it seems that there must be some population size at which the physiological processes can be considered at a normal or minimal level.

In this respect the data collected in this study reflect the findings of Pearson (1962) in that the levels of plasma corticosterone increased as a function of increasing population density. Again, it was observed that the most notable increase in adrenocortical function was found to exist between groups 4 and 8. The corticosterone levels between groups 8 and 16 were observed to be relatively the same.

These data were consistent with the findings of Louch and Higginbotham (1967) who have shown that mice which are dominant in a social hierarchy have lower plasma corticsterone levels than the subordinate animals. In this respect it is pertinent to note that these data parallel the results of several other studies of mice which detected an inverse relation between position in a social hierarchy and adrenal weight (Davis and Christian, 1957). More importantly, these data suggest that the enlarged adrenals which were observed were functioning at a higher level and secreting greater amounts of adrenocorticoids. An

interesting corollary is the observation that prolonged grouping among the mice was accompanied by an increased intragroup variability in the indices of pituitary-adrenal function, an effect presumably mediated by the marked differences which developed between the dominant and subordinate group members.

From the preceding it might be assumed that corticosteroid levels offer a panacea for the measurement of adrenocortical function. Such is not the case. First, it is very difficult to differentiate between acute and chronic stimuli with respect to these cortical hormone levels. The distinction between acute and chronic stimuli is important because the results differ markedly. An acute stimulus (wound, fright) produces an immediate chain of reactions resulting in the prompt release of hormones from the adrenal gland and then return to normal without a change in the average level. On the other hand, a chronic stimulus (daily conflict) results in a gradient increase of release of hormones until a new, higher level is attained. Second, it has been reported that glucocorticoid levels are influenced by sex steroids (Brain and Nowell, 1969b, 1971a, 1971b; Brain et al., 1971). An additional factor that adds complications is the fact that the output of corticoids from the adrenal follows a circadian rhythm. Important to the investigator's interest is that the collection of plasma or serum must be conducted at the same time of day to allow interpretation or comparison of the data.

CHAPTER VI

SUMMARY AND CONCLUSIONS

The present study involved the subjection of live-trapped male white-footed mice (Peromyscus leucopus) to densities of 1, 2, 4, 8, and 16 mice per cage (27.9 x 22.6 x 30.5 cm) for 20 days, following an acclimation period to the laboratory. At the end of this period of crowding, social rank was subjectively assigned to each mouse which was then sacrificed. A differential leukocyte count, induced granuloma weight, dried relative adrenal weights, and plasma corticosterone level was quantified for each mouse. Changes in these indices of stress for the various groups and their respective social ranks were examined.

It was found that the differential leukocyte count was not valid as an assessment of adrenocortical function due to large variation and the inability to replicate measurements consistently. The remaining indices indicated that isolatin was a stressful situation for male P. leucopus. Densities of 2 and 4 mice per cage were found to be characterized by indices indicative of optimum population density. The most marked increase in the adrenal responses was from group 4 to group 8. The increase in adrenocortical function from the density of 8 per cage to 16 per cage was not significant.

In conclusion, the ususal response to increased population density in natural populations is emigration. When the animals have no opportunity to emigrate, increased aggressive behavior, associated with changes

in dispersionand social structure, is the primary consequence of increased density. Secondary effects, the physiological consequence of fighting, occur as a result of the increased frequency of aggressive interactions.

Social organization plays a critical role in determining the effect which crowding has upon the health and welfare of the population. The most deleterious effects of crowding may be to disrupt normal behavior patterns and to create pathological social conditions. The main result of crowding in this study was the potential elimination of low ranking individuals. Many animals with highly evolved and adaptive social behavior show territorial and hierarchial patterns which act as intrinsic self-regulatory mechanisms. Members of the species Peromyscus leucopus are good examples, and in addition these animals rarely if ever develop eruptive growth and excessive densities.

LITERATURE CITED

Ader, R. 1967. The influence of psychological factors on disease susceptibility in animals. In M. L. Conalty (ed.), "The Husbandry of Laboratory Animals." Academic Press, London. Pp. 219-236.

Ader, R. and S. M. Plaut. 1968. Effects of prenatal maternal handling and different housing on offspring emotionality, plasma corticosterone levels, and susceptibility to gastric erosions. Psychosom. Med. 30:277-286.

Akin, D. P. 1972. Ultraradian, circadian, and infraradian rhythms of corticosterone, eosinophils and hematocrit in Rana pipiens. Unpublished Ph.D. thesis. Indiana University. 195 pp.

Bailey, E. D. 1966. Social interaction as a population-regulating mechanism in mice. Can. J. Zool. 44:1007-1012.

Baker, B. L. 1954. The connective tissue reaction around implanted pellets of steroid hormones. Anat. Record. 119:529-539.

Bamgbose, S. O. A. and D. M. James. 1969. The role of glucose in acute laboratory trypanosomal infection. J. Trop. Med. Hyg. 72:296-297.

Barrett, A. M. and M. A. Stockham. 1963. The effect of housing conditions and simple experimental procedures upon the corticosterone level in the plasma of rats. J. Endocrin. 26:97-105.

Barnett, S. A. 1958. Physiological effects of "social stress" in wild rats.--I. The adrenal cortex. J. Psychosomat. Res. 3:1-11.

Barr, A. J. and J. H. Goodnight. 1972. A user's guide to the statistical analysis system. North Carolina State University, Raleigh, N. C. 260 pp.

Barthe, P. L. and P. A. Desaulles. 1970. Influence of time factor on the restoration of adrenal weight and cortical functions in hypophysectomized rats. Hormones. 1:257-272.

Beisel, W. R. 1969. Inter-relations between adrenocortical functions and infectious illness. N. Engl. J. Med. 280:541-546.

Best, W. R. and M. Samter. 1951. Variation and error in eosinophil counts of blood and bone marrow. Blood. 6:61-74.

Bloch, E. and A. I. Cohen. 1960. In vitro steroid production by normal and adrenal tumor-bearing mice. J. Natl. Cancer Inst. 24:97-107.

Boring, W. O., D. M. Angevine and D. L. Walker. 1955. Factors influencing host-virus interactions. I. A comparison of viral multiplication and histopathology in infant, adult, and cortisone-treated adult mice infected with the Conn.-5 strain of Coxsackie virus. J. Exptl. Med. 102:753-766.

Brain, P. F. and N. W. Nowell. 1969a. Some behavioral and endocrine relationships in adult male laboratory mice subjected to open field and aggression tests. Physiol. Behav. 4:945-947.

Brain, P. F. and N. W. Nowell. 1969b. The effects of isolation as opposed to grouping on adrenal and gonadal functions in male and female mice. J. Endocrinol. 46:xvi-xvii.

Brain, P. F. and N. W. Nowell. 1971a. Isolation versus grouping effects on adrenal and gonadal function in albino mice. I. The male. Gen. Comp. Endocrinol. 16:149-154.

Brain, P. F. and N. W. Nowell. 1971b. Isolation versus grouping effects on adrenal and gonadal function in albino mice. II. The female. Gen. Comp. Endocrinol. 16:149-154.

Branson, E. D. 1968. Adrenal cortex. Ann. Rev. Physiol. 30:171-212.

Briggs, N. T. 1958. Serological response of cotton rats and white rats in relation to the extent of infection with Litomosoides carnii. Diss. Abstr. 19(5):1062. Univ. of Mich.

Bronson, F. H. and B. E. Eleftheriou. 1963. Adrenal response to crowding in Peromyscus and C57BL/10J mice. Physiol. Zool. 36:161-166.

Bronson, F. H. and B. E. Eleftheriou. 1964. Chronic physiologic effects of fighting in mice. Gen. Comp. Endocrinol. 4:9-14.

Bronson, F. H. and B. E. Eleftheriou. 1965. Behavioral, pituitary, and adrenal correlates of controlled fighting (defeat) in mice. Physiol. Zool. 38:406-411.

Bush, I. E. 1953. Species differences in adrenocortical secretion. J. Endocrinol. 9:95-100.

Calhoun, J. B. 1948. Mortality and movement of brown rats in artificially super-saturated populations. J. Wildl. Mgmt. 12:167-171.

Calhoun, J. B. 1949. A method for self-control of population growth among mammals living in the wild. Science. 109:333-335.

Calhoun, J. B. 1950. The study of wild animals under controlled conditions. Ann. N. Y. Acad. Sci. 51:1113-1122.

Canham, R. P. 1970. Sex ratios and survival in fluctuating populations of the deer mouse, Peromyscus maniculatus borealis. Can. J. Zool.

Cartwright, G. E. 1958. Diagnostic laboratory hematology. Grune and Stratton. New York. 250 pp.

Chang, S. S. and A. F. Rasmussen. 1965. Stress-induced suppression of interferon production in virus-infected mice. Nature. 205:623-624.

Chitty, D. 1952. Mortality among voles (Microtus agrestis) at Lake Vyrnwy, Montgomeryshire in 1936-39. Phil. Trans. Roy. Soc. London, Ser. B. 236:505-558.

Chitty, D. 1954. Tuberculosis among wild voles with a discussion of other pathological conditions among certain mammals and birds. Ecology. 35:227-237.

Chitty, D. 1957. Self regulation of numbers through changes in viability. Cold Springs Harbor Symp. Quant. Biol. 22:277-280.

Chitty, D. 1967. The natural selection of self-regulatory behavior in animal populations. Proc. Ecological Soc. Australia. 2:51-78.

Chitty, H. 1961. Variations in the weight of the adrenal glands of the field vole (Microtus agrestis). J. Endocrinol. 22:387-397.

Christian, J. J. 1950. The adreno-pituitary system and population cycles in mammals. J. Mammal. 31:247-259.

Christian, J. J. 1955a. Effect of population size on the weights of the reproductive organs of white mice. Am. J. Physiol. 181:477-480.

Christian, J. J. 1955b. Effect of population size on the adrenal glands and reproductive organs of male mice in populations of fixed size. Am. J. Physiol. 182:292-300.

Christian, J. J. 1956a. Adrenal and reproductive responses to population size in mice from freely growing populations. Ecology. 37: 258-273.

Christian, J. J. 1956b. Reserpine suppression of density-dependent adrenal hypertrophy and reproductive hypoendocrinism in populations of male mice. Am. J. Physiol. 187:353-356.

Christian, J. J. 1959a. Control of population growth in rodents by interplay between population density and endocrine physiology. Wildl. Dis. 1:1-38.

Christian, J. J. 1959b. The roles of endocrine and behavioral factors in the growth of mammalian populations. In "Comparative Endocrinology," Proc. Columbia University Symposium, 1958 (A. Gorman, ed.). Pp. 71-97. Wiley, New York.

Christian, J. J. 1959c. Lack of correlation between adrenal weight and injury in grouped male albino mice. Proc. Soc. Exp. Biol. Med. 101:166-168.

Christian, J. J. 1961. Phenomena associated with population density. Proc. Natl. Acad. Sci. U. S. 47:428-449.

Christian, J. J. 1963a. Endocrine adaptive mechanisms and the physiologic regulation of population growth. In "Physiological Mammalogy," (W. V. Mayer and R. G. Van Gelder, eds.). 1:189-353. Academic Press, New York.

Christian, J. J. 1963b. The pathology of overpopulation. Military Med. 128:571-603.

Christian, J. J. 1963c. Interrelated social and endocrine factors in populations of rodents. Proc. 16th Intern. Congr. Zool., Washington, D. C. 3:8-13.

Christian, J. J. 1967. ACTH-induced renal glomerular disease in intact, adrenalectomized and castrate male mice. Proc. Soc. Exp. Biol. Med. 126:152-157.

Christian, J. J. 1968. The potential role of the adrenal cortex as affected by social rank and population density on experimental epidemics. Am. J. Epidemiol. 87:255-266.

Christian, J. J. and D. E. Davis. 1964. Endocrines, behavior and population. Science. 146:1550-1560.

Christian, J. J., J. A. Lloyd and D. E. Davis. 1965. The role of endocrines in the self-regulation of mammalian populations. Rec. Prog. Hor. Res. 21:501-578.

Christian, J. J. and C. D. LeMunyan. 1958. Adverse effects of crowding on lactation and reproduction of mice and two generations of their offspring. Endocrin. 63:517-529.

Christian, J. J. and H. O. Williamson. 1958. Effect of crowding on experimental granuloma formation in mice. Proc. Soc. Exptl. Biol. Med. 99:385-387.

Clarke, J. R. 1953. The effect of fighting on the adrenals, thymus, and spleen of the vole (Microtus agrestis). J. Endocrinol. 9:114-126.

Clarke, J. R. 1955. Influence of numbers on reproduction and survival in two experimental vole populations. Proc. Roy. Soc. B144:68-85.

Coker, C. M. 1955. Effects of cortisone on Trichinella spiralis infections in non-immunized mice. J. Parasitol. 41:498-508.

Colfer, H. F., J. DeCroot and G. W. Harris. 1950. Pituitary gland and blood lymphocytes. J. Physiol. 111:328-334.

Cortes, J. M., F. G. Peron and R. I. Dorfman. 1963. Secretion of 18-hydroxydeoxy-corticosterone by the rat adrenal gland. Endocrinol. 73:713-717.

Crew, F. A. E. and L. Mirskaia. 1931. Effect of density on adult mouse populations. Biol. General. 7:239-250.

Cross, J. H. 1960. The natural resistance of the white rat to Nematospiroides dubius and the effect of cortisone on this resistance. J. Parasit. 46:175-178.

Crowcraft, P. and F. P. Rowe. 1957. The growth of confined colonies of the wild house mouse (Mus musculus). Proc. Zool. Soc. London. 129:359-370.

Crowcraft, P. and F. P. Rowe. 1963. Social organization and territorial behaviour in the wild house mouse (Mus musculus L.). Proc. Zool. Soc. London. 140:517-531.

Davis, D. E. 1953. The characteristics of rat populations. Quart. Rev. Biol. 28:373-401.

Davis, D. E. 1958. The role of density in aggressive behavior of house mice. Animal Behavior. 6:207-210.

Davis, D. E. and J. J. Christian. 1957. Relation of adrenal weights to social rank in mice. Naval. Med. Res. Inst. Rep. 15:311-316.

Davis, D. E. and Read, C. P. 1958. Effect of behavior on development of resistance in trichinosis. Proc. Soc. Exptl. Biol. Med. 99: 269-272.

DeFries, J. C. and G. E. McClearn. 1970. Social dominance and Darwinian fitness in the laboratory mouse. Am. Natur. 104:408-411.

Discombe, G. 1946. Criteria of eosinophilia. Lancet. 1:195-199.

Dorfman, A. 1953. The effects of adrenal hormones on connective tissue. Ann. N. Y. Acad. Sci. 56:598-703.

Dougherty, T. F. 1953. Some observations on mechanisms of corticosteroid action on inflammation and immunologic processes. Ann. N. Y. Acad. Sci. 56:748-756.

Dougherty, T. F. and G. L. Schneebeli. 1955. The use of steroids as anti-inflammatory agents. Ann. N. Y. Acad. Sci. 61:328-348.

Dreyfuss, F. and S. Feldman. 1952. Eosinopenia induced by emotional states. Acta Med. Scandinav. 144:107-113.

Eisenstein, A. B. 1967. The Adrenal Cortex. Little, Brown and Company: Boston. 685 pp.

Eleftheriou, B. E. 1964. Bound and free corticosteroid in the plasma of two subspecies of deer mice (Peromyscus maniculatus) after exposure to a low abient temperature. J. Endocrin. 31:751-780.

Elmadjian, F., J. M. Hope and E. T. Lamson. 1958. Excretion of epinephrine and norepinephrine under stress. Recent. Progr. in Hormone Res. 14:513-553.

Elton, C. 1942. Voles, mice, and lemmings: Problems in population dynamics. Oxford. 496 pp.

Emel'yantsev, A. M. and N. A. Sveshnikova. 1969. Effect of cortisone and insulin on hexokinase activity of rat blood lymphocytes. Byvl. E. Ksp. Biol. Med. 67:65-67

Errington, P. L. 1943. An analysis of mink predation upon muskrats in Northcentral United States. Iowa State College Agr. Expt. Sta. Bull. No. 320.

Errington, P. L. 1954. The special responsiveness of minks to epizootics in muskrat populations. Ecol. Monographs. 24:377-393.

Essellier, A. F., R. L. Jenneret and L. Merandi. 1954. The mechanism of glucocorticoid eosinopenia. Blood. 9:531-539.

Frank, F. 1953. Untersuchungen uber den Zusammenbruch von Feldmausplagen (Mictrotus arvalis Pallas). Zool. Zahrb. Jena. 82:95-136.

Friedman, S. B., L. A. Glasgow and R. Ader. 1969. Psychological factors modifying host resistance to experimental infections. Second Conference on Psychophysiological Aspects of Cancer. Ann. N. Y. Acad. Sci. 164:381-383.

Friedman, S. B., L. A. Glasgow and R. Ader. 1970. Differential susceptibility to a viral agent in mice housed alone or in groups. Psychosom. Med. 32:285-299.

Friedman, S. B. and L. A. Glasgow. 1973. Interaction of mouse strain and differential housing upon resistance to Plasmodium berghei. J. Parasitol. 59(5):851-854.

Gartner, K. and K. Bonath. 1971. Der einflub der gruppengrobe auf den kortikosteronoe halt der nebennieren und des serums mannlicher mause und ratten. Endokrinologie. 58:129-139.

Gartner, K., H. Reznik-Schuller and G. Reznik. 1973. The influence of overcrowding on spermatogenesis size of leydig-cell nuclei (histometrical investigation), and the adrenal corticosterone contents in mice. Acta Endocrinologica. 74:783-791.

Glasgow, L. A. 1965. Leukocytes and interferon in the host response to viral infections. I. Mouse leukocytes and leukocyte-produced interferon in vaccina virus infection *in vitro*. J. Exp. Med. 121:1001-1018.

Glasgow, L. A. and S. B. Friedman. 1969. Interferon and host resistance to Rauscher virus-induced leukemia. J. Virol. 3:99-105.

Glickman, S. E. and B. J. Morrison. 1969. Some behavioral and neural correlates of predation susceptibility in mice. Comm. Behav. Biol. 4:261-267.

Golley, F. G. 1961. Effect of trapping on adrenal activity in *Sigmodon*. J. Wildl. Mgmt. 25:331-333.

Gorbman, A. and H. A. Bern. 1962. Adrenal cortex and interrenal gland. In "A textbook of comparative endocrinology." Pp. 297-339. John Wiley and Sons, New York.

Gordon, A. S. and G. F. Katsh. 1949. The relation of the adrenal cortex to the structure and phagocytic activity of the macrophagic system. Ann. N. Y. Acad. Sci. 52:1-30.

Gordon, A. S. Some aspects of hormonal influences upon the leukocytes. Ann. N. Y. Acad. Sci. 59:907-927.

Greenberg, J., E. M. Nadel and G. R. Coatney. 1953. The influence of strain, sex, and age of mice on infection with *Plasmodium berghei*. J. Inf. Dis. 93:96-100.

Guillemin, R. 1974. The hormones of the hypothalamus. In "Integrative hypothalamic activity." Prog. Brain Res. 41:117-132.

Harley, J. P. and V. Gallicchio. 1970. Effect of cortisone on the establishment of *Nippostrongylus brasiliensis* in the rabbit. J. Parasitol. 56:271-276.

Hart, P. and R. T. W. Rees. 1950. Enhancing effect of cortisone on tuberculosis in the mouse. Lancet. ii:391-395.

Healey, M. C. 1967. Aggression and self-regulation of population size in deer mice. Ecology. 48:377-392.

Hepworth, J. L. 1966. Hematology of Sigmodon hispidus: average parameters compared with those under induced stresses. Stillwater, Oklahoma: Oklahoma State University. Unpubl. Ph.D. thesis. 71 pp.

Hofmann, F. G. 1956. Observations on *in vitro* adrenal steroid biosynthesis in the albino mouse. Endocrinol. 59:712-715.

Hofmann, F. G. 1957. The *in vitro* hydroxylation of 21-carbon steroids by rat adrenal glands. *Endocrinol*. 60:382-389.

Huang, K. Y., W. W. Schultz and F. B. Gordon. 1968. Interferon induced by Plasmodium berghei. Science. 162:123-124.

Ingle, D. J. 1954. Permissibility of hormone action: a review. Acta Endocrinol. 17:172-186.

Jackson, G. J. 1955. The effect of cortisone on Plasmodium berghei infections in the white rat. J. Inf. Dis. 95:152-159.

Jahiel, R. J., J. Vicek, R. Nussenzweig and J. Vanderberg. 1968. Interferon inducers protect mice against Plasmodium berghei malaria. Science. 161:803-804.

Jensen, M. M. 1968. Transitory impairment of interferon production in stressed mice. J. Inf. Dis. 118:230-234.

Kass, E. H., M. I. Kendrick and M. Finland. 1953a. Effects of certain corticosteroids and of growth hormone on nucleoproteins of lymph nodes. Ann. N. Y. Acad. Sci. 56:737-741.

Kass, E. H., M. M. Lundgren and M. Finland. 1953b. Observations on the effect of corticosteroids and growth hormones on resistance to experimental pneumococcal and influenza virus infection. Ann. N. Y. Acad. Sci. 56:765-771.

Kelsall, M. A. and E. D. Crabb. 1958. Lymphocytes and plasmocytes in nucleoprotein metabolism. Ann. N. Y. Acad. Sci. 72:293-338.

Kemper, B. W., W. B. Pratt and L. Aronow. 1969. Nucleic acid synthesis in intact nuclei isolated from mouse fibroblasts: Characterization of the system and effects of gluccocorticoids. Mol. Pharmacol. 5:507-531.

Kerr, A. C. 1956. The effect of mental stress on the eosinophil leukocyte count in man. Quant. J. Exper. Physiol. 41:88-93.

Keuning, F. J. and L. B. Van der Slikke. 1950. The role of immature plasma cells, lymphoblasts, and lymphocytes in the formation of antibodies, as established in tissue culture experiments. J. Lab. Clin. Med. 36:167-182.

Kligman, A. M., G. D. Baldridge, G. Rebell and D. M. Pillsbury. 1951. The effect of cortisone on pathologic responses of guinea pigs infected cultaneously with fungi, viruses, and bacteria. J. Lab. Clin. Med. 37:615-620.

Krebs, C. J. 1964. The lemming cycle at Baker Lake, Northwest Territories, during 1959-1962. Arctic Inst. N. Am. Tech. Paper 15, 104 pp.

Lack, D. 1954. The natural regulation of animal numbers. Oxford Univ. Press. 343 pp.

LeMaistre, C. A., R. Tompsett and W. McDermott. 1953. The effects of corticosteroids upon tuberculosis and pseudotuberculosis. Ann. N. Y. Acad. Sci. 56:772-778.

Les, Edwin P. 1968a. Cage population density and efficiency of feed utilization in inbred mice. Lab. Anim. Care. 18:305-313.

Les, Edwin P. 1968b. Environmental factors influencing body weight of C57BL/6J and DBA/2J mice. Lab. Anim. Care. 18:623-625.

Les, Edwin P. 1972. A disease related to cage population density: tail lesions of C3H/HeJ mice. Lab. Anim. Care. 22:56-60.

Lidicker, W. Z. 1965. Comparative sutdy of density regulation in confined populations of four species of rodents. Res. on Pop. Ecology. 7:57-72.

Lissak, K. and E. Endroczi. 1965. Anatomical and functional connexions between the adenophypophysis and the adrenal cortex. In "The Neuroendocrine Control of Adaptation." Pergamon Press, London. Pp. 1-5.

Louch, C. D. 1956. Adrenocortical activity in relation to the density and dynamics of three confined populations of Microtus pennsylvanicus. Ecology. 37:701-713.

Louch, C. D. 1958. Adrenocortical activity in two meadow vole populations. J. Mammal. 39:109-116.

Louch, C. D., R. K. Meyer and J. T. Emlen. 1953. Effect of stress on diurnal fluctuations in eosinophils of the laboratory mouse. Proc. Soc. Exptl. Biol. Med. 82:668-671.

Louch, C. D. and M. Higginbotham. 1967. The relation between social rank and plasma corticosterone levels in mice. Gen. Comp. Endocrinol. 8:441-444.

Luft, J. H. 1965. The ultrastructural basis of capillary permeability. In "The Inflammatory Process," Zweifach, Grant and McClusky (eds.). Academic Press: New York. Pp. 153-155.

Macleod, A. G. 1970. Aspects of acute inflammation. Upjohn Publ.

Mason, J. W. 1959. Central nervous system factors in the regulation of endocrine secretion. Recent. Progr. in Hormone Research. 15:345-389.

McBride, G. 1971. Theories of animal spacing: the role of flight, fright, and social distance. In "Behaviour and Environment: the use of space by animals and men," A. H. Esser, (ed.). Plenum Press: New York. Pp. 53-68.

McCabe, T. T. and B. D. Blancard. 1950. Three species of Peromyscus. Rood Associates: Santa Barbara, CA.

McKeever, S. 1964. Variation in the weight of the adrenal, pituitary, and thyroid gland of the white-footed mouse, Peromyscus maniculatus. Am. J. Anat. 114:1-15.

McKinney, T. D. and J. J. Christian. 1970. Incidence and effects of botfly parasitism in the eastern chipmunk (Tamias striatus). J. Wildl. Dis. 6:140-143.

Meier, R., W. Schuler and P. Desavlles. 1950. Zur Frage des Mechanismus der Hemmung des Bindegewebswachstume durch Corstisone. Experientia. 6:469-471.

Menkin, V. 1959. Factors concerned in the mobilization of leukocytes in inflammation. Ann. N. Y. Acad. Sci. 59:956-958.

Miller, D. H. and L. L. Getz. 1969. Botfly infections in a population of Peromyscus leucopus. J. Mammal. 50:277-283.

Moore, K. E. 1968. Studies with chronically isolated rats. Tissue levels and urinary excretion of catecholamines and plasma levels of cortisone. Can. J. Physiol. Pharmacol. 46:553-558.

Moos, W. S., R. K. Clark and H. C. Mason. 1970. Behavior and physiology of mice in a closely controlled environment. Int. J. Biometerol. 14:133-154.

Munday, K. A. 1961. Asepcts of stress phenomena. In "Mechanisms of biological competition," Hartsone (ed.). Symp. Soc. Exp. Biol. 15:168-189.

Nelson, D. H. 1953. Eosinophils, lymphocytes, and 17-hydroxycorticosteroids as indices of adrenocortical activity. In "Bioassay of Anterior Pituitary and Adrenocortical Hormones," (Ciba Foundation Colloquia on Endocrinology). Little, Brown: Boston.

Nelson, D. H. 1955. The significance of plasma corticosteroids as a measure of adrenal cortical function. In "Symposium on Adrenal Functions in Infants and Children," N. Y. Univ. Med. Ctr., Syracuse, 1954. NMRI 55-1.

Ogilvie, B. M. 1965. Use of cortisone derivatives to inhibit resistance to Nippostrongylus brasiliensis and to study the fate of parasites in resistant hosts. Parasitology. 55:723-730.

Ogunsa, A. O., A. F. DeNicola, H. Traikov and M. K. Birmingham. 1971. Adrenal steroid biosynthesis by different species of mouselike rodents. Gen. Comp. Endocrinology. 16:192-199.

Olson, L. J. 1957. Studies on the resistance of white rats and cotton rats to infection with Litomosoides carnii. Diss. Abstr. 17(11): 2574. Univ. of Mich.

Pearson, P. G. 1962. The relationship between population density and plasma corticosterone levels in mice. Bull. Ecol. Soc. Amer. 43:134-139.

Plaut, S. M., R. Ader, S. B. Friedman and A. L. Ritterson. 1969. Social factors and resistance to malaria in the mouse. Effects of group vs. individual housing on resistance to Plasmodium berghei infection. Psychosom. Med. 31:536-552.

Plaut, S. M. and L. J. Grota. 1971. Effects of differential housing on adrenocortical activity. Neuroendocrin. 7:348-360.

Plaut, S. M., S. B. Friedman and L. J. Grota. 1971. Plasmodium berghei: resistance to infection in group and individually housed mice. Exp. Parasit. 29:47-52.

Pollard, M. and B. R. Wilson. 1955. The prolongation of murine typus rickettsemia by cortisone. Texas Repts. Biol. and Med. 13:126-128.

Ramaley, J. A. 1972. Changes in daily serum corticosterone values in maturing male and female rats. Steroids. 20:185-197.

Randolph, T. G. and C. L. Stanton. 1945. A comparison of differential counts from the stained film and counting chamber with glycol stain. Am. J. Clin. Tech. Suppl. 9:17-21.

Rapp, J. P. and J. J. Christian. 1963. Splenic extramedullary hematopoiesis in grouped male mice. Proc. Soc. Exptl. Biol. Med. 114:26-28.

Ratcliffe, H. L. and M. T. I. Cronin. 1958. Changing frequency of arteriosclerosis in mammals and birds at the Philadelphia Zoological Gardens. Circulation. 18:41-52.

Retzlaff, E. G. 1938. Studies in population physiology with the albino mouse. Biol. Generalis. 14:238-265.

Riegle, G. D. 1973. Chronic stress effects on adrenocortical responsiveness in young and aged rats. Neuroendocrinol. 11:1-10.

Robert, A. and J. E. Nezamis. 1957. The granuloma pouch as a routine assay for antiphlogistic compounds. Acta Endocrinol. 25:105-112.

Robinson, H. J. and A. L. Smith. 1953. The effect of adrenal cortical hormones on experimental infection. Ann. N. Y. Acad. Sci. 56: 757-764.

Roman, E. 1956. Specific parasitism of Strongyloides ratti, to the surmount, effects of the cortisone on the infestation of other rodents by this nematade. Ann. Parasitol. 31:552-571.

Rosemberg, E., J. Cornfield, R. W. Bates and E. Anderson. 1954. Bioassay of adrenal steroids in blood and urine based on eosinophil response: a statistical analysis. Endocrin. 54:363-383.

Sadlier, R. M. S. F. 1965. The relationship between agonistic behaviour and population changes in the deer mouse (Peromyscus maniculatus Wagner). J. Animal Ecol. 34:331-352.

Santisteban, G. A. and T. F. Dougherty. 1954. Comparison of the influence of adrenocortical hormones on the growth and involution of lymphatic organs. Endocrin. 54:130-146.

Sayers, G. and M. Sayers. 1949. The pituitary-adrenal system. Ann. N. Y. Acad. Sci. 50:522-539.

Schachter, M. 1969. Kallikreins and kinins. Physiol. Rev. 49:510-514.

Schalm, O. W. 1975. Veterinary hematology. 7th ed. Lea and Febiger, Philadelphia. 664 pp.

Schmidt, L. H. and W. L. Squires. 1951. The influence of cortisone on primate malaria. J. Exptl. Med. 94:501-520.

Schwartzman, G. 1952. Poliomyelitis infection in cortisone treated hamsters induced by i.p.r. Proc. Soc. Exp. Biol. and Med. 59: 573-576.

Schwartzman, G. and S. M. Aronson. 1953. Poliomyelitis infection by parenteral routes made possible by cortisone. Ann. N. Y. Acad. Sci. 56:793-798.

Selye, H. 1946. The general adaptation syndrome and the diseases of adaptation. J. Clin. Endocrin. 6:117-230.

Selye, H. 1950. The physiology and pathology of stress. Acta, Inc., Montreal. 1025 pp.

Selye, H. 1951. The influence of STH, ACTH, and cortisone upon resistance to infection. Can. Med. Assoc. J. 64:489-494.

Selye, H. and P. Bois. 1954. On the design of experiments to appraise the role of the adrenals in biologic reactions. In "4th Annual Report on Stress," (H. Selye and G. Heuser, eds.), pp. 533-522. Acta, Montreal.

Selye, H. 1955. Interactions between corticoid and folliculoid hormones in the regulation of anabolism, lymphatic tissue and inflammation. Acta Endocrinol. 20:1-18.

Selye, H. 1956. The stress of life. New York: McGraw-Hill Company.

Sigg, E. B., C. Day and C. Colombo. 1966. Endocrine factors in isolation-induced aggressiveness in rodents. Endocrinol. 78:679-684.

Silber, R. H., R. D. Busch and R. Oslapas. 1958. Practical procedure for estimation of corticosterone or hydrocortisone. Clin. Chem. 4:278-285.

Singer, I. 1954. The effect of cortisone on infections with Plasodium berghei in the white mouse. J. Inf. Dis. 94:164-172.

Snedecor, G. W. and W. G. Cochran. 1967. Statistical Methods, 6th ed. Ames, Iowa: Iowa State University Press.

Solomon, G. F., T. C. Merigan and S. Levine. 1967. Variation in adrenal cortical hormones within psychologic ranges, stress, and interferon production in mice. Proc. Soc. Exp. Biol. Med. 126: 74-79.

Sorensen, B. M. 1966. Influence of glucocorticoid and anabolic steroid hormones on the wound healing process. In "Hormones and Connective Tissue," (G. Asboe-Hansen, ed.), pp. 106-123. Munksgaard Press: Copenhagen, Denmark.

Southwick, C. H. 1955a. The population dynamics of confined house mice supplied with unlimited food. Ecology. 36:212-225.

Southwick, C. H. 1955b. The regulatory mechanisms of house mouse population; social behavior affecting litter survival. Ecology. 36:627-634.

Southwick, C. H. 1959. Eosinophil response of C57BR mice to behavioral disturbance. Ecology. 40:156-157.

Southwick, C. H. 1964. Peromyscus leucopus: an interesting subject for studies of socially induced stress response. Science. 143: 55-56.

Speirs, R. S. 1955. Physiological approaches to an understanding of the function of eosinophils and basophils. Ann. N. Y. Acad. Sci. 59:706-731.

Speirs, R. S. and R. K. Meyer. 1949. The effects of stress, adrenal and adrenocorticotrophic hormones on the circulating eosinophils of mice. Endocrin. 45:403-429.

Speirs, R. S. and R. K. Meyer. 1951. A method of assaying adrenal cortical hormones based on a decrease in the circulating eosinophil cells of adrenalectomized mice. Endocrin. 48:316-326.

Stoner, R. D. and J. T. Godwin. 1953. The effects of ACTH and cortisone upon susceptibility to trichinosis in mice. Am. J. Pathol. 29: 943-950.

Strecker, R. L. 1954. Regulatory mechanics in house mouse populations: the effect of limited food supply on an unconfined population. Ecology. 35:249-253.

Strecker, R. L. and J. T. Emlen. 1953. Regulatory mechanisms in house mouse populations--the effect of limited food supply on a confined population. Ecology. 34:375-385.

Sundberg, R. D. 1955. Lymphocytes and plasma cells. Ann. N. Y. Acad. Sci. 59:671-689.

Taubenhaus, M. and D. Amromin. 1950. The effects of hypophysis, thyroid, sex, steroids and the adrenal cortex upon granulation tissue. J. Lab. Clin. Med. 36:7-18.

Tepperman, J. 1971. Metabolic and Endocrine Physiology. Year Book Medical Publishers, Inc.: Chicago. 250 pp.

Terman, C. R. 1965a. A study of population growth and control exhibited in the laboratory by prairie deermice. Ecology. 46: 890-895.

Terman, C. R. 1965b. Adrenal gland weights of prairie deermice from populations with a long duration of asymptote. Am. Zoologist. 5: 656.

Thiessen, D. D. 1966. Role of physical injury in the physiological effects of population density in mice. J. Comp. Physiol. Psych. 62:322-324.

Thomas, L. 1953. Cortisone and infection. Ann. N. Y. Acad. Sci. 56: 799-814.

Thorn, G. W., D. Jenkins and J. C. Laidlaw. 1953. The adrenal response to stress in man. Recent Progr. in Hormone Research. 8:171-215.

Tobach, E. and H. Block. 1956. Effect of stress by crowding prior to and following tuberculosis infection. Am. J. Physiol. 187:399-402.

Treiman, D. M. and S. Levine. 1969. Plasma corticosteroid response to stress in four species of wild mice. Endocrinol. 84:676-680.

Vandenbergh, J. G. 1960. Eosinophil response to aggressive behavior in CFW albino mice. Animal Behavior. 8:13-18.

Vessey, S. H. 1964. Effects of grouping on levels of circulatory antibodies in mice. Proc. Soc. Exp. Biol. Med. 115:252-255.

Visscher, M. B. and F. Halberg. 1955. Daily rhythms in numbers of circulating eosinophils and some related phenomena. Ann. N. Y. Acad. Sci. 59:834-849.

Weaver, J. A. 1955. Changes induced in the thymus and lymph nodes of the rat by the administration of cortisone and sex hormones and by other procedures. J. Pathol. Bacteriol. 69:133-139.

Weinstein, P. P. 1953. The effect of cortisone on the development of the immune response in the white rat to Nippostrongylus muris. J. Parasit. 39(Suppl.):86-90.

Weinstein, P. P. 1955. The effect of cortisone on the immune response of the white rat to Nippostrongylus muris. Am. J. trop. Med. Hyg. 4:61-74.

Weissman, G. and L. Thomas. 1964. The effects of corticosteroids upon connective tissue and lysosomes. Rec. Prog. Hormone Res. 20:215-245.

Welch, B. L. 1947. The generalization of student's problem when several different population variances are involved. Biometrika. 34:28-35.

Welch, B. L. and P. H. Klopfer. 1961. Endocrine variability as a factor in the regulation of population genetics. Am. Naturist. 95:256-260.

Welch, B. L. and A. Welch. 1969. Sustained effects of brief daily stress (fighting) upon brain and adrenal catecholamines and adrenal, spleen, and heart weights of mice. Proc. Nat. Acad. Sci. 64:100-107.

Wells, P. D. 1962. Mast cells, eosinophil, and histamine levels in Nippostrongylus brasiliensis infected rats. Exp. Parasitol. 12: 82-101.

Weltman, A. S., A. M. Sackler, S. B. Sparber and S. Opert. 1962. Endocrine aspects of isolation stress on female mice. Fed. Proc. 21:184-186.

Weltman, A. S., A. M. Sackler and S. B. Sparber. 1966. Endocrine, metabolic, and behavioral aspects of isolation stress in female albino mice. Aerospace Med. 37:804-810.

Weltman, A. S., A. M. Sackler, R. Schwartz and S. Roman. 1967. Effects of isolation on maternal aggressiveness and body growth rates of offspring. Experentia. 23:782-784.

Weltman, A. S., A. M. Sackler, R. Schwartz and H. Owens. 1968. Effects of isolation stress on female albino mice. Lab. Anim. Care. 18: 426-435.

Whitney, O. M. and L. Anigstein. 1953. Effect of cortisone on experimental ricketsioses and on antibody formation. Texas Repts. Biol. and Med. 11:561-569.

Wilson, E. D. and A. A. Dewees. 1962. Body weights, adrenal weights, and oestrous cycles of Nutria. J. Mammal. 43:362-368.

Wilson, H., J. J. Borris and R. C. Bahn. 1958. Steroids in the blood and urine of female mice bearing on ACTH-producing pituitary humor. Endocrinol. 62:135-149.

Wintrobe, M. M. 1961. Clinical hematology. Lea and Febiger, Philadelphia. 1186 pp.

Wynne-Edwards, V. C. 1962. Animal dispersion in relation to social behavior. Hatner Publishing Company, New York. 653 pp.

Wynne-Edwards, V. C. 1965. Self-regulating systems in populations of animals. Science. 147:1543-1548.

Wynne-Edwards, V. C. 1966. Regulation in animals societies and population. In "Regulation and Control in Living Systems (edited by H. Kalman). John Wiley and Sons Ltd., New York. Pp. 397-421.

Zweifach, B. W., L. Grant and R. T. McCluskey. 1965. The Inflammatory Process. Academic Press: New York. 931 pp.

APPENDIX

TABLE X

ADRENAL PARAMETERS FOR THE GROUP SIZE OF 1 MOUSE PER CAGE

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
01	A	01	13.0	0.0	52.0	13.5	21.5	628.4	11.3	39.5
02	A	02	36.0	2.0	30.5	11.0	20.5	643.9	11.7	56.0
03	A	03	11.0	0.0	27.5	15.5	46.0	569.2	11.6	48.0
04	A	04	20.5	0.0	44.0	17.0	18.5	684.3	11.2	31.0
05	A	05	9.0	0.5	59.5	4.5	26.5	578.8	11.4	42.0
06	A	06	13.5	1.0	67.0	0.0	18.5	705.4	10.9	27.0
07	A	07	11.5	2.0	38.5	15.0	33.0	618.8	10.8	29.5
08	A	08	30.5	0.0	23.0	3.0	43.5	553.1	12.0	62.5
09	A	09	7.5	0.0	57.5	11.0	24.0	596.0	11.5	41.0
10	A	10	24.0	0.0	36.0	12.0	18.0	649.4	11.2	33.0
11	A	11	4.5	2.0	42.0	11.5	40.0	645.9	11.5	47.5
12	A	12	28.5	0.0	58.0	7.0	6.5	637.5	11.6	45.0
13	A	13	12.0	0.0	47.5	14.5	26.0	692.4	11.8	58.0
14	A	14	14.5	0.0	25.5	17.5	42.5	611.7	11.6	51.0
15	A	15	22.0	1.5	61.0	13.0	2.5	554.6	11.7	52.0
16	A	16	21.0	1.0	41.5	5.5	31.5	622.9	11.2	36.5
17	A	17	6.5	0.0	33.0	7.5	53.0	707.3	10.9	29.5
18	A	18	28.0	0.0	37.0	2.0	33.0	524.2	11.9	62.5
19	A	19	4.0	0.0	35.5	14.0	46.5	550.8	11.8	59.0
20	A	20	15.5	1.0	11.0	13.0	59.5	629.6	10.8	28.0

TABLE X (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
21	A	21	34.0	0.0	31.0	6.5	28.5	602.4	11.3	44.0
22	A	22	20.5	0.5	25.0	2.5	51.5	671.7	11.1	36.0
23	A	23	8.0	1.0	2.5	16.0	72.5	628.0	11.6	46.0
24	A	24	13.5	0.5	4.5	10.0	71.5	653.2	11.5	40.5
25	A	25	19.5	0.0	51.5	8.5	20.5	599.9	11.8	54.0
26	A	26	25.0	0.0	35.0	15.5	24.5	590.6	11.7	48.0
27	A	27	11.0	1.0	20.5	13.5	54.0	685.1	10.6	24.0
28	A	28	29.5	0.5	39.0	12.5	18.5	545.3	11.9	58.0
29	A	29	10.0	0.0	37.0	11.0	42.0	701.4	10.6	26.0
30	A	30	35.5	0.5	32.0	1.5	30.5	628.2	11.2	37.0
31	A	31	1.0	0.0	31.5	17.0	50.5	640.1	11.9	59.0
32	A	32	6.5	1.5	39.0	1.0	52.0	580.4	11.6	52.0
33	A	33	17.5	0.0	21.0	3.5	58.0	667.7	11.4	47.0
34	A	34	8.0	0.0	17.5	6.5	68.0	645.7	11.6	63.0
35	A	35	13.5	0.0	62.5	17.5	7.0	583.0	11.5	55.5
36	A	36	2.0	0.0	27.5	12.0	58.5	619.5	12.1	63.5
37	A	37	18.0	1.0	39.0	15.5	26.5	629.2	11.9	58.5
38	A	38	27.0	2.0	48.0	7.5	15.5	676.2	10.4	24.5
39	A	39	30.0	1.5	54.0	3.0	11.5	652.7	11.3	42.0
40	A	40	6.0	0.0	25.5	13.0	55.5	654.2	11.3	43.0

TABLE X (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
41	A	41	3.0	0.0	34.0	15.0	48.0	695.7	11.1	40.0
42	A	42	4.0	0.0	24.0	9.5	62.5	607.0	11.5	53.5
43	A	43	21.0	1.0	33.5	18.0	26.5	672.1	11.4	43.0
44	A	44	9.5	0.0	37.0	14.5	39.0	639.1	11.4	45.5
45	A	45	11.5	0.5	56.5	6.5	25.0	676.6	11.6	45.5
46	A	46	17.5	0.5	68.0	1.0	13.0	673.8	11.8	52.5
47	A	47	14.0	2.0	10.0	14.0	55.0	693.3	12.0	60.0
48	A	48	5.5	0.0	31.0	17.0	46.5	685.1	11.3	41.5

TABLE XI

ADRENAL PARAMETERS FOR THE GROUP SIZE OF 2 MICE PER CAGE

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
01	A	01	24.5	0.0	60.0	8.0	7.5	666.6	7.9	36.0
02	B	01	13.5	0.5	59.5	15.5	11.0	654.4	10.1	42.0
03	A	02	19.0	0.0	63.0	7.5	10.5	689.6	8.0	39.0
04	B	02	13.0	1.5	38.0	15.0	31.5	639.2	8.2	44.0
05	A	03	22.0	1.5	55.5	10.0	11.0	628.4	9.3	45.5
06	B	03	27.5	0.0	26.5	11.0	35.0	682.8	9.4	47.0
07	A	04	20.5	0.0	58.5	10.0	11.0	679.1	7.6	27.5
08	B	04	6.5	0.0	38.0	14.0	41.5	662.1	7.9	41.0
09	A	05	35.5	0.5	51.5	6.0	7.0	635.9	7.8	25.0
10	B	05	6.0	0.0	23.5	6.5	64.0	606.4	8.2	45.0
11	A	06	21.5	0.0	45.5	18.0	15.0	767.6	7.0	32.0
12	B	06	14.0	0.5	34.0	5.0	46.5	754.9	8.1	36.0
13	A	07	23.5	0.0	67.5	4.5	4.5	685.5	7.8	26.5
14	B	07	4.5	0.5	20.5	16.5	58.0	669.5	9.2	36.0
15	A	08	36.5	0.0	44.0	15.0	4.5	708.1	8.1	38.0
16	B	08	32.0	0.0	29.5	1.5	37.0	673.6	8.4	43.0
17	A	09	22.0	1.0	25.0	17.5	24.5	744.0	9.6	34.5
18	B	09	36.0	0.0	34.0	13.0	17.0	656.3	10.6	46.0
19	A	10	20.5	0.5	48.5	18.5	12.0	709.5	9.4	30.0
20	B	10	33.0	2.0	36.5	2.0	26.5	677.0	9.8	35.0

TABLE XI (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
21	A	11	16.0	0.0	53.5	17.0	13.5	691.9	7.6	23.0
22	B	11	9.0	0.0	40.0	2.0	49.0	641.1	8.0	36.0
23	A	12	2.5	1.0	37.5	4.0	54.5	668.0	7.9	33.0
24	B	12	4.0	0.5	14.5	14.5	66.5	625.6	8.1	42.0
25	A	13	35.5	1.0	45.0	17.5	1.0	679.5	7.7	25.0
26	B	13	16.5	1.0	34.0	11.5	37.0	654.1	8.6	31.0
27	A	14	26.5	0.0	52.0	15.5	6.0	728.2	7.7	25.0
28	B	14	31.0	0.0	30.0	14.5	24.5	640.0	8.2	46.0
29	A	15	27.0	1.0	53.0	3.5	15.5	663.6	7.0	26.0
30	B	15	7.5	0.0	66.0	2.5	24.0	659.5	8.6	44.5
31	A	16	11.0	0.0	36.5	18.0	39.5	718.1	7.7	27.5
32	B	16	19.0	2.0	42.0	10.0	27.0	659.2	8.0	40.5
33	A	17	22.0	1.0	58.5	0.5	18.0	641.2	7.9	33.0
34	B	17	8.0	0.0	29.5	10.0	52.5	632.2	8.2	39.0
35	A	18	13.5	0.0	59.0	16.0	6.5	637.6	9.1	24.0
36	B	18	3.0	1.5	39.5	11.0	45.0	616.4	9.8	47.5
37	A	19	32.0	0.0	27.0	7.5	33.5	639.6	9.0	42.0
38	B	19	2.0	0.0	31.0	10.5	56.5	643.0	9.2	46.0
39	A	20	31.5	0.0	48.0	6.5	14.0	744.1	7.8	31.0
40	B	20	17.5	1.5	29.5	10.0	41.5	669.3	8.2	45.0

TABLE XI (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
41	A	21	24.0	0.5	22.5	6.0	47.0	680.3	7.9	27.5
42	B	21	26.5	0.0	51.0	15.5	7.0	670.9	8.0	30.0
43	A	22	32.5	0.0	48.5	16.0	3.0	613.4	7.8	26.0
44	B	22	3.0	0.0	37.5	0.5	59.0	621.8	8.4	38.0
45	A	23	31.0	0.0	47.0	14.0	8.0	717.1	8.1	33.0
46	B	23	20.0	2.0	38.0	12.0	28.0	624.2	8.7	42.0
47	A	24	28.5	0.0	45.5	11.5	14.5	638.7	9.6	32.0
48	B	24	5.5	0.5	69.5	8.5	16.0	674.5	9.6	32.5

TABLE XII

ADRENAL PARAMETERS FOR THE GROUP SIZE OF 4 MICE PER CAGE

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
01	A	01	30.0	0.0	52.0	6.5	11.5	705.6	7.9	27.0
02	B	01	18.0	0.0	60.0	16.5	5.5	699.8	9.3	44.0
03	B	01	21.0	0.0	64.0	7.5	23.0	699.5	9.5	42.5
04	W	01	5.5	2.0	17.0	17.0	58.5	643.9	10.1	52.5
05	A	02	26.0	0.0	30.0	15.0	29.0	722.2	7.6	21.0
06	B	02	28.5	0.5	25.5	8.0	37.5	684.1	9.2	45.0
07	B	02	9.0	1.0	24.0	6.0	60.0	667.5	8.9	36.0
08	W	02	10.0	0.0	23.5	9.0	57.5	637.9	9.7	47.0
09	A	03	34.0	2.0	36.0	8.5	19.5	700.3	7.8	29.0
10	B	03	15.0	0.0	55.5	6.5	23.0	671.2	9.5	34.0
11	B	03	9.5	0.5	12.5	16.0	61.5	672.6	9.4	39.0
12	W	03	10.5	1.0	21.5	15.5	51.5	646.7	10.3	55.5
13	A	04	29.0	0.0	47.0	7.0	17.0	705.8	8.1	26.0
14	B	04	9.0	0.0	23.5	9.5	58.0	689.0	9.2	35.0
15	B	04	8.0	0.0	44.0	1.0	47.0	654.0	9.6	43.5
16	W	04	3.0	0.5	42.5	3.5	50.5	639.9	9.9	51.0
17	A	05	33.5	0.0	38.5	9.5	18.5	727.8	8.4	27.0
18	B	05	32.0	0.0	32.5	14.0	21.5	676.3	9.3	32.0
19	B	05	13.0	2.0	52.5	15.5	17.0	640.6	9.7	47.0
20	W	05	24.5	0.0	51.0	5.0	19.5	655.3	10.5	53.0

TABLE XII (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
21	A	06	7.5	0.0	32.0	0.5	60.0	681.9	8.2	25.0
22	B	06	7.0	0.0	43.5	6.0	43.5	679.3	9.4	35.0
23	B	06	10.0	0.0	22.5	7.5	60.0	662.1	9.5	42.5
24	W	06	5.0	0.0	24.5	14.5	56.0	651.9	9.6	48.5
25	A	07	26.0	0.0	53.0	4.0	17.0	706.5	8.1	29.0
26	B	07	17.0	0.0	46.5	4.5	32.0	678.4	9.4	44.0
27	B	07	14.5	0.0	59.5	4.0	22.0	657.2	9.0	37.0
28	W	07	2.5	1.5	29.0	17.0	50.0	618.8	9.4	49.0
29	A	08	20.5	1.0	26.5	1.0	51.0	668.3	8.6	29.0
30	B	08	19.0	2.0	36.0	6.5	36.5	680.9	9.6	45.0
31	B	08	1.0	0.0	23.5	12.0	63.5	655.5	9.4	31.5
32	W	08	3.0	0.0	35.5	7.0	54.5	641.1	10.0	51.5
33	A	09	24.5	0.0	53.5	14.0	8.0	709.6	7.9	24.0
34	B	09	7.0	0.0	45.5	17.0	30.5	667.4	8.5	42.0
35	B	09	13.5	0.0	38.5	0.5	47.5	650.5	9.1	36.5
36	W	09	2.0	0.0	34.0	14.0	50.0	641.6	10.3	54.0
37	A	10	19.0	0.0	44.5	13.5	23.0	688.3	8.8	28.5
38	B	10	11.0	1.5	43.5	6.0	38.0	684.1	9.0	35.0
39	B	10	1.5	0.5	33.0	11.5	53.5	665.1	9.2	42.0
40	W	10	5.0	2.0	15.0	11.0	67.0	645.3	9.4	49.0

TABLE XII (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
41	A	11	28.5	0.0	48.5	15.0	8.0	717.1	8.9	30.0
42	B	11	25.5	0.5	52.5	9.0	28.0	679.0	9.3	42.5
43	B	11	14.0	0.0	30.5	15.5	40.0	636.0	9.2	37.0
44	W	11	6.5	0.0	21.5	3.0	69.5	624.4	9.5	46.0
45	A	12	33.0	0.0	28.5	15.5	23.0	690.4	9.1	32.5
46	B	12	16.0	0.5	54.5	8.5	20.5	682.3	9.4	35.0
47	B	12	19.5	0.0	53.0	9.5	18.0	654.8	9.5	39.0
48	W	12	6.0	2.0	31.0	5.5	55.5	621.6	9.8	48.0

TABLE XIII

ADRENAL PARAMETERS FOR THE GROUP SIZE OF 8 MICE PER CAGE

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
01	A	01	28.5	2.0	56.0	9.5	4.0	854.9	9.2	29.0
02	B	01	35.0	0.0	35.5	10.5	19.0	649.1	10.6	43.0
03	B	01	35.0	0.5	38.0	0.5	26.0	645.9	10.9	50.0
04	B	01	10.5	1.0	56.0	5.0	27.5	636.4	11.0	54.0
05	B	01	26.0	0.0	64.0	13.0	3.0	666.6	11.7	63.0
06	B	01	24.0	0.0	35.0	4.0	37.0	670.3	11.8	66.0
07	B	01	8.0	0.0	59.5	11.0	21.5	618.3	11.1	57.0
08	W	01	9.5	0.0	44.5	8.0	38.0	602.9	15.3	71.0
09	A	02	16.0	0.0	48.5	5.5	30.0	792.1	8.8	24.0
10	B	02	7.5	1.5	11.0	13.5	66.5	643.4	9.8	39.5
11	B	02	11.5	0.0	25.0	2.0	61.5	605.1	12.0	56.0
12	B	02	33.0	2.0	29.5	14.0	21.5	642.1	14.2	65.0
13	B	02	5.0	1.5	30.0	10.0	53.5	652.5	13.6	59.0
14	B	02	2.0	0.0	28.5	12.5	57.0	618.3	13.7	61.5
15	B	02	15.5	0.0	36.0	17.5	31.0	659.6	13.4	59.5
16	W	02	7.0	2.0	7.0	5.0	79.0	583.2	15.6	73.0
17	A	03	33.0	0.0	56.5	5.5	5.0	801.7	9.6	27.5
18	B	03	36.0	0.0	32.5	4.0	27.5	711.2	10.9	39.0
19	B	03	32.5	0.0	4.5	15.0	48.0	694.1	11.3	34.5
20	B	03	23.0	0.0	33.5	15.0	28.5	668.5	11.7	40.0

TABLE XIII (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
21	B	03	21.0	1.0	17.5	12.0	48.5	675.9	12.6	55.0
22	B	03	11.5	2.0	24.5	18.0	44.0	653.3	12.5	52.0
23	B	03	14.0	0.0	34.0	1.5	50.5	685.4	12.8	55.0
24	W	03	2.0	2.0	41.5	9.0	45.5	596.7	14.7	67.5
25	A	04	10.0	0.5	41.0	18.0	30.5	684.8	9.1	31.0
26	B	04	26.0	0.0	39.5	2.5	32.0	633.6	11.3	57.0
27	B	04	10.5	0.5	36.5	0.5	52.0	601.2	12.4	67.0
28	B	04	31.0	0.0	33.5	14.5	21.0	686.4	12.4	69.0
29	B	04	4.0	0.5	42.5	15.0	38.0	614.8	10.2	31.0
30	B	04	22.0	0.0	23.0	6.5	48.5	638.1	12.5	57.0
31	B	04	30.5	0.0	45.0	17.0	7.5	631.4	12.9	67.0
32	W	04	25.0	0.5	16.0	0.5	58.0	537.3	14.4	69.0
33	A	05	13.0	0.5	38.0	2.0	46.5	723.7	9.0	29.0
34	B	05	11.5	0.0	28.5	1.5	58.5	609.3	11.5	51.0
35	B	05	8.5	0.0	2.0	16.0	73.5	613.0	13.7	60.5
36	B	05	22.0	0.0	39.5	12.5	26.0	616.1	13.6	59.0
37	B	05	34.5	0.5	5.0	15.5	44.5	634.7	11.8	43.5
38	B	05	26.5	0.0	59.0	8.5	6.0	615.3	10.1	46.5
39	B	05	20.0	0.0	44.0	2.5	33.5	628.6	12.4	59.0
40	W	05	6.0	0.5	34.0	6.5	53.0	511.2	14.7	68.0

TABLE XIII (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
41	A	06	10.0	0.0	6.0	15.0	69.0	692.5	9.9	23.5
42	B	06	7.0	2.0	41.5	6.5	43.0	673.8	11.2	37.0
43	B	06	17.0	0.0	34.0	19.0	30.0	634.9	12.3	43.0
44	B	06	29.5	2.0	31.0	17.0	20.5	673.0	13.7	40.0
45	B	06	29.5	0.0	57.0	13.0	0.5	685.8	13.6	49.5
46	B	06	8.0	0.0	17.5	0.5	74.0	636.2	13.9	58.0
47	B	06	2.5	0.0	21.5	3.5	72.5	602.6	14.4	65.0
48	W	06	5.5	0.0	5.5	8.5	80.5	607.9	14.8	80.0

TABLE XIV

ADRENAL PARAMETERS FOR THE GROUP SIZE OF 16 MICE PER CAGE

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
01	A	01	14.0	0.5	60.0	1.5	24.0	830.5	9.1	25.0
02	B	01	27.0	0.0	33.5	0.5	39.0	757.8	9.7	35.0
03	B	01	3.0	0.0	54.0	5.0	38.0	683.9	10.2	37.0
04	B	01	34.5	0.1	31.0	14.0	19.5	643.8	10.0	32.5
05	B	01	5.5	0.0	17.0	15.5	62.0	686.1	10.4	29.0
06	B	01	21.5	0.0	22.0	5.5	51.0	608.4	11.8	40.5
07	B	01	31.0	0.0	39.0	3.0	27.0	654.8	11.5	37.0
08	B	01	24.0	0.5	10.5	6.0	59.0	630.3	12.2	47.0
09	B	01	30.0	0.0	30.0	10.5	29.5	673.2	11.4	45.0
10	B	01	3.5	0.0	40.5	6.5	49.5	675.1	12.0	61.0
11	B	01	8.0	0.0	45.0	16.5	30.5	640.6	11.7	60.0
12	B	01	1.0	0.0	31.0	12.0	56.0	627.8	10.5	44.5
13	B	01	22.0	0.0	29.0	15.0	34.0	699.3	10.8	47.0
14	B	01	11.5	1.0	24.0	17.5	46.0	663.5	11.9	53.5
15	B	01	30.5	1.0	29.5	11.5	27.5	655.4	10.6	48.0
16	W	01	25.0	0.0	25.5	12.5	37.0	573.5	15.6	79.0
17	A	02	31.0	0.0	24.5	0.5	44.0	805.0	9.3	27.0
18	B	02	24.0	1.0	24.5	12.0	38.5	763.5	10.1	36.5
19	B	02	28.5	0.0	29.5	7.5	34.5	730.3	10.0	39.5
20	B	02	24.5	1.0	37.5	14.5	22.5	670.8	11.4	45.0

TABLE XIV (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
21	B	02	34.0	0.0	11.5	8.0	46.5	619.9	11.2	45.5
22	B	02	8.0	0.0	10.5	10.5	71.0	671.7	11.3	48.0
23	B	02	36.0	0.0	34.5	9.0	20.5	662.5	13.7	62.0
24	B	02	0.5	1.0	19.5	18.5	60.5	676.8	13.6	62.0
25	B	02	24.5	0.0	18.5	10.5	46.5	659.0	12.9	55.0
26	B	02	5.0	0.5	25.0	12.0	57.5	657.1	13.5	68.0
27	B	02	7.5	0.0	41.5	10.0	41.0	680.4	14.4	72.5
28	B	02	15.0	2.0	32.5	15.0	35.5	648.2	13.7	65.0
29	W	02	19.0	0.0	22.0	13.0	46.0	546.9	14.8	77.5
30	W	02	11.0	2.0	23.5	16.5	47.0	556.6	16.1	80.5
31	W	02	23.5	0.0	42.5	8.0	26.0	523.3	15.9	83.0
32	W	02	18.0	0.0	32.5	14.5	35.0	532.4	14.8	68.0
33	A	03	36.0	0.0	25.5	1.0	37.5	785.6	9.8	30.0
34	B	03	20.5	0.0	50.0	11.5	18.0	635.4	10.3	33.0
35	B	03	29.0	1.5	38.0	11.5	20.0	540.2	14.8	71.0
36	B	03	13.0	2.0	37.5	6.0	41.5	625.7	12.5	45.0
37	B	03	13.5	0.5	40.5	12.0	33.5	656.3	11.9	49.0
38	B	03	34.0	0.0	12.0	1.0	53.0	679.2	12.6	51.0
39	B	03	5.0	0.0	29.5	16.5	49.0	686.0	12.5	55.5
40	B	03	27.0	0.0	25.0	2.5	45.5	642.6	12.1	36.0

TABLE XIV (Continued)

Mouse No.	Social Rank	Cage No.	Differentials in Percentages					Granuloma Wt. (mg)	Dried Adrenal Wts. (mg/100 g body wt.)	Plasma Corticosterone (µg/100 ml plasma)
			Eos.	Bas.	Lym.	Mon.	Neu.			
41	B	03	16.5	0.0	16.5	10.0	57.0	614.8	13.7	58.0
42	B	03	10.0	0.0	40.5	14.0	35.5	618.9	13.6	62.0
43	B	03	10.5	0.0	28.5	11.0	50.0	661.0	13.2	66.0
44	B	03	10.5	1.0	24.5	13.5	50.5	611.8	13.8	67.0
45	B	03	11.0	0.5	4.5	1.5	82.5	668.1	12.9	64.5
46	B	03	19.0	2.0	20.5	3.5	55.0	594.5	13.6	67.5
47	W	03	17.5	0.0	28.5	9.5	44.5	575.9	14.7	69.0
48	W	03	4.0	0.0	23.0	5.0	68.0	554.4	15.1	79.0

TABLE XV

RECOVERY OF STABLE CORTICOSTERONE

Tube Number	% Recovery of Stable Corticosterone
1	99.0
2	103.4
3	90.2
4	94.7
5	94.7
6	97.6
7	91.8
8	95.0
9	94.7
10	95.0
Mean $\pm$ S.E.	95.61 $\pm$ 3.69

VITA

James Steven Walker

Candidate for the Degree of

Master of Science

Thesis: THE EFFECTS OF CROWDING UPON SOME ADRENAL RESPONSES IN MALE WHITE-FOOTED MICE (PEROMYSCUS LEUCOPUS)

Major Field: Zoology

Biographical:

Personal Data: Born in Hobart, Oklahoma, 4 February 1951, the son of Mr. and Mrs. James C. Walker.

Education: Graduated from Snyder High School, Snyder, Oklahoma, in May, 1969; received the Bachelor of Science degree from Oklahoma State University with a major in Zoology in May, 1973; completed the requirements for the Master of Science degree from Oklahoma State University with a major in Zoology in July, 1975.

Professional Experience: Graduate Teaching Assistant, Department of Zoology, Oklahoma State University, Stillwater, Oklahoma, August, 1973 to May, 1974; Curator and Cataloger, Oklahoma State University Museum, Summer, 1974; Graduate Teaching Assistant, Department of Biological Sciences, Oklahoma State University, Stillwater, Oklahoma, August, 1974 to May, 1975.

Professional Organizations: American Society of Mammalogists.