THE EFFECTS OF DIFFERING PRE-PUBERTY EXERCISE PROGRAMS ON SELECTED MEASURES OF GROWTH IN THE MALE ALBINO RAT

Thesis for the Degree of Ph. D.
MICHIGAN STATE UNIVERSITY
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1966



This is to certify that the

thesis entitled

THE EFFECTS OF DIFFERING PRE-PUBERTY
EXERCISE PROGRAMS ON SELECTED MEASURES
OF GROWTH IN THE MALE ALBINO RAT

presented by

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has been accepted towards fulfillment of the requirements for

Ph.D. degree in Physical Education

Date __Ju

ABSTRACT

THE EFFECTS OF DIFFERING PRE-PUBERTY EXERCISE PROGRAMS ON SELECTED MEASURES OF GROWTH IN THE MALE ALBINO RAT

by Jerome Charles Weber

The purpose of this study was to determine the effects of differing pre-puberty exercise programs upon selected measures of growth in the male albino rat. These measures were total body weight, tibia length, serum cholesterol level, and the relative weights of the heart, spleen, liver, testes, kidneys, and adrenals.

Three groups of male albino rats were assigned to exxerimental regimens as follows: (1) a sedentary group with no access to activity, (2) a voluntary activity group with free access to a rotating exercise wheel, and (3) a voluntary activity plus forced exercise group under the same conditions as group two and also forced to swim for 30 minutes each day with an overload of two percent of the animal's body weight attached to the base of the tail. The animals continued their respective experimental procedures for a period of 35 days.

At the conclusion of the experimental period the rats were sacrificed by ether anesthesia, total body weight was determined, the organs were weighed after removal and cleaning, the tibia was excised and measured, and a blood

sample was taken from the orbital sinus for determination of serum cholesterol level.

A one-way fixed-effects model of the analysis of variance was used to determine if significant differences existed between groups at the .05 level of confidence. If the F ratio showed overall significance, the Scheffé test was used to determine between which means these differences existed.

The results of the present study indicate that the type of exercise program to which male albino rats are subjected in the pre-puberty period of life is a significant variable in determining total body weight at puberty, testes, kidneys, and adrenals weight as percentage of body weight at puberty, length of the tibia at puberty, and serum cholesterol level at puberty.

The results also indicate that the type of exercise program to which male albino rats are subjected in the prepuberty period of life is not a significant variable in determining heart, liver, or spleen weight as percentage of body weight at puberty.

THE EFFECTS OF DIFFERING PRE-PUBERTY EXERCISE PROGRAMS ON SELECTED MEASURES OF GROWTH IN THE MALE ALBINO RAT

By

Jerome Charles Weber

A THESIS

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

Department of Health, Physical Education and Recreation

DEDICATION

To Barbara, for all the things that make a man's life worthwhile, meaningful, and happy.

ACKNOWLEDGMENTS

The author wishes to acknowledge the aid given him by the members of the Department of Health, Physical Education and Recreation. Particular appreciation is expressed to Dr. Wayne D. Van Huss, Dr. William W. Heusner, and Dr. Gale E. Mikles who were of great help throughout the author's tenure as a student. In addition, the author also wishes to thank Dr. Stanley C. Ratner of the Department of Psychology and Dr. John C. Howell of the Department of Sociology for their aid and encouragement.

The author wishes to further acknowledge the abovenamed people for their instruction in the meaning of professionalism and integrity.

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CHAPTER I

INTRODUCTION AND STATEMENT OF THE PROBLEM

INTRODUCTION

The fact that children voluntarily exercise can be noted by observation of any normal, healthy group of youngsters in almost any informal setting. It has generally been assumed that exercise is essential for proper growth and development. (16, 25, 31, 38, 80, 94, 110) However, there has been little quantitative evidence which directly shows the effects of exercise upon such factors as the growth of the long bones, relative weight of body tissues, and cholesterol level during the period of life prior to puberty.

STATEMENT OF THE PROBLEM

The purpose of this study was to determine whether different exercise programs, engaged in between the weaning and puberty of male albino rats, have a significant effect on growth as determined by the following measures: total body weight, tibial length, serum cholesterol level, and the relative weights of the heart, spleen, liver, testes, kidneys, and adrenals.

IMPORTANCE OF THE PROBLEM

Exercise and activity, like all other facets of a swiftly changing world, are at the point where they must be critically examined and evaluated to determine the contribution they make to living. There is an ever-growing body of evidence which points out the importance of activity throughout life as a means of delaying the physiological changes that accompany the aging process. This evidence points to the possibility of further increasing life-span by reducing the severity and incidence of such severe manifestations of the aging process as atheroschlerosis.

In other research dealing with the relationships of exercise and aging, it has been suggested that physical activity is an important factor in delaying the mental phenomena of senescence as well as the physical phenomena.

Turning to the effects of exercise on growth and development, while it is generally conceded that exercise is beneficial, there has been little research done in terms of longitudinal studies to quantitatively define the effects of exercise. In reviewing the available literature, the reader is struck by the fact that many of the findings and interpretations are contradictory.

In an era when technology has changed Western man's patterns of living, the schools are charged with the responsibility of providing their students with the knowledge and

skills which will enable them to enter this changing world with the tools they need for maximal achievement. For the physical educator, as well as those in other disciplines, it must be proven that his area of study contributes something unique and valuable to the child's development or the area is unable to justify its existence. Thus, it remains for research to provide justification for a discipline and answers to the questions which that discipline recognizes as being within its rightful sphere of responsibility and purpose.

Ultimately, the job of the school is to enhance the student's mental and physical growth in as effective and efficient a manner as possible. Studies which determine the effects of exercise on growth and development, the aging process, and the physiological functioning of the organism throughout life are those which physical education, as an independent and meaningful discipline, must recognize as being its rightful responsibility.

In addition to serving the function of meeting the responsibilities of an independent discipline, research of the sort which establishes the unique contributions of exercise and activity to growth, health, and longevity must ultimately be the basis upon which physical education will survive as an independent, meaningful field of inquiry.

LIMITATIONS OF THE STUDY

Due to considerations of time, the present study was performed using male albino rats as subjects. It must be immediately noted that the results of animal studies, no matter how significant, cannot be used to draw direct inferences concerning human populations. However, it has been noted that in many ways the internal chemistry of man and the rat are similar and there is reason to believe that man and rat are in like developmental phases at relatively equivalent ages. (80)

In addition, studies of this type serve the function of determining directions for future research as well as providing information about the animal subjects and indicating possible consequences in humans. Thus, in a special sense, animal studies may be thought of as being pilot studies. It is with this understanding that this study was undertaken and conclusions were drawn. Also, the number of animals used as subjects in the study was limited by the amount of available equipment.

DEFINITION OF TERMS USED

The following terms were used in this study:

Spontaneous Exercise: Spontaneous exercise was used to denote an experimental regimen in which the subjects were allowed free access to activity by means of an exercise wheel directly adjacent to their living cages, and were

able to exercise as long as they wished and at whatever intensity they wished.

Forced Exercise: Forced exercise was used to denote an experimental regimen in which the subjects were allowed free access to activity by means of an exercise wheel directly adjacent to their living cages, and were able to exercise as long as they wished and at whatever intensity they wished. In addition, this regimen also required that the subjects be forced to swim for 30 minutes each day with an overload of two percent of the animal's body weight attached.

Sedentary: Sedentary was used to denote an experimental regimen in which the subjects were allowed no access to activity and spent the entire experimental period in their living cages except for being handled daily.

Pre-puberty: Pre-puberty was used to denote the period of the subject's life between weaning and sexual maturity. In this experiment the pre-puberty period was a forty-four day span from the rats' twenty-first day of life to the sixty-fifth day of life.

CHAPTER II

RELATED LITERATURE

The purpose of this experiment was to determine the effects of differing types of training, engaged in during the pre-puberty period of life, upon various measures of growth in the male albino rat. The results of previous experiments on the relationships between exercise and bone growth, organ growth, and serum cholesterol level were reviewed.

THE EFFECTS OF EXTRINSIC FACTORS ON GROWTH

It is generally believed that physical activity is essential for proper growth and that body parts will grow in size in proportion to functional demand. (26) Weiss (109), however, pointed that environment cannot bring new patterns of growth into being for which an organism was not already predisposed through genetic factors. Therefore, we can assume that no mechanical factor such as exercise can be considered as the sole determinant of the extent of an organism's growth. Still, hereditary predisposition is subject to influence by environmental factors and it is essential to determine if exercise acts in a positive or negative fashion in this regard.

There is, however, some feeling evident in the literature which maintains that extrinsic factors are not very important in determining growth. Olson (8) believed that an organism tends to hold to an original developmental line. By this, Olson meant that while extrinsic factors may momentarily accelerate or retard growth, these factors will not affect the ultimate size of the organism. Garn (44) found that genetically related children maintained similarities in body build and body proportions in spite of differences in diet which were both quantitative and qualitative. Dupertuis and Michael (35) analyzed the growth curves of ectomorphic and mesomorphic 21 year old males. When the growth curves for height and weight were examined for ages 2 to 17, the ectomorphic group was taller and lighter, both absolutely and relatively, at every age. Stuart and Dwinell (98) studied the growth of children 6 to 10 years of age and found that the rank orders of bone, muscle, and subcutaneous tissue were constant over this period of time. Meredith and Carl (71) observed children 5 to 9 years of age and measured hip width over this period of time. Their findings indicated that while there was a high correlation between hip width measures at 5 and 9 years of age, 58 percent of the children had a change in percentile rank during this period.

Even when we consider muscularity, we find that extrinsic factors may be only a small part of the total causative agents accounting for individual differences.

Rarick (80) discussed the work of Lindegard (6) who believed that the minimal and maximal muscularity attainable by the individual is relatively constant. In support of this view, Jones (60) found that he could account for 75 percent of the variance in strength of adolescent boys by five factors related to body build.

Extrinsic factors, however, cannot be considered as having absolutely no effect on growth. Nutrition is one extrinsic factor generally considered important in growth. Meredith and Meredith (72) showed that Canadian school boys 13 to 14 years of age were approximately nine centimeters taller in 1943 than 50 years before. The authors concluded that nutritional improvement was one of the factors which accounted for this difference. Conversely, Howe and Schiller (56) studied the average height and weight of German school children over a 40 year period of time which encompassed both World War I and World War II. During both these war periods there was a decrease in average height and weight. This was assumed to be due to a reduction in total calories, proteins, vitamins, and minerals in the diets which were available during the food-rationing periods of 1914-1920 and 1935-1945. Waters (108) reported a study in which animal subjects were fed a diet which contained all necessary foods but in quantities so small, the animals could not maintain increases in weight. While the animals continued to grow in length, adipose tissue

disappeared, the muscles became depleted, and a narrower skeleton resulted.

Rarick (80) stated that prolonged illnesses and chronic disease can have a profound effect upon the physical growth of children when there is accompanying loss of appetite and metabolic disturbances. He also states that illness of a temporary nature, while possibly affecting growth momentarily, is not believed to have an effect on the long-range pattern of growth.

Habits of sleep and rest are another extrinsic factor which is believed to be important in growth. Insufficient rest has a deleterious effect upon the general nutritional and health status of children. It is also believed that the anabolic processes of tissue building for growth occur primarily during periods of sleep and rest.

It is also thought that seasonal changes affect the individual's pattern of growth. Reynolds and Sontag (81) have shown that growth in height is greatest during the spring while growth in weight is greatest during the fall months.

In view of these reports, it seems reasonable to conclude that growth is a phenomenon affected by factors both extrinsic and intrinsic to the organism. Intrinsic factors of heredity may be considered as setting upper and lower limits on the growth of the organism, while extrinsic factors determine where in this range of possible growth

the individual shall be situated. It is within this perspective that we may examine exercise as a possible extrinsic factor in determining growth.

EXERCISE AND GROWTH

Donaldson and Meeser (34) used albino rats as subjects in an early experiment on the effects of exercise on growth carried through seven successive generations. The subjects included both male and female rats and were exercised by running. The results showed an increase in the weight of the heart, kidneys, and gonads of both male and female subjects in the experimental group as compared to the non-exercised controls. The liver was found to be smaller in the experimental group than in the controls. regard to total body growth, there was no difference in either length or weight between the control group and experimental group males. However, the experimental group females were larger than their controls. There was no evidence that quantitative changes in organ or skeletal size was carried from generation to generation. authors also hypothesized that this skeletal difference between males and females in the experimental groups might be due to earlier maturation of the female.

Another study by Donaldson and Meeser (33) examined the effects of varying periods of forced inactivity following a 90 day period of exercise. Again, organ weights were found to be larger in the experimental

animals. The authors also found that during the periods of inactivity the weight gained by the experimental animals was slowly lost and organ size eventually returned to the same level as that of the controls. This reduction in organ weight of the experimental group seemed to be directly proportional to the length of the period of inactivity. Rarick (80) suggested that these data indicate that a protracted exercise program has a ". . . a favorable effect on the growth of rats which is still apparent after long periods of inactivity." The work of Thomas and Miller (102) also indicated that the changes in body weight of rats brought about by exercise will disappear when the experimental group is no longer forced to exercise more than the controls.

Donaldson (31) summarized his studies with the conclusion that, in rats, body weight and body length both seem to improve slightly due to exercise. He also concludes that these effects are more obvious in the female than in the male.

Hatai (53) also found that exercised animals were larger than their non-exercised controls in weight, but were not larger in length. The experimental animals also possessed larger hearts, kidneys, gonads, livers, thymus, and adrenals. It was also noted that these changes were more marked in the female than in the male. Hatai also found that the spleens of his exercised animals were smaller than those of the controls.

Steinhaus (96) reported that dogs exercised by swimming showed an increase in weight of the kidneys, but that dogs exercised by running had kidneys quantitatively similar to the controls. He also found an increase in the size of the adrenals of dogs exercised by swimming and the opposite in dogs exercised by running.

Borovansky (22) described an experiment in which rats in an exercise group were compared with their non-exercised controls after having been grouped from the time they were six weeks old until sacrifice at 14 months of age. He reports no difference in bone lengths between exercised animals and their controls. He found an increase in the absolute and relative weight of the gonads and kidneys and a decrease in the absolute and relative weight of the liver and spleen of the experimental animals.

Steinhaus (94) surveyed much of the literature concerned with the effects of exercise and stated, "In man it is an unsettled question as to whether the growth of long bones is accelerated or retarded by an active life." He cited a study by Muller in which one leg of his rat subjects was skinned and was sewn up. The limb was kept inactivated by this procedure for a period of six weeks. The tibia and radius of the inactivated limbs were regularly found to be 1 to 1½ millimeters longer than their homologs of the active side.

Another study was quoted in which the leg of dog and rabbit subjects was kept inactivated by means of a

stiff bandage or denervation. When the active limb was compared to the inactivated experimental limb, the experimental limb was found to be lighter, have a higher water content, and less mineral content. These signs of atrophy were attributed to reduced blood supply as a result of less exercise. This is supported by Coolbaugh (28) who stated that if the blood supply to a bone is interrupted for any duration of time, the first reaction is a decrease in bone density. In the same study, the inactivated limb was found to be longer and more slender with a more actively growing epiphyseal line. The more slender bone of the inactivated limb was attributed to the absence of pulling muscles at their points of attachment which serves as a normal stimulus to lateral growth. The increased length of the inactivated limb was attributed to the absence of the growth slowing effect of pressure on the ends of the long bones.

In the same study Kohlrausch hypothesized that the effect of exercise is to produce a pressure effect on the epiphyses of the bones which has the effect of stimulating growth to an optimal length. He further hypothesizes that pressure beyond some optimal point would have the effect of retarding growth. In support of this view, Whedon (110) stated, "It seems generally accepted that mechanical stresses and strains logically stimulate processes of bone formation and calcium deposition."

Howell (57) cited a study comparing the length of bones from active and inactivated limbs. These results showed no differences in length. However, the bones of the active limb were generally thicker than the bones of the inactivated limb. Howell concluded that the length of a bone is determined by hereditary factors, whereas growth in diameter is influenced greatly by exercise.

Steinhaus (95) expressed the opinion that exercise may have the effect of limiting the growth of long bones.

Adams (16) reported an extensive study comparing 100 Negro women who were in occupations requiring heavy manual labor with 100 Negro women who did only light work. The anthropometric measurements showed that the heavy working group was taller and heavier at the conclusion of the growing years than the comparative group. Adams concluded that the factor which resulted in this difference was the heavy exercise. He feels that nutritional differences would have tended to favor the light-working group since they were from a higher socioeconomic level than the women engaged in heavy labor. Of course, hereditary factors were not evaluated and could not be considered in this study.

A study by Van Dusen (103) compared dominant and non-dominant arm sizes of children 1 to 4 years of age, children 5 to 8 years of age, and college age adults. The findings indicated that the dominant arm was longer more frequently for the college age group than the other two

groups, and more frequently for the 5 to 8 year old group than the 1 to 4 year olds. The author felt that this trend indicated that as the individual grows older, the dominant arm becomes longer as the result of having received more exercise. However, Van Dusen points out that heredity must be considered as a probable causative factor. In this connection he cites a study in which human embryos were X-rayed and which showed a longer humarus on the right side of the body in over half the cases.

A similar study by Buskirk, Anderson, and Brozek (25) compared the dominant and non-dominant forearms of nationally ranked tennis players. X-ray measurements showed increases in the length of the radius and ulna of the dominant forearm. These differences were in addition to increases in muscular development favoring the dominant side. The same comparisons were made on a group of non-tennis playing men and similar differences were found. However, these differences were not as marked as among the tennis players who had been very active in tennis over a period of years. The authors concluded that the amount of exercise was the differentiating factor.

These studied provide rather substantial evidence that exercise of the upper extremities does stimulate the growth processes in this region. Whether heavy use of the lower extremities with the added burden of weight-bearing would result in both linear and appositional growth is still open to question. (80)

EXERCISE AND CHOLESTEROL LEVEL

Statistical studies have shown contradictory results in regard to the relation of exercise to serum cholesterol level. Fulmer and Roberts (41) and Gsell and Major (47) found that high levels of physical activity seemed to be related to lower values of serum cholesterol level among Navajo Indians and Swiss villagers respectively. However, Brunner, Loebl, and Altman (23) found no such relationship to exist.

Karvonen (62) and Johnson, Wong, Shim, Liu, and Hall (59) found that crosscountry skiers and college swimmers showed serum cholesterol levels which were not significantly lower than those of controls of the same age.

Other statistical studies have shown that when men with proved coronary heart disease are compared to "clinically normal" controls, those with heart disease generally have significantly higher levels of serum cholesterol. (20) In addition, it has been found that "clinically normal" individuals with elevated serum cholesterol levels run a greater risk of coronary heart disease. (61)

Rochelle (84) reported a study in which six males were assigned to an experimental group which received four weeks of physical training and six other males were assigned to a non-exercised control group. The experimental group showed a significant decrease in cholesterol level after the training period while the controls exhibited no similar

change. Six weeks after cessation of training the cholesterol levels of the experimental group had returned to the pre-training level. Rochelle also reported that in every case the subjects in the experimental group showed an increase in cholesterol level during exercise. It was suggested that this indicates that during physical exercise the rate of mobilization and utilization of fat is increased thus supporting the hypothesis that exercise decreases serum cholesterol level.

Golding (45) also found that increased levels of physical activity resulted in decreases in weight and serum cholesterol levels.

Friedberg, Harlan, Trout, and Estes (40) concluded that exercise is effective in lowering serum cholesterol levels by serving as a stimulus to adipose tissue lipolysis.

In connection with this finding, Olson (77) reported that a change in caloric balance, due to a decrease in caloric intake, as opposed to an increase in caloric output, also was evidenced by a decrease in weight and often an early transient fall in cholesterol level.

Taylor, Anderson, and Keys (101) investigated the hypothesis that serum cholesterol is controlled by the percentage of total calories derived from fat, and is independent of the absolute fat intake when total caloric intake is increased by physical activity. It is also assumed in this hypothesis that body weight will remain

constant. The experiment utilized students on a 3,250 daily calorie diet which consisted of 42 percent fat.

After a two week control period, the subjects' activity was increased from its original level by 950 calories. The absolute intake of fat was increased from 155 to 201 grams per day but the percentage of fat in the diet remained constant. It was found that cholesterol concentration did not change as a result of increasing the absolute fat intake when exercise kept body weight constant.

In another report, Taylor (100) discussed two similar experiments where the additional food given to the subjects during the exercise period contained 100 and 0 percent fat respectively. In summarizing these experiments Taylor stated:

It is clear . . . that substantial amounts of fat . . . can be added to the diet to maintain caloric balance without raising the serum cholesterol concentration when the level of physical activity is increased. . . . The effects of exercise might be looked on as non-specific but, in the situation reported here, exercise has apparently prevented a rise in the serum cholesterol that might be expected on grounds other than that connected with the maintenance of caloric balance. This suggests that exercise may have a specific effect on cholesterol metabolism.

Mann, Teel, Hayes, McNally, and Bruno (68) reported that when an individual's caloric intake remains constant, but his activity level is lowered, serum cholesterol level rises. This is interpreted as meaning that a positive caloric balance increases cholesterol level. From this point of view, exercise may not have a specific effect on

cholesterol metabolism, but may be important in preventing a rise in cholesterol level by helping to maintain caloric balance. In this connection, Mayer (70) believes that to maintain close check on caloric balance in modern society, a man must either decrease his food intake and remain mildly or acutely hungry all his life, or raise his level of physical activity.

Montoye and Van Huss (75), in studying the effects of exercise on blood cholesterol in middle-aged men, concluded that exercise was helpful in decreasing serum cholesterol in some subjects, but that this was the indirect result of a reduction in weight.

Holloszy, Skinner, Toro, and Cureton (55) found that a six month program of exercise was not effective in lowering serum cholesterol level. However, they stated, "Although exercise does not appear to produce a persistent reduction in serum cholesterol, it evidently can prevent the rise which accompanies increased caloric intake."

Their findings also included the interesting observation that their subjects did not exhibit a significant loss of weight as the result of training, although caloric balance was definitely altered by the increased caloric output.

The authors hypothesized that, due to the higher caloric equivalent of one pound of adipose tissue as opposed to one pound of muscle tissue, body energy stores could have been depleted by a large number of calories without any

concurrent loss of weight if the pounds of adipose tissue lost were replaced by gains in muscle tissue.

Keys, Anderson, Aresu, Biorck, Brock, BronteStewart, Fidanza, Keys, Malmros, Poppi, Postelli, Swahn,
and del Vecchio (64) showed that men in heavy labor occupations tend to pay off the energy requirements of their
jobs with high carbohydrate foods. According to the hypothesis of Taylor, Anderson, and Keys (101) these individuals
would be expected to have lower cholesterol levels than the
rest of the population since their diet, while possibly
higher in total calories, contains a lower percentage of
fats. This hypothesis is borne out by the results of such
a study done in South Africa. (64) However, Taylor (100)
reported no difference between the cholesterol levels of
railroad clerks who are sedentary at their job and railroad switchmen who are active.

Myasnikov (76) found that rabbits which were fed a high cholesterol diet and were exercised, showed much lower cholesterol level than a control group on the same diet without exercise. Myasnikov states, "... physical exercise, by intensifying metabolism in the body, results in a more intensive assimilation of alimentary cholesterol thereby lowering its level in the blood." Similar results were reported by Warnock (107) in an experiment which used cockrels as subjects.

Rowsell, Downie, and Mustard (85), using swine, and Taylor, Cox, Counts, and Yogi (99), using monkeys, showed

that when diet is changed to include higher percentages of fats, and activity level is not altered, serum lipid level is elevated and coronary atherosclerosis may result.

No studies were found which used pre-puberty animals as subjects.

CHAPTER III

RESEARCH METHODS

The purpose of this study was to determine whether differing programs of pre-puberty exercise resulted in significantly different selected measures of growth in the male albino rat. Prior studies have shown contradictory results and have not been limited to this period of life.

EXPERIMENTAL DESIGN

Sixty male Sprague-Dawley rats, born the same day but not litter mates, were purchased from Hormone Assay Laboratories in Chicago, Illinois. These animals were received at the laboratory on their 25th day of life, which was four days after their weaning. Upon being received in the laboratory the animals were matched into trios on the basis of a two day sample of spontaneous activity and were then randomly assigned to three experimental regimens:

(1) sedentary housing, (2) voluntary activity, and (3) voluntary activity plus forced exercise. One week was allowed for assignment to groups, marking of the animals and cages, and adjustment of the animals, particularly the forced exercise group, to the experimental conditions to

which they would be subjected for the length of the experimental period. The first day of the experiment was equivalent to the 32nd day of life of the subjects.

The experimental period continued for 35 days until the 66th day of life of the rats. On the 36th day of the experiment (67th day of life of the subjects) the animals were sacrificed and the desired measures were taken.

EXERCISE REGIMENS

Sedentary Condition:

The animals which were assigned to the sedentary group were housed in standard 10 x 8 x 7 inch small-animal cages. This group received no exercise other than what they were able to obtain in this confined area. The only other specific procedure followed with this group was to insure that they were handled each day as much as the animals in the other groups since the amount of handling in early life has been shown to be an important variable in the later behavior of the rat and it was felt that this might also aid in keeping all known relevant variables as constant as possible for the three groups of subjects.

(36, 50, 92)

<u>Voluntary</u> <u>Activity</u>:

The animals which were assigned to the voluntary activity group were housed in standard 10 x 8 x 7 inch

small-animal cages which were equipped so that the rat had free access to a freely revolving drum five inches wide and 14 inches in diameter, in which he could run at his own volition. The activity wheel was constructed of one-half inch mesh hardware cloth around one-quarter inch thick plexiglass which served as the center disk. The housing cages and activity wheels were manufactured by the Unifab Corporation of Kalamazoo, Michigan. A revolving drum of this type is standard in providing voluntary activity for small animals. (83, 88, 90) This group was also handled daily for the same reason as the sedentary group.

Voluntary Activity Plus Forced Exercise:

The animals which were assigned to the voluntary activity plus forced exercise group were housed in a fashion identical to those in the voluntary activity group. In addition to exercising at their own volition, the animals in this group were also forced to swim for 30 minutes each day with an overload of two percent of their body weight attached to them in the form of lead weights which were taped to the base of the animals' tails prior to the swimming period. The animals were weighed every third day and the weights were adjusted when needed as the experiment continued. During the swimming period, the animals were placed in individual one foot square plexiglass compartments within a large steel tank. The water was held between 35 and 37 degrees Centigrade. After being exercised, the animals were placed in small holding cages under

lamps and were then dried off by toweling before being returned to their individual housing cages.

GENERAL ANIMAL CARE

The animals in all three groups were fed a prepared ground ration (Appendix K) ad libitum and records of food consumption were kept as part of a concurrent study. rats had constant access to water by means of an inverted eight ounce polyethylene bottle fastened above the living cage. A straight metal tube ran from the water bottle into the cage. Care was taken to insure that the animals in the sedentary and voluntary activity groups were handled as much as the animals in the voluntary activity plus forced exercise group. The windows in the room which housed all three groups of animals were blacked out and a timer was attached to the lighting system which turned the room lights on each morning at 7 A.M. and turned them off at 7 P.M. Room temperature and humidity varied but were the same for all three groups at any given time throughout the experimental period.

WEIGHING PROCEDURES

The animals were sacrificed at the end of the experimental period by ether anesthesia. Body weight was determined to the nearest whole gram on a Cenco triplebeam balance. After recording total body weight, a team procedure was employed to remove the desired organs, trim

them of excess connective tissue, and weigh them to the nearest milligram on a Mettler electronic balance. Body weights were expressed in absolute terms (grams) and were statistically treated in this form. Organ weights were expressed as percentage of body weight. These percentages were obtained by dividing organ weight by the animal's total body weight and expressing the resulting quotient as a percent. The values for total body weight are presented in Appendix A. The values for the percentage of total body weight each organ represents are presented in Appendices B through G. During the course of the experiment one rat in the voluntary activity plus forced exercise group died leaving only 19 subjects in this group.

BONE LENGTH

After ether anesthesia and determination of total body weight, the right hind limb of the animal was removed and the tibia was dissected out. All excess tissue was removed from the bone including all non-ossified material found at the epiphyses. The bones were then tagged for identification and stored for measurement at a later date. Approximately two weeks later, the bones were measured with a Cenco vernier caliper to the nearest tenth of a millimeter. The length of the tibia was measured from the most posterior presentation of the lateral condyle of the proximal tibial epiphysis to the most posterior point of the distal fibular epiphysis. Bone lengths were expressed

in absolute terms (millimeters) and were statistically treated in this manner. Due to a need for the total carcass for use in a concurrent study, the tibia was excised on only 30 of the rats. The values for tibial length are presented in Appendix H.

CHOLESTEROL LEVEL

Immediately prior to sacrifice by ether anesthesia, the animals were rendered unconscious by ether and a blood sample was taken from the orbital sinus of the rat after the method by Stone. (97) The sample was then spun down in a Cenco electric clinical centrifuge and the serum was pipetted off for testing. Determination of cholesterol level was done by the Schoenheimer and Sperry (86) method as adapted by Foldes and Wilson (39) for photoelectric instruments. Cholesterol levels were expressed in terms of milligrams per 100 cubic centimeters of blood (milligrams percent) and were statistically treated in this In addition to the subject lost in the forced exercise group, one subject in this same group could not be analyzed for serum cholesterol level due to the fact that a sufficiently large blood sample could not be ob-The values for cholesterol level expressed in milligrams percent are presented in Appendix I.

STATISTICAL TREATMENT

A one-way fixed-effects model of the analysis of variance was used to analyze the data. The probability of making a Type I error was held to the .05 level of confidence. If the F ratio indicated overall significant differences at this level, the Scheffé test was used to determine between which means these differences existed.

CHAPTER IV

RESULTS AND DISCUSSION

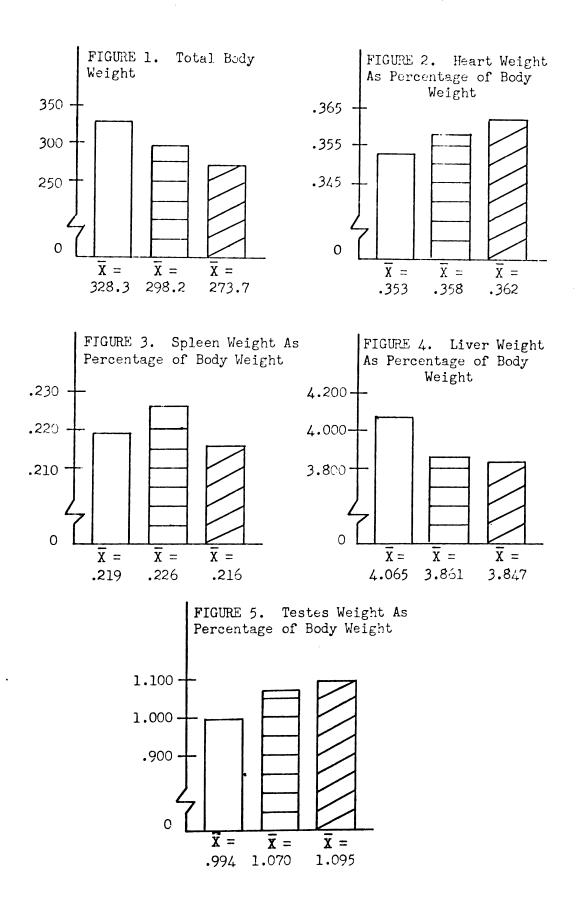
The purpose of this study was to determine the effects of differing pre-puberty exercise programs upon selected measures of growth in the male albino rat.

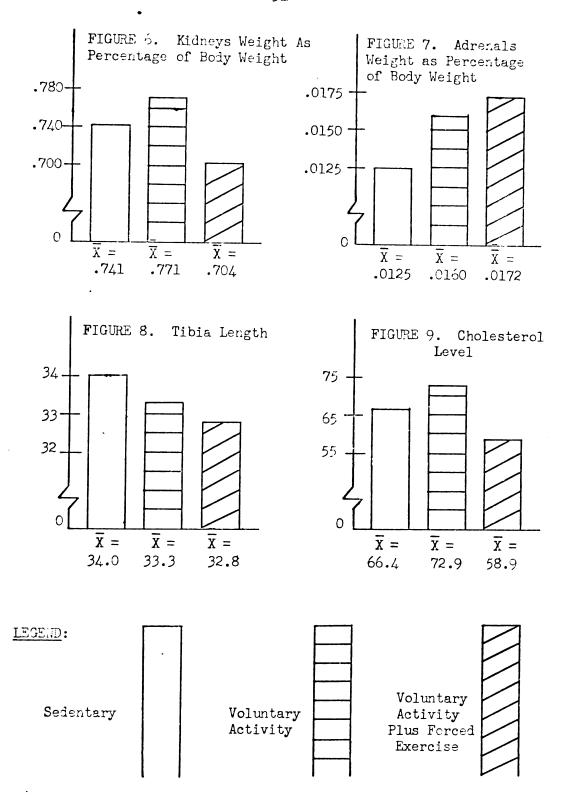
The subjects were divided into three groups as follows: (1) a sedentary group with no access to activity other than that allowed within the confines of a small living cage, (2) a voluntary activity group with free access to a freely rotating exercise wheel, and (3) a voluntary activity plus forced exercise group with free access to the exercise wheel and also forced to swim for 30 minutes each day with an overload of two percent of their body weight attached in the form of lead weights.

RESULTS

The group means of total body weight are presented graphically in Figure 1. The raw data are presented in Appendix A.

The analysis of variance detected significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 1. The Scheffe test gave a value of 5.32 grams.





This value indicates that all three means differed significantly in terms of total body weight. In ascending order the group means for total body weight were ranked in the following order: (1) voluntary activity plus forced exercise, (2) voluntary activity, (3) sedentary.

TABLE 1. Analysis of Variance for Total Body Weight

Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,56)
Between	29.67	2	14583.5	0.7. (2.76
Within	31802	56	567.8	25.6	3.16
Total	60969	58			

The group means of heart weight as percentage of total body weight are presented graphically in Figure 2. The raw data are presented in Appendix B.

The analysis of variance detected no significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 2. The power of the F test was calculated to be approximately .90 with mean differences on the order of one standard deviation. Therefore, the probability of committing a Type II error was approximately .10.

TABLE 2. Analysis of Variance for Heart Weight as Percentage of Total Body Weight

Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,56)
Between	.000891	2	.000446	06	2.16
Within	.097891	56	.001748	.26	3.16
Total	.098782	58			

The group means of spleen weight as percentage of body weight are presented graphically in Figure 3. The raw data are presented in Appendix C.

The analysis of variance detected no significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 3. The power of the F test was calculated to be

TABLE 3. Analysis of Variance for Spleen Weight as Percentage of Total Body Weight

Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,56)
Between	.000891	2	.0004455	60	2.16
Within	.040320	56	.0007200	.62	3.16
Total	.041211	58			

approximately .90 with mean differences on the order of one standard deviation. Therefore, the probability of committing a Type II error was approximately .10.

The group means of liver weight as percentage of total body weight are presented graphically in Figure 4. The raw data are presented in Appendix D.

The analysis of variance detected no significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 4. The power of the F test was calculated to be approximately .90 with mean differences on the order of one standard deviation. Therefore, the probability of committing a Type II error was approximately .10.

TABLE 4. Analysis of Variance for Liver Weight as Percentage of Total Body Weight

Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,56)
Between	.590	2	.295	1 00	2.16
Within	8.577	56	.153	1.93	3.16
Total	9.167	58			

The group means of testes weight as percentage of total body weight are presented graphically in Figure 5.

The raw data are presented in Appendix E.

The analysis of variance detected significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 5. The Scheffé test gave a value of .02259 percent. This value indicates that all three means differed significantly in terms of testes weight as percentage of total body weight. In descending order the group means for testes weight as percentage of total body weight were ranked in the following order: (1) voluntary activity plus forced exercise, (2) voluntary activity, (3) sedentary.

TABLE 5. Analysis of Variance for Testes Weight as Percentage of Total Body Weight

Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,56)
Between	.110646	2	.055323	,	
Within	•577505	56	.010312	5.36	3.16
Total	.688151	58			

The group means of kidneys weight as percentage of total body weight are presented graphically in Figure 6.

The raw data are presented in Appendix F.

The analysis of variance detected significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in

Table 6. The Scheffé test gave a value of .01757 percent. This value indicates that all three means differed significantly in terms of kidneys weight as percentage of total body weight. In descending order the group means for kidneys weight as percentage of total body weight were ranked in the following order: (1) voluntary activity, (2) sedentary, (3) voluntary activity plus forced exercise.

TABLE 6. Analysis of Variance for Kidneys Weight as Percentage of Total Body Weight

Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,56)
Between	.044148	2	.022074	2 (1	2.16
Within	.342732	56	.006120	3.61	3.16
Total	. 386880	58			

The group means of adrenals weight as percentage of total body weight are presented graphically in Figure 7.

The raw data are presented in Appendix G.

The analysis of variance detected significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 7. The Scheffé test gave a value of .00053 percent. This value indicates that all three means differed significantly in terms of adrenals weight as percentage of total

body weight. In descending order the group means for adrenals weight as percentage of total body weight were ranked in the following order: (1) voluntary activity plus forced exercise, (2) voluntary exercise, (3) sedentary.

TABLE 7. Analysis of Variance for Adrenals Weight as Percentage of Total Body Weight

Source	Sums of	Degrees of Freedom	Mean Squares	F	F.95(2,56)
Between	.000237	2	.0001185	00 50	2.16
Within	.000321	56	.0000057	20.79	3.16
Total	.000558	58			

The group means of tibia length are presented graphically in Figure 8. The raw data are presented in Appendix H.

The analysis of variance detected significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 8. The Scheffé test gave a value of .7511 millimeters. This value indicates that the tibia of the sedentary group was significantly longer than the tibia of the voluntary activity plus forced exercise group. The tibia of the voluntary activity group did not differ significantly

TABLE 8	 Analys 	is of	Variance	for	Tibia	Length
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Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,27)
Between	7.1687	2	3.5844	0 -	
Within	11.5580	27	.4280	8.37	3.35
Total	18.7267	29			

from either the sedentary group or the voluntary activity plus forced exercise group. In ascending order the group means for tibia length were ranked in the following order:

(1) voluntary activity plus forced exercise, (2) voluntary activity, (3) sedentary.

The group means of cholesterol level are presented in Figure 9. The raw data are presented in Appendix I.

The analysis of variance detected significant differences between groups at the .05 level of confidence. The results of the analysis of variance are presented in Table 9. The Scheffé test gave a value of 2.62 milligrams percent. This indicates that all three means differed significantly in terms of cholesterol level. In ascending order the group means for cholesterol level were ranked in the following order: (1) voluntary activity plus forced exercise, (2) sedentary, (3) voluntary activity.

TABLE 9. Analysis of Variance for Serum Cholesterol Level

Source	Sums of Squares	Degrees of Freedom	Mean Squares	F	F.95(2,55)
Between	1844.56	2	922.28	7 05	2.16
Within	7198.26	55	130.88	7.05	3.16
Total	9042.82	57			

DISCUSSION

The present results indicate that, in rats, weight seems to vary inversely with level of activity. This is as the literature indicates and is in keeping with current hypotheses concerning weight control. (70) It seems reasonable to infer that patterns of activity which are directed at the ultimate goal of maintaining proper weight should be initiated early in life. This would serve not only to firmly establish regular habits of exercise in the pattern of daily living, but would also aid in controlling weight early in life.

No significant differences between groups were found when heart, spleen, and liver weights were expressed as percentage of total body weight. In relation to the heart, it must be assumed that the level of activity used in the present study was not of sufficient intensity to

produce cardiac hypertrophy of the magnitude necessary to be reflected in a significant F ratio.

In relation to both the spleen and liver, both of these organs are primarily concerned with functions of the body which are not specifically related to adaptation to exercise and it is assumed that the observed results are the reflection of random effects not due to differences in treatment between groups.

Testes and kidneys weight as percentage of total body weight were both significantly different between all three groups. In the case of the testes, the linear ascendency of these percentages with increased activity seems to reflect the proportional differences that might be expected due to changes in total body weight with increased activity. No pattern was evident in regard to kidneys weight as percentage of total body weight. This may be due to the fact that with heavy exercise more body water is lost through the lungs and sweat and less body water is lost through the urine.

Weight of the adrenals as percentage of body weight was significantly different between all three groups and varied directly with level of activity. It seems reasonable to assume that exercise served as a stressor which acted upon the body to cause a general adaptation syndrome which was reflected in increases in the size of the adrenal glands which are directly involved in the process of adaptation to stress. (13, 87) It was also deemed

possible that the animals in the sedentary group might show a similar reaction to the stress of confinement but this was not borne out by the present results. It may be that confinement of the type used in this study did not constitute a stress to the subjects, or it may be that exercise is simply a more powerful stressor.

The longer tibia of the animals in the sedentary group as compared to the voluntary activity plus forced exercise group cannot be explained in terms of the effect of pressure since the forced exercise was in the form of swimming as opposed to running. There is the possibility that the difference reflects a negative effect of pressure due to the rats in the voluntary activity plus forced exercise group running on the revolving drum. this should also be reflected in the comparison of the voluntary activity and sedentary groups. It was thought possible that the swimming served as a stimulus to voluntary activity which would have raised the voluntary activity level of the rats receiving forced exercise and would account for the observed differences. However, a six day record of activity immediately prior to and including the 35th day of training, indicated that the voluntary activity group mean daily number of wheel revolutions was significantly higher than the mean daily number of wheel revolutions for the forced exercise group (see Appendix J). is also possible that exercise, by increasing metabolism and the rate of carbon dioxide formation and thus lowering

the pH of the body's extracellular fluids to a more acid level, may reduce the number of dibasic phosphate ions available for formation of bone salts and the intervening formation of calcium phosphate. Also, the increase in metabolic rate may possibly result in a greater increase in osteoclastic activity as opposed to osteoblastic activity. (2)

However, in view of the results which indicated that exercise served to elicit a general adaptation syndrome, and that adrenal size varied directly with level of activity, a more tenable hypothesis is that of a direct inhibition of growth due to a shift in hormone formation. In relation to this hypothesis, Selye (13) stated:

Somatic growth is inhibited during exposure to stress
. . . This has been ascribed to a 'shift in pituitary
hormone formation' . . . accomplished at the expense
of other, less urgently needed, hypophyseal principles
such as somatotrophin. . . "

In view of the finding that significant differences existed only between the sedentary and voluntary activity plus forced exercise groups, it must also be noted that the voluntary activity plus forced exercise group was forced to exercise by swimming. This form of exercise may have served as an additional stress agent, thus increasing the adaptive reactions of the subjects in this group and resulting in a shift in hormone formation and the observed changes in growth.

It should also be noted that the voluntary activity plus forced exercise group was also subjected to heat following swimming and then to rough toweling before being returned to their cages. These variables, while not considered important when designing the present study, may have served to further stress the forced exercise group.

Cholesterol level differed significantly between all three groups, yet this was not proportional to changes in physical activity. The forced exercise group was significantly lower in cholesterol level than either the sedentary or voluntary activity groups, but the sedentary group cholesterol level was lower than that of the voluntary activity group. These results indicate that there is a relationship between the effect of exercise on cholesterol level and the intensity of the exercise and that, in order to be effective, the exercise must be at some minimal level of intensity.

CHAPTER V

SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

SUMMARY

The purpose of this study was to determine the effects of differing pre-puberty exercise programs upon selected measures of growth in the male albino rat. The measures used were total body weight, tibia length, serum cholesterol level, and the relative weights of the heart, spleen, liver, testes, kidneys, and adrenals.

Three groups of male albino rats were assigned to experimental regimens as follows: (1) a sedentary group with no access to activity, (2) a voluntary activity group with free access to a rotating exercise wheel, and (3) a voluntary activity plus forced exercise group under the same conditions as group two and also forced to swim for 30 minutes each day with an overload of two percent of the animal's body weight attached to the base of the tail. The animals continued their respective experimental procedures for a period of 35 days.

At the conclusion of the experimental period the rats were sacrificed by ether anesthesia, total body weight was determined, the organs were weighed after removal and

cleaning, the tibia was excised and measured, and a blood sample was taken from the orbital sinus for determination of serum cholesterol level.

A one-way fixed-effects model of the analysis of variance was used to determine if significant differences existed between groups at the .05 level of confidence. If the F ratio showed overall significance, the Scheffé test was used to determine between which means these differences existed.

No significant differences between group means were found for heart weight as percentage of total body weight, spleen weight as percentage of total body weight, or liver weight as percentage of total body weight.

Total body weight differed significantly among the three groups. In ascending order the group means for total body weight were ranked in the following order: (1) voluntary activity plus forced exercise, (2) voluntary activity, (3) sedentary.

Testes weight as percentage of total body weight differed significantly among the three groups. In descending order the group means for testes weight as percentage of total body weight were ranked in the following order:

(1) voluntary activity plus forced exercise, (2) voluntary activity, (3) sedentary.

Kidneys weight as percentage of total body weight differed significantly among the three groups. In ascending order the group means for kidneys weight as percentage

of total body weight were ranked in the following order:
(1) voluntary activity plus forced exercise, (2) sedentary, (3) voluntary activity.

Adrenals weight as percentage of total body weight differed significantly among the three groups. In descending order the group means for adrenals weight as percentage of total body weight were ranked in the following order:

(1) voluntary activity plus forced exercise, (2) voluntary activity, (3) sedentary.

Tibia length was significantly greater in the sedentary group when compared to the voluntary activity plus forced exercise group. The voluntary activity group mean was not significantly different from the mean of the sedentary group or the voluntary activity plus forced exercise group. In ascending order the group means for tibia length were ranked in the following order: (1) voluntary activity plus forced exercise, (2) voluntary activity, (3) sedentary.

Serum cholesterol level differed significantly among the three groups. In ascending order the group means for serum cholesterol level were ranked in the following order:

(1) voluntary activity plus forced exercise, (2) sedentary,

(3) voluntary activity.

CONCLUSIONS

The results of the present study indicate that the type of exercise program to which male albino rats are

subjected in the pre-puberty period of life is a significant variable in determining total body weight at puberty, testes, kidneys, and adrenals weight as percentage of total body weight at puberty, tibia length at puberty, and serum cholesterol level at puberty.

The results also indicate that the type of exercise program to which male albino rats are subjected in the pre-puberty period of life is not a significant variable in determining heart, liver, or spleen weight as percentage of total body weight at puberty.

RECOMMENDATIONS

Future research concerned with the effects of exercise on growth should be directed along the following lines and should consider the following points:

- Research should be conducted to determine the intensity of exercise needed in the pre-puberty period to secure specific physiological results.
- 2. Research should be conducted on a longitudinal basis to determine if the effects of pre-puberty exercise continue throughout the life span of the organism.
- 3. Research should be conducted to determine if pre-puberty exercise is a significant variable in determining other measures of bone growth such as breaking strength, bone density, and bone weight.

- 4. Research should be conducted to determine if the effects of pre-puberty exercise on organ function reflect the observed effects on organ size.
- 5. Research should be conducted to determine if the observed effects of forced exercise on bone growth are reproduceable when forms of exercise other than swimming are used as the stress medium.
- 6. Research should be conducted using additional control groups which also receive immersion in water, heating, and toweling but not swimming, to determine if these factors are stressful.

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APPENDICES

APPENDIX A
TOTAL BODY WEIGHT

SEDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
303	275	275
298	293	244
308	277	247
332	303	247
295	329	286
329	288	267
327	322	274
334	320	261
337	274	271
279	271	246
371	267	300
346	358	300
334	315	271
344	308	285
311	278	282
352	326	303
349	323	278
359	271	274
308	316	290
350	251	-
Sum 6566	5965	5201
Mean 328.3	298.2	273.7

APPENDIX B

HEART WEIGHT AS PERCENTAGE OF TOTAL BODY WEIGHT

SED	ENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	.316	. 376	. 354
	. 346	.483	. 386
	.482	.312	. 366
	.428	. 346	.351
	.303	. 464	.311
	. 344	•439	•372
	• 377	. 365	• 373
	. 338	. 348	• 38 3
	. 336	. 368	. 358
	.423	• 347	.376
	.292	.316	. 382
	• 374	•295	. 348
	· 335	. 328	. 382
	. 334	. 362	• 393
	.312	.331	.341
	. 340	• 336	. 36 3
	• 335	• 327	. 357
	. 388	. 328	. 334
	. 304	. 326	. 356
	• 351	.338	_
Sum 7	.058	7.135	6.886
Mean	. 353	. 358	. 362

APPENDIX C

SPLEEN WEIGHT AS PERCENTAGE OF TOTAL BODY WEIGHT

s -	EDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	.248	.233	.206
	.273	.231	.228
	.254	.228	.219
	.236	.277	.277
	.225	.238	.166
	.229	.207	.252
	.234	.208	.206
	.233	.223	.238
	.209	.271	.253
	.218	.226	.187
	.219	.187	.201
	.200	.224	.192
	.222	.201	.215
	.193	.225	.180
	.144	.236	.232
	.226	.206	.178
	.240	.240	.215
	.183	.205	.233
	.195	.200	.235
	.200	.248	-
Sum	4.381	4.514	4.113
Mean	.219	.226	.216

APPENDIX D

LIVER WEIGHT AS PERCENTAGE OF TOTAL BODY WEIGHT

S	EDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	3.784	3.933	3.742
	3.515	3.731	3.584
	3.862	3.174	4.957
	3.917	3.902	4.059
	3.548	3.692	3.448
	3.780	4.360	3.603
	4.829	5.083	3.954
	4.037	3.523	3.441
	4.487	3.764	3.297
	3.633	3.930	3.183
	4.630	3.794	3.931
	4.107	4.317	4.178
	3.922	4.084	4.086
	4.192	3.750	3.797
	4.383	3.748	3.617
	3.881	4.038	3.948
	4.312	3.923	4.162
	4.250	3.489	3.838
	4.009	3.451	4.269
	4.222	3.537	-
Sum	81.300	77.223	73.094
Mean	4.065	3.861	3.847

APPENDIX E

TESTES WEIGHT AS PERCENTAGE OF TOTAL BODY WEIGHT

	SEDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	1.058	1.145	0.867
	1.034	1.183	0.762
	0.992	1.072	1.151
	1.031	0.989	1.073
	1.030	0.962	1.130
	0.989	1.049	1.241
	0.938	1.022	1.143
	1.005	1.058	1.123
	0.906	1.195	1.128
	1.219	1.220	1.179
	0.734	1.043	1.098
	1.061	0.945	1.046
	1.028	1.115	1.202
	0.921	1.048	1.169
	1.044	1.043	1.092
	0.982	1.025	0.967
	0.903	0.955	1.165
	0.992	1.212	1.121
	1.140	1.031	1.155
	0.864	1.091	_
Sum	19.871	21.403	20.812
Mean	•994	1.070	1.095

APPENDIX F

KIDNEYS WEIGHT AS PERCENTAGE OF TOTAL BODY WEIGHT

	SEDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	•773	.747	.634
	.758	.884	.734
	.831	.728	.850
	.854	.965	.856
	.714	.843	.731
	.710	.781	•709
	.838	1.007	.711
	.772	.802	.704
	.828	.849	.698
	.824	.782	.684
	.762	.641	.713
	•690	.699	.726
	.702	.714	.694
	.640	•799	.665
	.709	•713	.682
	.664	.693	. 589
	.692	.662	.697
	.689	.689	.635
	.685	.662	.658
	.690	. 758	_
Sum	14.825	15.418	13.370
Mean	.741	.771	.704

APPENDIX G

ADRENALS WEIGHT AS PERCENTAGE OF TOTAL BODY WEIGHT

:	SEDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	.013	.016	.018
	.014	.014	.021
	.010	.012	.018
	.012	.015	.017
	.012	.013	.013
	.011	.016	.019
	.013	.017	.014
	.013	.013	.014
	.011	.016	.019
	.016	.020	•015
	.014	•015	.018
	.011	.014	.015
	.012	.016	.016
	.012	.019	.017
	.013	•015	.020
	.012	.014	.015
	.013	.018	.017
	.014	.016	.021
	.012	.016	.020
	.012	.026	_
Sum	.250	.321	. 327
Mean	.0125	.0160	.0172

APPENDIX H

TIBIA LENGTH

s _	SEDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	34.3 34.0 33.0 33.8 33.7 35.1 33.8 34.3 34.1	33.5 33.7 33.0 34.2 33.4 32.3 33.5 34.4 32.5	33.2 32.2 33.6 31.7 33.5 33.5 32.8 33.0 32.6 32.0
Sum	340.0	332.9	328.1
Mean	34.0	33.3	32.8

APPENDIX I
SERUM CHOLESTEROL LEVEL

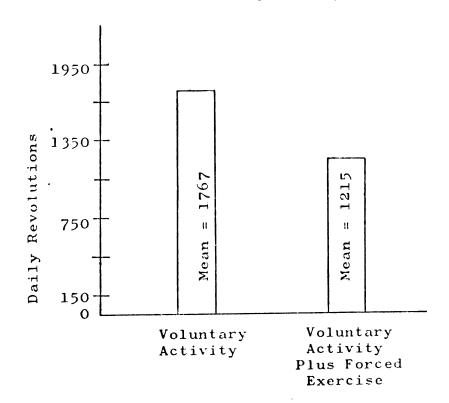
	SEDENTARY	VOLUNTARY	VOLUNTARY PLUS FORCED
	71.0	63.7	50.5
	85.7	49.4	61.7
	63.5	50.8	57.6
	65.5	65.5	63.5
	57.6	90.6	65.0
	78.0	79.5	62.0
	64.3	75.8	67.2
	73.5	96.0	47.2
	69.5	110.7	59.3
	68.5	87.0	
	57•3	68.7	55.2
	80.4	75.8	64.1
	78.4	78.1	57.8
	54.2	63.4	67.7
	52.2	69.7	45.3
	65.4	54. 0	54.2
	67.7	73.8	49.6
	50.9	54.5	64.6
	62.1	77.1	68 . 0
	61.6	73.3	
Sum	1327.3	1457.4	1060.5
Mean	66.4	72.9	58.9

APPENDIX J

EFFECT OF PRE-PUBERTAL EXERCISE ON DAILY VOLUNTARY ACTIVITY

OF RATS AT PUBERTY*

(Estimated by Mean of Six Days of Activity Immediately Prior to and Including 35th Day of Exercise)



^{*}A comparison of means showed the voluntary activity group ran significantly more than the voluntary activity plus forced exercise group (t = 2.230, t = 2.009).

APPENDIX K

RAT RATION - INGREDIENTS PER 500 POUNDS

303.5 lbs.
140.0 lbs.
10.0 lbs.
12.5 lbs.
12.5 lbs.
8.0 lbs.
8.7 lbs.
2.5 lbs.
225.0 gms.
182.0 gms.
19.0 gms.
56.0 gms.
113.0 gms.
159.0 gms.
1.25 gms.
.75 gms.
7.5 gms.
1.5 gms.
1.0 gms.
.5 gms.
113.5 gms.

