

EFFECT OF EXERCISE ON EARLY NORMAL-FED
AND OVERFED MICE

by

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INTRODUCTION

Obesity is a serious problem for many American children. It is estimated that between 6% to 15% of American children are obese (1). Unfortunately many of these children become obese adults.

Obesity and its control has been a concern primarily for adults in the United States. However, in recent years, childhood obesity has received more attention as research has illustrated the detrimental effects of obesity throughout the life span.

The key factor in weight control is energy balance. The energy balance equation states that body weight remains constant when caloric intake equals expenditure. Thus, to bring about a decrease in body weight, caloric intake must be decreased, caloric expenditure increased, or some combination of the two must be utilized.

The purpose of this study was to determine if over-nourished mice, with a high level of body fat, respond differently to the effects of exercise than mice receiving normal nutriture. Early normal and over-nutriture was accomplished by a manipulation of litter size in mice. These mice were then subjected to exercise or non-exercise programs to see if over-fed mice responded differently to exercise than normal-fed mice. Body weight, body composition, and feed intake were measured. It is hoped that the experiment will provide information leading to better treatment of childhood obesity.

REVIEW OF LITERATURE

I. Metabolic Alterations in the Obese

Alterations in Carbohydrate Metabolism. The obese individual exhibits several endocrine and metabolic abnormalities. Bray (2) and Kalkhoff et al. (3) found that fasting blood glucose was higher in chronically obese individuals and that this observation was frequently accompanied by impaired glucose tolerance. Sims and Horton (4) observed that dietary-induced obesity in man was followed by a reduction in the utilization of glucose for energy, and an increase in the amount of glucose available for synthesis of fat. Goldberg and Gordan (5) found that epinephrine resistance in obese individuals was highly correlated with abnormalities in peripheral glucose and oxidative metabolism. Knittle and Ginsberg-Fellner (6) observed an improvement in oral glucose tolerance after substantial weight reduction in obese individuals. Kalkhoff et al. (3) found that fasting plasma glucose levels decreased in obese subjects after substantial weight reduction.

Other metabolites including pyruvate, lactate, and citrate are elevated in the blood stream of obese individuals (7). West and Todd (8) have shown the higher blood levels of lactic and pyruvic acid in obese subjects reflect a reduced ability of tissues to oxidize pyruvic acid formed from the glycolytic phase of carbohydrate breakdown. Wertheimer (9) found that pyruvic acid inhibits the oxidation of fatty acids. He suggested that obesity may be caused by the inability of cells to break down carbohydrates beyond the pyruvic acid stage, thereby inhibiting fat utilization and promoting fat storage.

Hyperinsulinemia is another characteristic of obesity. Bray and York (10) stated that an increase in circulating insulin occurs in all forms of obesity, and could be attributed to one or several mechanisms. Insulin initiates its action on target tissues by interacting with specific receptors on the cell membrane. The insensitivity of large fat cells to insulin could result from a decrease in the number of insulin binding sites on the cell membrane, a lower affinity of binding, distortion of the cell membrane, or a defect following the insulin-cell interaction.

Amatruda et al. (11) showed that large adipocytes from obese subjects have receptor numbers and affinities for insulin similar to adipocytes from subjects of normal weight. Amatruda and coworkers examined the affinity of the insulin receptor for its hormone by measuring the displacement of (^{125}I) insulin by native insulin in adipocytes from obese and normal weight subjects. The concentrations of native insulin necessary to displace 50 percent of the maximum specific binding were 2.17×10^{-9} M for small adipocytes and 1.75×10^{-9} M for large adipocytes. From these findings, it was concluded that the parameter of adipocyte insulin-receptor interaction was unaltered in obesity. They then suggested that the hormonal insensitivity in obese human subjects might be caused by a "dilution" of insulin receptors over the surface area of large cells which may hinder the transmission of signals arising from the interaction between insulin and the insulin receptor.

Amatruda et al. (11) reviewed numerous studies which showed that intracellular carbohydrate and lipid metabolism is altered in large fat cells when compared to small ones in both animal and human adipocytes. It is possible that these metabolic alterations might render large fat

cells less sensitive to some of the actions of insulin which would in turn appear as insulin resistance.

Archer et al. (12) studied insulin-receptor binding in obese humans. With insulin at 0.1 ng/ml, the binding of (^{125}I) insulin in-vitro to circulating lymphocytes from 11 obese patients was less than that observed with cells from 10 thin volunteers. In obese cells, unlabeled insulin was less effective in competing with labeled hormone for binding, both at low and high concentrations of unlabeled insulin. Results indicate that the decrease in binding appears to be due to a lowering of the receptor concentration, but some loss of affinity was not excluded as a possibility.

Salans et al. (13) studied glucose metabolism and insulin sensitivity of isolated human adipose tissue. Adipose cells of obese individuals metabolized glucose to carbon dioxide and triglyceride at rates similar to adipose cells of nonobese subjects. In contrast, insulin responsiveness of adipose tissue was dependent upon adipose cell size. The larger its adipose cells, the less insulin sensitive was the tissue. Thus, adipose tissue of obese subjects, with enlarged cells, showed a diminished response to insulin. After weight loss and reduction in adipose cell size, insulin sensitivity of the adipose tissue of obese patients was restored to normal. When adipose tissue of obese individuals showed impaired responsiveness to insulin, their plasma insulin levels, after oral glucose, were elevated. Weight loss and reduction in adipose cell size restored plasma insulin concentration to normal, concomitant with the return of normal tissue insulin sensitivity.

Jungas (14) observed that the larger the adipose cell, the less responsive the tissue to insulin, possibly indicating that the amount of

lipid within the cell may influence its metabolic efficiency and responsiveness to insulin. "Stuffing" the cell with lipid may cause distortion of the cell membrane, resulting in a change in some receptor site(s) with which insulin interacts, or a change in some membrane constituent, such as adenylyl cyclase, which may be involved in the mediation of the intracellular action of insulin.

Salans et al. (13) suggested that it is possible that the elevated levels of plasma insulin are the cause rather than the result of the "insulin resistance" of adipose cells. Under the influence of prolonged, high concentrations of insulin, fatty acid esterification and triglyceride deposition in the adipose cell may proceed at an increased rate, resulting in enlarged, fat-filled adipose cells with diminished sensitivity to insulin. Thus, the insulin resistance of the adipose tissue may be a secondary phenomenon and not causal in the plasma insulin abnormalities of these individuals. That the adipose tissue is a primary factor in these abnormalities, however, is supported by the observation that plasma insulin concentration returns to normal after weight loss and reduction in adipose depots and cell size.

Hormonal Alterations. Hyperinsulinemia is usually observed in both the fasting state and in response to a glucose load. In smaller animals, Aubert et al. (15) found that mice overnourished during the suckling period showed an increase in circulating insulin at 29 weeks of age, although it was not significant in the fasting state, indicating hyperinsulinemia. This suggests that hyperinsulinemia and obesity might be a consequence of hyperphagia induced early in life.

Insulin is a protein secreted into the blood by the beta cells in the islets of Langerhans of the pancreas. The normal stimulus to insulin

secretion is the ingestion of carbohydrates and the consequent rise in blood sugar. Secretion is also stimulated by amino acids and by the intestinal hormone pancreozymin. Insulin facilitates glucose uptake and storage in body tissues.

Cuatrecasas (17) demonstrated that insulin receptors are located on the outer surface of the fat cell membrane and that approximately 10,000 molecules of insulin can be bound per fat cell.

The mechanism by which insulin acts to permit the entry of glucose into the fat cell is not clear. Since insulin promotes lipid synthesis and cyclic AMP (cAMP) promotes lipid breakdown, Butcher et al. (18) suggested that the action of insulin involves the inhibition of adenylyl cyclase activity thereby decreasing the concentration of cAMP in adipose cells. Illiano and Cuatrecasas (19) found that insulin may also directly modulate cAMP activity or modify adenylyl cyclase activity by producing a change in the conformation of the membrane. Still another hypothesis by Galton (20) explaining insulin action on the fat cell suggests that the insulin molecule combines with the plasma membrane at a site near the glucose carrier. An alteration in membrane structure takes place to enhance the carrier activity.

Davidson et al. (21) observed that obese subjects appear to handle a glucose load in a way quantitatively different from that of normal subjects. Glucose tolerance was often lower than normal, and this may be an early manifestation of the diabetes that frequently develops. However, when fasting, the plasma insulin concentration was higher in obese patients than in persons of normal weight.

Gordon and Goldberg (22) showed that after giving ^{14}C -glucose, more $^{14}\text{CO}_2$ appeared in the expired air of normal than in obese subjects.

After giving ^3H -glucose, more ^3HOH was found in the body water of the normal subjects. These findings indicated an altered rate of carbohydrate utilization in the obese.

Growth hormone metabolism is also affected in obesity. Growth hormone has a myriad of effects including an increase in muscle glycogen concentration, stimulation of FFA release from adipose tissue via hormone sensitive lipase, and stimulation of protein synthesis in for example, muscle tissue. In normal individuals factors increasing growth hormone are exercise, sleep, and a high protein or carbohydrate meal (7). However obese individuals seem to have the reverse growth hormone response to insulin (23), arginine (24), starvation (23) and sleep (25). Quabbe et al. (26) reported that the nocturnal rise in growth hormone during sleep in normal individuals was significantly lower in obese children and was nearly absent in obese adults. Schwarz et al. (27) showed that although exercise increased growth hormone levels in the blood of normal individuals it did not have this effect in the obese. Thus as a general rule, obesity blunts the responsiveness to all factors which normally stimulate the release of growth hormone.

Sims and Horton (4) found that as subjects gained weight there was a progressive impairment in the plasma growth hormone response that occurred 4-6 hours after glucose administration. Thus the diminishing growth hormone response following glucose ingestion appeared to be the result of the development of obesity rather than a primary cause. Decreased growth hormone secretion may, however, tend to perpetuate obesity once it has developed. Kalkhoff et al. (3) showed that after weight reduction the growth hormone response to insulin returned toward normal in obese patients.

The thyroid gland and its hormones also have important physiological effects on obesity. Bray (7) stated there was a positive correlation between excess weight and triiodothyronine (T_3). He hypothesized that the higher concentrations of T_3 were attributed to the resistance of peripheral tissue of obese people to the action of T_3 .

Alteration of Lipolysis and Lipogenesis. Lipolysis is the breakdown of triglyceride molecules into constituent fatty acids and glycerol. The first fatty acid is removed by a rate limiting step which is controlled by a "hormone sensitive" lipase. Removal of the second and third fatty acids occurs by a monoglyceride lipase (16).

Bortz (28) showed that serum fatty acids are frequently elevated in obesity. Dole (29) noted that 94 percent of his obese patients had fasting levels of free fatty acids which were above 600 $\mu\text{Eq/l}$, whereas only 18 percent of the normal subjects had values above 600 $\mu\text{Eq/l}$. Issekutz et al. (30) found that the elevated fasting levels of free fatty acids in obese subjects are probably the result of increased release of free fatty acids from adipose tissue, as well as increased peripheral rates of turnover.

Opie and Walfish (31) studied the plasma FFA concentration, which reflects a balance between the rate of FFA mobilization and FFA uptake by tissue, in obese and lean subjects. They found an early rise of the plasma FFA values in obese subjects during fasting, which probably indicated an increased rate of FFA mobilization. If, however, the greater adipose-tissue mass in obesity is taken into account, FFA mobilization may have been relatively defective.

Opie and Walfish (31) looked at the effect of exercise on plasma FFA concentrations. Four obese subjects and 4 lean subjects undertook a

standard exercise test after 21 hours of fasting. The test consisted of running up and down 90 stairs (each 16.5 cm high) within two minutes. The amount of work performed was proportional to the body weight. Immediately after exercise the subjects were rested, and a blood sample was taken. A second blood sample was taken fifteen minutes later.

Exercise reduced the plasma FFA concentrations of both lean and obese subjects to a similar extent from the resting values after 21 hours of fasting. However, fifteen minutes after exercise there was a significant difference in the plasma FFA response, with the lean group showing levels about 70 percent higher than the exercise values; in contrast to an increase of only 10 percent in the obese group.

FFA uptake by tissue appears to be adequate in obesity as judged by the fall in plasma FFA levels during exercise. Corvilain et al. (32) measured respiratory quotients of fasting obese subjects and found that they utilized lipid predominantly. Opie and Walfish (31) reviewed the study by Stuchlikova (33) who found elevations in blood lactate and pyruvate levels in fasting obese subjects indicating carbohydrate utilization. In contrast with the previous studies, Opie and Walfish (31) have indicated the results reported are compatible with a role for inadequate FFA mobilization and hence decreased lipolysis (during fasting) in the perpetuation and possibly genesis of obesity in human subjects.

The release of fatty acids from adipose tissue is also influenced by the size of the fat cell (34). Studies with animals (34) and man (35) showed that the rate at which glycerol is released by adipocytes is proportional to the size of the fat cell. Bray (7) reported that when adipocytes are reduced in size, their rate of basal lipolysis decreases in relation to the decreasing size. Conversely, large adipocytes show a

greater basal release of glycerol. In addition to basal lipolysis, the responsiveness of the hormone sensitive lipase is also related to the size of the adipose cell. The larger the fat cell, the greater its sensitivity to various lipolytic agents. Studies by Knittle and Ginsberg-Fellner (6) showed that when obese subjects reduced their weight, the lipolytic effects of hormones on the smaller adipocytes were smaller than those on the enlarged adipocytes obtained before weight loss.

Lipogenesis is the process by which glucose and fatty acids are transported into the cell where they are converted to triacyl glycerol (16). The addition of insulin to an incubation medium containing adipose tissue or adipose cells accelerates the rate at which glucose is metabolized by these cells (36). Bjorntorp et al. (37) showed that this effect of insulin resided in at least two loci: (1) insulin enhanced the entry of glucose (2) within the cell insulin enhanced the conversion of carbon from glucose into glycogen and fatty acids.

Aubert et al. (15) showed that obese mice, manipulated from a small litter size, had higher levels of liver lipogenesis than those of normal mice. This was suggested from the specific activity of the glucose-6-phosphate dehydrogenase measured in the fed state: 0.624 IU/mg protein for obese mice and 0.502 IU/mg for control groups. Glucose incorporation into triglyceride-glycerol and triglyceride-fatty acids was elevated in obese mice.

The initial step in the action of insulin on the fat cell is its binding to the plasma membrane (7). The relationship of obesity to the binding of insulin by cell membrane has been studied in the experimental animals. Kahn et al. (38) showed in mice with genetically inherited obesity, and in mice with obesity due to treatment with gold thioglucose,

there was a decrease in the number of insulin binding sites. Exercise also plays an important role in the response of adipocytes to insulin. Palmer and Tepton (39) demonstrated that in rats trained to run on a treadmill five days a week, lipogenesis from glucose was greater than in untrained animals. Bjorntorp (40) showed that obese individuals who were exercised showed evidence of improved glucose tolerance and decreased insulin levels.

Bray (7) found a correlation between plasma triglycerides and obesity in clinical studies. The high level of triglyceride is explained at least in part by the increased supply of free fatty acids to the liver which results from the increased rate of lipolysis associated with the large adipocytes.

A summary of metabolic alterations in the obese is shown in table 1.

II. Fat Cell Growth

In spite of all the research on obesity, its etiology is still not completely understood. Van Itallie and Campbell (41) proposed three causes for obesity based on a mechanistic approach: regulatory, metabolic, and constitutional. They suggested that regulatory obesity was caused by overeating as a result of psychologic or physiologic problems. Metabolic obesity, on the other hand, could result from a loss of enzymatic, hormonal, or neurologic control. Constitutional obesity may be caused by an increased number of fat cells (hyperplasia) during certain stages of growth.

Adipocyte Development. Fat cells are normally present in loose connective tissue either singly or in small groups. When large numbers of these cells are organized, the resulting lobules constitute adipose

TABLE 1

Summary of metabolic alterations in the obese

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- I. Fasting Blood Glucose Elevated in Chronically Obese
 - A. Impaired glucose tolerance
 - B. Increased amount of glucose available for synthesis of fat
 - C. Epinephrine resistance in obese correlated with abnormal glucose
 - II. Pyruvate, Lactate, and Citrate Levels are Increased
 - A. Inability to breakdown pyruvate thereby inhibiting fat utilization
 - III. Hyperinsulinemia
 - A. Insulin insensitivity
 - 1. "Dilution" of insulin receptors
 - 2. Alteration of intracellular metabolism in large fat cells
 - 3. Decreased insulin-receptor binding in obese
 - 4. Large adipose cells are less insulin sensitive
 - 5. Distortion of cell membrane causing changes in receptor site(s)
 - B. Increase in circulating insulin
 - 1. Consequence of hyperphagia induced early in life
 - 2. Diminished rate of carbohydrate utilization
 - IV. Decreased Growth Hormone
 - A. Obesity blunts responsiveness to stimulus
 - B. Decreased rate of lipolysis therefore tend to conserve fat tissue
 - V. Resistance of Peripheral Tissue to T_3
 - VI. Elevation of Serum Free Fatty Acids
 - VII. Increased Lipogenesis in Obese Experimental Animals
-

tissue (16). Adipose tissue, as other tissues in the body, may increase in size as a result of increases in cell number and/or cell size (42). Growth by cell replication or increase in cell number is called hyperplasia, whereas an increase in cell size is referred to as hypertrophy.

Bjorntorp (43) studied adipose tissue cellularity, and concluded from studies of humans of various ages, that fat cell number and size increases rapidly in children during the first years of life. The number of cells increases particularly from the thirtieth week of gestation through the first year. The fetus accumulates a great deal of fat in the last trimester of pregnancy. Apparently after the first year or two of life, there is little increase in cell number until the preadolescent growth spurt occurs. Adebajo (44) reported that human adipose cells derived from infants less than 1 year of age and adolescent children multiplied readily in culture. Cell division did not occur in cells obtained from 1- to 10-year old children, nor in fat-laden cells. He concluded that there are critical growth periods during which division of fat cells occurs. Hirsch (45) agreed with this concept and further speculated that the critical periods for adipocyte development in the human may be the last months of gestation, the first year of life, and during the preadolescent and adolescent growth period.

Development of adipose tissue cellularity in the Sprague-Dawley rat was studied by Hirsch and Han (46). The rat epididymal fat pad grew by increases in both cell size and cell number until week 15 of the rat's life, when cell number stabilized. Any further change in depot size was the result of change in the size of the adipocytes. The Zucker obese rat differs from the Sprague-Dawley in that adipocyte number, as well as adipocyte size, continues to increase beyond week 14 of the rat's life,

resulting in an animal with grossly enlarged adipose depots (46). Hirsch (45) showed that development of adipose tissue in the rat can be modified by overfeeding before weaning, resulting in hyperplasia. Thus, there appear to be "critical periods" of nutritionally-influenced permanent changes on adipose cellularity in experimental animals, as well as in human beings.

Methods of Measurement. To determine the size and number of adipocytes, two measurements are needed: the size of fat cells, and the total quantity of body fat. Bray (7) recommended three general methods for measuring the size of individual adipocytes: (1) the microscopic method, (2) the collagenase method, and (3) the osmium tetroxide method. In the microscopic method, the diameter of individual fat cells is measured optically after isolation from adipose tissue with collagenase. This enzyme breaks down the intercellular collagen bridges and releases free adipocytes. The diameter of the adipocyte can then be used to calculate fat cell volume mathematically. The third and most commonly used method was developed by Hirsch and Gallian (47). To perform the osmium tetroxide method in humans, small fragments of adipose tissue are obtained from subcutaneous sites by means of needle aspiration. In laboratory animals, adipose tissue samples are dissected from a fat depot. In both cases, shreds of tissue are washed, weighed, and fixed in 2% osmium tetroxide in 0.05 Mcollidine buffer. The osmium tetroxide combines with lipid and the adipocytes harden and fall away from the tissue matrix. Cells are then separated by serial filtration through nylon mesh, suspended in a known amount of saline, and counted in a Coulter electronic counter. In another tissue sample, percent lipid is determined and cell size is estimated in terms of average amount of lipid per cell. In

animal studies, total fat is determined directly by lipid extraction from the total tissue. In man, an indirect method is used, such as administration of tritiated water to determine water space. Lean body mass and fat content can then be calculated. Total cell number is calculated by dividing total fat by average cell size.

Using the latter method, Hirsch and Knittle (48) reported that non-obese individuals have an average cell size of 0.6 μ g triglyceride/cell and an average fat cell number of 25 billion. Severely obese individuals had three times as many adipose cells while their fat cells were 40% larger than those of nonobese persons. Knittle (49) reported that obese humans may have a normal or increased number of fat cells.

Effects of Obesity on Cell Number and Size. Bjurlef (50) examined the number and size of adipocytes in the abdominal panniculus of obese men at autopsy and found two groups: those with increased size and number of fat cells, and those with only increased size of adipocytes. He concluded that genetic factors are of prime importance in determining an individual's number of fat cells, and that environmental factors such as nutrition are probably the major determining factor in the enlargement of adipocytes in men who do not have an increased number of cells.

Hirsch and Knittle (48) also indicated that human obesity is accompanied by a marked increase in adipose cell number. They demonstrated that weight loss is achieved by a decrease in cell size while cell number remains high.

Sims and co-workers (51) showed that adult men made experimentally obese by overfeeding and restriction of activity had increased adipose cell size but not an increase in cell number. When overfeeding was discontinued, weight reduction occurred and adipocytes returned to normal size.

Salans et al. (52) observed significant variation in the size of fat cells between two depots in the same individual, as well as a significant variation between obese individuals. Brook et al. (53) examined this variability in children and suggested that most of the hyperplasia of fat cells probably occurs in the first year of life. Brook later found that the number of adipose cells of children who became obese in the first year of life was higher than those who became obese in later childhood. He also found that those children who were light for their age had a profound deficit in adipose cell number (54).

Knittle (55) found that obese children have a greater number of adipose cells, and generally larger cells than nonobese children.

Eid (56) reported that infants who gained excessive amounts of weight during the first six weeks of life had a significantly greater likelihood of being obese later in childhood. Huenemann (57) confirmed this finding, and showed that children who grew most rapidly during the first six months were most likely to be obese.

Hypercellularity in Different Degrees of Obesity. Gurr et al. (58) found no clear distinction between types of obese patients on the basis of their estimated total number of measurable body fat cells. The number of fat cells was weakly related to the degree of obesity. Weight reduction caused no appreciable change in fat cell number.

Jung et al. (59) did not observe hypercellularity in moderately obese patients. However, as obesity increases, fat cells enlarge, eventually reaching a plateau in cell size. From this point on, the number of observed fat cells increase; whether by filling pre-existing 'empty cells' or by increases in cell division has not been established. Thus, hypercellularity would be expected in the severely obese and should be

regarded as a normal physiological process resulting from obesity and not as a pathological entity. The measurement of cellularity is unlikely to assist in the assessment or treatment of simple obesity.

Genetic Factors. In humans many studies have shown that heredity plays a role. In families where both parents are of normal weight, less than 10% of their children will be obese. Yet if one parent is obese, 40% of their offspring will be obese and this increases to 80% if both parents are obese (60). Furthermore, the weight of adopted children bears little relationship to that of their adopted parents (61). Identical twins who have been separated and raised in different homes have been found to have similar body weights (62).

Bray (7) stated that both genetic and environmental factors influence the size and number of adipocytes. The genetic factors include differences in species and strains of animals, differences due to sex, and differences in anatomic location of fat deposits. A difference in adipose cell number and cell size has been found in genetically-transmitted obesity in rodents.

Several scientists have examined the interaction of genetic and nutritional components in animals with hypercellular obesity. Johnson et al. (63) studied this interaction in Zucker obese male rats. During the first 30 days of life, nutrition had a major effect on cell number. If dietary intake was reduced, cell proliferation was diminished. After 30 days, however, genetic factors became more important than nutritional factors in determining cellularity of the adipose tissue. By 12 weeks of age, genotypic differences became the major determinant of body weight. While early overfeeding significantly increased adipocyte number in both obese and nonobese rats, early underfeeding resulted in a smaller increase in

adipocyte number in the nonobese rats only. Thus it is possible that in the obese animal there is failure to regulate adipose cell number. The obese rat could continue to synthesize new fat cells long after cell proliferation has shut down in the normal rat.

Environmental Factors. Environmental factors also play an important role in modifying the size and number of adipocytes. If rodents are fed a diet restricted in calories, they will have fewer adipocytes than litter-mates exposed to optimal nutrition. Knittle and Hirsch (64) found that 4 rats suckled with one mother had significantly heavier epididymal fat pads and weighed more than animals suckled 20 per mother. The animals from these large litters had a smaller number of adipocytes in all depots than animals fed 4 per mother.

Dietary excesses may also increase the number of fat cells. Lemonnier (65) showed that 32-week-old female mice fed a high fat diet had an increase in the number of fat cells in the perimetrial and perirenal depots, but not in the subcutaneous depot. Female rats fed a high fat diet from age 5 months to one year also showed a significant increase in the number of fat cells in the perirenal depot.

Faust et al. (66) examined diet-induced obesity and demonstrated that adult rats of various strains became obese when they were fed a highly palatable diet for several months. They found that increased adipocyte number persists during subsequent weight loss and thus suggests new cell formation rather than lipid filling of differentiated cells normally too small to detect, and there is an orderly morphologic sequence of adipose depot enlargement so that initially only mean adipocyte size increases. After a certain size is reached, most further depot enlargement is due to adipocyte number increase.

Similar results were also shown by Mandenoff et al. (67) when studying adipose tissue cellularity in rats fed a "cafeteria" diet which is a free choice of highly palatable items. At 8 weeks, the increase in adipose stores was attributed to hypertrophy. Between the 8th and 20th week hypertrophy persisted, but hyperplasia appeared in the retroperitoneal and epididymal sites.

Peckham et al. (68) examined the effect of a high-fat diet on gross body and adipose tissue composition in the rat. The researchers found that although the average body weight attained by rats fed a high-fat diet for 62 weeks was 37% greater than that of rats fed a standard pellet diet, the total amounts of body water, protein, and ash were the same for both groups. However the discrepancies in weight were attributed to the differences in fat content.

III. Effect of Exercise on Obesity

Effect of Exercise on Food Consumption and Body Weight in Normal

Subjects. Katch et al. (69) investigated the effect of two different levels of exercise of equal caloric cost on food consumption and body weight changes in 24 normal male rats. For the low intensity exercise, the speed of the treadmill was set at 5 m/min for 1 hour at zero percent grade. For the high intensity work the speed was set at 16 m/min for 50 minutes at 10 percent grade. Food consumption was measured following exercise sessions at 1 hour, and every 4 hours thereafter for 24 hours during alternate weeks of exercise and rest. Results showed that rats exercised at high intensity levels exhibited a greater depression in food consumption and body weight gain than those exercised at low intensity levels. Both exercise groups had lower feed consumption and weight gain than nonexercised animals.

Mayer et al. (70) studied the effects of exercise duration on feed intake and body weight of normal rats and genetically obese adult mice. The mature rats, accustomed to a sedentary existence, were exercised on a treadmill at 1 mph for different lengths of time. It was observed that rats exercised for 20 minutes to 1 hour had lower feed intakes and body weights than sedentary mice. For longer durations of exercise (1-6 hours) feed intake increased proportional to the length of time they exercised and weight was maintained. At exercise intervals above 6 hours, the animals lost weight and feed intake decreased. These three ranges of activity were termed sedentary, normal, and exhaustion. Both the sedentary and the exhaustion ranges were considered 'non-responsive ranges,' since an increase in activity was not accompanied by a corresponding increase in feed intake.

In the same study (70) normal and genetically-obese mice were exercised on a treadmill (0.27 mph) for 1 hour daily. The 1-hour period of exercise forced on the animals resulted in greater feed intakes of the nonobese group only slightly, but it increased that of the obese animals considerably. However, the weight of the exercised nonobese animals did not differ from that of their nonexercised controls; while the rate of weight gain of obese animals was considerably decreased by the 1-hour exercise period.

Effect of Exercise on Food Consumption and Body Weight in Obese

Subjects. Woo et al. (71) examined the effect of increased physical activity on energy intake and balance in six obese women (167% above ideal weight) voluntarily hospitalized for metabolic balance studies. Three 19-day treatment periods were applied to each subject. A sedentary period was followed by two periods of treadmill exercise. The exercise

treatments increased daily energy expenditure to 110% (mild) and 125% (moderate) of sedentary expenditure. The daily energy expenditure was raised to the moderate level by setting a 2.5% grade on the treadmill. Subjects chose their own rate of walking (approximately 3 mph), and were given daily assignments (min/day) based on individual expenditure rates. Individual daily expenditure and ad libitum intake were determined. Subjects did not change intake level which allowed for energy balance during the sedentary period only. Thus, the difference between intake and expenditure between treatments was significantly different. Mean total body weight, fat, and lean body mass did not decrease significantly with treatment. The negative balance indicated no compensatory increase in intake due to increased physical activity.

In a later study by Woo et al. (72) the effect of prolonged exercise on voluntary food intake was examined in three obese women (187% of ideal weight). The metabolic balance study consisted of 57 days of moderate physical activity. Food was offered in extra quantity and individual treadmill assignments were given to subjects to increase daily expenditure to 125% of sedentary levels. Mean daily intake (1903 kcal/day) and expenditure (2882 kcal/day) did not change with time. As a result, negative energy balance was obtained and sustained as shown in the previous study. A consistent rate of weight loss (0.12 kg/day) at a reasonable cost (8200 kcal/kg) occurred. Thus, results indicate that obese women doing long-term moderate exercise do not compensate by an increase in caloric intake.

Rolls and Rowe (73) studied the effect of exercise on body weights of male and female rats. Adult male and female rats were maintained either in standard cages or in cages with a running wheel attached. Both groups

were offered either the mixed diet with lab chow or lab chow alone. Under both conditions of feeding, exercise had little effect on female body weight. However both sedentary and exercised females became obese on the mixed diet. But with males, both mixed diet and lab chow-fed exercised groups weighed less than the sedentary groups; only the sedentary males fed the mixed diet became obese. The mixed diet was withdrawn after 10 weeks. The body weights of the sedentary females were maintained at the mixed diet level, but in the exercised females body weights fell, but was regained 4-6 weeks later. The researchers concluded that there are sex differences in the effects of exercise on the maintenance of body weight in rats.

Effects of Exercise on Body Composition and Metabolism in Normal Subjects. Crews et al. (74) showed that exercised animals gained less weight than free-eating sedentary controls as a result of both an increase in caloric expenditure and a decrease in feed intake. Exercise resulted in a significant decrease in the percentage of the carcass fat, with a proportional relative increase in lean body mass. Because sedentary calorie-restricted animals had a higher fat content than exercised animals of the same weight, a fat-mobilizing effect of exercise was suggested.

Oscai and Holloszy (75) investigated factors influencing weight change in rats including exercise, feed restriction, and overeating. The body weight of rats was reduced by exercise or by restriction of feed intake over a period of 18 weeks. At 6 weeks of age, a sedentary group and an exercised group (which was subjected to a program of swimming) were provided with feed ad libitum. Two sedentary, paired-weight subgroups were calorie restricted so that they lost weight at the same rate as the exercisers. One paired-weight subgroup received the same protein intake

as did the exercised group. The other sedentary paired-weight animals ate a standard diet.

There were no significant differences in body composition between the two sedentary, paired-weight subgroups. The exercisers lost 182 ± 19 g as a result of both an increase in caloric expenditure and a decrease in feed intake. The sedentary, feed restricted animals lost an average of 182 ± 18 g. The sedentary, free-eating animals gained 118 ± 13 g. The carcasses of the exercised animals contained significantly less fat and more lean tissue than those of the sedentary, paired-weight animals, providing evidence for a fat mobilizing and protein conserving effect of exercise.

The composition of the body substance lost by the exercised animals was 78% fat, 5% protein, 1% minerals, and 16% water; compared to 62% fat, 11% protein, 1% minerals, and 26% water for the sedentary, feed-restricted rats. Fat accounted for 87% and water for 10% of the weight gained by the sedentary, free-eating animals. Results demonstrated that obese rats losing weight by caloric restriction had a loss of lean tissue. They also showed that a comparable reduction of weight induced by means of exercise in normal rats was associated with the loss of significantly less lean tissue and significantly more fat.

Oscai and Holloszy reviewed the study of Rodahl et al. (76), who reported that prolonged exercise increased fatty acid mobilization markedly. It appeared possible that the cumulative effects of daily, exercise-induced bouts of lipolysis could be responsible for the greater fat loss of the exercised animals. Furthermore, he concluded that this fat-mobilizing effect, which persisted for a considerable time after the cessation of exercise, could play a role in the conservation of lean

tissue by making available to muscle and organ cells more of the energy stored as fat.

Effects of Exercise on Body Composition and Metabolism in Obese

Subjects. Bjorntorp et al. (77) studied the effect of long-term physical training on the body composition of obese patients. Eight severely obese patients with adipose tissue hypercellularity, elevated body cell mass, and a juvenile onset obesity were subjected to scheduled physical training during 6 months without dietary restrictions. Each training session lasted for 35 minutes and started with 5 minutes of warming up by jogging and light gymnastics, followed by three periods of rather strenuous exercise of fixed intensity. Two periods of jogging were inserted between exercise periods. The exercise was finished with a 5-minute interval of jogging and light gymnastics, and the exercise was gradually decreased in intensity down to walking. Body weight, body cell mass, and body fat showed no significant changes after 3 or 6 months of training, suggesting that the lack of body fat decrease after the long training period might be characteristic for subjects with the type of severe obesity studied. These findings were in contrast to those of an earlier study by Bjorntorp et al. (78), in which a similar, although lighter, training procedure produced a marked body fat decrease in myocardial infarction patients without severe obesity.

In a later study, Bjorntorp et al. (79) examined the effect of a shorter but slightly more intense period of physical training on obese subjects. Severely obese subjects and sex- and age-matched controls underwent physical training during a 6-week period. The training consisted of an initial warming-up period, three 5-minute periods of cycling on an ergometer and walking, jogging, and light gymnastics in-between,

followed by a relaxation period with jogging down to walking. The intensity of bicycle training was adapted just below the individual maximal working capacity, so that almost complete exhaustion was reached each 5-minute period. Training sessions had a duration of about 1 hour and took place 3 times/week. In controls there was evidence for the beginning of a decrease in adipose tissue mass. However, in the obese group, body weight, body fat, and fat cell size did not decrease during training even though there was no apparent change in dietary energy intake. The authors suggested that the extra energy output was balanced by an extra energy input that was not discovered.

In another study, Bjorntorp et al. (80) investigated the effect of intense physical training on 10 obese subjects. The group was trained in a gymnasium under the leadership of a physiotherapist during an eight-week period; four hours/day five days a week during the first two weeks, and thereafter three hours/day three days a week. Each training program followed a fixed schedule, but the intensity of the different parts was proportioned according to the individual level of physical fitness. An initial warmup period on the ergometer bicycle lasted four minutes, followed by physical training on the bicycle and static and dynamic training of muscle power in arm, abdominal, back, and leg muscles.

Three measured changes occurred after physical training. The first was an increase in body fat, which the authors proposed resulted from a small increase in fat cell size. The second change was an increase in body cell mass in some of the patients; although this was not significant in the whole group. Increased cell mass suggested an increase in muscle mass during physical training. The final alteration was an increase in working capacity and muscular strength.

Effect of Exercise on Adipose Cellularity. Deb and Martin (81)

studied the effect of exercise and feed restriction on the development of spontaneous obesity in rats. Lean and Zucker obese rats were divided into four experimental groups. The groups were: Group 1, lean, control, no exercise with ad libitum feeding; Group 2, obese, no exercise, pair-fed to group 1; Group 3, lean, exercised, pair-fed to group 1; Group 4, obese exercised, pair-fed to group 1.

The exercise program started when the animals were 8 weeks of age. Week 1 of exercise was a conditioning learning phase of running. In week 2, the animals ran for 1 hour at a speed of 1 mph with a 2% grade, 5 days/week. This was repeated for the following 2 weeks. In week 5 of exercise, the animals ran for 1 hour at the rate of 1 mph with a 4% grade, 5 days/week. The last exercise program was continued until the rats had a total period of 11 weeks of exercise.

Results of the study showed that liver weights of the obese animals were heavier than those of the lean nonexercised animals and were not influenced by exercise. The fat pads of obese animals were significantly heavier than those of the lean animals. Exercise significantly reduced the fat pad weights only in the obese rats. Exercise had no effect on adipose cellularity of the obese rats. Although exercise reduced fat content significantly, obese rats were still fatter (27.7% fat) than the lean controls (6.4% fat). The protein content of obese rats was significantly increased by exercise, indicating that physical activity affects protein synthesis.

Physical Training in Obese Humans. Bjorntorp et al. (80) examined the effect of physical training on insulin production in obesity. Two obese subjects were subjected to physical training. Before training the

obese subjects had more body fat and larger fat cells than control subjects. The patients were told not to restrict their diet during the whole investigation. The training, as described in a previous section, resulted in an increased maximal oxygen consumption and an increased isometric muscle strength. Body weight increased, due primarily to an increase in body fat, but also in several cases from an increase in body cell mass. Fat cell diameter was unchanged. Peroral glucose tolerance test showed no changes in blood glucose values after training but a marked decrease in insulin levels. This was interpreted as an increased sensitivity of tissues to insulin. Since the body fat mass was not decreased, it was not considered likely that the increased insulin sensitivity was due to adipose tissue factors. Furthermore, the increase of insulin sensitivity was not found to be related to the increase in body cell mass and therefore probably not to an increase in muscle mass. Therefore, it was concluded that the effect of physical training producing a decrease in insulin resistance in the obese patients was caused by a factor in muscle metabolism.

In a later study by Bjorntorp et al. (79), the effect of exercise on hormonal status in human hyperplastic obesity was examined. Severely obese subjects and sex- and age-matched controls underwent physical training during a 6-week period. Before training the obese subjects exhibited decreased glucose tolerance, hyperinsulinemia, elevated blood glycerol, and plasma-free fatty acids. During the training period all subjects had increased aerobic power. As in the previous study, patients and controls were told to eat freely and not adhere to any kind of restriction in food intake before or during the physical training period. Results from the experiment showed a decrease in insulin with a corresponding increase in

plasma glycerol. The investigators suggested that the increase of plasma glycerol might be due to an increased adipose tissue lipolysis associated with the decreased insulin concentration after training. Glucose tolerance was not changed, and this, together with decreased plasma insulin, indicated an increased insulin sensitivity of the peripheral tissues. Urinary cortisol output was found to decrease after training in the obese. The researchers concluded that there were several significant correlations between urinary cortisol output and plasma insulin levels, but this is not fully understood.

In a third study, Bjorntorp et al. (77) studied the effect of long-term physical training on human obesity. Eight severely obese patients with adipose tissue hypercellularity, elevated body cell mass, and juvenile onset obesity were subjected to physical training during 6 months without dietary restrictions. The maximum work loads during these training sessions were individualized according to each subject's maximal working capacity and kept 10-15 pulse beats per minute below this level. Aerobic power increased in all subjects after three and six months. Fasting plasma insulin decreased after 3 months of training, but fasting blood glucose was not changed. After 6 months, plasma insulin values were still decreased, and glucose tolerance had improved. Glucose tolerance may indicate a primary effect of physical training on glucose removal mechanisms rather than on insulin secretion.

In summary, Bjorntorp's studies showed that hyperinsulinemia was reduced in severely obese subjects but they decreased their body fat abnormally slow or not at all when subjected to physical training on an ad libitum diet. He emphasized that in all the studies patients had been selected who were severely obese, who usually had been obese since

childhood, and who had an elevated body cell mass and a hypercellular adipose tissue.

Nonobese subjects usually show a decrease in body fat during physical training. They encounter a negative caloric balance during this training period in spite of the fact that food intake might well be increased during this period, particularly in training athletes. They have, however, an appetite regulation which is set to decrease adipose tissue mass during training. It seems an attractive possibility that appetite regulation is in some way dearranged in hyperplastic obese subjects with physical training on ad libitum diet (40).

The main difference between the results of physical training in severely obese and nonobese subjects seem to be the lack of decrease in body fat in the former. It is not yet possible to state whether a normalization of plasma insulin occurs in obesity.

Deb and Martin (81) showed that Zucker obese rats pair-fed to lean littermates gained more body fat on the same intake; indicating greater efficiency of diet utilization. Exercise significantly reduced the fat pad weights, increased protein content and decreased fat content in the obese rats. The experimental obese animals had significantly higher serum insulin levels. Though exercise tended to lower serum insulin levels in obese rats to a certain extent, the decrease was not significant. These findings are in contrast to those of Bjorntrop et al. (77). Liver tissue in-vitro lipogenic capacity and lipogenic enzyme activities were significantly elevated in obese rats. In obese rats, exercise increased citrate cleavage enzyme significantly. The enzyme 6-phosphogluconate dehydrogenase and glucose-6-phosphate-dehydrogenase were significantly reduced by exercise. These enzymes are normally associated with lipogenesis and

their reduction possibly aids in the decreased fat content of the exercised obese rats.

Richard et al. (82) investigated the effect of exercise-training on insulin levels in exercise-trained female rats made obese by a palatable diet. The exercise-training program consisted of forced swimming 6 days per week 2 hours per day. Results of the study showed that the high-energy diet and exercise-training had opposite effects on both glucose tolerance and insulin secretion, with the high-energy diet impairing glucose tolerance and increasing insulin levels, and the exercise-training leading to the opposite adaptation. Exercise training improved glucose tolerance in animals fed standard laboratory chow and reduced plasma insulin levels, regardless of diet. However, when rats were fed supermarket foods with simultaneous exposure to the physical training program, they became heavier, fatter, were less glucose tolerant, and more insulinemic than the control sedentary rats fed standard laboratory chow. From these results Richard and coworkers (82) suggested that the reduced insulin response to glucose observed in exercise-trained animals may be related to accompanying reduction of adiposity rather than to exercise per se. This suggestion is in contrast to the findings of Bjorntorp et al. (80), and Bjorntorp et al. (79), who observed reduced plasma insulin levels in obese subjects after physical training, without significant loss of body weight.

In contrast to earlier studies by Bjorntorp (79, 80), Oscai and Holloszy (75) showed that regularly-performed exercise produced a marked decrease in body weight in obese (free eating) male rats subjected to 2 hours of daily swimming for 18 weeks. Weight loss, which amounted to approximately 25% of the animals' initial weight, resulted from both a

suppression of appetite and an increase in caloric expenditure, with the latter contributing roughly three times as much as the former to the negative energy balance. The major portion (78%) of the weight loss that occurred in the exercised animals was due to a loss of fat. The exercise provided significant, but not complete, protection against the loss of protein and fat-free weight was associated with a negative caloric balance. Although the weight loss of the exercised and the sedentary, calorie-restricted animals was the same, the exercisers lost significantly more fat and less than one-half as much protein. The finding that the exercised animals were leaner than the sedentary controls of the same weight suggests a lipid-mobilizing effect of exercise, in addition to the increase in caloric expenditure and decrease in feed intake.

A study by Larsson and Jonsson (83) also showed a decrease in weight gain in obese mice as a result of exercise. In the experiment, spontaneous activity in "squirrel wheels" (mild exercise) was conducted for 30 days with gold thioglucose-induced obese male mice of the Swiss, NMRI and CBA strains. Results showed that weight gain of the controls was more than twice that of exercised mice. The body fat content in all strains after activity was significantly lower than that in the controls, and in obese animals, the carcass fat content was similar to that of normal animals. The protein-fat ratio in these animals also was similar to that of the nonobese mice.

MATERIALS AND METHODS

Animals and Their Care

The experimental animals were Swiss albino male mice obtained from our departmental breeding colony.¹ Early normal nutriture and over-nutriture were accomplished by manipulation of litter size during the nursing period (birth to 3 weeks of age). It had previously been shown that litter size for "normal-fed" nursing Swiss mice is 8 pups/litter (84); this litter size was used for our normally nourished mice. Overfed mice were raised 4 pups/litter. Litter size was adjusted to 1 day of age and only male pups were used. There were 8 litters of normal-fed mice (8/litter) and 8 litters of overfed mice (4/litter) in our study.

At 4 weeks of age one mouse per litter was sacrificed and body composition of those animals was determined. The remaining litters of normal-fed and overfed mice were subdivided into sedentary or exercise groups on a littermate basis. At this time the normal-fed mice weighed 7.6-14.2 g and the overfed mice weighed 14.4-19.4 g. They were housed individually in wire bottom cages and fed a stock diet² and water ad libitum throughout the study. Room conditions were maintained at $(20^{\circ} \pm 2)$ with an automatic 12-hour light-dark cycle.

The exercise groups were exercised 2 hours a day by running on a

¹Original breeding stock for this colony was the Ha(ICR) Swiss Albino strain obtained from Harlan Sprague Dawley (Indianapolis, IN).

²Purina® Rodent Laboratory Chow 5001, St. Louis, MO. This diet contains approximately 23.4% protein, 4.5% fat, 5.0% fiber, 7.3% ash, and 49.8% nitrogen-free-extract (by difference).

motor-driven zero-grade treadmill.³ The sedentary mice remained in their cages during this time.

The exercise periods were conducted 5 days a week (Mon.-Fri.) in the late morning and early afternoon hours for 6 weeks. The first two weeks were set aside as a "training" period in which the animals gradually adapted to a maximum speed of 0.5 mph (table 2). For the remaining 4 weeks, the exercised mice ran at a speed of 0.5 mph.

During the 6-week period body weight was measured twice a week (Mon. and Fri.), and feed intake was measured daily. Mice were weighed on Monday and Friday to see if different activity levels had a temporary effect on body weight (mice were rested on weekends). Weekday weight gain was calculated by subtracting Monday body weight from Friday body weight. Weekend weight gain was calculated by subtracting the previous Friday body weight from Monday body weight. Daily feed intake for each mouse was calculated by subtracting residual feed left each day from the feed weight of the previous day.

Sacrifice Procedures

At the end of the 6-week exercise study, the mice were sacrificed by cervical dislocation. A hair removal mixture used previously by Crews et al. (74) which is composed of barium sulfide (200 g), detergent (Tide, 50 g), and 10% glycerol (500 ml mixture), was used to remove the mouse fur. After washing the mice, the digestive tract from the gastro-esophageal sphincter to the anus were then removed from each mouse and discarded. This was done to minimize variations in body weight and

³Radiotrol Treadmill, 1/2 h.p., Boston Gears, Quincy, MA.

TABLE 2

Preliminary training schedule: progressive exercise by speed
acceleration during the two-week period

Week	Day	% of Target speed	Speed (mph)
1	Mon.	(10)	.05
	Tue.	(20)	.10
	Wed.	(30)	.15
	Thur.	(40)	.20
	Fri.	(50)	.25
2	Mon.	(60)	.30
	Tue.	(70)	.35
	Wed.	(80)	.40
	Thur.	(90)	.45
	Fri.	(100)	.50

composition caused by periodic food intake. The mouse without the digestive tract was considered for analytical purposes, the mouse carcass.

The measurements taken at sacrifice included body weight, weight without fur, carcass weight, and epididymal fat pad weight. The fat pads removed for weighing were returned to the rest of the carcass which was placed in a pint-size labeled canning jar and stored at -18° for later analysis.

Preparation and Homogenization of Mouse Carcasses

The flow chart for carcass homogenization is shown in figure 1. The jars containing the frozen carcasses were allowed to sit at room temperature for 1 hour. Four volumes ($4 \times$ carcass weight) of deionized water were added to each jar and a blade, rubber gasket, and cap were placed on the jar opening. The caps were closed tightly on each jar and then unscrewed about $1/4$ turn to allow the steam to escape during autoclaving. Jars were autoclaved in batches of 18 in a stainless steel tray for one hour at 121° and 15 psi. After autoclaving, jars were cooled in a 37° water bath for one hour. The eighteen jars included at least one mouse from every treatment group.

Each jar was removed from the water bath and prepared for homogenization. To prevent leakage, 2 rubber gaskets were placed between the jar mouth and blade, and another gasket was placed between the blade and an adaptor. The adaptor was screwed onto the jar and the whole unit was fitted onto an Osterizer blender. The mouse carcass was then homogenized one minute at low speed (stir) and 4 minutes at high speed (liquify).

The jar containing the homogenate was placed on a magnetic stirrer and was mixed for about 5 minutes to keep the homogenate in suspension.

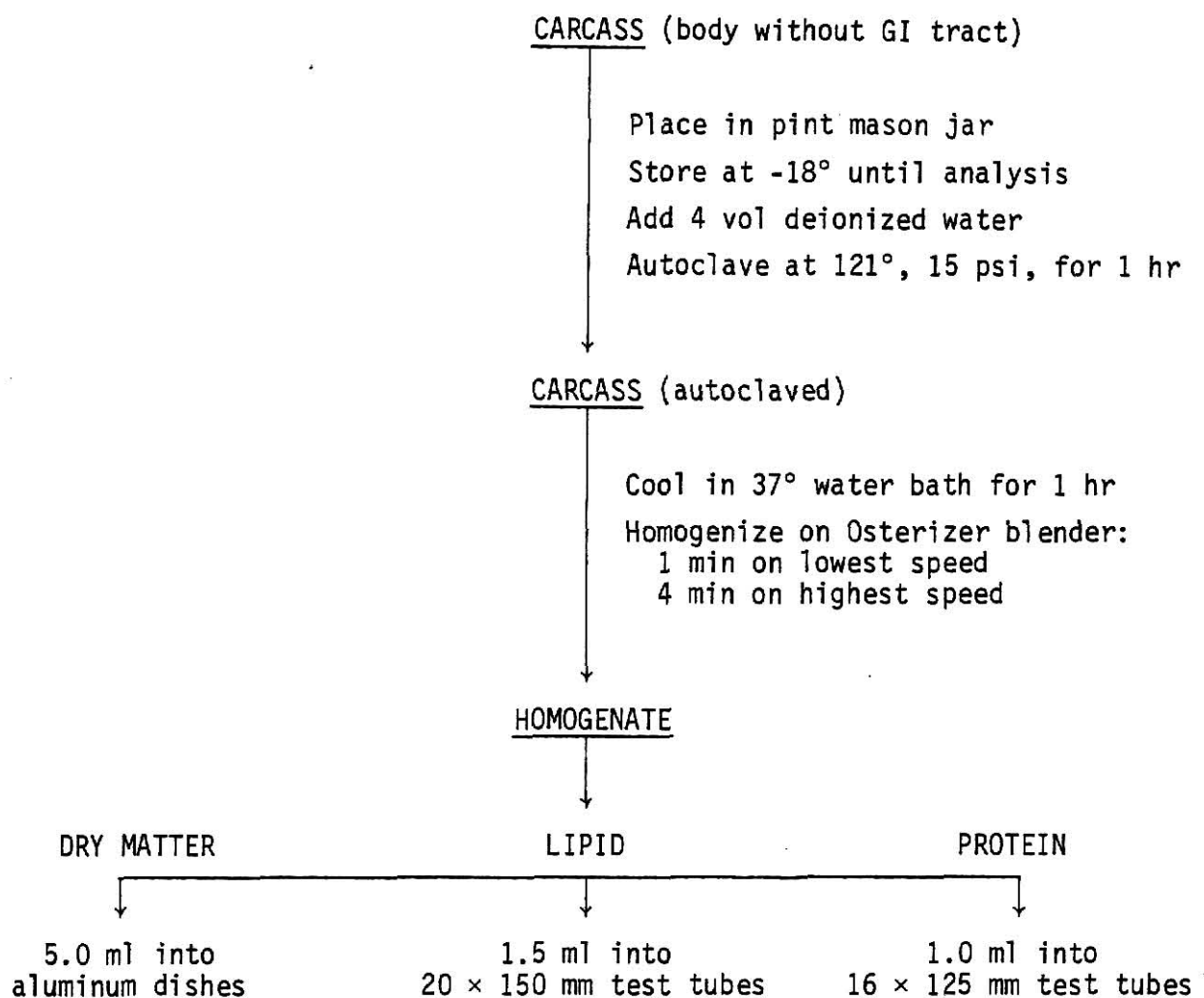


Fig. 1 Preparation and homogenization of mouse carcasses.

Aliquots were pipetted into prelabeled test tubes for body composition analyses. Volumes of duplicate aliquots were as follows: 5.0 ml into pre-weighed aluminum dishes for dry matter determination, 1.5 ml into 20 × 150 mm test tubes for lipid analysis and 1.0 ml into 16 × 125 mm test tubes for protein analysis. Dry matter and protein determinations were performed on the day of homogenization. Samples for lipid analysis were stored at -18° before analysis. Analytical procedures for dry matter, protein, and lipid are shown in figure 2 (85), and are explained in greater detail in the following paragraphs.

Determination of Dry Matter

Aluminum dishes (S/P catalog #D2165) were labeled, dried 2 hours in a 103° forced-air draft oven, cooled 1 hour in a desiccator and weighed to the nearest .001 g. Dishes were then arranged in the work area and 5 ml of each homogenate was pipetted into each pre-weighed dish while the homogenate was mixing on the magnetic stirrer. The dishes containing the homogenate samples were then weighed and dried at 103° for 5 hours. Dishes were cooled in a desiccator for 1 hour and weighed. The percent dry matter was calculated by dividing the weight of the dried homogenate by the weight of the 5 ml homogenate before drying and multiplying by a correction factor of 500. This correction factor was obtained by multiplying 5 (initial dilution for homogenization) by 100 (to obtain percentage). Percent body water for each animal was calculated at 100 minus the percent dry matter.

Determination of Protein

Protein analysis was performed on 1.0 ml homogenate on the day of homogenization using the Folin-Lowry procedure (86). Each 1.0 ml sample

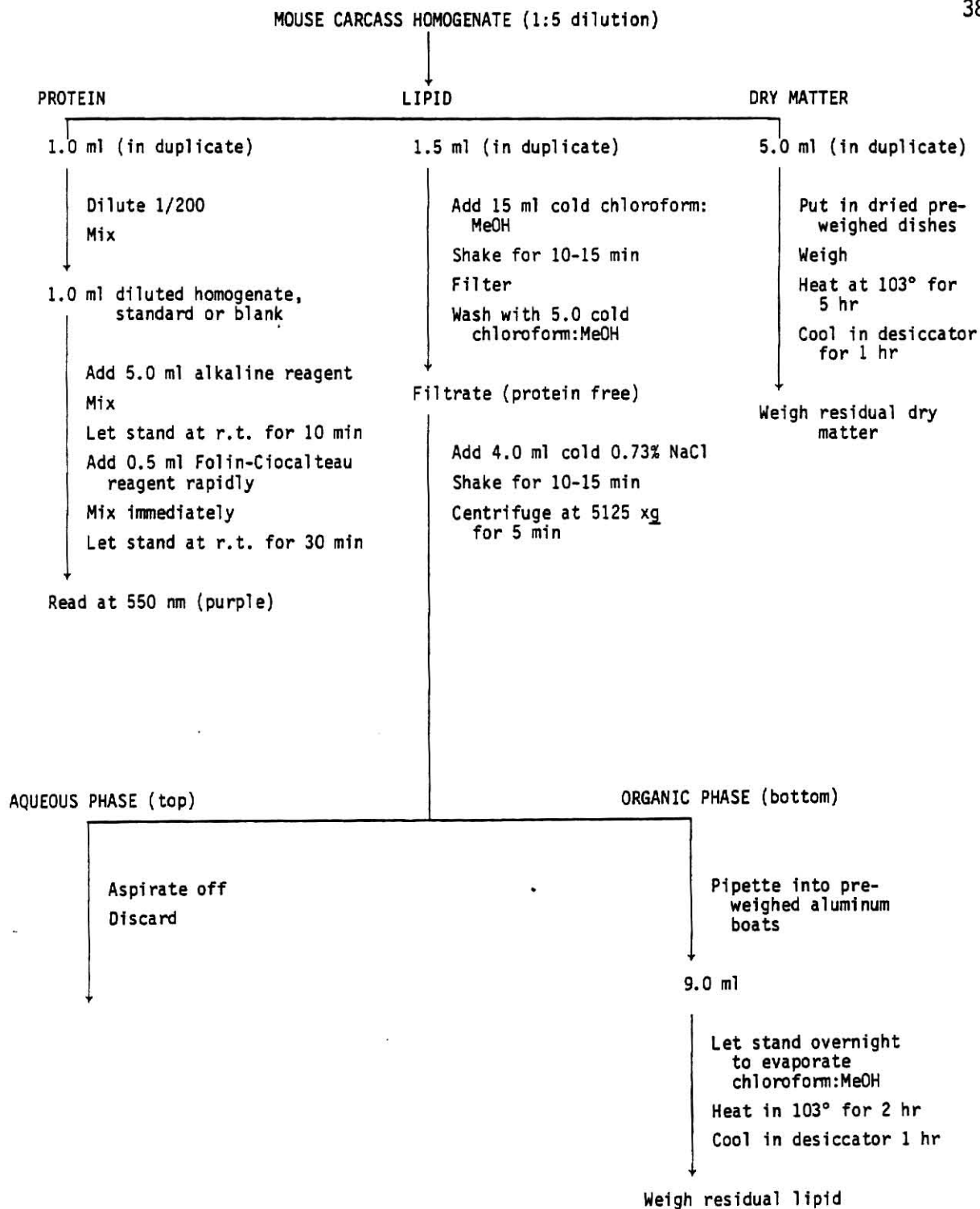


Fig. 2 Analyses for protein, lipid, and dry matter in mouse carcass homogenate.

was diluted 1/100 with distilled water and one ml of this mixture was used for further analysis. To each tube containing the diluted homogenate was added 5 ml alkaline reagent (see appendix table 1 for preparation of reagents). After mixing, each tube was allowed to stand at room temperature for 10 minutes. Then 0.5 ml Folin-Ciocalteu reagent was added rapidly and the tube contents were mixed immediately on a vortex mixer. After 30 minutes the optical density (purple color) was read at 500 nm on a spectrophotometer. Eight samples could be analyzed at a given time with a distilled water blank (for zeroing the spectrophotometer) and a protein standard solution (0.2 mg bovine serum albumin/ml). To correct for non-colored turbidity observed in each sample, we prepared a "sample blank" containing 1.0 ml diluted homogenate (1/100) and 5 ml alkaline reagent without the copper sulfate, and subtracted the absorbance reading of the "sample" tube. The resultant value was considered the corrected absorbance reading. Protein contents of each tube were determined by linear regression after comparing the corrected absorbance reading of each sample to those of the distilled water blank and protein standard. These values were then multiplied by a correction factor of 50 to get percent body protein. This factor was obtained by multiplying 5 (initial dilution for homogenization) by 100 (dilution of homogenate), divided by 1000 (conversion of mg to g), times 100 (to obtain percentage).

In this method of assay a purple-colored complex is formed when the copper in the alkaline reagent reacts with the peptide bonds in protein and the reduction of phosphomolybdate by tyrosine and tryptophan in the protein.

Determination of Lipids

The Folch method (87) was used to analyze fat content in the 1.5 ml homogenate. Fifteen ml cold chloroform-methanol (2:1, v/v) was added to the homogenate in each 20 × 150 mm test tube. Tubes were shaken 10-15 minutes on a wrist-action shaker. Samples were filtered through Whatman #1 filter paper into glass conical centrifuge tubes. An additional 5.0 ml chloroform-methanol was used to wash the filter paper. Four ml cold 0.73% NaCl was then added to each centrifuge tube and tubes were shaken again for 10-15 minutes.

After centrifuging in a clinical centrifuge at highest speed (5125 xg), the tube contents had separated into 2 phases: the top layer being the aqueous phase and lower layer the organic phase containing the lipids. The upper phase was aspirated off by a vacuum pump into an erlenmeyer flask trap. Nine ml of the organic phase was pipetted into pre-weighed aluminum dishes (prepared as for dry matter determination). The dishes were allowed to stand overnight during which time the chloroform-methanol evaporated leaving the lipid residue in the dishes. The following day the dishes were heated in a 103° oven for 2 hours, cooked in a desiccator 30 minutes, and weighed. Percent body fat (lipid) was calculated by multiplying 5 (initial dilution for homogenization), by 1.7777 (16 ml total organic phase/9 ml organic phase used), divided by 1.5 ml (because 1.5 ml homogenate was used), times 100 (to get percent).

Statistical Analyses

The effects of early nutriture on sacrifice measurements of weanlings (4-week-olds) before exercise treatments were determined by Analysis of Variance procedures (88). The sources were as follows:

<u>Source</u>	<u>d.f.</u>	
OBNOB	1	OBNOB = litter size
mom(OBNOB)	16	mom(OBNOB) = litter size within litter

The remaining mice were subdivided into exercise and nonexercise groups to form a split plot design. Early nutriture (litter size) was the main plot and exercise the subplot. Analysis of Variance procedures were performed with sources as follows:

<u>Source</u>	<u>d.f.</u>	
OBNOB	1	
mom(OBNOB)	16	
EXNEX	1	EXNEX = exercise treatment
EXNEX*OBNOB	1	EXNEX*OBNOB = interaction between exercise and litter size

The mean square for litter size within litter (mom(OBNOB)) was used as the error term.

Statistical procedures were carried out using the computerized SAS (Statistical Analysis System) (89). A sample computer program for our data is shown in appendix table 2.

RESULTS AND DISCUSSION

Effect of Litter Size on Body Weight and Body Composition at Weaning

Early nutrition and growth can be influenced by a manipulation of litter size. Because the nursing mother has a limited amount of milk, mice from small litters receive an abundant supply of milk, whereas those from large litters receive smaller amounts. Mice are usually nursed for 3 to 4 weeks and then weaned onto solid food.

Epstein (84) showed that average pup weight at weaning is inversely related to litter size down to 4 pups/litter. In her study the litter size for "normal-fed" nursing Swiss mice was 8 pups/litter. She found that mice from four-pup litters weighed an average of 70% more than those for the eight-pup litters at 23 days of age. In our study mice from four-pup litters were 69.3% heavier than those from the eight-pup litters at 28 days of age.

The effect of litter size on mean body weight, weight without fur, and carcass weight of male mice at weaning (4 weeks) are presented in table 3. Mice from the 4/litter group were significantly heavier ($P < 0.05$) than those from the 8/litter group when all three measurements of weight were compared.

The effect of litter size on body composition at weaning is shown in tables 4 and 5. Weanling mice in the 4/litter group had significantly more ($P < 0.05$) body fat and significantly less ($P < 0.05$) lean body mass than those in the 8/litter group. Hirsch (45) observed that when rats are overfed before weaning, there is an increased percentage of body fat. Investigators (63, 64) have shown that rat adipose cellularity is affected by both an increase in cell number and cell size. Up to 15-20

Effect of litter size on body weight (g) of mice at weaning¹

Measurement	8/litter	4/litter	Source of variation	df	Mean square and significance ²		
					Whole body weight	Weight without fur	Carcass weight
Whole body weight	11.3 ± 0.5	16.3 ± 0.6	OBNOB ³	1	101.0*	205.2*	163.8*
Body weight without fur	9.0 ± 0.5	16.2 ± 0.6	Residual	14	2.3	30.4	21.7
Carcass weight	7.2 ± 0.4	13.6 ± 0.5					

TABLE 4

Effect of litter size on percent body fat, body protein, and lean body mass of mice at weaning¹

Measurement	8/litter	4/litter	Source of variation	df	Mean square and significance ²		
					Body fat	Body protein	Lean body mass
Body fat	12.4 ± 1.2	15.8 ± 0.8	OBNOB ³	1	46.2*	7.8 NS	46.2*
Body protein	15.9 ± 1.1	14.5 ± 0.5	Residual	14	7.9	5.4	102.7
Lean body mass	87.6 ± 1.2	84.2 ± 0.8					

TABLE 5

Effect of litter size on percent dry matter and body water of mice at weaning¹

Measurement	8/litter	4/litter	Source of variation	df	Mean square and significance ²	
					Dry matter	Body water
Dry matter	39.0 ± 1.4	34.8 ± 0.8	OBNOB ³	1	71.0*	48.0*
Body water	61.0 ± 2.2	65.2 ± 0.8	Residual	14	11.1	39.1

¹ Mean ± SEM for 16 animals per group.² * P < 0.05, NS P > 0.05.³ Litter size.

weeks of age adiposity in rats is determined by an enlargement of cell size and cell number. Aubert et al. (15) reported that litter size in mice affects both cell number and size. Mice raised in litters of four had fat depots with an increase in cell number and size. In contrast, mice raised in large litters had fewer and smaller cells. The increase in percentage of body fat observed in our study is probably due to both an increase in adipose cell size and number, although this was not studied.

Early nutrition showed no effect on percent protein in either of the groups.

The 8/litter mice had significantly more ($P < 0.05$) dry matter than the 4/litter mice (table 5). Litter size did not affect percent body water at the time of sacrifice.

Exercise and Feed Intake

The effect of exercise on feed intake of the early normal-fed and overfed mice during the 6-week study is shown in table 6. Weeks 1 and 2 were used as a training period where mice were not exercised at the target speed of 0.5 mph, but were on the progressive exercise schedule shown in table 2.

Feed consumption was measured during the training period and the following four weeks of target-level exercise. Average daily feed consumption was measured as the average per weekday (Monday to Friday/5), and as the average per weekend (Saturday and Sunday/2). Because the 4/litter mice were initially heavier than the 8/litter mice it was necessary to express feed intake per unit body weight. Feed/body weight ratios are listed in table 6.

During the initial training period the 4/litter mice consumed more feed per unit body weight than the 8/litter mice. This suggests that

TABLE 6
Effect of exercise on feed/body weight ratio¹ (g/g) of early
normal-fed and overfed mice (week 1-6)²

	8/litter		4/litter		source of variation	df	Mean square and significance ²	
	-Exer.	+Exer.	-Exer.	+Exer.			Ave. weekday	Ave. weekend
Training (weeks 1&2) ³					OBNOB ⁴	1	0.080*	0.111*
Ave. weekday	0.252±0.004	0.290±0.008	0.245±0.005	0.341±0.061	MOM(OBNOB) ⁵	16	0.016 NS	0.004 NS
Ave. weekend	0.294±0.021	0.390±0.021	0.334±0.027	0.396±0.019	EXNEX ⁶	1	0.009 NS	0.009 NS
					EXNEX*OBNOB ⁷	1	0.015 NS	0.005 NS
					Residual	16	0.017	0.006
Target (weeks 3-6)								
Ave. weekday	0.229±0.005	0.239±0.007	0.211±0.008	0.226±0.005	OBNOB ⁴	1	0.005*	0.002 NS
Ave. weekend	0.256±0.012	0.247±0.007	0.249±0.010	0.239±0.010	MOM(OBNOB) ⁵	16	0.001 NS	0.002 NS
					EXNEX ⁶	1	0.009*	0.001 NS
					EXNEX*OBNOB ⁷	1	0.000 NS	0.000 NS
					Residual	16	0.001	0.003

¹ Ratios calculated as average daily feed consumption over body weight for that period. ² Results are expressed as means ± SEM for 9 mice in each group. ³ During weeks 1 and 2, mice were not exercised at the target speed, but were on the progressive exercise schedule shown in table 2. ⁴ Litter size. ⁵ Effect of individual mothers. ⁶ Exercise. ⁷ Interaction between litter size and exercise.

the 4/litter mice were influenced by factors which influenced their greater feed consumption. It is possible that they were somehow "programmed" early in life (pre-weaning) to consume more feed. Exercise did not seem to affect feed intake during the training period, possibly because the exercise at this time was at an insufficient intensity and/or duration to elicit a response.

The effects of early nutrition and exercise were also studied during the following 4-week target exercise period. In contrast to the training period, the 4/litter mice had lower adjusted feed intakes than the 8/litter mice. Although the reasons for this reversal are not clear, one explanation is that once the 4/litter mice attained a certain weight, they became more energy efficient, and did not require as much energy for maintenance. This was noted only during the weekday periods when the mice were exercised.

During weekdays the exercised mice consumed more feed/body weight than nonexercised mice. This suggests that the level of exercise in this study resulted in a greater feed intake than if the mice were not exercised. There was no interaction between exercise and litter size on feed intake suggesting that the 8/litter and 4/litter mice responded similarly to the effects of exercise.

Exercise and Weight Gain

The measurement of weight gain is one way to assess growth. At weaning the mice were not fully grown, so a net weight gain was anticipated throughout the 6-week study. The effect of exercise on weight gain on early normal-fed and overfed mice is shown in table 7 and figure 3. After the 2-week training period there were no significant differences due to litter size or exercise. But by the end of the 6-week study there

TABLE 7
Effect of exercise on weight gain (g) after 2 weeks and end of study of early
normal-fed and overfed mice¹

Measurement	8/litter		4/litter		Source of variation	df	Mean square and significance ²	
	-Exer.	+Exer.	-Exer.	+Exer.			Weight gain after 2 weeks	Weight gain at end of study (6 weeks)
Weight gain (training, weeks 1&2)	10.5±0.6	11.0±0.7	10.7±1.4	9.2±0.5	OBNOB ³	1	5.4 NS	24.2*
Weight gain during 6-week study	20.4±0.6	18.1±0.6	18.9±0.8	16.2±0.6	MOM(OBNOB) ⁴	16	6.2	5.6
					EXNEX ⁵	1	2.4 NS	57.0*
					EXNEX*OBNOB ⁶	1	7.9 NS	0.3 NS
					Residual	16	7.6	2.5

TABLE 8
Effect of exercise on final body weights (g) of early normal-fed and overfed mice¹

Measurement	8/litter		4/litter		Source of variation	df	Mean square and significance ²		
	-Exer.	+Exer.	-Exer.	+Exer.			Whole body weight	Weight with- out fur	Carcass weight
Whole body weight	32.6±0.7	30.1±0.7	36.1±0.7	33.2±0.9	OBNOB ³	1	96.4*	84.3*	55.0*
Weight without fur	32.1±0.7	29.7±0.7	35.5±0.7	32.4±0.9	MOM(OBNOB) ⁴	16	7.2	7.8	5.3
Carcass weight	26.7±0.6	24.7±0.5	29.7±0.7	26.6±0.8	EXNEX ⁵	1	63.2*	70.3*	57.5*
					EXNEX*OBNOB ⁶	1	0.3 NS	1.1 NS	2.3 NS
					Residual	16	2.6	2.3	2.3

¹ Mean ± SEM for 9 animals per group. ² *P < 0.05, NS P > 0.05. ³ Litter size. ⁴ Effect of individual mothers. ⁵ Exercise. ⁶ Interaction between litter size and exercise.

were significant differences between the 4/litter and 8/litter mice. The 8/litter groups gained significantly more ($P < 0.05$) than the 4/litter. It is possible that the 8/litter mice "caught up" to the 4/litter mice as a compensatory response to early nutrient restriction. The exercised group as a whole gained significantly less weight ($P < 0.05$) than the sedentary group. Crews et al. (74) showed that exercised rats gained less weight than the sedentary controls. There was no significant interaction between obesity and exercise, indicating exercise did not affect weight gain of 8/litter mice differently than 4/litter mice.

Final weights at sacrifice are listed in table 8. Data show that mice from the 4/litter group remained significantly heavier ($P < 0.05$) than those of the 8/litter group after the 6 week study period regardless of exercise treatments. The exercise group as a whole was significantly lighter ($P < 0.05$) than the sedentary group. This is supported by Walberg et al. (90), who reported that exercise resulted in significantly lighter body weights in both lean and obese rats compared with their appropriate controls. No significant interaction was seen between exercise and litter size.

Exercise and Body Composition

The effect of the 6-week exercise program on percent body fat and epididymal fat pad (EFP) weight of the early normal-fed and overfed mice is shown in table 9 and figure 3. The 4/litter group had a significantly higher ($P < 0.05$) percent body fat than the 8/litter group at sacrifice. We found no significant differences in percent fat between the exercise and sedentary groups. Bjorntorp and coworkers (77) found similar results showing that severely obese subjects decrease their body fat abnormally slowly or not at all when subjected to physical training on an ad libitum

TABLE 9

Effect of exercise on percent fat and epididymal fat pad weight (g)
of early normal-fed and overfed mice¹

Measurement	8/litter		4/litter		Source of variation	df	Mean square and significance ²	
	-Exer.	+Exer.	-Exer.	+Exer.			Fat	Epididymal fat pad
Fat	20.4±1.7	16.5±1.6	21.5±1.7	22.1±1.9	OBNOB ³	1	99.7*	0.5*
Epididymal fat pad	0.7±0.1	0.5±0.1	1.0±0.1	0.7±0.1	MOM(OBNOB) ⁴	16	19.6	0.1
					EXNEX ⁵	1	23.2 NS	0.7*
					EXNEX*OBNOB ⁶	1	46.9 NS	0.4 NS
					Residual	16	33.6	0.0

TABLE 10

Effect of exercise on percent lean body mass and protein of
early normal-fed and overfed mice¹

Measurement	8/litter		4/litter		Source of variation	df	Mean square and significance ²	
	-Exer.	+Exer.	-Exer.	+Exer.			Lean body mass	Protein
Lean body mass	79.7±1.7	83.5±1.6	78.5±1.7	77.9±1.9	OBNOB ³	1	101.7*	0.1 NS
Protein	14.3±0.5	14.3±0.6	14.0±0.6	14.4±0.5	MOM(OBNOB) ⁴	16	19.7	5.1
					EXNEX ⁵	1	22.2 NS	0.4 NS
					EXNEX*OBNOB ⁶	1	45.6 NS	0.5 NS
					Residual	16	33.4	0.5

TABLE 11

Effect of exercise on percent dry matter and body water
of normal-fed and overfed mice¹

Measurement	8/litter		4/litter		Source of variation	df	Mean square and significance ²	
	-Exer.	+Exer.	-Exer.	+Exer.			Dry matter	Body water
Dry matter	38.7±1.0	35.1±1.2	39.7±1.7	40.3±1.8	OBNOB ³	1	86.2*	86.2*
Body water	61.3±1.0	64.9±1.2	60.3±1.7	59.7±1.8	MOM(OBNOB) ⁴	16	10.1	10.1
					EXNEX ⁵	1	20.7 NS	20.7 NS
					EXNEX*OBNOB ⁶	1	39.5 NS	39.5 NS
					Residual	16	27.9	27.9

¹ Mean ± SEM for 9 animals per group.
individual mothers.

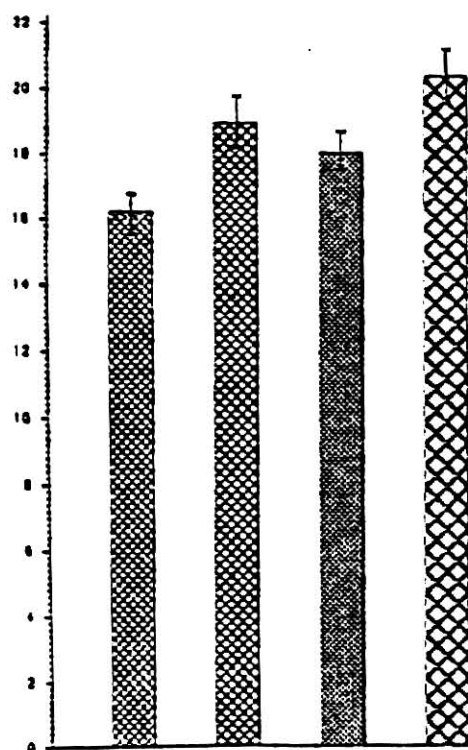
² *P < 0.05, NS P > 0.05.

³ Litter size.

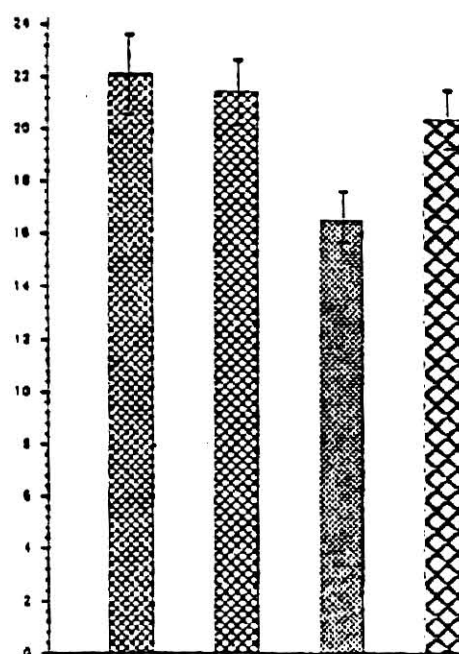
⁴ Effect of

⁵ Exercise. ⁶ Interaction between litter size and exercise.

WEIGHT GAIN (GMS)



BODY FAT (%)



EPIDIDYMAL FAT PAD WEIGHTS (GMS)

LEGEND: GROUP

- 4/L-EXER
- 4/L-NOEXER
- 8/L-EXER
- 8/L-NOEXER

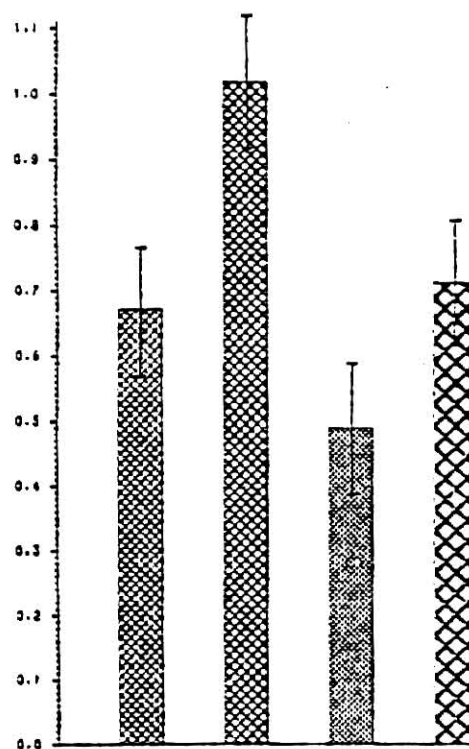


Fig. 3 Effect of exercise on weight gain, body fat, and epididymal fat pad weight of early normal-fed and overfed mice.

diet. In contrast to our results, Walberg et al. (90) showed that both lean and obese exercised rats had lower percent body fat than the sedentary freely-eating control animals.

The 4/litter group had significantly heavier ($P < 0.05$) EFP weights than the 8/litter group. The exercised group as a whole had significantly lighter ($P < 0.05$) EFP weights than the sedentary group but there was no interaction between litter size and exercise. Other investigators (91, 92) also reported a reduction in EFP weights in response to exercise. Therefore, it seems that exercise has an effect on EFP weights but not on total body fat. These findings are similar to those reported by Gelhorn and Benjamin (93). They found that the weight of the EFP was not a reliable index of percentage of fat in the carcass, as fat is deposited in the EFP at a different rate from the deposition of fat in the carcass as a whole. This suggests that total carcass fat is not indicative of the fat content of individual fat pads.

The effect of exercise on lean body mass and protein at sacrifice is shown in table 10. The 8/litter mice had significantly higher ($P < 0.05$) percent lean body mass than the 4/litter mice. However, we observed no significant difference in percent lean body mass (100 minus % body fat) between the exercise and sedentary groups. In contrast with our results, Crews et al. (74) found that exercise in rats resulted in an increase in lean body mass. Percent protein was not significantly affected by either litter size or exercise. Walberg et al. (90) reported that exercise appeared to have a protein-sparing effect in the obese Zucker rat. The protein sparing effect of exercise is possibly the reason for no significant difference in percent lean body mass between the exercise and sedentary groups.

The effect of exercise on dry matter and body water of the normal-fed and overfed mice are shown in table 11. Mice from the 8/litter group had significantly less ($P < 0.05$) dry matter and significantly more ($P < 0.05$) body water than those from the 4/litter group. Exercise did not affect these measurements. The higher % body water of 8/litter mice was expected because these mice also had significantly lower percent body fat which is inversely proportional to body water (16). Exercise did not have a significant effect on percent body water.

SUMMARY

The effects of exercise were evaluated in mice which had previously received either normal- or over-nutrition during the nursing period. Early nutriture was accomplished by a manipulation of litter size where normal-fed mice were reared in litters of 8, and overfed mice in litters of 4. At weaning the normal-fed mice weighed an average 11.3 g and overfed mice, 16.3 g. Litters were then subdivided into exercise and non-exercise groups ($n = 9$) and placed on a 2-week training program followed by 4 weeks of target exercise at 0.5 mph on a zero-grade treadmill.

During the exercise period the normal-fed mice gained more weight ($P < 0.05$) and consumed more feed/body weight ($P < 0.05$) than the overfed mice; but they still remained lighter than the overfed mice ($P < 0.05$). The normal-fed mice also had lower percent body fat ($P < 0.05$) and lighter epididymal fat pads ($P < 0.05$) than the overfed mice.

Exercised mice within each normal-fed or overfed group gained less weight ($P < 0.05$) and had lighter epididymal fat pads ($P < 0.05$) than their sedentary counterparts, but they did not have significantly different percentages of body fat.

Throughout the study there were no significant interactions between early nutriture and exercise suggesting the normal-fed and overfed mice respond similarly to the effects of exercise.

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LITERATURE CITED

1. Powers, P. (1980) Obesity: The Regulation of Weight, pp. 29-31, Williams & Wilkins Co., Baltimore, MD.
2. Bray, G.A. (1974) Metabolic effects of corpulence. In: Recent Advances in Obesity Research (Howard, A., ed.), pp. 56-65, Newman Publishing LTD.
3. Kalkhoff, R.K., Kim, H.J., Cerletty, J. & Ferrou, C.A. (1971) Metabolic effects of weight loss in obese subjects. *Diabetes* 20, 83-91.
4. Sims, A.H. & Horton, E.S. (1968) Endocrine and metabolic adaption to obesity and starvation. *Am. J. Clin. Nutr.* 21, 1455-1470.
5. Goldberg, M. & Gordon, E.S. (1964) Energy metabolism in human obesity. *JAMA* 189, 616-623.
6. Knittle, J.L. & Ginsberg-Fellner, F. (1972) Effect of weight reduction on in vitro adipose tissue lipolysis and cellularity in obese adolescents and adults. *Diabetes* 21, 754-761.
7. Bray, G.A. (1976) The Obese Patient, pp. 252-299, W.B. Saunders Company, Philadelphia.
8. West, E.S. & Todd, W.R. (1951) Textbook of Biochemistry, p. 974, Macmillan, New York.
9. Wertheimer, E. & Ben-Tor, V. (1952) Fat utilization in muscle. *Biochem. J.* 50, 573-576.
10. Bray, G.A. & York, D.A. (1971) Genetically transmitted obesity in rodents. *Physiol. Rev.* 51, 598-646.
11. Amatruda, J.M., Livingston, J.N. & Lockwood, D.H. (1975) Insulin receptor: role in the resistance of human obesity to insulin. *Science* 188, 264-266.
12. Archer, J.A., Gorden, P. & Roth, J. (1975) Defect in insulin binding to receptors in obese man. *J. Clin. Invest.* 55, 166-174.
13. Salans, L.B., Knittle, J.L. & Hirsch, J. (1968) The role of adipose cell size and adipose tissue insulin sensitivity in the carbohydrate intolerance of human obesity. *J. Clin. Invest.* 47, 153-165.
14. Jungas, R.L. (1966) Role of cycle-3',5'-AMP in the response of adipose tissue to insulin. *Proc. Nat. Acad. Sci.* 56, 757-763.

15. Aubert, R., Suquet, J. & Lemmoneir, D. (1980) Long-term morphological and metabolic effects of early under- and over-nutrition in mice. *J. Nutr.* 110, 649-661.
16. Pike, R.L. & Brown, M.L. (1975) *Nutrition: An Integrated Approach*, pp. 714-716, Wiley & Sons, Inc., New York.
17. Cuatrecasas, P. (1973a) Insulin receptor of liver and fat cell membranes. *Fed. Proc.* 32, 1838-1846.
18. Butcher, R.W., Baird, C.E. & Sutherland, E.W. (1968) Effects of lipolytic and antilipolytic substances on adenosine 3',5'-monophosphate levels in isolated fat cells. *J. Biol. Chem.* 243, 1705-1712.
19. Illiano, G. & Cuatrecasas, P. (1972) Modulation of adenylate cyclase in liver and fat cell membranes by insulin. *Science* 175, 906-908.
20. Galton, D.J. (1971) *The Human Adipose Cell: A Model for Errors in Metabolic Regulation*. Appleton-Century-Crofts, New York.
21. Davidson, S., Passmore, R., Brock, J.F. & Truswell, A.S. (1975) *Human Nutrition and Dietetics*, pp. 288-301, Churchill Livingstone, Edinburgh, London and New York.
22. Gordon, E.S. & Goldberg, M. (1964) Studies of energy metabolism in human subjects using carbon 14-labeled compounds. I. Effect of sex, state of nutrition and body weight. *Metabolism* 13, 775-790.
23. Beck, P., Koumans, J.H.T. & Winterling, C.A. (1964) Studies of insulin and growth hormone secretion in human obesity. *J. Lab. Clin. Med.* 64, 654-667.
24. Copinschi, G., Wegienka, L.C. & Hane, S. (1967) Preliminary report: effect of arginine on serum levels of insulin and growth hormone in obese subjects. *Metabolism* 16, 385-491.
25. Vander Laan, W.P., Parker, D.C. & Rossman, L.G. (1970) Implication of growth hormone release in sleep. *Metabolism* 19, 891-897.
26. Qualbe, H.J., Helge, H. & Kubicki, S. (1971) Nocturnal growth hormone secretion: correlation with sleeping EEG in adult and pattern in children and adolescents with non-pituitary dwarfism, overgrowth and with obesity. *Acta Endocrinol.* 67, 767-783.
27. Schwarz, F., ter Haar, D.J., van Riet, H.G. & Thijssen, J.H.H. (1969) Response of growth hormone (GH), FFA, blood sugar and insulin to exercise in obese patients and normal subjects. *Metabolism* 18, 1013-1020.
28. Bortz, W.M. (1969) Metabolic consequences of obesity. *Ann. Intern. Med.* 71, 833-843.

29. Dole, V.P. (1956) A relationship between non-esterified fatty acids in plasma and the metabolism of glucose. *J. Clin. Invest.* 35, 150-154.
30. Issekutz, B., Jr., Paul, P., Miller, H.J. & Bortz, W.M. (1968) Oxidation of plasma FFA in lean and obese humans. *Metabolism* 17, 62-73.
31. Opie, L.H. & Walfish, P.G. (1963) Plasma free fatty acid concentrations in obesity. *N. Engl. J. Med.* 268, 757-760.
32. Corvilain, J., Loeb, H., Champenois, A. & Abramow, M. (1961) Effect of fasting on levels of plasma-nonesterified fatty acids in normal children, normal adults, and obese adults. *Lancet* 1, 534.
33. Stuchlikova, E. (1961) Some changes in intermediary metabolism of obese patients. *Clin. Chem. Acta* 6, 571-577.
34. York, D.A. & Bray, G.A. (1973) Genetic obesity in rats. II. The effect of food restriction on the metabolism of adipose tissue. *Metabolism* 22, 443.
35. Smith, U. (1970) Effects of glucose and insulin on lipolysis rates in human fat cells of different sizes. *F.E.B.S. Letter* 11, 8-10.
36. Kahlenberg, A. & Kalant, N. (1964) The effect of insulin on human adipose tissue. *Canad. J. Biochem.* 42, 1632-1625.
37. Bjorntorp, P., Berchtold, P., Holm, J. & Larsson, B. (1971) The glucose uptake of human adipose tissue in obesity. *Europ. J. Clin. Invest.* 1, 480-485.
38. Kahn, C.R., Soll, A.H., Neville, D.M., Goldfine, J.D., Archer, J.A., Gorden, P. & Roth, J. (1975) The insulin receptor in obesity and other states of altered insulin sensitivity. In: *Obesity in Perspective, Part 2* (Bray, G.A., ed.), DHEW Publication No. (NIH) 301-311.
39. Palmer, W.K. & Tipton, C.M. (1974) Effect of training on adipocyte glucose metabolism and insulin responsiveness. *Fed. Proc.* 33, 1964-1968.
40. Bjorntorp, P. (1973) Effects of physical conditioning in obesity. In: *Obesity in Perspective, Part 2* (Bray, G.A., ed.), DHEW Publication No. (NIH) 116.
41. Van Itallie, T.B. & Campbell, R.G. (1972) Multi-disciplinary approach to the problem of obesity. *J. Am. Diet. Assoc.* 61, 385.
42. Bennion, M. (1979) *Clinical Nutrition*, pp. 92-115, Harper & Row, New York.

43. Bjorntorp, P. (1974) Effects of age, sex, and clinical conditions on adipose tissue cellularity in man. *Metabolism* 23, 1091.
44. Adebajo, F.O. (1975) Studies on human adipose cells in culture: relation of cell size and cell multiplication to donor age. *Yale J. Biol. Med.* 48, 9-16.
45. Hirsch, J. (1972) Can we modify the number of adipose cells? *Postgrad. Med.* 51, 83.
46. Hirsch, J. & Han, P. (1969) Cellularity of rat adipose tissue: effects of growth, starvation on obesity. *J. Lipid Res.* 10, 77.
47. Hirsch, J. & Gallian, E. (1968) Methods for determination of adipose cell size in man and animals. *J. Lipid Res.* 9, 110-119.
48. Hirsch, J. & Knittle, J.L. (1970) Cellularity of obese and non-obese human adipose tissue. *Federation Proc.* 29, 1516-1521.
49. Knittle, J.L. (1972a) Obesity in childhood: a problem in adipose tissue cellular development. *J. Pediatr.* 81, 1048-1059.
50. Bjorntorp, P. (1959) Atherosclerosis and body build with special reference to size and number of subcutaneous fat cells. *Acta Med. Scand. Suppl.* 349, 166-199.
51. Sims, E.A., Goldman, R.F., Gluck, C.M., Horton, E.S., Kelleher, P.C. & Rowe, D.W. (1968) Experimental obesity in man. *Trans. Assoc. Am. Physicians* 81, 153-170.
52. Salans, L.B., Horton, E.S. & Sims, E.A. (1971) Experimental obesity in man: cellular character of the adipose tissue. *J. Clin. Invest.* 50, 1005-1011.
53. Brook, C.G.D., Lloyd, J.K. & Wolf, O.H. (1972) Relationship between age of onset of obesity and size and number of adipose cells. *Brit. Med. J.* 2, 25-27.
54. Brook, C.G.D. (1973) Cell growth in man. *Am. Heart J.* 86, 571.
55. Knittle, J.L. (1972) Obesity in childhood: a problem in adipose tissue cellular development. *J. Pediatr.* 81, 1048.
56. Eid, E.E. (1970) Follow-up study of physical growth of children who had excessive weight gain in first six months of life. *Br. Med. J.* 2, 74-76.
57. Huenemann, R.L. (1974) Environmental factors associated with pre-school obesity. II. Obesity and food practices of children at successive age levels. *J. Am. Diet. Assoc.* 64, 488-491.

58. Gurr, M.I., Jung, R.T., Robinson, M.P. & James, W.P.T. (1982) Adipose tissue cellularity in man: the relationship between fat cell size and number, the mass and distribution of body fat and the history of weight gain and loss. *Intern. J. of Ob.* 6, 419-436.
59. Jung, R.T., Gurr, M.I., Robinson, M.P. & James, W.P.T. (1978) Does adipocyte hypercellularity in obesity exist? *Br. Med. J.* 2, 319-321.
60. Johnson, M.L., Burke, B.S. & Mayer, J. (1956) Relative importance of inactivity and overeating in the energy balance of obese high school girls. *Am. J. Clin. Nutr.* 4, 37.
61. Withers, R.F.J. (1964) Problems in the genetics of human obesity. *Eugenics Reviews* 56, 81.
62. Newman, H.H., Freeman, F.N. & Holzinger, J.J. (1937) *Twins, a Study of Heredity and Environment*. University of Chicago Press, Chicago.
63. Johnson, P.R., Stern, J.S., Greenwood, M.R.C., Zucker, L.M. & Hirsch, J. (1973) Effect of early nutrition on adipose cellularity and pancreatic insulin release in the Zucker rat. *J. Nutr.* 103, 738-743.
64. Knittle, J.L. & Hirsch, J. (1968) Effect of early nutrition on the development of rat epididymal fat pads: cellularity and metabolism. *J. Clin. Invest.* 47, 2091-2098.
65. Lemonnier, D. (1972) Effect of age, sex, and site on the cellularity of the adipose tissue in mice and rats rendered obese by a high-fat diet. *J. Clin. Invest.* 51, 2907-2915.
66. Faust, I.M., Johnson, P.R., Stern, J.S. & Hirsch, J. (1978) Diet-induced adipocyte number increase in adult rats: a new model of obesity. *Am. J. Physiol.* 235, E279-E286.
67. Mandenoff, A., Lenoir, T. & Affelbaum, M. (1982) Tardy occurrence of adipocyte hyperplasia in cafeteria-fed rat. *Am. J. Physiol.* 242, R349-R351.
68. Peckman, S.C., Entenman, C. & Carroll, H.W. (1962) The influence of a hypercaloric diet on gross body and adipose tissue composition in the rat. *J. Nutr.* 77, 187-197.
69. Katch, V.L., Martin, R. & Martin, J. (1979) Effects of exercise intensity on food consumption in the male rat. *Am. J. Clin. Nutr.* 32, 1401-1407.
70. Mayer, J., Marshall, N.B., Vitale, J.J., Christensen, J.H., Mashayeki, M.B. & Stare, F.J. (1954) Exercise, food intake and body weight in normal rats and genetically obese adult mice. *Am. J. Physiol.* 177, 544-548.

71. Woo, R., Garrow, J.S. & Pi-Sunyer, F.X. (1982) Effect of exercise on spontaneous caloric intake in obesity. *Am. J. Clin. Nutr.* 36, 470-477.
72. Woo, R., Garrow, J.S. & Pi-Sunyer, F.X. (1982) Voluntary food intake during prolonged exercise in obese women. *Am. J. Clin. Nutr.* 36, 478-484.
73. Rolls, B.J. & Rowe, E.A. (1977) Dietary obesity: permanent changes in body weight. *J. Physiol.* 272, 2P.
74. Crews, E.L., III, Fuge, K.W., Oscai, L.B., Holloszy, J.O. & Shank, R.E. (1969) Weight, food intake, and body composition: effects of exercise and of protein deficiency. *Am. J. Physiol.* 216, 359-363.
75. Oscai, L.B. & Holloszy, J.O. (1969) Effects of weight changes produced by exercise, food restriction, or overeating on body composition. *J. Clin. Invest.* 48, 2124-2128.
76. Rodahl, K., Miller, H.I. & Issekutz, B., Jr. (1964) Plasma free fatty acids in exercise. *J. Appl. Physiol.* 19, 489.
77. Bjorntorp, P., de Jonge, K., Krotkiewski, M., Sullivan, L., Sjostrom, L. & Stenber, J. (1973) Physical training in human obesity. III. Effects of long-term physical training on body composition. *Metabolism* 22, 1467-1475.
78. Bjorntorp, P., Gumby, G., Sanne, L., Sjostrom, L., Tibblin, G. & Wilhelmsen, L. (1972) Adipose tissue fat cell size in relation to metabolism in weight-stable, physically active men. *Horm. Metab. Res.* 4, 182.
79. Bjorntorp, P., Holm, G., Jacobsson, B., Schiller-de Jonge, K., Lundberg, P., Sjostrom, L., Smith, U. & Sullivan, L. (1977) Physical training in human hyperplastic obesity. IV. Effects on the hormonal status. *Metabolism* 26, 319-328.
80. Bjorntorp, P., de Jonge, K., Sjostrom, L. & Sullivan, L. (1970) The effect of physical training on insulin production in obesity. *Metabolism* 19, 631-638.
81. Deb, S. & Martin, R.J. (1975) Effects of exercise and of food restriction on the development of spontaneous obesity in rats. *J. Nutr.* 105, 543-549.
82. Richard, D., Labrie, A., Lupien, D., Tremblay, A. & Le Blanc, J. (1982) Interactions between dietary obesity and exercise-training on carbohydrate metabolism. *Intern. J. Obes.* 6, 359-367.
83. Larsson, S. & Jonsson, S. (1967) The effect of muscular activity on fat mobilization and body fat content in normal and obese mice. *Acta Physiol. Scand. Suppl.* 294, 49-58.

84. Epstein, H.T. (1978) The effect of litter size on weight gain in mice. *J. Nutr.* 108, 120-123.
85. Haack, R.R. (1983) Effect of exercise duration on food intake, body weight, and body composition of mice. M.S. Thesis, Kansas State University.
86. Plummer, D.T. (1971) An Introduction to Practical Biochemistry: The Folin-Lowry Method of Protein Assay, p. 156, McGraw-Hill Book Co. Limited, England.
87. Folch, J., Lees, M. & Sloane Stanley, G.H. (1957) A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.* 226, 497-509.
88. Snedecor, G.W. & Cochran, W.G. (1980) Statistical Methods, Iowa State University Press, Ames.
89. Helwig, J.T. & Council, W.G. (eds.) (1979) SAS User's Guide, SAS Institute, Inc., Cary.
90. Walberg, J.L., Mole, P.A. & Stern, J.S. (1982) Effect of swim training on development of obesity in the genetically obese rat. *Am. J. Physiol.* 242, R204-R211.
91. Askew, E.W. & Hecker, A.L. (1976) Adipose tissue cell size and lipolysis in the rat: response to exercise intensity and food restriction. *J. Nutr.* 106, 1351-1360.
92. Oscai, L.B., Spirakis, C.N., Wolff, C.A. & Beck, R.J. (1972) Effects of exercise and of food restriction on adipose tissue cellularity. *J. Lipid Res.* 13, 588-592.
93. Gellhorn, A. & Benjamin W. (1965) Effect of aging on the composition and metabolism of adipose tissue in the rat. In: Handbook of Physiology (Renold, A.D. & Cahill, G.F., eds.), p. 399, American Physiological Society, Washington DC.

APPENDIX

APPENDIX TABLE 1

Reagents for analysis of protein and lipid

Protein

1. Alkaline sodium carbonate solution (2% Na_2CO_3 in 0.1 N NaOH). Dissolve 20 g Na_2CO_3 and 4 g NaOH in a liter of deionized water. This was used in place of the "alkaline solution" for sample blanks.
2. Copper sulfate-sodium-potassium tartrate solution (0.5% CuSO_4 in 1% Na, K tartrate). Dissolve 0.25 g CuSO_4 in 50 ml 1% Na, K tartrate. Prepare fresh daily.
3. "Alkaline solution." Prepare fresh daily by mixing 50 ml of (1) and 1 ml of (2).
4. Folin-Ciocalteu reagent. Dilute commercial reagent (Fisher Scientific, St. Louis, MO) with an equal volume of deionized water on the day of use.
5. Standard protein solution (0.2 mg/ml). Dissolve 50 mg bovine serum albumin (Sigma Chemical Co., St. Louis, MO) in 250 ml deionized water. Refrigerate.

Lipid

1. Chloroform-methanol (2:1, v/v). Mix 667 ml chloroform with 333 ml methanol. Refrigerate and keep cold during analysis by placing automatic pipettor in a container of crushed ice.
 2. 0.73% NaCl. Dissolve 0.73 g NaCl in 100 ml water. Refrigerate and keep cold during analysis as for chloroform.
-

APPENDIX TABLE 2

Sample SAS¹ computer program for ANOVA procedure

```
// EXEC SAS
// SYSIN DD *
DATA:
INPUT;
CARDS;
PROC ANOVA;
CLASSES MOM EXNEX OBNOB;
MODEL (VARIABLES);
TEST H=OBNOB E=MOM(OBNOB);
MEANS EXNEX EXNEX*OBNOB/LSD;
MEANS OBNOB/LSD E=MOM(OBNOB);
/*
```

¹ Statistical Analysis System.

APPENDIX TABLE 3

Effect of litter size on body weight of mice at weaning

Measurement	8/litter	4/litter
Whole body weight, g	9.7	16.4
	11.5	17.1
	11.9	16.5
	11.9	17.7
	9.6	13.6
	12.4	18.0
	9.8	14.2
	<u>13.6</u>	<u>17.1</u>
	11.3 ± 0.5^1	16.3 ± 0.6
Body weight (without fur), g	9.5	16.2
	8.9	17.0
	9.3	16.4
	9.4	17.5
	7.0	13.4
	9.8	17.9
	7.2	14.1
	<u>11.0</u>	<u>16.9</u>
	9.0 ± 0.5	16.2 ± 0.6
Carcass weight (body weight without fur and gastrointestinal tract), g	7.9	13.8
	7.2	14.4
	7.5	13.4
	7.4	15.3
	5.6	11.1
	8.0	14.1
	5.8	11.9
	<u>8.3</u>	<u>14.9</u>
	7.2 ± 0.4	13.6 ± 0.5

¹ Results are expressed as means $\pm$ SEM for 8 mice in each group.

APPENDIX TABLE 4

Effect of litter size on dry matter and body water of mice at weaning

Measurement	8/litter	4/litter
Dry matter, %	29.7	33.6
	40.2	34.2
	41.5	32.1
	39.0	37.8
	43.0	32.7
	37.2	34.8
	40.8	34.4
	40.5	38.6
	39.0 ± 1.4^1	34.8 ± 0.8
Body water, %	70.3	66.4
	59.8	65.8
	58.5	67.9
	61.0	62.2
	57.0	67.3
	62.8	65.2
	59.2	65.6
	59.5	61.4
	61.0 ± 2.2	65.2 ± 0.8

¹Results are expressed as means $\pm$ SEM for 8 mice in each group.

APPENDIX TABLE 5

Effect of litter size on body fat, body protein, and
lean body mass of mice at weaning

Measurement	8/litter	4/litter
Body fat, %	11.3	15.4
	14.8	16.0
	16.0	14.2
	14.2	20.1
	7.1	14.2
	13.0	18.4
	8.3	13.6
	<u>14.8</u>	<u>14.8</u>
	12.4 ± 1.2^1	15.8 ± 0.8
Body protein, %	14.1	13.5
	17.8	13.5
	17.8	14.5
	18.7	14.7
	11.7	15.7
	18.0	12.5
	11.4	14.6
	<u>17.5</u>	<u>16.8</u>
	15.9 ± 1.1	14.5 ± 0.5
Lean body mass, %	88.7	84.6
	85.2	84.0
	84.0	85.8
	85.8	79.9
	92.9	85.8
	87.0	81.6
	91.7	86.4
	<u>85.2</u>	<u>85.2</u>
	87.6 ± 1.2	84.2 ± 0.8

¹ Results are expressed as means $\pm$ SEM for 8 mice in each group.

APPENDIX TABLE 6

Effect of exercise on feed intake of early normal-fed
and overfed mice: WEEK 1 (training)

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Average daily consumption, g	4.6	4.9	4.6	5.9
	3.6	4.8	5.9	5.4
	5.1	5.2	5.6	5.2
	4.8	5.2	5.7	5.1
	4.6	4.4	5.7	5.1
	4.9	4.9	6.4	5.5
	5.0	5.5	6.2	5.2
	4.8	4.4	5.1	5.4
	5.0	4.6	4.7	5.9
	4.7 ± 0.2^1	4.9 ± 0.1	5.5 ± 0.2	5.4 ± 0.1
Average weekday consumption, ² g	4.1	4.6	4.6	5.9
	4.7	4.5	5.4	5.2
	3.6	4.7	5.1	5.0
	4.6	5.1	5.4	4.9
	4.3	4.2	5.3	4.9
	4.5	4.5	4.8	4.8
	4.5	5.1	5.6	5.2
	4.4	4.1	4.6	5.2
	4.7	4.4	4.4	5.7
	4.4 ± 0.1	4.6 ± 0.1	5.0 ± 0.1	5.2 ± 0.1
Average weekend consumption, ³ g	5.8	5.7	4.7	6.1
	3.6	5.6	7.3	6.0
	6.0	6.4	6.8	5.8
	5.4	5.4	6.4	5.8
	5.5	4.8	6.6	5.6
	5.8	5.8	10.4	7.4
	6.2	6.5	7.6	5.4
	5.6	5.0	6.2	5.9
	5.9	5.0	5.4	6.3
	5.5 ± 0.3	5.6 ± 0.2	6.8 ± 0.5	6.0 ± 0.2

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

² Average consumption during exercise days (Monday through Friday).

³ Average consumption during nonexercise days (Saturday and Sunday).

APPENDIX TABLE 7

Effect of exercise on feed intake of early normal-fed
and overfed mice: WEEK 2 (training)

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Average daily consumption, g	6.2	5.4	6.7	6.8
	5.1	6.2	6.7	4.6
	6.4	6.7	6.4	6.7
	6.2	6.0	6.5	6.3
	5.7	5.5	6.7	6.0
	6.6	6.3	6.5	6.5
	6.4	6.5	6.3	5.8
	6.1	6.3	6.7	6.7
	6.5	6.9	6.6	6.3
	6.1 ± 0.2^1	6.2 ± 0.2	6.6 ± 0.1	6.2 ± 0.2
Average weekday consumption, ² g	6.1	5.6	6.1	7.0
	5.2	6.1	7.4	5.4
	7.2	6.8	6.4	6.7
	6.1	6.0	5.8	6.5
	5.7	5.0	6.9	6.8
	6.4	6.2	7.3	6.9
	6.3	6.9	7.0	5.5
	5.8	6.6	7.2	6.6
	6.5	7.2	6.3	7.8
	6.1 ± 0.2	6.3 ± 0.2	6.7 ± 0.2	6.6 ± 0.2
Average weekend consumption, ³ g	6.2	4.8	8.4	6.4
	4.9	6.2	5.0	2.6
	4.4	6.4	6.4	6.7
	6.7	6.2	8.3	5.8
	5.8	6.7	6.2	4.0
	7.2	6.4	4.4	5.4
	6.6	5.3	4.5	6.4
	6.8	5.5	5.5	6.8
	6.4	6.2	7.6	2.6
	6.1 ± 0.3	6.0 ± 0.2	6.2 ± 0.5	5.2 ± 0.6

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

² Average consumption during exercise days (Monday through Friday).

³ Average consumption during nonexercise days (Saturday and Sunday).

APPENDIX TABLE 8

Effect of exercise on feed intake of early normal-fed
and overfed mice: WEEK 3

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Average daily consumption, g	6.5	5.7	6.6	6.2
	5.4	6.5	8.0	6.8
	6.1	7.0	6.5	6.1
	6.1	6.3	6.8	5.7
	5.5	6.0	6.6	7.4
	6.8	5.8	7.4	7.4
	6.8	8.4	8.1	6.9
	6.4	6.4	6.5	7.3
	<u>5.9</u>	<u>6.0</u>	<u>7.9</u>	<u>7.9</u>
	6.1 ± 0.2 ¹	6.4 ± 0.3	7.2 ± 0.2	6.8 ± 0.2
Average weekday consumption, ² g	6.4	5.5	8.2	6.6
	5.6	6.6	8.8	6.5
	6.9	7.4	6.7	6.3
	6.9	6.4	6.4	5.7
	5.5	6.3	8.4	8.2
	6.7	5.8	6.9	7.6
	6.2	9.0	8.2	6.9
	7.7	6.8	6.5	7.4
	<u>6.7</u>	<u>6.2</u>	<u>7.5</u>	<u>8.3</u>
	6.5 ± 0.2	6.7 ± 0.3	7.5 ± 0.3	7.1 ± 0.3
Average weekend consumption, ³ g	5.6	6.2	5.8	5.4
	5.4	6.2	6.2	7.4
	2.0	6.2	6.2	5.5
	5.9	5.9	8.0	5.8
	5.2	5.4	6.8	5.3
	6.9	5.7	7.2	6.8
	6.6	6.8	7.6	7.1
	6.5	5.4	2.2	6.8
	<u>2.8</u>	<u>5.5</u>	<u>7.3</u>	<u>6.9</u>
	5.2 ± 0.6	5.9 ± 0.2	6.4 ± 0.6	6.4 ± 0.3

¹ Results are expressed as means ± SEM for 9 mice in each group.

² Average consumption during exercise days (Monday through Friday).

³ Average consumption during nonexercise days (Saturday and Sunday).

APPENDIX TABLE 9

Effect of exercise on feed intake of early normal-fed
and overfed mice: WEEK 4

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Average daily consumption, g	7.5	6.5	6.5	6.2
	5.4	4.3	7.7	6.9
	6.5	5.8	6.1	6.7
	6.4	5.3	6.6	7.2
	5.8	5.6	6.3	5.6
	6.6	5.7	8.8	7.3
	6.2	7.4	6.6	6.7
	7.0	6.9	7.5	7.6
	<u>5.8</u>	<u>6.6</u>	<u>6.9</u>	<u>7.7</u>
	6.3 ± 0.2^1	6.0 ± 0.3	7.0 ± 0.3	6.9 ± 0.2
Average weekday consumption, ² g	6.6	6.2	6.1	5.6
	5.5	4.0	6.5	6.3
	6.1	5.4	5.8	4.7
	7.0	5.1	6.1	6.3
	5.3	5.6	6.2	7.4
	5.7	5.6	6.5	6.8
	6.3	7.5	6.7	6.3
	6.0	6.6	6.1	7.3
	<u>6.6</u>	<u>5.8</u>	<u>8.7</u>	<u>7.5</u>
	6.1 ± 0.2	5.8 ± 0.3	6.5 ± 0.3	6.5 ± 0.3
Average weekend consumption, ³ g	8.7	7.3	6.6	7.8
	5.7	5.2	10.7	8.6
	7.0	6.8	6.8	11.5
	7.2	5.9	7.8	9.4
	6.4	5.6	7.0	1.0
	6.6	6.0	9.2	8.6
	6.5	7.4	7.6	7.8
	7.9	7.6	11.0	8.4
	<u>6.1</u>	<u>8.8</u>	<u>7.6</u>	<u>8.3</u>
	6.9 ± 0.3	6.7 ± 0.4	8.3 ± 0.5	7.9 ± 0.9

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

² Average consumption during exercise days (Monday through Friday).

³ Average consumption during nonexercise days (Saturday and Sunday).

APPENDIX TABLE 10

Effect of exercise on feed intake of early normal-fed
and overfed mice: WEEK 5

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Average daily consumption, g	7.8	7.4	6.8	7.0
	6.6	6.0	7.4	7.7
	6.5	6.6	6.4	7.3
	7.5	7.0	6.8	6.4
	6.8	7.1	5.1	7.7
	7.2	6.2	6.0	7.8
	6.7	8.3	6.4	7.5
	7.7	7.4	6.9	7.0
	<u>6.7</u>	<u>7.4</u>	<u>4.7</u>	<u>8.2</u>
	7.0 ± 0.2^1	7.0 ± 0.2	6.3 ± 0.3	7.4 ± 0.2
Average weekday consumption, ² g	7.7	7.9	6.5	5.8
	6.4	6.0	7.3	7.9
	6.4	6.5	6.0	6.9
	7.7	6.6	6.5	6.0
	6.2	6.8	6.0	8.3
	6.3	6.1	6.5	7.5
	6.1	9.4	3.4	7.6
	7.0	6.9	2.8	6.8
	<u>6.0</u>	<u>7.4</u>	<u>4.6</u>	<u>7.9</u>
	6.6 ± 0.2	7.1 ± 0.4	5.5 ± 0.5	7.2 ± 0.3
Average weekend consumption, ³ g	8.3	6.0	7.6	10.2
	7.8	6.2	7.4	7.2
	7.4	7.0	7.2	8.4
	8.8	8.2	7.6	7.2
	7.8	7.6	10.8	6.2
	10.0	6.2	9.6	8.7
	7.4	5.6	7.2	7.0
	7.8	8.4	8.0	7.6
	<u>7.8</u>	<u>7.4</u>	<u>8.0</u>	<u>8.8</u>
	8.1 ± 0.3	7.0 ± 0.3	8.2 ± 0.4	7.9 ± 0.2

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

² Average consumption during exercise days (Monday through Friday).

³ Average consumption during nonexercise days (Saturday and Sunday).

APPENDIX TABLE 11

Effect of exercise on feed intake of early normal-fed
and overfed mice: WEEK 6

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Average weekday consumption, ² g	7.9	7.6	9.7	6.2
	9.5	8.7	8.1	7.3
	8.9	9.0	8.0	8.3
	8.3	7.9	8.6	9.0
	8.0	7.8	10.4	6.9
	8.5	8.1	6.7	7.6
	7.9	5.5	9.3	9.0
	7.9	7.2	8.1	8.5
	<u>8.7</u>	<u>8.1</u>	<u>9.3</u>	<u>7.9</u>
	8.4 ± 0.2 ¹	7.8 ± 0.3	8.7 ± 0.4	7.9 ± 0.3

¹ Results are expressed as means ± SEM for 9 mice in each group.

² Average consumption during exercise days (Monday through Friday).

APPENDIX TABLE 12

Effect of exercise on body weight of early normal-fed
and overfed mice: WEEK 1 (training)

Day measured	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Monday body wt, g	11.5	11.4	17.0	17.5
	11.6	11.8	17.7	16.5
	12.9	12.3	17.7	17.4
	13.1	13.0	15.1	14.4
	11.1	9.6	18.5	19.4
	12.8	13.2	17.0	17.5
	13.9	14.2	15.2	15.0
	11.1	11.4	18.0	17.1
	<u>12.0</u>	<u>11.9</u>	<u>18.0</u>	<u>18.2</u>
	12.2 ± 0.3 ¹	12.1 ± 0.4	17.1 ± 0.4	17.0 ± 0.5
Friday body wt, g	15.8	13.1	22.1	21.7
	2.6	15.6	22.1	21.1
	16.8	16.7	21.0	20.3
	15.3	16.3	18.7	18.5
	14.8	12.9	21.6	21.1
	15.9	17.3	20.5	20.6
	16.5	14.5	21.5	20.4
	12.9	13.6	21.2	18.7
	<u>15.8</u>	<u>15.0</u>	<u>18.2</u>	<u>21.6</u>
	14.0 ± 1.5	15.0 ± 0.5	20.8 ± 0.5	20.4 ± 0.4

¹ Results are expressed as means ± SEM for 9 mice in each group.

APPENDIX TABLE 13

Effect of exercise on body weight of early normal-fed
and overfed mice: WEEK 2 (training)

Day measured	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Monday body wt, g	19.0	16.5	22.5	23.2
	14.6	18.7	25.0	23.8
	20.3	20.2	24.1	22.8
	17.7	17.2	22.4	21.0
	17.9	15.7	25.2	20.4
	20.4	20.7	23.7	24.1
	20.0	18.3	25.0	21.6
	16.7	20.8	25.1	21.7
	<u>17.0</u>	<u>24.2</u>	<u>20.6</u>	<u>24.7</u>
	18.2 ± 0.6 ¹	19.1 ± 0.9	23.7 ± 0.5	22.6 ± 0.5
Friday body wt, g	24.0	21.4	23.9	27.1
	18.9	23.7	30.3	23.8
	24.9	25.4	27.5	26.1
	23.1	22.1	25.5	24.5
	22.2	20.2	25.0	26.7
	20.9	24.2	29.3	29.0
	24.7	22.5	29.7	23.8
	21.9	21.2	28.6	26.2
	<u>24.2</u>	<u>26.7</u>	<u>24.2</u>	<u>29.0</u>
	22.8 ± 0.7	23.0 ± 0.7	27.1 ± 0.8	26.2 ± 0.7

¹ Results are expressed as means ± SEM for 9 mice in each group.

APPENDIX TABLE 14

Effect of exercise on body weight of early normal-fed
and overfed mice: WEEK 3

Day measured	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Monday body wt, g	28.2	21.6	25.5	27.3
	20.8	25.1	26.0	21.8
	23.5	26.6	28.9	27.4
	24.3	24.6	26.6	25.9
	23.0	22.1	25.5	24.5
	22.5	27.1	29.8	28.2
	26.0	24.0	26.6	25.1
	25.6	21.2	27.5	21.8
	<u>26.7</u>	<u>26.7</u>	<u>24.4</u>	<u>28.0</u>
	24.5 ± 0.8 ¹	24.3 ± 0.8	26.8 ± 0.6	25.6 ± 0.8
Friday body wt, g	29.7	23.6	28.5	29.8
	23.3	27.0	32.8	27.2
	28.8	28.8	30.0	29.5
	27.0	25.4	27.6	26.7
	26.8	24.9	28.7	28.2
	24.5	27.8	29.8	31.8
	29.3	26.8	33.6	27.0
	27.5	26.4	32.3	29.0
	<u>29.2</u>	<u>27.2</u>	<u>31.3</u>	<u>33.3</u>
	27.3 ± 0.7	26.4 ± 0.5	30.6 ± 0.7	29.2 ± 0.7

¹ Results are expressed as means ± SEM for 9 mice in each group.

APPENDIX TABLE 15

Effect of exercise on body weight of early normal-fed
and overfed mice: WEEK 4

Day measured	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Monday body wt, g	29.4	25.1	29.7	29.7
	24.8	28.3	32.2	29.8
	28.9	28.4	31.2	29.7
	27.5	25.4	27.7	26.7
	27.5	24.8	29.5	28.5
	26.5	27.8	29.1	31.3
	30.2	27.4	33.5	27.8
	27.9	26.5	33.6	28.3
	<u>28.9</u>	<u>27.4</u>	<u>33.2</u>	<u>33.7</u>
	28.0 ± 0.5 ¹	26.8 ± 0.5	31.1 ± 0.7	29.5 ± 0.7
Friday body wt, g	32.4	26.5	31.8	30.7
	26.7	27.2	36.3	31.5
	31.2	29.6	32.2	29.1
	29.3	27.8	29.8	28.6
	29.2	26.8	30.8	30.5
	30.0	29.5	32.3	33.2
	32.3	29.4	35.9	28.9
	29.4	28.1	35.5	31.0
	<u>30.4</u>	<u>27.7</u>	<u>35.0</u>	<u>35.6</u>
	30.1 ± 0.6	28.1 ± 0.4	33.3 ± 0.8	31.0 ± 0.7

¹ Results are expressed as means ± SEM for 9 mice in each group.

APPENDIX TABLE 16

Effect of exercise on body weight of early normal-fed
and overfed mice: WEEK 5

Day measured	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Monday body wt, g	33.8	26.8	33.2	31.8
	24.6	27.2	30.7	33.4
	32.0	30.4	36.7	30.5
	36.5	27.8	33.2	28.6
	28.4	27.1	32.0	32.0
	36.6	30.6	32.1	34.2
	33.3	30.4	36.7	30.3
	30.7	27.9	33.6	30.3
	<u>31.2</u>	<u>29.9</u>	<u>36.4</u>	<u>36.0</u>
	31.9 ± 1.3^1	28.7 ± 0.5	33.8 ± 0.7	31.9 ± 0.8
Friday body wt, g	34.2	29.9	34.5	30.3
	28.5	26.1	31.1	33.6
	32.2	31.6	35.6	31.8
	37.7	28.2	34.5	28.5
	29.8	29.1	32.4	32.8
	34.3	31.0	36.4	34.9
	34.8	31.0	35.6	31.6
	31.7	27.0	34.2	25.2
	<u>32.4</u>	<u>30.8</u>	<u>37.2</u>	<u>36.0</u>
	32.8 ± 0.9	29.4 ± 0.6	34.6 ± 0.6	31.6 ± 1.1

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

APPENDIX TABLE 17

Effect of exercise on body weight of early normal-fed
and overfed mice: WEEK 6

Day measured	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Monday body wt, g	35.3	28.1	35.0	31.6
	29.0	27.2	35.5	34.4
	33.0	31.2	34.3	33.0
	30.0	29.4	31.8	29.9
	31.0	29.1	32.6	33.2
	32.3	31.1	34.5	35.2
	34.4	31.1	35.2	31.4
	32.2	29.5	36.9	34.2
	33.2	32.3	37.1	36.7
	32.3 ± 0.7^1	29.9 ± 0.6	34.8 ± 0.6	33.3 ± 0.7
Friday body wt, g	35.2	28.7	36.2	32.0
	28.6	26.3	37.4	36.0
	33.3	30.8	34.7	33.5
	31.2	29.0	32.7	29.9
	31.9	29.7	32.7	34.2
	33.0	32.3	38.6	35.6
	35.3	32.7	37.3	28.8
	32.3	30.3	37.5	33.3
	32.7	31.5	37.5	35.8
	32.6 ± 0.7	30.1 ± 0.7	36.1 ± 0.7	33.2 ± 0.9

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

APPENDIX TABLE 18

Effect of exercise on weight gain of early normal-fed
and overfed mice

Time period	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Preliminary training period (2 wk)	12.5	10.0	6.9	9.6
	7.3	11.9	19.4	7.3
	12.0	13.1	9.8	8.7
	10.0	9.1	10.1	10.1
	11.1	10.6	6.5	7.3
	8.1	11.0	12.3	11.5
	10.8	8.3	14.5	8.8
	10.8	9.8	10.6	9.1
	<u>12.2</u>	<u>14.8</u>	<u>6.2</u>	<u>10.8</u>
	10.5 ± 0.6 ¹	11.0 ± 0.7	10.7 ± 1.4	9.2 ± 0.5
Total study period (6 wk)	23.7	17.3	19.2	14.5
	17.0	14.5	19.7	19.5
	20.4	18.5	17.0	16.1
	18.1	16.0	17.6	15.5
	20.8	20.1	14.2	14.8
	20.2	19.1	21.6	18.1
	21.4	18.5	22.1	13.8
	21.2	18.9	19.5	16.2
	<u>20.7</u>	<u>19.6</u>	<u>19.5</u>	<u>17.6</u>
	20.4 ± 0.6	18.1 ± 0.6	18.9 ± 0.8	16.2 ± 0.6

¹ Results are expressed as means ± SEM for 9 mice in each group.

APPENDIX TABLE 19

Effect of exercise on feed/body weight ratio of early
normal-fed and overfed mice: WEEK 1 (training)

Time period	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Weekday ²	0.26	0.35	0.21	0.27
	1.38	0.29	0.24	0.25
	0.28	0.28	0.24	0.25
	0.30	0.31	0.29	0.26
	0.29	0.32	0.25	0.23
	0.28	0.26	0.24	0.23
	0.27	0.35	0.26	0.25
	0.34	0.30	0.22	0.28
	0.29	0.29	0.24	0.26
	<u>0.41 ± 0.12¹</u>	<u>0.31 ± 0.01</u>	<u>0.24 ± 0.01</u>	<u>0.25 ± 0.00</u>
Weekend ³	0.51	0.50	0.28	0.35
	0.31	0.47	0.41	0.36
	0.47	0.52	0.39	0.34
	0.41	0.42	0.43	0.40
	0.50	0.51	0.36	0.29
	0.45	0.44	0.61	0.42
	0.44	0.46	0.50	0.36
	0.50	0.44	0.35	0.35
	0.49	0.42	0.30	0.35
	<u>0.45 ± 0.02</u>	<u>0.46 ± 0.01</u>	<u>0.40 ± 0.34</u>	<u>0.36 ± 0.01</u>

¹ Results are expressed as means ± SEM for 9 mice in each group.

² Calculated as average weekday consumption for week 1 divided by Friday body weight of that week.

³ Calculated as average weekend consumption

divided by Monday body weight of the following week.

APPENDIX TABLE 20

Effect of exercise on feed/body weight ratio of early
normal-fed and overfed mice: WEEK 2 (training)

Time period	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Weekday ²	0.26	0.26	0.26	0.26
	0.27	0.26	0.24	0.23
	0.29	0.27	0.23	0.26
	0.26	0.27	0.23	0.26
	0.26	0.25	0.28	0.25
	0.31	0.26	0.25	0.24
	0.26	0.31	0.24	0.23
	0.26	0.31	0.25	0.25
	0.27	0.27	0.26	0.27
	0.27 ± 0.01^1	0.27 ± 0.01	0.25 ± 0.00	0.25 ± 0.00
Weekend ³	0.33	0.29	0.37	0.38
	0.34	0.33	0.20	0.12
	0.22	0.31	0.26	0.29
	0.38	0.36	0.37	0.28
	0.32	0.43	0.24	0.20
	0.35	0.31	0.18	0.22
	0.33	0.29	0.18	0.30
	0.41	0.26	0.22	0.22
	0.38	0.26	0.37	0.11
	0.34 ± 0.02	0.32 ± 0.02	0.27 ± 0.03	0.23 ± 0.03

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

² Calculated as average weekday consumption for week 2 divided by Friday body weight of that week. ³ Calculated as average weekend consumption divided by Monday body weight of the following week.

APPENDIX TABLE 21

Effect of exercise on feed/body weight ratio of early
normal-fed and overfed mice: WEEK 3

Time period	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Weekday ²	0.23	0.23	0.24	0.22
	0.24	0.24	0.27	0.24
	0.27	0.26	0.22	0.21
	0.23	0.25	0.30	0.21
	0.21	0.25	0.23	0.29
	0.27	0.21	0.25	0.24
	0.23	0.34	0.25	0.25
	0.23	0.26	0.25	0.26
	0.23	0.23	0.26	0.25
	0.24 ± 0.01^1	0.21 ± 0.01	0.24 ± 0.01	0.24 ± 0.01
Weekend ³	0.20	0.29	0.23	0.20
	0.26	0.25	0.24	0.34
	0.09	0.23	0.22	0.20
	0.24	0.24	0.23	0.23
	0.23	0.24	0.27	0.22
	0.31	0.21	0.24	0.24
	0.25	0.28	0.28	0.28
	0.25	0.25	0.08	0.31
	0.10	0.21	0.30	0.25
	0.21 ± 0.02	0.24 ± 0.01	0.24 ± 0.02	0.25 ± 0.02

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

² Calculated as average weekday consumption for week 3 divided by Friday body weight of that week.

³ Calculated as average weekend consumption divided by Monday body weight of the following week.

APPENDIX TABLE 22

Effect of exercise on feed/body weight ratio of early
normal-fed and overfed mice: WEEK 4

Time period	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Weekday ²	0.22	0.24	0.20	0.18
	0.20	0.15	0.18	0.20
	0.20	0.18	0.18	0.16
	0.20	0.18	0.20	0.22
	0.19	0.21	0.20	0.24
	0.22	0.19	0.27	0.20
	0.19	0.25	0.17	0.22
	0.22	0.23	0.17	0.24
	<u>0.19</u>	<u>0.21</u>	<u>0.19</u>	<u>0.21</u>
	0.20 ± 0.00 ¹	0.20 ± 0.01	0.20 ± 0.01	0.21 ± 0.01
Weekend ³	0.30	0.29	0.22	0.26
	0.23	0.18	0.33	0.29
	0.24	0.24	0.22	0.39
	0.26	0.23	0.28	0.35
	0.23	0.23	0.24	0.35
	0.25	0.22	0.32	0.27
	0.22	0.27	0.23	0.28
	0.28	0.29	0.33	0.30
	<u>0.21</u>	<u>0.32</u>	<u>0.23</u>	<u>0.25</u>
	0.25 ± 0.01	0.25 ± 0.01	0.27 ± 0.02	0.27 ± 0.03

¹ Results are expressed as means ± SEM for 9 mice in each group.

² Calculated as average weekday consumption for week 4 divided by Friday body weight of that week.

³ Calculated as average weekend consumption divided by Monday body weight of the following week.

APPENDIX TABLE 23

Effect of exercise on feed/body weight ratio of early
normal-fed and overfed mice: WEEK 5

Time period	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Weekday ²	0.22	0.26	0.19	0.19
	0.22	0.23	0.24	0.24
	0.19	0.21	0.17	0.22
	0.19	0.23	0.19	0.21
	0.21	0.23	0.09	0.25
	0.18	0.20	0.13	0.21
	0.18	0.30	0.17	0.24
	0.24	0.26	0.19	0.27
	<u>0.19</u>	<u>0.24</u>	<u>0.09</u>	<u>0.22</u>
	0.20 ± 0.01 ¹	0.24 ± 0.01	0.16 ± 0.20	0.23 ± 0.01
Weekend ³	0.24	0.22	0.23	0.32
	0.32	0.23	0.24	0.22
	0.23	0.23	0.20	0.27
	0.24	0.29	0.23	0.25
	0.27	0.28	0.24	0.19
	0.27	0.20	0.30	0.25
	0.22	0.18	0.20	0.23
	0.26	0.30	0.24	0.25
	<u>0.25</u>	<u>0.25</u>	<u>0.22</u>	<u>0.25</u>
	0.26 ± 0.01	0.24 ± 0.01	0.24 ± 0.02	0.25 ± 0.01

¹ Results are expressed as means ± SEM for 9 mice in each group.

² Calculated as average weekday consumption for week 5 divided by Friday body weight of that week.

³ Calculated as average weekend consumption divided by Monday body weight of the following week.

APPENDIX TABLE 24

Effect of exercise on feed/body weight ratio of
normal-fed and overfed mice: WEEK 6

Time period	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Weekday ²	0.22	0.26	0.27	0.19
	0.23	0.33	0.22	0.20
	0.27	0.29	0.23	0.25
	0.27	0.27	0.26	0.30
	0.25	0.26	0.32	0.20
	0.26	0.25	0.17	0.21
	0.22	0.17	0.25	0.31
	0.25	0.24	0.22	0.26
	<u>0.27</u>	<u>0.26</u>	<u>0.25</u>	<u>0.22</u>
	0.26 ± 0.01 ¹	0.26 ± 0.01	0.24 ± 0.01	0.24 ± 0.01

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

² Calculated as average weekday consumption for week 6 divided by Friday body weight of that week.

APPENDIX TABLE 25

Effect of exercise on final body weights of early
normal-fed and overfed mice

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Whole body weight, g	35.2	28.7	36.2	32.0
	28.6	26.3	37.4	36.6
	33.3	30.8	34.7	33.5
	31.2	29.0	32.7	29.9
	31.9	29.7	32.7	34.2
	32.3	32.3	38.6	35.6
	35.3	32.7	37.3	28.8
	32.3	30.3	37.5	33.3
	<u>32.7</u>	<u>31.5</u>	<u>37.5</u>	<u>35.8</u>
	32.6 ± 0.7 ¹	30.1 ± 0.7	36.1 ± 0.7	33.2 ± 0.9
Body weight (without fur), g	34.4	28.3	35.6	31.6
	28.1	25.8	36.9	35.4
	32.8	30.4	34.1	30.0
	30.8	28.4	32.2	29.4
	31.4	29.2	32.1	33.6
	33.0	31.8	38.2	35.0
	34.9	32.2	36.7	28.4
	31.9	30.0	36.9	32.8
	<u>32.2</u>	<u>31.0</u>	<u>37.1</u>	<u>35.3</u>
	32.1 ± 0.7	29.7 ± 0.7	35.5 ± 0.7	32.4 ± 0.9
Carcass weight (body weight without fur and gastrointestinal tract), g	28.8	24.0	29.5	24.7
	23.3	21.7	30.7	27.6
	27.4	24.6	28.9	28.9
	25.2	23.9	26.1	24.0
	25.4	24.1	27.0	27.7
	32.6	26.0	31.6	29.2
	29.4	26.8	30.5	24.3
	26.9	25.1	31.6	23.9
	<u>26.9</u>	<u>25.7</u>	<u>31.0</u>	<u>29.3</u>
	26.7 ± 0.6	24.7 ± 0.5	29.7 ± 0.7	26.6 ± 0.8

¹ Results are expressed as means ± SEM for 9 mice in each group.

APPENDIX TABLE 26

Effect of exercise on dry matter and body water of
early normal-fed and overfed mice

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Dry matter, %	39.3	32.1	27.8	50.6
	38.7	31.2	38.7	41.9
	40.0	33.3	40.7	34.1
	34.2	39.4	42.1	37.9
	34.7	32.6	41.0	40.8
	36.8	33.0	45.8	37.0
	41.2	40.9	38.2	40.7
	44.0	37.1	41.5	45.9
	<u>39.8</u>	<u>36.6</u>	<u>41.9</u>	<u>34.0</u>
	38.7 ± 1.0^1	35.1 ± 1.2	39.7 ± 1.7	40.3 ± 1.8
Body water, %	60.7	67.9	72.2	49.4
	61.3	68.8	61.3	58.1
	60.0	66.7	59.3	65.9
	65.8	60.6	57.9	62.1
	65.3	67.4	59.0	59.2
	63.2	67.0	54.2	63.0
	58.8	59.1	61.8	59.3
	56.0	62.9	58.5	54.1
	<u>60.2</u>	<u>63.4</u>	<u>58.1</u>	<u>66.0</u>
	61.3 ± 1.0	64.9 ± 1.2	60.3 ± 1.7	59.7 ± 1.8

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

APPENDIX TABLE 27

Effect of exercise on body fat and epididymal fat pad
weights of early normal-fed and overfed mice

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Fat, %	20.7	14.8	11.3	30.8
	18.4	11.3	21.9	26.1
	22.5	12.4	21.9	13.6
	13.0	21.3	23.1	17.2
	13.6	13.0	21.9	22.5
	18.4	13.0	28.4	18.4
	24.3	25.5	21.9	24.9
	27.9	19.0	16.6	27.9
	<u>24.9</u>	<u>18.4</u>	<u>26.1</u>	<u>17.8</u>
	20.4 ± 1.7^1	16.5 ± 1.6	21.5 ± 1.7	22.1 ± 1.9
Epididymal fat pad, g	0.7	0.4	1.2	0.4
	0.6	0.2	1.1	1.0
	0.8	0.4	0.9	0.9
	0.5	0.6	0.8	0.7
	0.5	0.4	0.9	0.9
	0.6	0.4	1.2	0.7
	0.7	0.8	0.5	0.2
	1.1	0.5	1.3	0.8
	<u>0.9</u>	<u>0.6</u>	<u>1.3</u>	<u>0.5</u>
	0.7 ± 0.1	0.5 ± 0.1	1.0 ± 0.1	0.7 ± 0.1

¹ Results are expressed as means $\pm$ SEM for 9 mice in each group.

APPENDIX TABLE 28

Effect of exercise on lean body mass and body protein
content of early normal-fed and overfed mice

Measurement	8/litter		4/litter	
	-Exer.	+Exer.	-Exer.	+Exer.
Lean body mass, %	79.3	85.2	88.7	69.2
	81.6	88.7	78.1	73.9
	71.1	87.6	78.1	86.4
	87.0	78.7	76.9	82.8
	86.4	87.0	78.1	77.5
	81.6	87.0	71.6	81.6
	75.7	74.5	78.1	75.1
	72.1	81.0	83.4	72.1
	<u>75.1</u>	<u>81.6</u>	<u>73.9</u>	<u>82.2</u>
	79.7 ± 1.7 ¹	83.5 ± 1.6	78.5 ± 1.7	77.9 ± 1.9
Protein, %	13.0	12.8	11.9	14.6
	12.5	12.5	12.5	11.9
	13.6	13.3	13.0	13.6
	13.1	13.1	13.6	13.9
	15.3	14.1	13.1	14.4
	16.2	15.3	15.7	15.0
	13.9	13.8	14.4	13.9
	13.8	14.7	14.2	15.3
	<u>17.3</u>	<u>18.8</u>	<u>17.3</u>	<u>17.1</u>
	14.3 ± 0.5	14.3 ± 0.6	14.0 ± 0.6	14.4 ± 0.5

¹ Results are expressed as means ± SEM for 9 mice in each group.

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ABSTRACT

The effects of exercise were evaluated in mice which had previously received either normal- or over-nutrition during the nursing period. Early nutriture was accomplished by a manipulation of litter size where normal-fed mice were reared in litters of 8, and overfed mice in litters of 4. At weaning the normal-fed mice weighed an average 11.3 g and overfed mice, 16.3 g. Litters were then subdivided into exercise and non-exercise groups ($n = 9$) and placed on a 2-week training program followed by 4 weeks of target exercise at 0.5 mph on a zero-grade treadmill.

During the exercise period the normal-fed mice gained more weight ($P < 0.05$) and consumed more feed/body weight ($P < 0.05$) than the overfed mice; but they still remained lighter than the overfed mice ($P < 0.05$). The normal-fed mice also had lower percent body fat ($P < 0.05$) and lighter epididymal fat pads ($P < 0.05$) than the overfed mice.

Exercised mice within each normal-fed or overfed group gained less weight ($P < 0.05$) and had lighter epididymal fat pads ($P < 0.05$) than their sedentary counterparts, but they did not have significantly different percentages of body fat.

Throughout the study there were no significant interactions between early nutriture and exercise suggesting the normal-fed and overfed mice respond similarly to the effects of exercise.