

# THE EFFECT OF GROWTH RATE, SEX AND AGE ON SKELETAL MUSCLE AND ADIPOSE TISSUE GROWTH AND DEVELOPMENT 

## By

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Sixty ewes with the fastest and 60 ewes with the slowest growing lambs from past history were mated to Suffolk and Dorset rams respectively. Three rams and three ewe lambs of each growth rate were slaughtered at each age (birth, 35, 70, 105, 140 and 175 days). The lambs were weaned at 82 days of age and then divided into four groups; fast growing rams and ewes, and slow growing rams and ewes, respectively. Muscle and fat samples were removed at slaughter, weighed and powdered. Perirenal, subcutaneous and intramuscular adipose tissues were assayed for glyceride synthetase, cellularity and chemical composition. Gastrocnemius (GT) muscle was analyzed for nucleic acid and protein fractions. Both GT and longissimus (LD) muscles were analyzed for fat, protein and moisture content.

Rams had more subcutaneous but less perirenal fat than ewes. Except for percentage protein, age affected the
chemical composition of adipose tissues. Perirenal fat of ewes had higher percentages of lipid and lower percentages of moisture than rams. Results of glyceride synthetase activity depend on the method of expressing the activities. In general, both on a protein and cell basis, the enzyme activities increased while on a per gram of fat basis activities decreased with age. Compared to the slow growing group, fast growing lambs had higher enzyme activities on a protein and a cell basis in perirenal fat and on a protein or gram of adipose tissue basis in the subcutaneous depot. Compared to ewes, rams had higher enzyme activities on a gram of adipose tissue or the cell basis in intramuscular and on a gram basis in perirenal fat. Lipid content per cell of adipose tissues increased with age. Ewe lambs had higher lipid per cell than rams.

With advancing age, the number of adipocytes per gram tissue decreased while the total number per fat depot increased. Neither growth rate nor sex affected the number of adipocytes per gram or total in the adipose tissues. Adipocyte diameter and volume of perirenal and subcutaneous fat increased with age. Neither growth rate nor sex affected the cell diameter or volume. Rams and ewes had similar frequency distributions of adipocytes. Growth and development of the adipose tissues is as follows: perirenal>subcutaneous> intramuscular. At 175 days of age hyperplasia was completed in perirenal while both hyperplasia and hypertrophy were
responsible for the increase in subcutaneous fat at that age.
Although muscle DNA and RNA concentrations decreased, the total in the GT muscle increased with age. Neither growth rate nor sex affected nucleic acid concentrations. Compared to the slow growing group, fast growing lambs had greater average daily gains, heavier GT and LD muscles, more total DNA and RNA, more nuclei per GT but a lower protein/ DNA ratio. Rams had higher total DNA and more nuclei in the GT muscle than ewes. Both weight/nucleus and protein/DNA were not affected by sex.

The percentage moisture in the GT and LD muscles decreased while the percentage fat increased with age. Compared to the slow growing group, fast growing lambs had more marbling in the GT and LD muscles, lower protein in the GT and lower concentrations of total nitrogen and nonprotein nitrogen in GT muscle. Rams had lower percentages of protein and total nitrogen concentrations but higher values for each of the total nitrogen fractions and percentage fat compared to ewes.

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## INTRODUCTION

The efficient production of a high proportion of muscle relative to fat is the principal objective of meat animal production. Considerable research effort is and has been expended on the contribution of nutritional and endocrine criteria to body composition and growth. Yet the question remains as to why the dietary nutrients in one animal are shunted toward muscle growth and in another, even among littermates, toward fat growth and development. Limited data (Holmes and Ashmore, 1973; Allen et al., 1974; Bergen et al., 1975) with several animal species tend to suggest that muscle cell number and probably more importantly, age of maximum myofiber hypertrophy determines the stage of rapid adipose tissue development. These studies suggest that rate and extent of muscle development has a significant effect upon the age of onset and the development of adipose tissue. Skeletal muscle and adipose tissue mass at any given stage of growth and development is determined by the extent of hyperplasia (cell number) and hypertrophy (cell size) of the respective tissue cells. These parameters are frequently estimated by DNA (hyperplasia) and protein/RNA (hypertrophy) analyses. Muscle RNA increases prior to the period of myofiber hypertrophy and it decreases during the
period of maximum growth rate. Since the point of maximum growth attainment cannot be ascertained, the exact sampling time for assessing muscle cell hypertrophy cannot be accurately determined. Likewise, DNA of muscle or adipose tissue cannot be selectively determined and the assay procedures used include the DNA from all cells present. Thus, hyperplasia and hypertrophy of muscle and adipose tissue can only be approximated by DNA and RNA analyses. Data are needed to determine the relationship of muscle tissue hyperplasia and hypertrophy to that of adipose tissue development by actual cell counts and size measurements of each tissue. To date adipose tissue and muscle growth and protein synthetase capacity have not been studied simultaneously in the same animals. Thus, the objectives of this study were to determine skeletal muscle and adipose tissue growth and development in the same group of animals. An additional objective was to study the effects of growth rate and sex on skeletal muscle and adipose tissue growth and development from birth to 175 days of age.

## LITERATURE REVIEW

General Aspects of Growth and Development in Meat Animals

The phenomenon of growth is the central focus of the livestock and meat industry from both the standpoint of animal growth and production of all other food materials. According to Fowler (1968), growth has two general aspects. The first is measured as an increase in mass per unit times. The second involves changes in form and composition which results from differential growth of the component parts of the body. The major attempt in the study of growth of the animals is to produce carcasses that have a high quantity of muscle combined with a desirable amount of carcass fat and a minimum of bone. Significant efforts are also being made to produce animals that gain more rapidly. These attempts are paralleled by the trend toward the production of less fat.

The order of tissue growth and development follows an outward trend starting with tissues comprising vital organs and physiological processes (central nervous system) followed by bone, tendon, muscle, intermuscular and subcutaneous fat (Palsson and Verges, 1962). However, in the case of limited nutrient supply, the tissues are affected in
reverse order of physiological importance. The tissues of the body of growing animals which have been retarded in development by restricted environment may exhibit a remarkable compensatory growth when changed to favorable conditions. Growth and development of the central nervous system is essentially completed at birth, therefore, postnatal growth mainly involves increases in bone, muscle and fat. Each tissue in its growth and development follows a sigmoidal curve, but the maximum growth of the various tissues occurs at different ages (Palsson, 1955).

A major portion of bone growth and development is completed in the early stages of postnatal life (McMeekan, 1959). Due to early development of bone and later developing of muscle, the ratio of muscle to bone at birth may be as low as 2:1. The growth pattern shows that bone grows at a steady, but slow rate, while muscle grows relatively fast; therefore, the ratio of muscle to bone increases (Berg and Butterfield, 1976). Weiss et al. (1971) reported that bone decreased from 32 to 15 percent as body weight increased from 1 to 137 in pigs.

With the exception of excessively fat animals, skeletal muscle ranges between 35 to 65 percent of carcass weight of the meat animals (Forrest et al., 1975). On the basis of muscle fiber number Burleigh (1976) suggested a two-phase pattern for muscle growth from embryonic to adult development. In the first phase cells destined to form muscle are
actively replicating for a significant portion of the animals embryonic development, and a second phase in which the amount of muscle protein per cell increases and cell replication is slow or negligible. On the basis of muscle mass, Berg and Butterfield (1976) proposed a four phase pattern for muscle growth. During the first phase (prenatal phase) muscle is mainly under genetic control. In the second phase (immediate postnatally) there is a great change in muscle weight which is completed during the doubling of the birth weight of the muscle mass, but in some muscles this phase continues to a quadrupling of the muscle weight. Phase three (pre-pubertal and adolescent phase) is characterized by uniform growth of muscle in both males and females and it is the product of gene expression and muscle function. During phase four (maturing phase), relative growth of the musculature changes dramatically in the male which results in male animals becoming much more muscular when compared to females. This phase is probably triggered by androgens.

Adipose tissue is the most variable carcass tissue component both in amount and in distribution. During postnatal growth and development, adipose tissue mass increases by either hyperplasia, hypertrophy, or a combination of the two. In the pig, adipocyte hyperplasia appears to be completed in the subcutaneous depot before 5 to 6 months of age (Anderson and Kauffman, 1973; Hood and Allen, 1977). Morphological development of adipose tissue in fetal lamb,

Calves, and pigs are generally similar, but with a different Eime sequence. It has been found that the depot sequence of adipocyte development in red meat animals from early to late Is perirenal, subcutaneous, intermuscular and intramuscular (Lee and Kauffman, 1974a).

Adipose Tissue Cellurlarity

Methodology of Adipocyte Sizing

Techniques for estimating adipocyte size and number has provided valuable information about the fattening proce 5 s and the study of lipid metabolism. Quantitation of DNA comitent of adipose tissue has been used to estimate adipose ce Il number. The limitation of this method is that it overesteimates the number of adipose cells because of the large number of stromal cells which are difficult to separate from adiepocytes and will contribute to tissue DNA level (Stern an a Greenwood, 1974). Although treatment of adipose tissue Wi Ch collagenase has been reported to improve the DNA estimation of adipose cells (Smith et al., 1972; Ashwell et al., $19>6$ ), it has been concluded that collagenase preparation Wi 11 rupture adipose cells, especially large ones (Ashwell et al., 1976). The microscopic technique of measuring fat Cell diameter from conventional thick or thin frozen sections (Ashwell et al., 1975) and stained sections (Ashwell et al., 1976), has been criticized (Sjostrom et al., 1971) because
the fixation procedure may cause shrinkage and some mathemaEical assumptions have to be applied to account for variaDility in shape. In addition, there would be error due to the fact that not all cells have been cut through their equator. Finally, only a small proportion of cells may be counted which might not be the representative of the population of fat cells in the respective depot.

The osmium fixation method of Hirsch and Gallian (1968)
is more objective than other methods. However, it requires an expensive apparatus and the cost of osmium tetroxide is $r e$ Iatively high. In addition, this method fails to measure small cells, generally those less than $25 \mu \mathrm{~m}$ in diameter. Mo elifications to this method have been suggested to improve the isolation of fat cells in osmium (Etherton et al., 1977). They reported that treatment of osmium fixed adipocytes with 8 I Urea and mild heat (50C) solubilized the connective tissue and resulted in a debris-free suspension of fixed adipoCyces. In addition, this modification greatly reduced the time for removing the adipocytes from the connective tissue matcix and appeared to have no effect in the structural in cegrity or size of the fixed cells.

EF Eect of Species and Anatomical Locations

Several investigations have confirmed that adipose $t_{i}$ ssue mass increases in cell number (hyperplasia) and CeII enlargement (hypertrophy) or a combination of both
(Hirsch and Han, 1969; Hubbard and Matthew 1971; Anderson and Kauffman, 1973; Hood and Allen, 1977). Different fat Ceposits of the animal body have different fat cell sizes. Haugebak et al., (1974) observed that the pattern of adipocyte volume in lamb depots were: perirenal > subcutaneous > intermuscular. Hood and Allen (1973) showed that the mean diameter of bovine adipocytes from the intramuscular depot is smaller than those in subcutaneous, intermuscular and perirenal depots. The observations of Moody and Cassens (1 968) with bovine intramuscular fat and those of Lee and Kauffman (1971) with porcine intramuscular fat also showed that the fat cell size of this depot is smaller than subCutaneous fat. Lee and Kauffman (1974a) concluded that the $\mathbf{P F}$ esence of small adipocytes in intramuscular fat indicates that this depot is later developing than other adipose tisswe depots. Moody and Cassens (1968) reported that the increase in marbling score was associated with both hypertrophy and hyperplasia of intramuscular adipocytes. These arthors suggested that once a muscle begins to increase fat COntent, both size and number of fat cells increase. They al so reported that the largest average adipose cell diameter in the intermuscular depot was associated with the largest adipose cell mass within a particular muscle. Anderson and Kauffman (1973) reported that the changes in total carcass Adipose tissue in 1 to 2 month old pigs were due primarily to increase in the number of adipose cells. Between 2 and 5
months, changes in intramuscular fat depot mass were due to a combination of hypertrophy and hyperplasia. After 5 months, Ehere was no increase in adipose cell number, and adipose Eissue mass increased solely by hypertrophy. Similar results were reported by Hood and Allen (1977). Hirsch and Han (1969) concluded that the plateau of adipose cell number in rats was reached at 15 weeks of age. More recently Greenwood and Hirsch (1974) reported that the majority of adiPO cyte hyperplasia is completed by the fifth postnatal week in rats. In a comparison between rats, guinea pigs and hamsters, Di Girolamo and Mendlinger (1971) found that rats ars $d$ hamsters had a considerable capacity to enlarge the fat ce $\mathcal{I l}$ size with increasing age between 6 weeks and 1 year, wh ile guinea pigs showed limited capacity in this respect. I2 the same interval, guinea pigs had a marked increase in the number of fat cells in the epididymal fat pads, while the rat and hamster had a limited increase. These results suggest species differences in that guinea pigs increased $i t s$ epididymal adipose tissue mass mainly by an increase in the number of fat cells with little change in cell size, Wh ile the rat, hamster and pig (Hood and Allen, 1977) do so mainly by an enlargement of individual adipose cells.

## Greowth Rate Effect

Comparing obese and non-obese humans, Hirsch and Knittle (1970) found that, both adipose cell size and number
were greater in obese subjects compared to non-obese humans. However, this observation was mainly because of the difference in adipocyte number which had a higher correlation with the degree of fatness. Hirsch and Knittle (1970) and Salans et al. (1971) concluded that childhood onset obesity is primarily associated with a hyperplastic increase in adipose depots, while adult onset obesity is accompanied mainly by hypertrophic changes in the adipocytes.

Hood and Allen (1977), reported a relationship between adipose cell number and the true body size in different $s t$ rains of pig. They concluded that at any live weight, the 1 e aner pigs had a larger number of extramuscular adipose $c e$ Ils than the fat group. They attributed this observation to the fact that fat pigs had fewer adipocytes than lean Pi ss due to the smaller true body size of these animals. They also suggested that there is a physiological relationsh ip between the number of adipose cells and the true body size of the animal. Johnson et al. (1971) suggested two gemeral classifications for obesity: (1) hypertrophic which WO uld be a model for adult obesity and (2) hypertrophichyperplastic which would be a model for early onset extreme Obesity in humans. Hood and Allen (1977) in their work sug$\boldsymbol{g} \boldsymbol{e}$ sted that excessive accumulation of fat in pigs is of the first type, that is, caused mainly by hypertrophy. They al so concluded that the carcass fat had a higher correlation with adipose cell size than adipose cell number. These
reports are confirmed with the results in human adipose tissue (Bjorntorp el al., 1971; Bjorntorp and Ostman, 1971; Salans et al., 1971).

Adipocyte size distribution (diameter) in pigs of difEerent quantities of backfat was studied by Allen et al. (1974). They reported that two groups of fatter pigs had biphasic diameter distribution for subcutaneous fat (Mersmann et al., 1973; 1975), bovine intramuscular (Hood and Allen, 1973 ) and bovine subcutaneous (Allen, 1976). It has been Concluded that the biphasic fat cell size distribution were due to either a reinitiation of hyperplasia or multiphasic $\mathrm{P} £$ riods in differentiation from preadipocytes and subsequent 1 i pid folling (Allen, 1976).
$S \in x$ Effect

Lee et al. (1973b) reported that even though barrows were fatter than gilts, barrows had fewer adipocytes than gills at constant body weight. It appears that adipocyte number and fat free-carcass weight have some physiological Te lationship. The possible reason for this relationship may be related to the fact that pigs with larger true body size require a large number of fat cells in order to reach the Same degree of fatness as pigs with a smaller true body size. Merkel et al. (unpublished data) found that at eight weeks, ewes had a larger fat cell size than either wethers or rams, although both of the latter sex groups were heavier than
ewes. At 16 weeks, fat cell size of the three group were not different, but at 32 weeks rams had smaller adipocytes than either wethers or ewes.

Nutrition Effect

Effect of level of nutrition of lambs has been studied by Haugebak et al. (1974). They reported that perirenal adiPocyte number was not changed by dietary treatment in main$t e n a n c e$ or ad libitum fed groups. This indicates that hyper$P$ Iasia in perirenal fat was completed before the animal was SLbjected to treatments. This is in agreement with Waters < ( 909) who concluded that perirenal adipose tissue developed $V 』 r y ~ e a r l y$ with regard to adipocyte number. However, hyperP Lasia in the subcutaneous and intermuscular depots were A』layed in maintenance diet groups as compared with ad 1 Ibitum fed group (Haugebak et al., 1974). These authors a 1 so reported that the mean adipocyte volume in any depot ODserved was smaller in the maintenance group than in the al libitum fed group of lambs.

The effect of early nutrition in pigs was studied by Lee et al. (1973a). The result of their experiments showed that although the age constant control pigs had about 1.5 to 4 times as much subcutaneous fat as the underfed pigs, the number of adipocytes in the subcutaneous depot was not Significantly different. On the other hand, the total number of adipocytes in intramuscular and visceral depots in
the control pigs were larger than underfed groups. In the weight constant experiment (Lee et al., 1973b), the total number of the adipocytes, adipocyte volume or the weight of the fat in various depots were not different between treatment groups. The exception was the intramuscular depot Which was smaller in underfed pigs. Studies of dietary restrictions with rats (Knittle and Hirsch, 1968) are in substantial agreement with reports in pigs (Lee et al., $1973 a$ ) on age constant basis. However, on a weight constant basis different conclusions have been reported (Lee et al., $1 \boldsymbol{\Omega} \mathbf{7 3 b}$ ). This disagreement confirms the fact that animals I ach compositional maturity at a body weight rather than aee (Bergen, 1974).

## Lipid Metabolism

Up take of Triglycerides

Quantitatively, the major lipid constituent in adipose ti ssue is triglyceride. The uptake of triglyceride from blood is believed to depend on the hydrolysis of triglycex ide in the capillary beds of the extrahepatic tissues by the enzyme lipoprotein lipase or so called clearing factor Iipase (Robinson, 1970). The activity of this enzyme has been identified in adipose tissue (Rodbell, 1964; Pokrajac et al., 1967; Parr, 1973; Haugebak et al., 1974; Cryer et al., 1975; Lithell and Boberg, 1978), muscle (Hollenberg, 1960;

Parr, 1973), heart and lung (Anfinson et al., 1952) and in Lactating mammary tissue (McBride and Korn, 1963), but not in liver (O1son and Alaupovic, 1966).

Uptake of triglyceride from blood is proportional to the rate of lipopretein lipase activity in adipose tissue (Bezman et al., 1962), therefore, this enzyme plays an important role in controlling lipid deposition in adipose tissue. Li poprotein lipase is a diet dependent enzyme. Its activity is lowered in starvation (Cherkes and Gordon, 1959; Hollenberg, 1959; Robinson, 1960; Wing and Robinson, 1968) and dii abetes (Pav and Wenkeova, 1960; Schnatz and Williams, 19 63; Brown et al., 1967) and is increased in refeeding ( $S$ alaman and Robinson, 1966; Reich1, 1972; Scow et al., 19 72; Haugebak et al., 1974). Haugebak et al. (1974) $\mathbf{r e p}$ ported that lipoprotein lipase activity was very low or non-detectable in adipose tissues from lambs fed at maintemance and slaughtered at the end of the growth period. However, when the lambs fed at maintenance were subsequently Gi ven a finishing diet ad libitum, the increase in carcass adipose tissue was paralleled by an increase in total lipo$\mathbf{P r}$ otein lipase activity in all adipose tissues. Results of experiments (Haugebak et al., 1974; Merkel et al., unpub1i shed data) have indicated that lipoprotein lipase activity Varies among anatomical fat depots with dietary manipulation. Li poprotein lipase activity as expressed on a cell number Or soluble protein basis was greater in subcutaneous adipose
tissue of lambs than in perirenal and intramuscular depots (Haugebak et al., 1974).

In an experiment with lambs (Merkel et al., unpublished data), lipopretein lipase activity in subcutaneous fat did not change as lambs grew from 8 to 32 weeks. Activity of the enzyme in perirenal fat was similar in 8- and 16week lambs, but decreased markedly in 32 -week lambs. In a study with pigs (Lee and Kauffman, 1974a) lipoprotein lipase activity increased in subcutaneous fat from birth to 4 weeks OE age, but only slightly thereafter up to 16 weeks. It Enen declined in subcutaneous fat, while it remained arechanged in muscle tissue (Lee and Kauffman, 1974a). Quantitatively, lipoprotein lipase activity in adipose tisSce of lambs was greater than in muscle (Parr, 1973; Lee 표 Kauffman, 1974a). Lipoprotein lipase activity in rat Pididymal fat decreased with increasing body weight $\ll 凡$ louverakis, 1962; Nestel el al., 1969). Results of lipoP $\mathcal{O}$ (ein lipase activities in different species of animals $5 \Omega$ ow that the activity correlated with the fat deposition of LE animals. (Nestel et al., 1969; Lee and Kauffman, 1974a, $19>4 \mathrm{~b}$ )
$E \in t y$ Acid Synthesis

Adipose tissue is the major site for de novo fatty
$\therefore<\boldsymbol{i} \mathbf{d}$ synthesis in ruminants (Payne and Masters, 1971; Ingle 르를., 1972a, 1972b; Martin et al., 1973). The pattern of
fatty acids synthesized is similar to the fatty acid composition of the tissue (Pothoven et al., 1974). The pathways of fatty acids syntheses in ruminant adipose tissue are different from nonruminants. The classic experiments of Hanson and Ballard (Hanson and Ballard, 1967, 1968; Ballard et al., 1972) first showed that adipose tissue from mature cows and sheep utilized acetate but not glucose as a carbon source for de novo synthesis of fatty acids. The pathways of de novo fatty acid synthesis from glucose and acetate is shown in figure 1 (Bauman, 1976). These pathways are also confirmed by other UOrkers who have studied adipose tissues from ovine, bovine ancl caprine species (Hood et al., 1972; Ingle et al., 1972b; Eldwin et al., 1973; Young and Baldwin, 1973). Fatty acid $\leq$ Snthesis can be described in a two step reaction by the ne lonyl CoA pathway (Kumar et al., 1972). In the first step, ER $\Rightarrow$ lonyl CoA is formed from acetyl CoA plus $\mathrm{HCO}_{3}^{-}$by the erz zyme acetyl coA carboxylase. In the second step, one HR 1 ecule of so called "primer" acetyl CoA condenses with $s \Longleftrightarrow$ Ven molecules of malonyl-CoA to form palmitic acid. The $\leq e<$ ond step is catalyzed by a multienzyme complex (fatty
 <MIayes, 1977). The overall reaction for this synthesis wlez Ch yields palmitic acid from acetyl CoA is:

$$
\begin{aligned}
A \_e t y l \mathrm{CoA}+ & 7 \text { malonyl CoA }+14 \mathrm{NADPH}+14 \mathrm{H}^{+} \longrightarrow \\
& \text { palmitic acid }+7 \mathrm{CO}_{2}+8 \mathrm{CoA}+14 \mathrm{NADP}^{+}+6 \mathrm{H}_{2} \mathrm{O}
\end{aligned}
$$



Figure 1. De novo synthesis of fatty acids from glucose and acetate (Bauman, 1976). (1) Pyruvate carboxylase. (2) Pyruvate dehydrogenase. (3) Citrate synthetase. (4) Citrate cleavage enzyme. (5) NAD-malate-dehydrogenase. (6) Malic enzyme. (7) AcetylCoA synthetase. (8) Acetyl-CoA carboxylase. (9) Fatty acid synthetase. (10) Hexokinase. (11) Glucose phosphate isomerase. (12) Aconitase. (13) NADP-isocitrate dehydrogenase. $B H B A=B$-hydroxybutyrate. The negligible activities in ruminant adipose tissues are denoted by $X$.

Some of the enzymes which are necessary for lipogenesis to occur from glucose as substrate are absent in ruminants. The enzymes which are absent in ruminant liver (Ballard and Oliver, 1964) and ruminant adipose tissues (Hanson and Ballard, 1967; Ingle et al., 1972b) are malic enzyme and citrate cleavage enzyme. The activities of these two key enzymes have been shown to be 50 -fold higher in rat adipose tissue than in ruminants (Hanson and Ballard, 1968). The two enzymes show dramatic changes in activity during Cevelopment of young ruminants. Ballard et al. (1969) found a considerable quantity of these two enzymes in fetal calf I I-ver which does possess the capability for lipogenesis from P ucose. The activities of these enzymes diminish as the工 mmen develops (Hardwick, 1966). Bauman et al. (1970) also $\tau \Longleftarrow$ ported very low activities of malic and citrate cleavage
er zyme in mammary tissue of ruminants which is indicative of $\mathbf{V} \sum$ Fy little incorporation of glucose into fatty acids in t12 m tissue. The inability of ruminants to incorporate glu$\infty \infty \in \mathbb{\infty}$ as a substrate for de nono fatty acid synthesis seems me t abolically adapted to conserve glucose for metabolic Frmetions such as energy production in nervous tissue and $\sum_{\sim}>$ throcytes, lactose synthesis in mammary glands, and proClecion of NADPH and $\alpha$-glycerol phosphated for triglyceride $s T 3$ thesis in lipogenic tissues (Allen et al., 1976).

The source of reducing equivalents for de novo fatty
$a<-\mathbb{Z}$ synthesis in ruminant adipose tissue also differs from
non-ruminants. In ruminant adipose tissue, NADPH is generated in the hexose monophosphate shunt pathway (via glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase). The activity of NADP-isocitrate dehydrogenase has been reported to be extremely high in ruminant adipose tissue relative to its activity in non-ruminants (Bauman, 1976). The advantage of the isocitrate cycle in ruminants is that acetate can be utilized to generate NADPH. It has been estimated that at least 25 percent of the NADPH necessary For lipogenesis in ruminants is supported by the isocitrate CScle, with the remainder generated from the pentose phosphate © Scle (Young and Baldwin, 1973).

Adipose tissue appears to be the major organ for de TR Qvo synthesis of fatty acids in ruminants (Ingle et al., I 972 b ) and nonlactating pigs (O'Hea and Leveille, 1969), wrile the liver is more important in birds (Leveille et al., I $968 ; 0$ Hea and Leveille, 1968). In rats both organs conE = bute significantly for de novo synthesis of fatty acids <Teveille, 1967; Chakrabarty and Leveille, 1968).

The intracellular sites of fatty acid synthesis are
C
A 工 hough ctyoplasmic and mitochondrial enzyme systems are $s=$ milar, they have different end products which are palmitic ata stearic acid for cytoplasmic and mitochondrial systems $\mathcal{T} e s$ pectively (Masoro, 1968).

Fatty acid synthesis has been shown to be influenced
by breed (Chakrabarty and Romans, 1972; Hood and Allen, 1975), age (Ingle et al., 1972b; Pothoven et al., 1975), diet (Allee et al., 1972; Ingle et al., 1973; Pothoven and Beitz, 1973) and anatomical site of the depot (Anderson et al., 1972; Hood and Allen, 1975). In contrast to non-ruminants, the fatty acid composition of adipose tissue in ruminants is not markedly affected by the fatty acid composition of the diet. This is because rumen microorganisms are able to hydrogenate the unsaturated fatty acids. Therefore, the Preformed fatty acids which are taken up by adipose tissue a re predominantly saturated (Dawson and Kemp, 1970).

## $\lessdot$ Iyceride Synthesis

There are two known pathways in the mammalian system $\mathcal{F}$ ©r triglyceride synthesis. The 2 -monoglyceride pathway <Clark and Hubscher, 1961) and glycerol 3-phosphate pathway <Weiss et al., 1960). Biosynthesis of triglycerides via the P1. $工$ cerol phosphate pathway appears to be the major route in alizpose tissue (Shapiro, 1965; Vaughan and Steinberg, 1965), - 12 liver (Weiss and Kennedy, 1956; Marinetti, 1970) and in L2e mammary gland (Howard and Lowenstein, 1965). In intesc Inal mucosa both pathways are functional, but, studies have KRown that the monoglyceride pathway is more important than P 1 Cerol phosphate pathway for re-synthesis (Johnston and BTC Own, 1962; Senior and Isselbacher, 1962; Senior, 1964).

The pathways and enzymes for triglyceride biosynthesis
are shown in figure 2. Triglyceride formation from fatty acids is dependent on ATP, CoA and $\mathrm{Mg}^{2+}$. Glycerol cannot replace $\alpha$-glycerol phosphate in the glycerol phosphate pathway, however, it has been shown that there is the possibility of some ester formation when millimolar amounts of glycerol are added (Margolis and Vaughan, 1962). Even in the absence of $\alpha$-glycerol phosphates, a small amount of fatty acids has been shown to be incorporated into triglyceride (Steinberg et al., 1961). This has been suggested to be due to endogenous $\alpha$-glycerol phosphate which might not have been Femoved completely upon dialysis or also may be due to e Strification of diglycerides, preformed or generated by 1 Enpolysis during incubation (Vaughan and Steinberg, 1965). ILerefore, formation of glycerol phosphate is an obiligatory $=$ Eep in the synthesis of triglycerides. This may be formed B phosphorylation of glycerol (derived from hydrolytic b $\mathcal{F}$ eakdown of lipids) with ATP in a reaction controlled by $\mathcal{L}$ cerol kinase (ATP-glycerol phosphotransferase), and also D reduction of dihydroxy -acetone phosphate which is gene a ted by the glycolytic sequence of reations, with the NAD1 Inked dehydrogenase as the enzyme. It has been reported c-at glycerol kinase has limited distribution in animal tisscres, but it is found in liver (Bublitz and Kennedy, 1954), le Aney (Wieland and Suyter, 1957) and intestinal mucosa $\lll$ ark and Hubscher, 1962) mainly in the cell sap. It is $e_{s} s$ entially absent from adipose tissue (Margolis and Vaughan,

## 2-monoglyceride pathway

Glycerol 3-phosphate pathway


[^0]1962), therefore, glycerol phosphate must be formed from the dihydroxyacetone phosphate via glycolysis of glucose. It has been reported that the level of $\alpha$-glycerol phosphate may be a key control for triglyceride synthesis in adipose tissue (Leboeuf, 1965). This suggests that triglyceride synthesis in adipose tissue is closely associated with carbohydrate metabolism including gluconeogenesis. This latter is particularly important in fat metabolism of ruminants. Results reported by Packter (1973) indicated that the rate of triglyceride synthesis is increased following Eeeding, which is accompanied by increased levels of blood E Iucose and insulin.

Although the subcellular site of triglyceride synthesis
EI $\sum$ adipose tissue in not known, the subcellular site of E Y Yceride biosynthesis in mammary gland, liver and intestinal mincosa of several species has been identified. Studies on marmary gland of cow (Gross and Kinsella, 1973), goat and $s \infty$ (Bickerstaffe and Annison, 1971), rat (Tanioka et al., $1 \gg 3)$ and guinea pig (Kuhn, 1967) indicate that the main sub$\subset \Longleftrightarrow 1$ Iular site of triglyceride synthesis in the above mentioned $S \sum \&$ cies is the microsomal fraction. The microsomal fraction 12 been found to be responsible for glyceride synthesis in m\&2, guinea pig and rat livers (Daae, 1973) and sheep, chicken ars pig intestinal mucosa (Bickerstaffe and Annison, 1969).

Lipid Mobilization

Lipid synthesis and mobilization in ruminant adipocytes are not independent, but their control must be coordinated. These two functions tend to be reciprocal process (figure 3 ).

Synthesis and mobilization of fat depot triglycerides are in a dynamic state. After feeding, a hyper-insulin state ensues especially in non ruminants. Insulin increases fatty acid synthesis and ultimately triglyceride synthesis. Insulin is believed to involve at least two possible mechanism of action in the synthesis of triglyceride. First it 표creases glucose permeability which stimulates glycolysis End hexose monophosphate shunt. The former yields acetyl CoA and glycerol phosphate and the latter produces NADPH. ©onsequently, the tricarboxylic acid cycle produces more ATP. A 11 of these processes result in higher rates of fatty acid ATP triglyceride synthesis. The second mechanism of action 0 F insulin is believed to be at the level of gene expression <MIErinetti, 1970). Injection of insulin has been shown to i-2 己rease the activity of certain enzymes such as acetyl CoA © (zbboxylase and citrate cleavage enzyme, both of which are $\overline{\text { i morntant for fatty acid synthesis (Tepperman and Tepperman, }}$ 1 - 5 ; 01son, 1966). Insulin also leads to increased glucose6 - Dhosphate dehydrogenase and 6-phosphogluconate dehydro$\mathcal{E}$ \&rase activity.

The rate of influx and efflux of non-estrified fatty


PI $\underset{\sim}{2}$ ma

FIER Pe 3. Synthesis/mobilization in ruminant adipocytes (Buuman, 1976)
$<2\}$ De novo fatty acid synthesis
$<2\} \begin{aligned} & \text { Uptake of plasma fatty a } \\ & \lll l\end{aligned}$
$\lll$ Fatty acid mobilzation NEFA $=$ non estrified fatty acid
acids in adipose tissue is under hormonal control, particularly insulin and epinephrine (Newsholme and Start, 1973). Insulin increases influx of NEFA while epinephrine increases efflux of NEFA from adipocytes. The effects of epinephrine are always opposed to those of insulin. The primary effect of epinephrine is to increase the hydrolysis of triglycerides in adipose tissue by a mechanism in which the hormone (epinephrine) is believed to stimulate the adenyl cyclase system by direct interaction at or near the cell membrane (Robinson et al., 1967). This interaction results in proCuction of more cyclic AMP which in turn stimulates the a Ctivation of hormone sensitive lipase.

Feedback control of triglyceride synthesis has been $\boldsymbol{C} \ell$ scribed by Newsholme and Start (1973). They indicated t1at fatty acids or fatty acyl-CoA esters have a negative $\boldsymbol{E} \Longleftarrow 巳$ dback effect on acetyl CoA carboxylase. The rate of

CI Crate formation and its effect on acetyl CoA carboxylase i $s$ another control point affecting the formation of acetyl CoA carboxylase which is believed to be a rate limiting ers z Sme in fatty acid synthesis. However, in vitro studies 51 亿 that the level of citrate needed to convert acetyl CoA C D Doxylase from a monomer to a more active trimer is much Hz Her than the physiological level of citrate in the cell $<V A$ zelos, 1964).

The control of fatty acid synthesis in adipose tissue $\mathrm{FR}_{2}$ dietary carbohydrate or lipid has a two-fold effect.

Ingestion of these nutrients influences the hormonal state of the animal together with a favorable substrate concentration and causes enzyme activities to increase and hence the rate of fatty acid synthesis. In short, the activity of the esterifying enzymes in starvation and refeeding parallels the activities of other enzyme systems involved in lipogenesis and lipid mobilization. Enzymes of the hexose monophosphate shunt pathway (Hollifield and Parson, 1965), fatty acid desaturating enzymes (Benjamin and Gellhorn, 1966) and Iipoprotein lipase activity (Hollenberg, 1959) are all
decreased in adipose tissues of fasted rats. These activi_ $\mathcal{i}$ ies have been shown to be restored to levels close to, or above normal with refeeding (Hollenberg, 1959; Hollifield arid Parson, 1965; Benjamin and Gellhorn, 1966).

Release of fatty acids from triglycerides is catalyzed
by triglyceride (hormone sensitive), diglyceride and mono8 $\mathbf{~} \boldsymbol{\sim}$ ceride lipases. The rate limiting step in lipolysis is the hormone sensitive lipase reaction. This enzyme has been Snezed to be a cytoplasmic enzyme in lipid rich matrix ce I Is (Khoo et al., 1972). The regulation of this enzyme $0 \& D E n d s$ on the intracellular level of cyclic AMP (Patton, $18>0$; Robinson et al., 1971). In this mechanism, cyclic AMP $s t=\mathcal{L}$ mulates protein kinase which in turn activates hormone Sensitive lipase by converting from the non-phosphorylated in a etive form to a phosphorylated active state (Robinson et Al., 1971). Lipolysis in adipose tissues removed from
lambs differing in propensities to fatten has been measured by Sidhu et al. (1973). They reported that lipolysis increased with age and fatness in lambs. In contrast to nonruminants, relatively few studies have been conducted with ruminants. However, it is apparent that ruminant adipose tissue is much less sensitive to lipolytic hormones than non-ruminants (Prigge and Grande, 1971).

Postnatal Muscle Growth

Changes in Muscle Mass During Growth

Regardless of size, muscle tissue constitutes approxinmately 25 percent of human and rat body weight at birth <Elliott and Cheek, 1968). This percentage changes to 45 $P @ \mathcal{P c e n t}$ in the adult mammal (Young, 1970). Therefore, there $\boldsymbol{i}=$ a substantial increase in the proportion of muscle during $p \infty=$ tembryonic period.

Postnatal growth of mammalian muscle fibers is almost ET irely due to hypertrophy of pre-existing muscle fibers 2t- not by hyperplasia (Stromer et al., 1974). However, $s<m$ e postnatal increase in muscle fiber number has been $\mathcal{F} \&$ orted (Goldspink, 1962; Chiakulas and Pauly, 1965; Bridge ATA Allbrook, 1970), which appears to depend on the state of mat urity of animal at birth, which can be considered as an exte ension of the embryonic differentiation of the tissue.

The increase in the length of muscle fiber is primarily associated with an increase in the number of sarcomeres along myofibrils (Goldspink, 1968), as well as a small increase in the length of the individual sarcomeres (Aronson, 1961; Shafiq, 1963). However, the increase in the length of individual sarcomeres is more important in invertebrates than vertebrates (Aronson, 1961). The changes in sarcomere length may vary in different species and strains of animals according to their rate of growth.

There are different schools of thoughts concerning the addition of sarcomeres to the myofibrils. Some authors
(Ruska and Edwards, 1957) have suggested that the myofibrils EFow interstitially; in other words, new sarcomeres are a dded to the myofibrils at some point along their length. IRey have based their theory on the fact that the sarcomeres O $\mathcal{F}$ adjacent myofibrils are often out of register because of $s$ I Ight differences in sarcomere length. In this case, there $\boldsymbol{\omega} \boldsymbol{\sim}=1$ be some of the myofibrils with additional sarcomeres for E Siven length of muscle which is taken as evidence that the $s$ zcomere has been inserted. However, in order to insert a 2 2 $\sim$ sarcomere in this way, it is necessary for the myofiber $T$ © only to divide transversally, but, also it should involve maifications of sarcoplasmic reticulum and transverse tub1 - Zr system (Goldspink, 1972). Other workers (Holtzer et al., $1 S 57$; MacKay et al., 1969) have suggested that the lengthen$i<2$ of myofibrils occurs by serially adding sarcomeres to
the myofibrils.
The mechanism by which the new sarcomeres are added has been discussed by Goldspink (1972). He suggested that the ends of fibers are the regions of longitudinal growth and that the new sarcomeres are most probably added serially to the ends of the pre-existing myofibrils. This hypothesis fits with fact that the terminal sarcomeres of myofibrils are shorter than those in the middle. Presumably the terminal sarcomeres are the most recently formed ones which have not had time to increase in length. The latter hypothesis is COnfirmed with the experiments (Williams and Goldspink, 1971)

In which tritiated adenosine was injected into growing mice. Antoradiography and scintillation counting from these expe-I- ments showed that most of the label was incorporated into t1e end regions of the muscle fibers, suggesting that these Ie $\mathcal{E}$ ions are more active in the synthesis of actin and ribosomal RNA.

The mechanism of sarcomere assembly is not well under$s<\infty$ od. Legato (1970) suggested that in cardiac muscle, Z$\boldsymbol{A}=\leq \mathbf{k s}$ are the centers for the assembly of the new sarcomeres. T1 $\overline{-}$ s assembly is accomplished by hypertrophy of Z-disk mate$\mathcal{T}$ - $1 s$ which occupy the areas where the sarcomere will ultim $\Rightarrow$ ely develop and then by gradual replacement of $Z$-substance, CT $\mathcal{L}$ ck and thin filament form the new sarcomere (Ezekwe and Matetin, 1975).

The increase in girth of muscle fiber is almost
entire $l y$ by the increase in the number and size of myofibrils (Goldspink, 1972). Studies by Goldspink (1970) showed that the number of myofibrils in mouse biceps brachii muscle may increase up to 15 -fold during postnatal growth. Goldspink (1972) suggested that when the myofibril reaches a certain thickness, it splits longitudinally by the force originated from stress placed on the Z-disk by the oblique pull of the actin $\mathbf{E i}_{\text {ilaments during contraction. This tension is suffi- }}^{\text {a }}$ cient to tear a small hole in the center of the disk which then spreads longitudinally and causes splitting of the entire myofibril.

Learge animals tend to have larger muscle fibers than those OI small animals (Luff and Goldspink, 1967; Byrne et al., 1973 ; Hanrahan et al., 1973; Ezekwe and Martin, 1975); but the difference in muscle size between the large line and small line is mainly due to difference in the total number of fibers in the muscle and not to difference in the F iber size (Luff and Goldspink, 1970). Although there is 2 ) difference in the total number of fibers between the same Ematomical muscles of males and females in mice, the mean F Jiber diameter in the male is greater than in females (Rowe - zid Goldspink, 1969).

Adrian et al. (1969) reported that from a physiological
$s$ Candpoint, it is not feasible to have development of fiber 0 Eyond a certain diameter, because the distance from the © $\Longleftarrow$ nter $O f$ the fiber would be too great to allow for oxygen
diffus ion and also impulse transmission down the T-system, to the center of the myofibril. This confirms the fact that somehow during the evolution of the larger animals it has been $n$ ecessary for the fiber to increase in number rather than size.

It has been shown in rodents (Rowe and Goldspink, 1969) that muscle fibers grow in a discontinuous way rather than in a gradual and continuous manner. Very soon after birth all of the fibers are approximately the same size. As the animal §rows postnatally some of the muscles such as the biceps brachii will undergo extensive hypertrophy as compared to other muscles such as soleus and extensor digitorum longus, which will essentially retain their original size throughout the life of the animal.

The population of small and large fibers can be Changed by exercise or changing the level of nutrition of © he animal (Goldspink, 1970). The stimulation for hyperE rophy of muscle fibers is believed to be due to the inten-
$s$ ity of the work load to which the fiber is exposed. This I-s apparant, because as the animal grows there will be a E onsiderable increase in body weight in the animal, thereore, the work load on skeletal muscle will be increased. Hypertrophy of striated muscle fiber due to exercise has $\geq$ een accepted for many years (Morpurgo, 1895). Morpurgo (1895) attributed muscle fiber hypertrophy to an increase in searcoplasm rather than myofibrils. Later, cytological
studies showed that hypertrophy was mainly associated with incre ases in myofibrillar portion of the muscle fiber (Richter and Kellner, 1963; Goldspink, 1964, 1970). However, under certain conditions of exercise some hypertrophy of the fibers has been found to be partly or wholly due to increase in mitochondrial and sarcoplasmic proteins (Gordon et al., 1967).

The effect of exercise on hypertrophy of muscle fibers has been studied at the molecular level by Goldberg (1968, 1969) and Hamosh et al. (1967). Goldberg $(1968,1969)$ has reported that during work induced hypertrophy, the incorporation OE leucine - ${ }^{14} \mathrm{C}$ into both sarcoplasmic and myofibrilLar proteins is enhanced and also the rate of degradation of these proteins is reduced.

Hamosh et al. (1967) reported that during hypertrophy Of muscle fibers, there is an increase in RNA concentration end also there is a greater ability for the cell-free system to synthesize proteins. They found that L-phenylalanine was - ncorporated into microsomal protein at a faster rate by the nelicrosomal fractions prepared from hypertrophied muscle, $\int$ oth in the presence or absence of artificial RNA (Poly U.). T- hey also reported an increased RNA content in the micro$\approx$ omal Eractions. .

Hormones may exert a direct effect on muscle growth or $\sum$ indirect via regulation of food intake in the animal. Seve zral hormones affect protein metabolism and the growth of
skele $E$ muscle. Insulin and growth hormone are considered to have the greatest effect on protein synthesis of mammalian skeletal muscle. Insulin stimulates amino acid uptake in muscle via its interaction with the cell membrane (Figure 4). Individual amino acids are taken into the intracellular compartment in proportion to the amino acid composition of the muscle protein, rather than in proportion to the amino acid in the extracellular compartment (Turner and Munday, 1976). Insulin also stimulates the translation process independent 1 y of amino acid intake, by an action possibly mediated by inhibition of adenyl cyclase and stimulation of guanOsyl cyClase (Cuatrecasas, 1974).

Although in vitro experiments using hypophysectemized animals have lead to the conclusion that growth hormone stimulates the transport of both amino acids and glucose, as well ans stimulating the incorporation of amino acids into proteins, I- $t$ should not be concluded that growth hormone mimics the a ctions of insulin. It has been shown that administration (f growth hormone to whole animals results in both protein Enabolism in muscle and lipolytic effects in adipose tissue <Reeds et al., 1971). The anti-insulin action of growth hormone on adipose tissue in the absence of an increase in [- nsulin is a protective mechanism for body protein during Fasting, exercise and stress in that they prevent the exces$S$ ive use of amino acids or substrate for the generation of P $\mathcal{P}$ ucose and metabolic energy. Nevertheless, when insulin


120
$<$ figure 4 . Diagram for the control of muscle protein metabolism - Stimulation; - OInhibition; $\sum \sum$ Cell membrane
and growth hormone secretion occur simultaneously, such as in the case of after feeding, the action of growth hormone on muscle is truly anabolic, which leads to the conclusion that here may be an important interaction between insulin and growth hormone in stimulation of muscle protein synthesis (Reed et al., 1971). Just as growth hormone needs insulin for its protein anabolic effect, insulin also depends on growth hormone for its protein systhetic action, which indicates that insulin and growth hormone are mutually dependent for increasing protein systhesis. In addition, it seems that they have a synergestic effect on protein synthetic action when the concentration of both hormones increases (Turner and Munday, 1976).

Androgens have direct or at least an indirect effect On muscle development (Goldspink and Rowe, 1968; Grigsby et al., 1976). The degree of responsiveness of different muscles to androgens varies considerably. Kochakian et al. $\subset$ 1961) working with guinea pigs reported that temporal and measter muscles are very sensitive to castration and subseQ uent replacement therapy. However, the response of muscle t $\omega$ androgens is more uniform (Kochakian, 1966), with the e sxception of lavator ani muscles (Venable, 1966, a,b). A-ndrogens increase the rate of protein synthesis in most metuscles. Results of experiments (Novak, 1957; Kochakian,
I Q66) indicate that following administration of adrogens, - Ie incoxporation of labelled amino acid into muscle proteins
is increased in both intact or castrated animals. Florini and BIeuer (1966) reported that ribosomes obtained from castrated animals are less active in protein synthesis than those of intact animals. They also showed that the combination of testosterone and growth hormone can modify the protein synthesis ability of ribosomes. They concluded that the main factor for increasing protein synthesis by these hormones is the increase in messenger RNA production.

The influence of early nutrition on growth and development of muscle fibers has received considerable attention in recent years. Muscle is one of later developing tissues and may be affected by nutritional deprivation imposed during hyperplasia. In an experiment with pigs, Robinson (1969) reported that undernutrition during pregnancy does not affect muscle cell number, while stress during pregnancy and Iactation caused the termination of muscle cellular hyperplasia to be earlier than those of the control. It is well L-snown that the reduction in food intake by animal or human causes a considerable reduction in muscle mass (Allison et ㄹ1., 1962). The decrease in muscle mass is shown to be Essociated with a decrease in muscle mean fiber diameter <Joubert, 1956; Montgomery, 1962; Goldspink, 1964, 1965). Goldspink (1964) and Rowe (1968) reported that the starvaCion effect on those mouse muscles that are normally comEosed Of large and small fibers, causes a reduction in the Zumber of large phase fibers in the muscle so that the fiber
size distribution tend to become unimodal again. Goldspink (1965) has also reported that the increase in the fiber size is due to reduction in the number of myofibrils in the fiber and that this accounts for the decrease in the contractile strength which is normally associated with starvation or atrophy. The mechanism of reduction of myofibrils from muscle fiber during starvation is not clear, however, Bird et al. (1968) reported that levels of cathepsins was increased five days after reduction of food intake. On the other hand, DeDuve et al. (1962) postulated that lysosomal enzymes were functioning in the normal economy of cell catabolism or renewa 1. Therefore it seems that there should be a mechanism under Which the release of these enzymes could be increased in case of fasting (Bird et al., 1968) or retarded in case of refeeding.

Changes in Muscle Protein and Nucleic Acids During Growth

True cellular growth is estimated by measuring weight, protein, DNA and RNA content of tissues and organs (Mirsky And Ris, 1949). The increase in mass of protein during Fypertrophy of muscle cells may rise from changes in the Zcates Of either protein synthesis or degradation or changes In both. The contribution that changes in the degradation Fate made during hypertrophy of skeletal muscle has not Deen clearly established. Turner and Garlick (1974) and

Millward et al. (1975) calculated that protein degradation rate doubled during the period of rapid muscle growth in rats. In contrast Goldberg (1969) reported a decrease in degradation rate which contributed towards the increased protein mass in rat Soleus muscle during hypertrophy, because more radioactivity was retained in the proteins of hypertrophying muscle than of the controls eight days after pulse 1 abeling with ${ }^{3} \mathrm{H}$-leucine.

The amount of DNA per diploid nucleus is generally considered to be consistent in tissues (Mirsky and Ris, 1949; Vendrely, 1955) and since there is no evidence of polyploidy during skeletal muscle growth (Enesco and Puddy, 1964), the increase in DNA reflects an increase in number of nuclei.

Results of experiments show that the total content of DNA is increased during growth (Enesco and Puddy, 1964; Gordon et al., 1966; Buchanan and Pritchard, 1970; Johns and Bergen, 1976; Harris et al., 1977; Laurent and Sparrow, Z977). Harbison et al. (1976) reported that total DNA =ncreased approximately 2.0 (obese pigs) - 2.7 (muscular Digs) fold between 23 and 118 kg of live weight. These Cesults agree with the data for rats (Enesco and Puddy,工 964; Enesco and LeBlond, 1962; Harris et al., 1977), mice <Robinson and Bradford, 1969), pigs (Powell and Aberle, Z 975), Cattle (LaFlamme et al., 1973) and chickens (Moss, Z 968; Moss et al., 1964). Gordon et al. (1966) working

With rats, reported that there was no increase in cell weight per unit DNA during the period of nuclear prolifexation. However, after 90 days, hypertrophy alone continued to occur which was mainly due to increases in myofibrillar and sarcoplasmic proteins.

Although some of the DNA increase in muscle may originate from an increase in the number of total nuclei associated with connective tissue and other cell types that $1 \mathbf{i e}$ between adjacent muscle fibers (Jablecki et al., 1973), Enesco and Puddy (1964) reported that a major proportion of this increase must originate from an increase in the number Of nuclei within the muscle fiber. Studies have show that nuclei within the multinucleated skeletal muscle do not undergo mitotic division, therefore, the increase in muscle DNA cannot originate from mitotic division of muscle nuclei.

The most likely answer to the question of origin of new nuclei that appear postnatally in muscle fibers are the satellite cells. The presence of these cells has been reported by Mauro (1961) who described them as mononucleated fusiform cells, lying between the basement membrane and the plasmalemma of multinucleated skeletal muscle fibers. Recent studies (Shafiq et al., 1968; Moss and LeBlond, 1970, 1971) have shown that satellite cells will incorporate labeled thymidine into their nuclei, which is indicative of mitotic ability. Results (Reger and Craig, 1968; Reznik, 1969; Moss and LeBlond, 1971; Schultz, 1976) show that
satellite cells present in even mature muscle fibers may undergo mitosis and that one or both of the daughter cells Erom this mitosis may be incorporated onto or fused with the mu1tinucleated skeletal muscle fiber and in this way add one OI both nuclei to the fiber.

In addition to total DNA, the total content of RNA per muscle also increases with age (Young and Alexis, 1968;

SI ivastava and Chandhary, 1969; Howarth and Baldwin, 1971; Johns and Bergen, 1976; Harris et al., 1977). However, the Concentration of DNA and RNA in muscle decreases with age <Powell and Aberle, 1975; Aberle and Doolittle, 1976; Harbison et al., 1976). Likewise the percentage of ribosomes in polyribosome aggregates also decreases during growth (Breuer and Florini, 1965; Srivastava, 1969), which is probably due to diluting of polyribosomes by the rapidly accumulating myofibrils (Goldspink, 1972; Tsai et al., 1973). Srivastava (1969) concluded that the decrease in ribosomal concentration is probably due to production of messenger RNA that becomes the limiting factor in myofibrillar production. Giovannetti and Stothers (1975) reported that RNA concentrations were inversely related to the Gastrocnemii muscle and body weight gains in rats. The decrease in concentration of RNA with increasing age was considerably influenced by diet (Giovannetti and Stothers, 1975). Harbison et al. (1976) reported a 54 percent decrease in muscle DNA concentration during growth. Similar results were obtained by other investigators
(LaFlamme et al., 1973; Powell and Aberle, 1975). Between
1ive weights of 23 kg and 68 kg , Harbison et al. (1976)工eported no difference in either muscle DNA or muscle RNA COncentration between obese and muscular genetic lines of Pigs. However, between 68 kg and 118 kg , animals from the muscular line had greater muscle DNA and muscle RNA concenCIations than animals for the obese line. Similar results were reported by Robinson and Bradford, 1969; Ezekwe and Martin, 1975; Powell and Aberle, 1975.

Munro (1969) and Goldberg (1967) have reported that the higher RNA concentration in muscle tissues are associated
with higher protein synthesis rates as measured by uptake Of labeled amino acids. Millward et al. (1973) reported that for rats in a variety of nutritional states, there was a linear relationship between the RNA concentration and the protein synthesis rate of hind limb muscles. The RNA concentration may increase through an increased de novo synthesis. This increase may occur in two ways: new nuclei may be produced which will synthesize new RNA, or alternatively, the rate at which pre-existing nuclei produce RNA may increase (Laurent and Sparrow, 1977).

Evidence for an increased RNA synthesis per nucleus can be obtained by measuring RNA/DNA ratio. Topel (1971) reported an increased RNA/DNA ratio in the longissimus muscle of a muscular genetic strain of pigs, suggesting increased protein synthesis. Ezekwe and Martin (1975)
reported a 4-fold increase in total muscle RNA and a greater RNA/DNA ratio in heavily muscled pigs which is indicative of a greater capacity of protein synthesis. Laurent and Sparrow < 1 977) reported a RNA/DNA ratio which was 39 percent higher ater two days of hypertrophy of the anterior latissimus Corsi muscle of adult fowl compared to the no hypertrophy Controls. They also reported that the RNA concentration of the muscle increased by 76 percent during hypertrophy. Similar increases have been reported during compensatory hypertrophy of rat soleus muscle (Goldberg, 1971).

Changes in Muscle Proteins During Growth

The contractile proteins increase during growth. In most animals, there is an increase in percentage dry weight and percentage nitrogen content of muscle during the period shortly after birth (Lawrie, 1961; Schwartz, 1961; Goldspink, 1962). This nitrogen content increase is mainly due to the increase in the number of myofibrils per muscle fiber. The relative changes in muscle myofibrillar and sarcoplasmic proteins with age are somewhat consistent among various animals as reported by different investigators (Helander, 1957; Dickerson and Widdowson, 1960; Gordon et al., 1966).

The myofibrillar proteins consisting principally of myosin, actin and tropomyosin and the less characterized minor proteins represent about 55 to 65 percent of the total
nitrogen in adult skeletal muscle (Perry, 1970). Helander
(1961) found an increase in myofibrillar protein concentration From $9.86 \mathrm{~g} / 100 \mathrm{~g}$ of muscle to $11.10 \mathrm{~g} / 100 \mathrm{~g}$ of muscle without any change in sarcoplasmic protein content in guinea pigs OE 4 to 6 weeks of age, after they had run 1000 meters daily For three months. These results were in constrast to the PIevious works which denied any increase in myofibrils while strongly supporting the point that hypertrophy was totally that of the sarcoplasmic proteins (Holmes and Rasch, 1958). Dickerson and Widdowson (1960) working with humans and pigs, quantitated changes in nitrogen constituents of muscle of these species during growth and development. They Found that non-protein nitrogen increased in the early stages Of development but reached its adult level soon after birth. Even though the newborn human had a higher initial non-protein nitrogen concentration than swine, the concentration of total protein nitrogen increased to the same minimal level during development of both species. During fetal life, the concentration of sarcoplasmic protein changed very little in human muscle but apparently decreased in porcine muscle. Postnatally, the concentration of sarcoplasmic and myofibrillar protein nitrogen increased, with greater percentage increases in myofibrillar than in sarcoplasmic nitrogen during most stages of development in both species. The Changes of protein components of muscle tissue continued to occur in both species until maturity. The concentration of
myofibrillar proteins were higher than sarcoplasmic at most stages in human and porcine muscle. Similar changes have been reported in rabbit Longissimus muscle (Perry, 1970) and in chicken (Hermann et al., 1970).

Weiss et al. (1971) reported that fat strain pigs had more sarcoplasmic and less myofibrillar protein than did 1 ean strain pigs. Sarcoplasmic and myofibrillar protein solubility increased as weight increased. Similar trends were also observed in human and pigs by Dickerson and Widdowson (1960) and in cattle by Helander (1957). Those results reflect increased myofibrillar protein deposition for a more meat-type pig as compared to fat type.

According to Dickerson and Widdowson (1960) non-protein nitrogen changed very little during development of human muscle and formed a lower proportion of total nitrogen than in pig muscle. In the pig the peak value was shown in the newborn animal. However, Weiss et al. (1971) reported that strain did not affect the quantity of non-protein nitrogen.

Since the intramuscular fat varied considerably, Lawrie (1961) expressed the data for total nitrogen, myofibrillar sarcoplasmic and non-protein nitrogen on the fat free basis. He showed that although the adult values for myofibrillar and sarcoplasmic nitrogen in cattle were attained at about 5 months of age, total nitrogen was reached somewhat later and appeared to reflect an increase in non-protein nitrogen
at this time.
The proportion of stromal nitrogen relative to total ITtrogen in newborn beef animals has been found to be larger Enan in the adult. However, the solubility of stromal pro$t e$ in diminishes throughout growth (Helander, 1957; Dickerson and Widdowson, 1960). The decrease in stromal nitrogen has

Deen shown to be due to dilution by the other fractions which show a proportionally greater increase during growth.

## Experimental Design

A commercial flock of 160 ewes served as the genetic base for differences in growth rate based on their records Of the previous 3 years. The 60 ewes with the fastest growing lambs in previous years were mated to 2 Suffolk rams which also had fast growth records. Likewise the 60 ewes with the slowest growing lambs were mated to Dorset rams which had slow weight gain records. Only lambs from ewes with twins were included in the study and only one of the twin pair was used in each case. Three ram and three ewe lambs of each growth rate at each age were slaughtered as shown in the experimental design in table 1.

Newborn lambs were removed from their dams shortly after birth and not allowed to suckle. The remainder of the lambs nursed at will and also creep fed ad libitum. Composition of the creep ration is shown in table 2. The remaining lambs were weaned at 82 days of age and brought to the University barns and divided into four groups (fast growing rams and ewes, and slow growing rams and ewes). Each group was penned and fed separately until slaughter time

TABLE 1. ALLOCATION OF THE LAMBS TO THE EXPERIMENT

|  |  | Grow | rat |  |
| :---: | :---: | :---: | :---: | :---: |
|  |  | st |  |  |
| Age of lambs (in |  |  | S |  |
| days) at slaughter time | Ram | Ewe | Ram | Ewe |
| 0 | 3 | 3 | 3 | 3 |
| 35 | 3 | 3 | 3 | 3 |
| 70 | 3 | 3 | 3 | 3 |
| 105 | 3 | 3 | 3 | 3 |
| 140 | 3 | 3 | 3 | 3 |
| 175 | 3 | 3 | 3 | 3 |
| Total | 18 | 18 | 18 | 18 |
|  | $=72$ lambs total |  |  |  |

TABLE 2. COMPOSITION OF THE CREEP RATION

| Ingredient | Percentage of total |
| :--- | :---: |
| Alfalfa Meal | 25 |
| Corn | 28 |
| Soybean Meal (49\% protein) | 29.5 |
| Crimped Oats | 10 |
| Molasses | 6 |
| High Zn-Trace-Mineral Salt | 1 |
| Bone Meal | 100.0 |

${ }^{\text {The }}$ vitamin and mineral supplement furnished the following quantities per kg of feed: Vitamin A, 2200 I.U.; Vitamin $\mathrm{D}_{3}$, 660 I.U.; Vitamin E, 11 I.U. In addition approximately $4.4^{3} \mathrm{~g}$ ASP (250) was added per kg feed.
and group feed intake was recorded at 140 and 175 days of age. The composition of ration for growing sheep is shown in table 3.

Slaughter Procedure

The neonatal lambs were slaughtered within 10 or 12 hr of birth. The rest of the lambs were fasted approximately 15 hr prior to slaughter. In order to avoid the effect of electrical stimulation, the animals were not immobilized prior to exsanguination. Bleeding was accomplished by severing the carotid artery and jugular vein. Following exsanguination the pelt was removed as rapidly as possible.

## Tissue Collection and Preparation

The desired samples were rapidly removed from the animals and weighed. These samples included the following: gastrocnemius (left leg) and longissimus muscles, perirenal, subcutaneous and intramuscular fats. The entire left longissimus muscle was removed and then freed of adhering surface fat before it was weighed. For fat, protein and moisture determinations, one third of the muscle from the lumbar region was saved (except for neonatal lambs the whole longissimus muscle was saved in order to obtain sufficient samples). Subcutaneous fat was removed from the dorsal thoracic and

TABLE 3. COMPOSITION OF THE RATION FOR GROWING SHEEP ${ }^{a}$

| Ingredients | Percentage of total |
| :--- | :---: |
| Dehydrated alfalfa (17\% protein) | 30.0 |
| Corn, grain | 32.5 |
| Oats, grain | 19.5 |
| Soybean Meal (50\% protein) | 12.5 |
| Molasses | 5.0 |
| High Zn-Trace-Mineral Salt | .5 |
|  | 100.0 |

$a_{\text {The }}$ vitamin and mineral supplement furnished the following quantity per kg of feed: Vitamin A, 5500 I.U.; Vitamin $D_{3}$, 687 I.U.; Vitamin K, 11 I.U.
lumbar regions of the carcass except at birth where the absence of external fat made it impossible to obtain subcutaneous fat samples. Intramuscular fat was obtained only from the lambs at 140 and 175 days of age. This fat was physically separated from the entire right longissimus muscle. The whole sample or subsamples were placed in polyethylene bags, frozen in a mixture of dry ice and 2-methylbutane, and stored at -85 C for subsequent analyses.

Powdering of Frozen Muscle and Fat Samples

The frozen muscle and fat samples (except for the intramuscular fat) were powdered in a -25 C room as described by Borchert and Briskey (1965). Chipped dry ice and shattered pieces of frozen muscle or fat were pulverized in a Waring Blendor jar for apporximately 30 to 60 sec . After sifting the samples, the coarse material which remained on the sieve was again placed in the blendor and the process repeated. After the second pulverization and sifting, the coarse material was discarded. The powdered samples were placed in polyethylene bags and were not sealed until 12 hr after filling to allow carbon dioxide sublimation. After sealing, the samples were stored at -85 C for later analyses.

Glyceride Synthetase Assay

A modification of the assay method of Bennink (1973) was used to determine glyceride synthethase.

Preparation of Crude Homogenate Preparation of the crude homogenate was carried out at 2 to 3 C . Depending on the adipose tissue, approximately 1 to 2 g were sliced with a razor blade and weighed while frozen. The sliced tissue was homogenized in three volumes of Tris-sucrose buffer (Appendix 1) in a Brinkmann Polytron (Model PCU-2-110, and saw-tooth model PT-10-ST) for 45 sec at setting, 5 ( $50 \%$ of full speed). The sample was further homogenized by three strokes in a Thomas teflon glass homogenizer. In order to separate cell debris and nuclei from crude homogenate, the hemogenate was filtered through glass wool (prewashed with Tris-sucrose buffer, pH 6.6 ).

Esterification In a preliminary study, the time, pH , and concentration of cofactors necessary for a maximum rate of glyceride synthesis were determined by varying the concentration of each cofactor while the other cofactors were held constant. The labeled precursor used in this assay was L-glycerol-14C(U)3-phosphate with a specific activity of approximately $20,000 \mathrm{dpm} / \mathrm{mole}$. The cofactor concentrations for the 3 fat depots as determined in the preliminary
studies were 1.75 mM ATP; $3.3 \mathrm{mM} \mathrm{Mg.Cl} 2 ; .1 \mathrm{mMCoA} ; 20 \mathrm{mg} /$ reaction tube BSA; 3.3 mM glycerol 3 -phosphate; 100 mM potassium phosphate buffer ( pH 6.6 ) and .67 mM mixture of fatty acids. In addition 15 mM glutathione (GSH) was used for subcutaneous and perirenal but, 7.5 mM GSH was used for intramuscular fat. The fatty acid mixture (Appendix 2) was solubilized and neutralized with KOH ( 1 ml of 2 M KOH per 100 ml of free fatty acid mixture) and sonicated for 1 min with a Bronson Sonifier (Model 350 ; duty cycle setting 2 ). The fatty acid mixture was immediately pipetted into 25 ml Erlenmeyer flasks which contained all cofactors. This mixture was sonicated as described above and .5 ml of the crude homogenate (enzyme) was added and the flasks were stoppered. The enzyme assay was conducted in a total volume of 3 ml at 37 C for 45 min with gentle shaking in a Eberbach Shaker Bath (Eberbach Corporation, Ann Arbor, Michigan). Duplicate flasks without ATP and CoA were run as blanks along with the samples.

Stopping the Reaction After the 45 min incubation period, the reaction was stopped by adding 8 ml of a solvent mixture consisting of isopropanol and heptane ( $1: 1 \mathrm{v} / \mathrm{v}$ ), and then shaken vigorously. Five ml of .03 M NaOH were added to the flasks and shaken to wash the solvent and the upper layer (heptane) allowed to separate from the lower aqueous layer.

Scintillation Counting A 2 ml aliquot of the heptane layer was transferred to scintillation vials containing 10 ml of scintillation cocktail (.5\% PPO in toluene). The
scintillation vials were counted in a Packard Tri-Carb Liquid Scintillation Spectrometer (Model 3310, Packard Instrument Company, Downers Grove, Illinois). Counting efficiencies were calculated by channels-ratio-method.

Protein Determinations Protein concentrations of the crude homogenates were determined by the Lowry method (Lowry et al., 1951). Bovine serum albumin was used as standard for these determinations.

Determination of Adipocyte Size and Number

Fixation Fresh samples of perirenal, subcutaneous and intramuscular fat were fixed in 5 ml of $3 \%$ osmium tetroxide (Appendix 3) in scintillation counting vials which contained 3 ml of 50 mM collidine buffer (Appendix 3). Fixation was allowed to proceed for 72 hr under a hood.

Filtration and Separation After fixation, adipocytes were filtered through two different pore sizes of Nitex nylon screens (Tetko Inc., Elmsford, N.Y.) with the smaller pore size ( $15 \mu \mathrm{~m}$ ) being placed on the bottom and the larger pore size (either 150 or $250 \mu \mathrm{~m}$ ) on top. The $150 \mu \mathrm{~m}$ pore size top screen was used for perirenal and subcutaneous adipocytes from the birth, 35 and 70 day age groups and also for the intramuscular fat cells from the 140 day age group. The top pore size screen for the remainder of the samples was $250 \mu \mathrm{~m}$. The pore size screens were chosen based on preliminary studies using microscopic and Coulter Counter observations.

The osmium fixed adipose tissue was transferred to the upper filtration screen and the fat cells were washed free of connective tissue with a stream of distilled water and gentle prodding with a blunt glass stirring rod. The released cells were collected on the lower $15 \mu \mathrm{~m}$ screen while the very small particles and cell fragments passed through. The fixed cells which remained on the bottom screen were transferred to a tared 250 ml beaker and weight of the suspension was brought to 240 g by adding $.9 \%$ sodium chloride. The suspended cells were then ready for sizing and counting on the Coulter Counter.

Counting and Sizing Adipocytes The procedure of Hirsch and Gallian (1968) was followed and counting was done on a Model B Coulter Counter. Calibration of the Coulter Counter was made with corn (large particle size) and pecan (small particle size) pollen. Appropriate settings for the Coulter Counter were determined by prior trials. Two aperture tubes were used. The aperture diameters used were $250 \mu \mathrm{~m}$ and 400 $\mu \mathrm{m}$ for samples that filtered through $150 \mu \mathrm{~m}$ and $250 \mu \mathrm{~m}$ top screens, respectively.

In a series of calculations (Appendix 4) the number and the volume of the cells per gram of adipose tissue for different selected size ranges were calculated.

Determination of RNA and DNA

A modification of the method Munro and Fleck (1969) was used to determine RNA and DNA in gastrocnemius muscle samples. Approximately .2 g of powdered muscle were weighed in duplicates in Corex test tubes and 2 ml of cold deionized water were added. The tubes were stoppered and vortexed. After adding 5 ml of cold $2.5 \%$ perchloric acid (PCA) (w/v) the tubes were stoppered, vortexed and placed in an ice bath for at least 10 min and then centrifuged at $34,800 \mathrm{x} \mathrm{g}$ for 15 min. The supernatant was discarded. The pellet was broken up with an applicator stick and 5 ml of cold $1 \%$ PCA were added. The tubes were stopped, vortexed and centrifuged at $34,800 \mathrm{x} \mathrm{g}$ for 15 min and the supernatant was discarded. The pellet was broken up and 4 ml .3 N potassium hydroxide were added and the tubes stoppered, vortexed and sealed with tape to prevent popping. The tubes were incubated at 37 C in a water bath for 2 hr . At the end of the incubation time, the tubes were vortexed and placed on ice for 5 min . Five ml of cold $5 \%$ PCA were added and the tubes stoppered, vortexed and placed on ice for 15 min . The tubes were centrifuged at $34,800 \times \mathrm{g}$ for 10 min . The supernatant was decanted into 25 ml graduated tubes and saved. The pellet was broken up and washed twice with 5 ml of $5 \%$ PCA each time followed by stoppering, vortexing and centrifuging at $34,800 \times \mathrm{g}$ for 10 min. The supernatant from each of these 2 centrifugations
was added to the 25 ml graduated tubes and the total volume was brought to 20 ml with $5 \%$ PCA and then mixed. This fraction contained RNA. The pellets were saved for DNA extraction.

For DNA extraction, the pellet remaining from the RNA isolation was broken up and 5 ml of cold $10 \%$ PCA were added and the tubes were vortexed and marbles were placed on the top of the tubes to act as condensers. The suspension was digested in a water bath at 70 C for 25 min . At the end of digestion, the tubes were placed on ice for 5 min , then centrifuged at $34,800 \times \mathrm{g}$ for 10 min . The supernatant was decanted into 15 ml graduated tubes and saved. The pellets were broken up and washed with 4.75 ml of $10 \%$ PCA, stoppered, vortexed and centrifuged at $34,800 \mathrm{x} \mathrm{g}$ for 10 min and the supernatant was added to the 15 ml graduated tubes and the total volume was brought to 10 ml with $10 \%$ PCA and mixed. This fraction contained the DNA.

For determination of RNA concentration, orcinol was utilized in a calorimetric procedure. Two ml of the RNA fraction were pipetted into 16 mm pyrex test tubes in duplicates, as well as a reagent blank using 2 ml of $5 \%$ (w/v) PCA instead of sample, and a set of duplicate test tubes containing RNA standards (Appendix 5) of $12.5,25.0,37.5$ and 50 mg RNA/ml was used. To all of the above tubes 2 ml of $1 \%$ (w/v) fresh orcinol reagent (Appendix 7) which were made up just prior to use, were added.

Marbles were placed on the top of the tubes to act as condensers and the tubes were placed in a boiling water bath for 30 min . After boiling, the tubes were cooled in running cold water for 5 min and allowed to reach room temperature and then read immediately at 680 nm on a Beckman Model 24 Spectrophotometer.

For determination of DNA concentration, diphenylamine and acetaldehyde were utilized in a colorimetric procedure. Two ml of the DNA fraction were pipetted into 16 mm pyrex test tubes in duplicates. In addition, a reagent blank using 2 ml of $10 \%$ PCA instead of sample and a set of duplicate test tubes containing DNA standards (Appendix 6) of 12.5 , $25.0,37.5$ and $50 \mathrm{mg} \mathrm{DNA} / \mathrm{ml}$ were used. To all of the above tubes 2 ml of $4 \%$ (w/v) diphenylamine in glacial acetic acid (Appendix 8) and .1 ml of acetaldehyde solution (Appendix 9) were added and vortexed. Marbles were placed on top of the tubes to act as condenser and the tubes were incubated overnight at 30 C in a water bath. After incubation, the tubes were cooled to room temperature and read at 595 nm on a Beckman Model 24 Spectrophotometer.

## Protein Fractionation

The protein fractionation procedure was a modificaiton of the method of Helander (1957). All fractionation procedures were carried out at 2 to 3 C with cold extraction solutions.

Sarcoplasmic Protein Five g of powdered frozen muscle were weighed in 250 ml polyethylene wide mouth centrifuge bottles equipped with screw caps. Fifty ml . 015 M potassium phosphate buffer (Appendix 10) were added to the bottles and extracted on a magnetic stirrer for 3 hr . After centrifugation at 1400 xg for 20 min , they were filtered through eight layers of cheese cloth into 100 ml graduated cylinders. The residue was re-suspended in 50 ml of potassium phosphate buffer and extracted on a magnetic stirrer for 3 hr . After extraction, they were centrifuged at 1400 xg for 20 min and filtered as described above. The volume of the combined supernatant was recorded. Duplicate 15 ml samples were used to determine the amount of sarcoplasmic protein nitrogen present in the sample by the Kjeldahl method. The residues were saved for myofibrillar protein nitrogen determination.

Non-Protein Nitrogen Fifteen ml duplicate aliquots of sarcoplasmic protein supernatants were pipetted into 50 ml polyethylene centrifuge tubes to which 5 ml of $10 \%$ (w/v) trichloroacetic acid (TCA) were added. The solution was allowed to stand for 2 to 4 hr , then centrifuged at 12,100 $\mathbf{x} g$ for 20 min . The supernatant was carefully decanted into Kjeldahl flasks for non-protein nitrogen determination by the micro-Kjeldahl method.

Myofibrillar Protein The residue from the sarcoplasmic protein extraction was suspended in 50 ml 1.1 M potassium iodide (KI) phosphate buffer (Appendix 10) and
extracted on a magnetic stirrer for 3 hr . After extraction, the bottles were centrifuged at $1400 \mathrm{x} g$ for 20 min and filtered through eight layers of cheesecloth into 100 ml graduated cylinders. The residue was resuspended in 50 ml of 1.1 M KI phosphate buffer, extracted for 3 hr , centrifuged at 1400 x g for 20 min and filtered as described above, and the combined volume of the supernatants was recorded. Duplicate 15 ml samples of the suspension were used to determine the amount of myofibrillar protein nitrogen in the sample by the micro-Kjeldahl method.

Total Nitrogen Total nitrogen was determined on approximately . 5 g of powdered muscle by the micro-Kjeldah1 method.

Stroma Protein Nitrogen Stromal protein nitrogen was calculated by subtracting the sum of sarcoplasmic, myofibrillar and non-protein nitrogen from the total nitrogen. Total nitrogen was expressed as milligrams per gram of fresh muscle tissue. All protein fraction nitrogen values were expressed as a percentage of total nitrogen.

Kjeldah1 Method

The American Instrument Company (1961) Micro-Kjeldah1 method was used for nitrogen determinations.

Moisture Determinations

Moisture and ether extract determinations were performed on powdered gastrocnemius and longissimus muscle samples, powdered perirenal and subcutaneous fat and on dissected intramuscular fat from the longissimus muscle. Approximately 1 to 5 g samples were weighed into previously dried aluminum dishes and dried in a 100C oven for 24 hr . Weight loss was recorded after cooling the samples in a desiccator and the moisture was calculated as percentage of fresh tissue (A.O.A.C., 1970). The dried samples were saved for the ether extract determinations.

Ether Extraction

The fat content was determined by extraction of the dried samples with anhydrous ether for 4 hr in a Goldfisch fat apparatus as outlined by A.O.A.C. (1970) and the data were expressed as percentage of fresh tissue. The fat content of adipose tissues was also expressed as grams per cell.

## Statistical Analysis

A factorial experiment was designed with two growth rates, two sexes and six ages. There were also three replicates per experimental unit. The main effects and their interactions were analyzed by the analysis of variance
method (Steel and Torrie, 1960).
When significant differences were observed between more than two means, Duncan's Multiple Range test (Duncan, 1955) was performed to determine which means were significantly different. In addition, linear correlation coefficients were calculated between pairs of dependant variables (Steel and Torrie, 1960). The statistical analysis was performed at the Michigan State University Computer Center.

## RESULTS AND DISCUSSION

Average Daily Gain, Feed Intake and Feed Conversion

Average daily gain, daily feed intake and feed conversion for the lambs are presented in table 4 . Since the lambs were group fed, individual feed intake could not be obtained. Therefore, average daily gain is the only feed lot characteristic that could be analyzed statistically. Since the Suffolk-sired lambs grew faster than Dorset-sired lambs in this study, they were catagorized as fast and slow growth rate groups, respectively. This reference to growth rate groups will be followed thoughout the remainder of results and discussions. Between 105 and 140 days, average daily gain of the fast growing rams and ewes and slow growing rams were similar and all were significantly ( $p<.05$ ) higher than the slow growing ewes. However, between 140 and 175 days only the fast growing rams gained more rapidly ( $\mathrm{p}<.05$ ) than the other groups. As would be expected, in both age periods (105 to 140 days and 140 to 175 days) feed intake tended to be related to body weight gain. Average feed intake of the lambs between 140 and 175 days was higher than between 105 and 140 days (1835 vs 1442 g/day/lamb, respectively). Feed
to gain ratio during both periods were similar for fast and slow growing lambs (table 4). Although rams and ewes had similar feed conversion between 105 and 140 days, ewes were superior to rams in feed conversion (5.52 vs 4.81 for rams and ewes, respectively) between 140 and 175 days of age. These data indicate that the fast growing lambs had higher feed to gain ratios than the slow growing rams.

TABLE 4. AVERAGE DAILY GAIN, FEED INTAKE AND FEED CONVERSION DATA OF THE EXPERIMENTAL LAMBS ${ }^{a}$

| Growth Rate |  |  |  | Growth Rate |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| t | 1n | , | in | st | win | low | win |
| Sex |  | Sex |  | Sex |  | Sex |  |
| Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe |

$$
105 \text { to } 140 \text { days } \quad 140 \text { to } 175 \text { days }
$$

| Number of lambs | 7 | 9 | 7 | 9 | 4 | 6 | 4 | 6 |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Average daily gain, g | $262^{\mathrm{b}}$ | $242^{\mathrm{b}}$ | $263^{\mathrm{b}}$ | $200^{\mathrm{c}}$ | $411^{\mathrm{b}}$ | $346^{\mathrm{c}}$ | $350^{\mathrm{c}}$ | $302^{\mathrm{c}}$ |
| Feed intake, g/day/lamb | 1592 | 1392 | 1519 | 1265 | 2324 | 1725 | 1888 | 1405 |
| Feed/gain | 6.08 | 5.79 | 5.77 | 6.32 | 5.65 | 4.98 | 5.39 | 4.65 |

${ }^{\text {a Means }}$ of the average daily gains for each period having the same superscripts are not significant (p>.05).

Adipose Tissue Growth

Adipose tissue accretion and its percentage of live body weight are presented in tables 5 and 6 and figure 5. The weights and the percentages of both subcutaneous and


Figure 5. Growth curves of perirenal, subcutaneous and intramuscular adipose tissues.
table 5. effects of growth rate, sex and age on weight and percentage of
PERIRENAL, SUBCUTANEOUS AND INTRAMUSCULAR ADIPOSE TISSUES

| Measurements | Growth rate ${ }^{\text {a }}$ |  | Sex ${ }^{\text {a }}$ |  |  | Pr. | Age(days) ${ }^{\text {b }}$ |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast growing | Slow growing | Pr. | Ram | Ewe |  | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Live weight (kg) | $26.43{ }^{\text {c }}$ | $22.82{ }^{\text {d }}$ | <. 001 | $126.62^{\text {C }}$ | $22.63{ }^{\text {d }}$ | <. 001 | $3.95{ }^{\text {c }}$ | $13.04{ }^{\text {d }}$ | $20.24{ }^{\text {e }}$ | 28.09 f | 36.258 | 46.19 | <.001 |
| Adipose tissue weight(g): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 216 | 208 | . 71 | 194 | 230 | . 09 | $18^{\text {c }}$ | $73^{\text {cd }}$ | $148{ }^{\text {de }}$ | $117^{\text {e }}$ | 327 f | 5908 | <. 001 |
| Subcutaneous | 901 | 935 | . 74 | 1023 | 814 | . 05 | - | $127^{\text {c }}$ | $296{ }^{\text {c }}$ | $430^{\text {c }}$ | $1313{ }^{\text {d }}$ | $2426{ }^{\text {e }}$ | < 2001 |
| Intramuscular | 8.26 | 9.17 | . 42 | 9.23 | 8.19 | . 35 | $.27{ }^{\text {c }}$ | $2.59{ }^{\text {cd }}$ | $5.96{ }^{\text {d }}$ | 5.99 d | $13.64{ }^{\text {e }}$ | $23.82{ }^{\text {f }}$ | <. 001 |
| Adipose tissue percentage: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | . 71 | . 74 | . 52 | $.61{ }^{\text {c }}$ | . $84{ }^{\text {d }}$ | <. 001 | .45 ${ }^{\text {c }}$ | . $54{ }^{\text {C }}$ | . 77 d | . $41{ }^{\text {c }}$ | . $88{ }^{\text {d }}$ | $1.29{ }^{\text {e }}$ | <. 001 |
| Subcutaneous | 2.33 | 2.75 | . 06 | 2.55 | 2.52 | . 88 | - | . $97{ }^{\text {c }}$ | $1.46{ }^{\text {c }}$ | $1.50{ }^{\text {c }}$ | $3.57{ }^{\text {d }}$ | $5.20{ }^{\text {e }}$ | <. 001 |
| Intramuscular | . 026 | . 030 | . 16 | . 029 | . 028 | . 78 | . $007{ }^{\text {c }}$ | . $023{ }^{\text {d }}$ | .029 ${ }^{\text {d }}$ | .022 ${ }^{\text {d }}$ | . $037{ }^{\text {e }}$ | . $052{ }^{\text {f }}$ | <. 001 |

a Mean of 36 lambs for perirenal or subcutaneous and 12 lambs for intramuscular adipose tissues.
$\mathrm{b}_{\text {Means }}$ are the average of 12 lambs.
cdef $g_{\text {Means }}$ within each main effect on the same row bearing the same superscripts are not statistically significant (p> . 05) .
Pr. $=$ Probability for level of significance.
TABLE 6. INTERRELATIONSHIP OF GROWTH RATE, AGE AND SEX ON WEIGHT AND PERCENTAGE
OF PERIRENAL, SUBCUTANEOUS AND INTRAMUSCULAR ADIPOSE TISSUESa

| Meausurements | Sex | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  |  | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Live weight(kg) | Ram | 3.88 | 14,81 | 24.72 | 32.00 | 43.24 | 53.24 | 4.46 | 11.94 | 19.81 | 27.42 | 38.10 | 45.66 |
|  | Ewe | 4.11 | 13.15 | 19.09 | 29.78 | 34.77 | 44.30 | 3.32 | 12.24 | 17.39 | 23.17 | 28.88 | 41.43 |
| Adipose tissue weight(g): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 18 | 84 | 95 | 99 | 359 | 502 | 19 | 44 | 144 | 107 | 392 | 467 |
|  | Ewe | 21 | 94 | 164 | 145 | 319 | 691 | 13 | 69 | 191 | 120 | 236 | 699 |
| Subcutaneous | Ram | - | 143 | 239 | 355 | 1533 | 2820 | - | 73 | 312 | 382 | 1674 | 2644 |
|  | Ewe | - | 156 | 268 | 529 | 1018 | 1897 | - | 137 | 310 | 452 | 1028 | 2343 |
| Intramuscular | Ram | . 27 | 3.04 | 8.47 | 6.34 | 18.72 | 18.39 | . 35 | 2.19 | 6.16 | 5.69 | 12.20 | 28.97 |
|  | Ewe | . 26 | 2.86 | 4.20 | 6.35 | 11.06 | 19.18 | . 24 | 2.29 | 5.01 | 5.59 | 12.56 | 28.67 |
| Adipose tissue percentage: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | . 44 | . 54 | . 39 | . 30 | . 82 | . 94 | . 43 | . 36 | . 70 | . 36 | 1.01 | . 99 |
|  | Ewe | . 52 | . 70 | . 89 | . 46 | . 92 | 1.57 | . 41 | . 56 | 1.30 | . 50 | . 80 | 1.69 |
| Subcutaneous | Ram | - | 1.01 | 1.15 | 1.07 | 3.50 | 5.25 | - | . 59 | 1.54 | 1.40 | 4.40 | 5.66 |
|  | Ewe | - | 1.18 | 1.39 | 1.69 | 2.90 | 4.24 |  | 1.11 | 1.76 | 1.89 | 3.48 | 5.65 |
| Intramuscular | Ram | . 006 | . 035 | . 035 | . 019 | . 042 | . 034 | . 008 | . 018 | . 031 | . 024 | . 033 | . 062 |
|  | Ewe | . 007 | . 022 | . 022 | . 021 | . 032 | . 043 | . 007 | . 018 | . 029 | . 024 | . 043 | . 070 |

${ }^{\text {a Means }}$ are the average of 3 lambs.
perirenal adipose tissues increased with age (p<.01, table 5). The differences in weight between perirenal and subcutaneous adipose tissues were lowest at the young ages, but as age and body weight increased, the differences between these fat depots became progressively greater, with the final subcutaneous fat being approximately four-fold greater than perirenal fat (figure 5). No dissectable subcutaneous fat was present in the lambs at birth. However, between 35 and 175 days of age, subcutaneous fat increased approximately 18 fold; whereas perirenal fat only showed a seven fold increase during this same period. Intramuscular adipose tissue (obtained from left longissimus muscle) showed the greatest increase ( 87 fold) between birth and 175 days. However, only at 140 and 175 days of age was sufficient adipose tissue present so it could physically be dissected from the muscle. Johnson et al. (1972) found preferential sites of fat deposition during growth of calves from 210 days of gestation to 1,200 days postnatally with the predominance of fat deposition occurring in the following order: intermuscular>subcutaneous>intramuscular>perirenal>pelvic fat. In their study, the 140 to 152 g of fat dissected from fetal calves was quite evenly distributed between intermuscular and perirenal fat. The most notable observation in their study was the complete lack of subcutaneous fat which agrees with the findings of the present study. There was a high correlation ( $p<.01$ ) between live
weight and perirenal fat weight ( $\mathrm{r}=.82$ ) as well as with subcutaneous fat weight ( $r=.89$ ). Several investigators including Barton and Kirton (1958) and Kirton and Barton (1962) have also observed a high positive correlation between weight of lambs and amount of carcass fat.

The pattern of perirenal and subcutaneous adipose tissues weight and percentage increases in relationship to the live weight were similar (figure 5). However, between 70 and 105 days (when body weight increased from 20.2 to 28.1 kg), there was a decrease in perirenal fat weight from 148 to 117 g , while subcutaneous fat increased from 296 to 430 g . Nevertheless, neither of these changes was significant (p>.05). These nonsignificant changes in perirenal and subcutaneous fat are probably due to the effect of the stresses associated with weaning. The data (table 5) suggests that perirenal fat is more sensitive to these stresses than subcutaneous fat.

Growth rate of the lambs did not significantly (p>.05) affect weight of perirenal and subcutaneous fat. However, the percentage of subcutaneous fat was higher in slow growing than fast growing lambs ( $\mathrm{P}=.06$ ). Makarechian et al. (1978) found that when breed of sire influenced growth rate, carcass composition of the progeny was not necessarily affected. They observed that Dorset-sired lambs grew slower, had less bone and more fat than Suffolk-sired lambs. These data agree with the results of the present study. However, their
results were based on carcass weight rather than live weight, as in the present experiment. They also reported a high dressing percentage for Dorset-sired lambs, which would be expected because of the greater amount of fat. Lambuth et al. (1970) reported that fast gaining lambs had no significant increase in total retail yield or edible portion, but had lower percentages of total fat trim and higher percentages of total bone than slow growing lambs.

Sex affected the perirenal and subcutaneous adipose tissue weights differently (table 5). Rams had more ( $\mathrm{P}=.05$ ) subcutaneous, but less ( $\mathrm{P}=.09$ ) perirenal fat than ewes. On a percentage basis, perirenal fat of ewes was significantly ( $\mathrm{P}<.01$ ) higher than rams. This is in agreement with the results reported by others (Shelton and Carpenter, 1972; Kemp et al., 1976). In general, female cattle and sheep fatten at lighter live weights than castrated males, whereas castrated males fatten at lighter weights than intact males (Bradley et al., 1963; Prescott and Lamming, 1964; Wilson et al., 1969). Berg and Butterfield (1976) reported that fat had the greatest effect on carcass composition between sexes.

Weight and percentage of perirenal fat were affected by the interaction between age and sex ( $\mathrm{P}<.05$, Appendix 13). No other interactions were significant ( $P>.05$ ).

Chemical Composition of Adipose Tissue

Proximate analysis of the fat depots are shown in tables 7 and 8. Except for percentage protein of intramuscular fat, age had a significant ( $\mathrm{P}<.01$ ) effect on the percentage lipid, protein and moisture of the three fat depots (table 7). During the 175 days of the experiment, percentage lipid in the perirenal and subcutaneous adipose tissues increased 1.7 and 22 fold, respectively (figure 6). The reason for this large difference in lipid deposition during postnatal growth is due to the difference in the fat content of the two adipose tissues already present at birth. Perirenal adipose tissue had accumulated $34 \%$ lipid prenatally, while subcutaneous adipose tissue had only $3.5 \%$ at birth. The correlation between percentage lipid and adipose tissue weight was higher in perirenal ( $\mathrm{r}=.82, \mathrm{P}<.01$ ) than that in subcutaneous ( $\mathrm{r}=.53, \mathrm{P}<.01$ ) or in intramuscular ( $\mathrm{r}=.38, \mathrm{P}>.05$ ). Body weight was also significantly ( $\mathrm{P}<.01$ ) correlated with percentage lipid in perirenal ( $\mathrm{r}=.72$ ) and subcutaneous ( $\mathrm{r}=.76$ ) fat but not intramuscular ( $\mathrm{r}=.38, \mathrm{P}>.05$ ) fat. The pattern of increase in percentage lipid of perirenal and subcutaneous fat was highly correlated ( $\mathrm{r}=.96, \mathrm{P}<.01$ ). Both of these fat depots were affected by the stresses associated with weaning (figure 6). The decrease in fat deposition immediately following weaning may be due to caloric reduction that resulted from reduced feed intake during the first


Figure 6. Percentage lipid in the three fat deoots as affected by age.
TABLE 7. EFFECTS OF GROWTH RATE, SEX AND AGE ON CHEMICAL COMPOSITION OF

| Measurements | Growth rate ${ }^{\text {a }}$ |  | Pr. | Sex ${ }^{\text {a }}$ |  | Pr. | Age(days) ${ }^{\text {b }}$ |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | growing | growing |  | Ram | Ewe |  | 0 | 35 | 70 | 105 | 140 | 175 | Pr. |
| Percentage lipid: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 76.5 | 76.6 | . 92 | $74.9{ }^{\text {c }}$ | $78.1{ }^{\text {d }}$ | <. 001 | $34.0{ }^{\text {c }}$ | $79.6{ }^{\text {d }}$ | $85.2{ }^{\text {ef }}$ | $81.6^{\text {de }}$ | $87.5^{\text {fg }}$ | $91.2{ }^{\text {g }}$ | <. 001 |
| Subcutaneous | 61.6 | 59.6 | . 32 | 60.2 | 61.0 | . 69 | $3.5{ }^{\text {c }}$ | $59.0^{\text {d }}$ | $74.1{ }^{\text {ef }}$ | $69.2{ }^{\text {e }}$ | $77.1{ }^{\text {f }}$ | $80.7{ }^{\text {f }}$ | <. 001 |
| Intramuscular | 53.2 | 54.0 | . 86 | 51.4 | 55.8 | . 33 | - | - | - | - | $48.1{ }^{\text {c }}$ | $59.1{ }^{\text {d }}$ | . 02 |
| Percentage Protein: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 3.56 | 3.62 | . 71 | 3.72 | 3.46 | . 14 | $11.17^{\text {c }}$ | $2.47{ }^{\text {d }}$ | $1.80{ }^{\text {e }}$ | $2.66{ }^{\text {d }}$ | $1.81{ }^{\text {e }}$ | $1.61{ }^{\text {e }}$ | <. 001 |
| Subcutaneous | 6.36 | 6.68 | . 48 | 6.39 | 6.65 | . 59 | $17.58{ }^{\text {c }}$ | $5.90{ }^{\text {d }}$ | $4.34{ }^{\text {de }}$ | $4.75{ }^{\text {de }}$ | $3.52^{\text {e }}$ | $3.40{ }^{\text {e }}$ | <. 001 |
| Intramuscular | 17.33 | 16.41 | . 30 | 15.70 | 18.04 | . 26 | - | - | - | - | 16.62 | 18.43 | . 31 |
| Percentage moisture: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 19.7 | 19.8 | . 95 | 21.2 | 18.2 | <. 001 | $53.1{ }^{\text {c }}$ | $18.1{ }^{\text {d }}$ | $12.6{ }^{\text {e }}$ | $16.2^{\text {d }}$ | $11.2{ }^{\text {e }}$ | $7.2{ }^{\text {f }}$ | <. 001 |
| Subcutaneous | 32.3 | 24.1 | . 26 | 33.8 | 32.7 | . 53 | $80.0^{\text {c }}$ | $35.2{ }^{\text {d }}$ | $21.7^{\text {ef }}$ | $26.6{ }^{\text {e }}$ | 19.9 fg | $15.9{ }^{\text {g }}$ | <. 001 |
| Intramuscular | 40.4 | 37.6 | . 40 | 41.2 | 36.8 | . 19 | - | - | - | - | $44.9{ }^{\text {c }}$ | $33.1{ }^{\text {d }}$ | <. 001 |

${ }^{\text {a Mean of }} 36$ lambs for perirenal or subcutaneous and 12 lambs for intramuscular adipose tissue.
$\mathrm{b}_{\text {Mean of }} 12$ lambs.
significant ( $\mathrm{p}>.05$ ).
Pr=Probability for level of significance.
74
TABLE 8. INTERRELATIONSHIP OF GROWTH RATE, AGE AND SEX ON CHEMICAL COMPOSITION OF PERIRENAL, SUBCUTANEOUS AND INTRAMUSCULAR ADIPOSE TISSUES ${ }^{a}$

| Measurement | Sex | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  |  | Age (days) |  |  |  |  |  | Age (days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Percentage lipid: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 30.0 | 81.1 | 80.7 | 76.9 | 87.2 | 89.1 | 32.5 | 74.2 | 85.7 | 82.6 | 88.6 | 90.3 |
|  | Ewe | 36.9 | 85.4 | 88.0 | 82.3 | 86.8 | 93.1 | 36.8 | 77.7 | 86.3 | 84.7 | 87.2 | 92.1 |
| Subcutaneous | Ram | 3.7 | 69.3 | 71.9 | 66.9 | 78.9 | 79.1 | 3.9 | 52.3 | 71.9 | 66.8 | 77.4 | 80.7 |
|  | Ewe | 1.6 | 66.0 | 78.9 | 69.0 | 76.9 | 76.7 | 5.0 | 48.3 | 73.7 | 74.3 | 75.0 | 86.3 |
| Intramuscular | Ram | - | - | - | - | 53.4 | 54.3 | - | - | - | - | 39.2 | 58.8 |
|  | Ewe | - | - | - | - | 45.7 | 59.6 | - | - | - | - | 54.2 | 63.9 |
| Percentage protein: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 11.68 | 2.47 | 2.03 | 3.17 | 1.70 | 1.63 | 11.02 | 2.90 | 1.91 | 2.54 | 1.74 | 1.90 |
|  | Ewe | 10.29 | 2.18 | 1.60 | 2.37 | 2.08 | 1.48 | 11.67 | 2.36 | 1.68 | 2.58 | 1.74 | 1.44 |
| Subcutaneous | Ram | 18.98 | 4.23 | 4.64 | 4.72 | 3.19 | 3.70 | 14.92 | 5.86 | 4.57 | 5.21 | 3.34 | 3.36 |
|  | Ewe | 16.83 | 4.72 | 3.70 | 4.65 | 3.16 | 3.80 | 19.59 | 7.27 | 4.46 | 4.41 | 4.39 | 2.77 |
| Intramuscular | Ram | - | - | - | - | 16.45 | 16.02 | - | - | - | - | 14.70 | 15.61 |
|  | Ewe | - | - | - | - | 17.79 | 18.87 | - | - | - | - | 17.34 | 17.98 |
| Percentage moisture: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 57.0 | 17.1 | 15.7 | 20.6 | 11.28 | 8.6 | 55.2 | 23.0 | 12.8 | 15.4 | 10.00 | 8.0 |
|  | Ewe | 49.7 | 13.9 | 10.2 | 15.5 | 11.2 | 5.6 | 50.4 | 18.4 | 11.5 | 13.3 | 12.2 | 6.6 |
| Subcutaneous | Ram | 78.8 | 26.5 | 23.6 | 28.2 | 16.6 | 17.7 | 82.4 | 42.0 | 23.8 | 28.7 | 19.7 | 15.8 |
|  | Ewe | 81.0 | 29.2 | 18.0 | 27.0 | 21.0 | 18.7 | 77.9 | 43.2 | 21.3 | 22.4 | 21.3 | 11.5 |
| Intramuscular | Ram | - | - | - | - | 41.7 | 37.6 | - | - | - | - | 48.4 | 36.9 |
|  | Ewe | - | - | - | - | 48.9 | 33.3 | - | - | - | - | 40.5 | 24.5 |

${ }^{a_{\text {Means }}}$ are the average of 3 lambs.
two weeks after weaning. The results of weaning stresses probably caused mobilization of fatty acids from the fat depots. Leat (1976) observed that the fat depots of fasted animals showed a reduction of $32 \%$ in clearing-factor lipase activity, a $99 \%$ depression in fatty acid synthesis and increased lipolysis. Mayerle and Havel (1969) also found that the blood flow rate and the loss of triglycerides was significantly increase in adipose tissues of fasted animals.

The percentage lipid in perirenal fat of ewes was higher than that of rams ( $\mathrm{P}<.01$, table 7 ). This was observed in most experimental periods. These data indicate that the perirenal fat of ewe lambs were more mature at most ages than that of rams. Because percentage lipid and moisture in perirenal adipose tissue were highly negatively correlated ( $\mathrm{r}=-.99, \mathrm{P}<.01$ ), the effect of sex resulted in a lower ( $\mathrm{P}<.01$ ) percentage moisture in perirenal adipose tissue of ewes than those of rams. Percentage lipid, moisture and protein was not influenced ( $\mathrm{P}>.05$ ) by sex for either subcutaneous or intramuscular adipose tissues (table 7). Also, percentage protein in perirenal fat was not affected by sex ( $P>.05$ ). Chemical composition of fast and slow growing lambs were similar ( $\mathrm{P}>.05$ ). There was a significant ( $\mathrm{P}<.05$ ) interaction between age and growth rate on the percentage lipid and moisture of the subcutaneous adipose tissue (Appendix 12). No other significant interaction was observed ( $\mathrm{P}>.05$ ) .

Measurements of Glyceride Synthetase Activity

Conditions for Optimum Glyceride Synthesis

A study to determine the assay conditions for optimum glyceride synthesis was conducted on sheep perirenal, subcutaneous and intramuscular adipose tissue. The conditions studied included pH of the assay medium, concentrations of ATP, CoA, glycerol 3-phosphate, $\mathrm{MgCl}_{2}$, glutathione, fatty acid mixture, length of the assay in minutes and levels of BSA and the adipose tissue homogenates. The results of these experiments are presented and discussed below.

The optimum pH for glyceride synthesis of the three fat depots was similar (figure 7). Maximum rates of glyceride synthesis occured at pH 6.6. This value is similar to the results reported by Bennink (1973) for rabbit mammary gland. An optimum pH of 7.0 has been reported for adipose tissue glyceride synthesis in the bovine (Benson, 1969) and rats (Steinberg et al., 1961; Daniel and Rubinstein, 1968). However, optimum pH values above 7.0 have been reported in mammary gland tissue of several different species (Askew et al., 1971; Bickerstaffe and Annison, 1971; Bennink, 1973; Gross and Kinsella, 1973).

Triglyceride formation was highly dependent upon the presence of ATP (figure 8). This finding agrees with the data reported for bovine adipose tissue homogenates (Benson,


Figure 7. Glyceride synthesis as a function of pH .


Figure 8. Glyceride synthesis as a function of ATP concentrations.
1969) and those for rats (Steinberg et al., 1961; Angel and Roncari, 1967). Glyceride synthesis without added ATP was approximately $5 \%$ of that for the optimum concentration (1.75 mM ). Concentrations of ATP greater than 2.5 mM inhibited esterification. The results confirm other work performed on bovine (Benson, 1969) and rat (Angel and Roncari, 1967) adipose tissue. This inhibition of higher concentrations of ATP can be partially reversed by increasing $\mathrm{Mg}^{2+}$ concentration (McBride and Korn, 1963). The concentration of ATP needed per milliliter of homogenate for triglyceride formation in sheep (present study) and bovine adipose tissue (Benson, 1969) seems to be considerably lower than that of mammary tissue (Askew et al., 1971). This observation may be due to the high requirement of ATP for triglyceride formation in mammary glands compared to that in adipose tissue because of the greater quantity of fat synthesized per unit of time.

There is an absolute requirement for CoA in the triglyceride formation (figure 9). The essentiality of CoA (and ATP) confirms the fact that triglyceride synthesis in sheep adipose tissue occurs via the $\alpha$-glycerol phosphate pathway since the initial reaction of this pathway requires ATP and CoA for its activity. The assay system is very sensitive to the added ATP and CoA (figures 8 and 9). Similar results have been found in bovine adipose tissues (Benson, 1969), rats (Steinberg et al., 1961) and also in mammary


Figure 9. Glyceride synthesis as a function of Coenzyme A concentration.
glands of cows (Askew et al., 1971; Bennink, 1973) and rabbits (Bennink, 1973).

In the absence of $\alpha$-glycerol phosphate there is no triglyceride formation (figure 10). This could be expected, becasue $\alpha$-glycerol phosphate is probably the main fatty acid acceptor in adipose tissue. However, Steinberg et al. (1961) showed that even in the absence of $\alpha$-glycerol phosphate there is a low level of incorporation. This may be due to the presence of endogenous $\alpha$-glycerol phosphate in the adipose tissue homogenate. Steinberg (1962) reported that the requirement for $\alpha$-glycerol phosphate in adipose tissue could be replaced by ADP + NADH but not by glycerol or monoolein. These results demonstrate biosynthesis of triglycerides in adipose tissue goes through the glycerol phosphate pathway and that the monoglyceride pathway is not important. This inability of adipose tissue to utilize glycerol can be explained by the fact that glycerol kinase is essentially absent in adipose tissue (Margolis and Vaughan, 1962).

When the mixture of fatty acids as described in materials and methods was added to the assay medium, there was a sharp increase in $\alpha$-glycerol phosphate incorporated into triglycerides (figure 11). In the absence of fatty acids, there was no incorporation of a-glycerol phosphate into triglycerides in the intramuscular fat, but in perirenal and subcutaneous fat there was approximately $20 \%$ incorporation (figure 11). This observation may be explained by the fact


Figure 10. Glyceride synthesis as a function of $\alpha$-glycerol 3-phosphate concentration.


Figure 11. Glyceride svnthesis as a function of fatty acids concentration.

1
$a$
p?
1
fa
ex
th
tr
po

BS
Ho
ti
$2 n$
Sut
$s t$
$h_{a}$
$s t_{t}$
that perirenal and subcutaneous fat depots are early developing adipose tissues compared to intramuscular fat, and they may have more endogeneous fatty acids than intramuscular fat. In addition, intramuscular fat is more sensitive to small increments of fatty acid concentrations (figure 11). Due to the presence of endogeneous fatty acids, Bennink (1973) concluded that acyltransferase activities must be measured with labeled glycerol 3-phosphate rather than labeled exogeneous fatty acids if true acylating capacities are to be determined. The use of labeled a-glycerol 3-phosphate has also been emphasized by Davidson and Stanacev (1972). Despite different sensitivities of intramuscular fat versus perirenal and subcutaneous fat to the addition of exogeneous fatty acid concentrations, the optimum level for the three fat depots was the same (. 67 mM ). Higher concentrations of fatty acids inhibited a-glycerol phosphate incorporation into triglycerides.

The response of the enzyme system to the addition of BSA was different in the 3 adipose tissues (figure 12). However, they had the same concentrations for optimum conditions of the enzyme assay. High levels ( $>20 \mathrm{mg}$ ) of BSA had an inhibitory effect on glyceride synthesis of adipose tissue homogenates. This might be due to enhancement of substrate emulsification. Similar results of BSA inhibition have been reported in other experiments (Daniel and Rubinstein, 1968; Benson, 1969).


Figure 12. Glyceride synthesis as a function of BSA level.

The amount of esterification was increased when $\mathrm{MgCl}_{2}$ was added to the assay medium (figure 13). However, even in the absence of $\mathrm{MgCl}_{2}$ there was approximately $50 \%$ к-glycerol phosphate incorporation, which suggests that endogenous $\mathrm{Mg}^{2+}$ was present. There was a rather broad plateau after the optimal concentration ( 3.3 mM ) of $\mathrm{Mg}^{2+}$. Endogenous $\mathrm{Mg}^{2+}$ has been reported to be higher in liver (Benson, 1969) than in adipose tissue.

The concentration of glutathione (GSH) needed for optimum enzyme assay conditions in the intramuscular fat was different than perirenal and subcutaneous depots (figure 14). High concentrations of GSH inhibited triglyceride synthesis as shown in figure 14. The function of GSH in the process of esterification is to increase activity of the enzyme system by protecting susceptible thiol groups of CoA from oxidation and it also protects lipids from autooxidation. Glutathione can be reversibly oxidized by the loss of two hydrogens, which results in formation of a disulfide bond. The latter functions as a hydrogen donor in oxidation-reduction reactions.

The amount of $\alpha$-glycerol phosphate esterifed increased linearly from 0 to 45 min of incubation time followed by a plateau from 45 to 60 min (figure 15).

The relationship between enzyme source (adipose tissue homogenate) and glyceride synthesis was linear between 0 and 1.0 ml of perirenal and subcutaneous homogenate, but between 0 and .6 ml for the intramuscular homogenate (figure 16).


Figure 13. Flvceride synthesis as a function of $\mathrm{MgCl}_{2}$ concentration.

m M Glutathione
Figure 14. Glyceride synthesis as a function of glutathione concentration.


Figure 15. Glvceride synthesis as a functic. $\begin{aligned} & \text { of }\end{aligned}$ time.

Homogenafe(ml)
synthesis as a function of homogenate volume.

Figure 16. Glvceride


In the present experiment .5 ml of crude adipose tissue homogenate was used per reaction vial for all depots.

The optimum assay conditions observed in these experiments were presented in the materials and methods section. Glyceride Synthetase Activity

Changes in glyceride synthesis activity are shown in tables 9 and 10. The pattern of enzyme activity changes with age, depends on the method of expressing the activities. When expressed per milligram protein, enzyme activities increased ( P <.05) from birth to 35 days in both perirenal and subcutaneous adipose tissues (table 9, figure 17). The increase in enzyme activity was much greater in subcutaneous fat as compared to perirenal fat (. 77 fold vs 113 fold increase). The enzyme activity in perirenal adipose tissue plateaued at age 35 days and remained essentially unchanged thereafter. However, there was a significant ( P <.05) increase in the activity between 70 and 105 days for subcutaneous adipose tissue which was then followed by a plateau through 175 days of age. The effect of age on the enzyme activity of intramuscular adipose tissue was nonsignificant ( P >.05). Even at the last period of the experiment (175 days) the enzyme activity of intramuscular adipose tissue was negligible. This fact necessitates the lenghtening of experimental periods if the pattern of enzyme activity in this adipose tissue is of interest.


Figure 17. . Glyceride synthetase activity of perirenal, subcutaneous and intramuscular adinose tissues expressed on a soluble protein basis.
table 9. effects of growth rate, sex and age on glyceride synthetase activity of perirenal, subcutaneous and intramuscular adipose tissues

|  |  |
| :---: | :---: |
| Noteres | 20, |
| Adipose tissue: Perirenal Subcutaneous |  |
|  |  |

- 

$b_{\text {Means }}$ are the average of 12 lambs.
cdef Means within each main effect on the same row bearing the same superscripts are not statistically significant(p).05).
Pr=Probability for level of significance.


| Measurements | Sex | GRQWTH_BATE |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  |  | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Nmoles substrate utilized per minute per: mg Protein: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 1.20 | 3.33 | 2.90 | 2.91 | 2.66 | 2.87 | 1.73 | 2.99 | 2.38 | 3.19 | 3.05 | 1.89 |
|  | Ewe | 1.48 | 2.87 | 3.89 | 2.39 | 3.44 | 2.70 | 1.60 | 1.43 | 1.81 | 2.68 | 2.62 | 3.08 |
| Subcutaneous | Ram | . 02 | 1.59 | 1.48 | 1.34 | 1.68 | 1.99 | . 01 | . 59 | . 61 | 1.67 | 1.75 | 1.28 |
|  | Ewe | . 01 | 1.52 | 1.37 | 1.40 | 1.32 | 1.36 | . 01 | . 80 | . 98 | 1.30 | 1.53 | 1.66 |
| Intramuscular | Ram | - | - | - | - | . 014 | . 015 | - | - | - | - | . 017 | . 004 |
|  | Ewe | - | - | - | - | . 004 | . 005 | - | - | - | - | . 010 | . 008 |
| g Adipose tissues: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 65.3 | 29.3 | 29.7 | 34.0 | 22.0 | 27.3 | 77.0 | 41.0 | 19.0 | 31.0 | 23.7 | 17.0 |
|  | Ewe | 67.3 | 24.7 | 26.3 | 22.3 | 29.0 | 16.3 | 85.3 | 15.3 | 13.3 | 26.7 | 21.3 | 18.3 |
| Subcutaneous | Ram | . 20 | 11.50 | 11.27 | 11.37 | 11.70 | 21.33 | . 11 | 5.57 | 4.30 | 17.67 | 13.13 | 10.97 |
|  | Ewe | . 05 | 13.60 | 8.40 | 11.40 | 12.60 | 10.07 | . 09 | 5.73 | 7.60 | 12.70 | 12.73 | 13.93 |
| Intramuscular | Ram | - | - | - | - | . 014 | . 015 | - | - | - | - | . 017 | . 004 |
| $10^{7}$ Adipocytes: 00.000 |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram Ewe | 14.3 17.4 | 19.3 | 19.6 20.3 | 54.9 21.5 | 51.0 54.0 | 71.2 73.4 | 18.3 21.43 | 20.1 8.8 | 13.3 | 33.2 | 31.6 | 80.8 |
| Subcutaneous | Ram | - | 6.9 | 5.9 | 19.2 | 33.3 | 75.0 | - | 3.8 | 2.8 | 17.8 | 45.0 | 34.1 |
|  | Ewe | - | 9.8 | 6.1 | 17.3 | 27.4 | 38.5 | - | 2.6 | 4.6 | 25.6 | 32.1 | 48.2 |
| Intramuscular | Ram | - | - | - | - | . 180 | . 527 | - | - | - | - | . 413 | . 113 |
|  | Ewe | - | - | - | - | . 110 | . 180 | - | - | - | - | . 127 | . 143 |



Glyceride synthesizing activity per milligram protein wes significantly correlated with fat content per adipocyte EOr the perirenal $(r=.26, P<.05)$ and subcutaneous fat ( $r=.39$, $P<.01$ ) and with percentage extractable fat for the perirenal $\mathcal{L}=.58, \mathrm{P}<.01$ ) and subcutaneous ( $\mathrm{r}=.82, \mathrm{P}<.01$ ) depots. None o F the above correlations was significant for intramuscular $E \Rightarrow t(P>.05)$. During all periods of the experiment, enzyme $a \subset$ tivities were higher for perirenal than for subcutaneous Eat (irrespective of how the activities were expressed). This difference was especially marked at birth because the enzyme activity (per milligram protein) for perirenal adipose tis sue was approximately $57 \%$ of its adults value while that EOT subcutaneous adipose tissue was $.6 \%$ of adult value. In other words, the postnatal increase in enzyme activity per mi 1 ligram protein was greater for the subcutaneous depot tham for perirenal adipose tissue. The accumulation of fat was also much greater for subcutaneous depot than for periren al fat during the 175 days of the experiment.

In both perirenal and subcutaneous fat, but not intramus Cular adipose tissue, fast growing lambs had higher $(\mathbf{P}<.01)$ enzyme activities (per milligram protein) compared to the slow growing group (table 9).

Merkel et al. (unpublished data) concluded that subcutaneous adipose tissue from Southdown lambs (higher propenSity to fatten) synthesized more glycerides than Suffolk (lower propensity to fatten) sired lambs. Effect of sex on
enzyme activity per milligram protein was not significant (P>.05). However, Merkel et al. (unpublished data) observed h표 gher glyceride enzyme activities (per milligram protein) E®r ewes than for wethers or rams.

Results of interaction between growth rate and sex are $p$ Fesented in Appendix 11. Only in perirenal adipose tissue $\omega \equiv s$ the interaction between growth rate and sex significant $(\mathbb{P}<.05)$. In this adipose tissue, the fast and slow growing ewe lambs had the highest and lowest average values for glyceride synthesizing activities (per milligram protein) (2.79 VS 2.20 respectively). There was a significant interaction ( $\mathbf{P}<.01$ ) between growth rate and age in both the perirenal anc subcutaneous adipose tissues (Appendix 12). In both of these adipose tissues the fast growing lambs had higher en < yme activities at most ages compared to slow growing lambs. G1 $工$ ceride synthesizing activity per milligram protein in pe I- irenal adipose tissue was also affected by the interaction between sex and age of the lambs (Appendix 13).

When the enzyme data are expressed per gram of adipose tissue, a different pattern was observed (table 9 and 10 , fiesure 18). The enzyme activity (per gram tissue) decreased dre amatically (64\%) in perirenal adipose tissue between birth and 35 days (table 9). This marked decrease in enzyme activity per gram of adipose tissue (figure 18) was also accomPanied by the period of greatest accumulation of lipid (figure 6) from birth to 35 days of age. The decrease in


Figure 18. Glyceride synthetase activity of perirenal, subcutaneous and intramuscular adipose tissues expressed on adipose tissue weight basis.
enzyme activity continued from 35 to 70 days followed by an increase between 70 and 105 days and then gradually decreased again and to a low value at 175 days. The reason for these changes may be explained as follows: the number of adipocytes of perirenal fat per gram of adipose tissue of new born lambs was 2.4 fold larger than those at 35 days (table 11). On the other hand, enyzme activity per gram of perirenal adipose tissue of new born lambs was 2.67 fold larger than at 35 days. Also there is a high correlation coefficient ( $\mathrm{r}=.89, \mathrm{P}<.01$ ) between the number of adipocytes and the enzyme activity per gram of perirenal adipose tissue. Since the number of adipocytes per unit weight of adipose tissue decreased between birth and 35 days of age because of the increase in their size, expression of the enzyme activity per unit weight decreased at 35 days when fewer fat cells were present per unit weight of tissue. However, when the data were expressed on adipocyte number basis, higher enzyme activities were observed for larger adipocytes than small cells (table 9). This general trend of decreased enzyme activity per gram of adipose tissue was seen over the 175 days of the present experiment. However, between 70 and 105 days, the enzyme activity increased ( $\mathrm{P}<.05$ ). This observation can be explained by the fact that the effects of weaning and the consequent restricted caloric intake suggests that fat mobilization has occured as shown by the decrease in weight of this tissue (table 5). Thus, the percentage of small adipocytes ( $<25 \mu \mathrm{~m}$ )
should have increased at age 105 days and this trend was actually observed (figures 22 through 25). However, the number of adipocytes per gram should also have increased because of the higher percentages of small cells. But the data in table 11 show a decrease ( $\mathrm{P}<.05$ ) in adipocytes per gram had occurred between 70 and 105 days of age. This observation may possibly be explained as follows: as mentioned earlier, the effects of weaning stress (82 days of age) resulted in mobilization of lipid and consequently the size of adipocytes was reduced. Reduction in size of the adipocytes may have resulted in some of the very small fat cells ( $<25 \mu \mathrm{~m}$ ) having passed through the filter screens and thus they were not counted. Consequently, the proportion of large cells had increased between 70 and 105 days which resulted in an artificially greater diameter or volume of the adipocytes. Therefore the number of cells per gram of tissue probably was underestimated in both the subcutaneous and perirenal depots at 105 days of age (table 11). The underestimated fat cells per gram of tissue also resulted in a decrease in numbers of total cells in each adipose tissue depot (total number of fat cells is the product of weight of the adipose tissue depot and number of fat cells per gram) between 70 and 105 days (table 11). Another possibility which might have contributed to the underestimation of total fat cells is that at age 105 days the aperture size used in the Coulter Counter was changed from $250 \mu \mathrm{~m}$ to $400 \mu \mathrm{~m}$. This
change in aperture size was made because from trial and error, past experiences and other works (Hirsch and Gallian, 1968) it was believed that at 105 days the diameter of some adipocytes was so large that they created problem with passage through $250 \mu \mathrm{~m}$ orifice. However, this decision was made without considering the effects that weaning might have on the results. The $400 \mu \mathrm{~m}$ orifice probably underestimated the number of fat cells because clumps of small cells which are not uncommon and are difficult to estimate, would not be individually counted. This difficulty also affects the size data (table 11).

In contrast to perirenal fat, the enzyme activity per gram of subcutaneous adipose tissue was very low in newborn lambs. This observation was not unexpected, because at birth no dissectable subcutaneous adipose tissue was present (table 5). The connective tissue layer in which the subcutaneous fat would develop later was physically dissected from the carcass of lambs at birth for use for the enzyme studies. This connective tissue layer was also fixed with osmium tetroxide and the cells separated as described in the materials and methods for the adipose tissues. Microscopic examination of the tissue showed that very few adipocytes were present and all were very small. Insufficient numbers of adipocytes precluded any Coulter Counter data on this tissue at birth. However, in this fat depot there was a marked increase (82 fold) in the glyceride enzyme activity
per gram of adipose tissue between birth and 35 days (table 9). This 82 fold increase in enzyme activity was accompanied by over a hundred fold increase in weight (table 5) and a 17 fold increase in the percentage lipid in the subcutaneous fat depot (table 7) during this dame 35 day period.

Growth rate of the lambs did not have an effect on enzyme activity per gram of perirenal and intramuscular fat ( $\mathrm{p}>.05$ ). However, enzyme activity was higher ( $\mathrm{P}=.06$ ) in the subcutaneous adipose tissue of fast growing lambs as compared to the slow groups (table 9). In both perirenal and intramuscular adipose tissues the values of glyceride synthesis activity per gram of tissue were higher for rams than for ewes ( P <.05) . However, the sex effect on the enzyme activity of subcutaneous adipose tissue was not significant ( $P>.05$ ). The enzyme activity per gram of both perirenal and subcutaneous fat was affected by the interaction between growth rate and age ( $\mathrm{P}<.01$, Appendix 12). In addition, enzyme activities of perirenal adipose tissue were affected ( $\mathrm{P}<.05$ ) by the interaction of sex and age (Appendix 13). No other interactions were significant ( $\mathrm{P}>.05$ ) .

When enzyme activities were expressed on a per cell basis, in both the perirenal and subcutaneous depots the enzyme activities increased with age ( $\mathrm{P}<.01$, tables 9 and 10 and figure 19). However, no significant change in enzyme activity occured for the intramuscular fat between 140 and 175 days (table 9). The enzyme activity expressed on the


Figure 19. Glyceride synthetase activity of perirenal, subcutaneous and intramuscular adipose tissues on a per cell basis.
basis of $10^{7}$ cells in perirenal adipose tissue did not change significantly ( $P>.05$ ) from birth to 70 days or in the subcutaneous fat from 35 to 70 days (table 9). Between 70 to 105 days, enzyme activities per $10^{7}$ cells in perirenal and subcutaneous depots increased ( $P<.05$ ) 2.2 and 4.2 fold, respectively. Between 105 and 140 days the enzyme activity in both adipose tissue depots increased but only that for the subcutaneous fat was significantly different ( $\mathrm{P}<.05$ ). The enzyme activity for both perirenal and subcutaneous fat expressed on a cell basis showed further increases between 140 and 175 days ( $P<.05$ ). The sharp increase between 70 and 105 days in enzyme activity is probably attributed to the problems of the adipocyte counts at 105 days as already explained.

Fast growing lambs had higher ( $P$ < . O5) enzyme activities per $10^{7}$ cells of perirenal adipose tissues than the slow growing group (table 9). The enzyme activities of subcutaneous and intramuscular adipose tissues expressed on a per cell basis were not significantly different ( $\mathrm{P}>.05$ ) between fast growing lambs and the slow growing group. Similar results were observed by Merkel et al. (unpublished data) in subcutaneous adipose tissue in lambs.

Effect of sex on the enzyme activity on a per cell basis of perirenal and subcutaneous adipose tissue was also not significant ( $\mathrm{P}>.05$ ). However, rams had higher enzyme activities per cell in the intramuscular adipose tissue
compared to ewes. In contrast, Merkel et al. (unpublished data) found higher glyceride enzyme activities on a per cell basis in subcutaneous fat of ewes compared to rams. Enzyme activity per cell in both subcutaneous and intramuscular adipose tissues was affected by the interaction between growth rate and age ( $\mathrm{P}<.05$, Appendix 12). The enzyme activities on a per cell of perirenal adipose tissue were also affected ( $\mathrm{P}<.05$ ) by the interaction of sex and age, (Appendix 13). None of the other interactions was significant ( $\mathrm{P}>.05$ ).

When comparing the three basis of expressing the glyceride synthetase activity, the data per unit of protein and per gram of perirenal and subcutaneous adipose tissues are quite constant after 70 days of age, but when the data are expressed on a cell basis, enzyme activities increased with fat accretion and adipocyte hypertrophy between 70 and 175 days of age. Thus these data indicate that adipocytes of perirenal and subcutaneous depots maintained the capacity for the glyceride synthesis throughout the experimental period in the present study. Additionally, glyceride synthesis activities on a cell basis of the large cells at 175 days of age (figure 19) were greater than at all other ages and the activity parallelled the increase in lipid accumulation (figure 21).

## Lipid Content Per Adipocyte

Changes in lipid content per adipocyte of the three adipose tissues with age are shown in tables 11 and 12 (also figures 20 and 21). As would be expected, in all adipose tissues, lipid content per cell increased with age. The lipid content per cell was significantly ( $\mathrm{P}<.01$ ) correlated with the percentage lipid in the perirenal ( $r=.55$ ) and subcutaneous ( $\mathrm{r}=.48$ ) depots but not for intramuscular ( $\mathrm{r}=.27$, P>.05) fat. The greatest increase in lipid content per cell for the perirenal fat ( 16 fold) occurred during the first 35 days. Undoubtedly, the subcutaneous fat increased similarly between birth and 35 days even though no observations of the birth adipocytes could be made. Fat content per cell increased significantly ( $\mathrm{P}<.05$ ) between all periods for the three adipose tissues, except no difference ( $P>.05$ ) occurred between 35 and 70 days for subcutaneous fat.

Growth rate did not significantly ( $P>.05$ ) affect the fat content per cell of the three adipose tissues. However, the fat content per cell of perirenal adipose tissue of ewe lambs was higher ( P <.05) than that of rams. This observation is consistent with the larger volume ( $\mathrm{P}=.04$ ) and diameter ( $\mathrm{P}=.07$ ) of the fat cells in ewes as compared to rams (table 11).

Lipid content per cell was highly correlated ( $r=.96$, $\mathrm{P}<.01$ ) with the volume of the perirenal fat cells (figure 10).


Figure 20. Lipid content and cell volume of perirenal adipose tissue as affected by age.


Figure 21. Linid content and cell volume of subcutaneous (S?) and intramuscular (IM) adioose tissues as affected by age.
table 11. EFFECTS OF GROWTH RATE, SEX AND AGE ON CELLULARITY AND ADIPOCYTE LIPID CONTENT OF PERIRENAL, SUBCUTANEOUS AND INTRAMUSCULAR ADIPOSE TISSUES

| Measurements | Growth Rate ${ }^{\text {a }}$ |  | Sexa |  |  | Pr. | Age(days) ${ }^{\text {b }}$ |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast growing | Slow growing | Pr. | Ram | Ewe |  | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Number of Adipocytes per: |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 14.9 | 15.9 | . 29 | 16.0 | 14.7 | . 18 | $41.7^{\text {c }}$ | $17.3{ }^{\text {d }}$ | $14.4{ }^{\text {d }}$ | $9.8{ }^{\text {c }}$ | $5.7{ }^{\text {f }}$ | $3.3{ }^{\text {f }}$ | <. 001 |
| Subcutaneous | 9.3 | 10.3 | . 37 | 10.0 | 9.6 | . 72 | - | $19.1{ }^{\text {c }}$ | $16.7^{\text {c }}$ | $6.5{ }^{\text {d }}$ | $3.8{ }^{\text {d }}$ | $2.9{ }^{\text {d }}$ | < . 001 |
| Intramuscular 7 | 3.52 | 3.33 | . 66 | 3.1 | 3.7 | . 16 | - | - | - | - | $4.26{ }^{\text {c }}$ | $2.58{ }^{\text {d }}$ | <. 001 |
| Separated adipose tissue ( $\times 10^{7}$ ) : |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 138.7 | 143.3 | . 73 | 138.8 | 143.1 | . 75 | $74.2{ }^{\text {c }}$ | $114.4{ }^{\text {c }}$ | 209.6 | $105.4{ }^{\text {c }}$ | $171.4^{\text {d }}$ | $170.9^{\text {d }}$ | <. 001 |
| Subcutaneous | 413.3 | 436.3 | . 65 | 444.3 | 405.4 | . 44 | - | $195.1^{\text {C }}$ | $491.0^{\text {d }}$ | $269.6^{\text {c }}$ | $478.2^{\text {d }}$ | $690.3^{\text {e }}$ | <. 001 |
| Intramuscular | 222.4 | 172.7 | . 16 | 200.9 | 194.2 | . 85 | - | - | - | - | 226.7 | 168.4 | . 10 |
| Adipocyte diameter ( mm ) : |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Subcutaneous | 69.5 | 69.6 | . 97 | 69.2 | 69.9 | . 66 | 37.0 | $51.6{ }^{\text {c }}$ | $52.1{ }^{\text {c }}$ | $70.6{ }^{\text {d }}$ | $83.1{ }^{\text {e }}$ | $90.3{ }^{\text {f }}$ | <.001 |
| Intramuscular | 56.9 | 57.3 | . 82 | 57.6 | 56.6 | . 61 | - | - | - | - | $50.1^{\text {c }}$ | $64.1{ }^{\text {d }}$ | <. 001 |
| Adipocyte volume $\left(\mu \mathrm{m}^{3} \times 10^{4}\right)$ : $\quad$ |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 15.4 | 13.8 20.8 | . 87 | 20.6 | 16.0.7 | . .96 | 2.7 | 6.8 7.3 | $7.1{ }^{\text {c }}$ | $19.0{ }^{\text {d }}$ | $30.9{ }^{\text {e }}$ | 38.9 f | <.001 |
| Intramuscular | 10.4 | 10.5 | . 89 | 10.5 | 10.4 | . 83 | _ | - | 7.1 | 19.0 | $6.8{ }^{\text {c }}$ | $14.1{ }^{\text {d }}$ | <. 001 |
| Lipid content per adipocyte(ng): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 126.4 | 114.7 | . 34 | $106.7^{\text {c }}$ | $134.4{ }^{\text {d }}$ | . 03 | $8.3{ }^{\text {c }}$ | $50.0{ }^{\text {d }}$ | $61.8{ }^{\text {e }}$ | $103.3{ }^{\text {f }}$ | $166.1^{8}$ | $333.8{ }^{\text {h }}$ | <. 001 |
| Subcutaneous | 135.0 | 129.5 | . 67 | 139.1 | 125.4 | . 30 | - | $38.3^{\text {c }}$ | $45.6{ }^{\text {c }}$ | $104.4{ }^{\text {d }}$ | $217.3^{\text {e }}$ | $255.8{ }^{\text {f }}$ | <.001 |
| Intramuscular | 9.5 | 11.7 | . 21 | $12.4{ }^{\text {c }}$ | $8.8{ }^{\text {d }}$ | . 04 | - | - | - | - | $7.0^{\text {c }}$ | $14.1{ }^{\text {d }}$ | <. 001 |

$a_{\text {Mean of }} 36$ lambs for perirenal or subcutaneous and 12 lambs for intramuscular adipose tissues.
$b_{\text {Means }}$ are the average of 12 lambs.
cdefgh means within each main effect on the same row bearing the same superscripts are not statistically significant ( $\mathrm{p}>$. 5) .
Pr. $=$ Probability for level of significance.
TABLE 12. INTERRELATIONSHIP OF GROWTH RATE, AGE AND SEX ON CELLULARITY AND ADIPOCYTE
LIPID CONTENT OF PERIRENAL, SUBCUTANEOUS AND INTRAMUSCULAR ADIPOSE TISSUES ${ }^{\text {a }}$

| Measurements | Sex | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  |  | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Number of adipocytes per: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| 8 Adipose tissue(x $10^{6}$ ): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 45.21 | 15.47 | 15.06 | 7.35 | 4.82 | 3.58 | 41.94 | 21.19 | 15.31 | 12.49 | 5.60 | 4.45 |
|  | Ewe | 38.83 | 15.41 | 13.56 | 10.81 | 5.50 | 2.92 | 41.04 | 17.31 | 13.68 | 8.63 | 6.88 | 2.27 |
| Subcutaneous | Ram | - | 17.33 | 20.63 | 6.68 | 3.44 | 2.91 | - | 19.12 | 15.72 | 8.01 | 2.95 | 3.24 |
|  | Ewe | - | 14.17 | 14.31 | 6.18 | 4.48 | 2.62 | - | 25.84 | 15.99 | 5.06 | 4.33 | 2.92 |
| Intramuscular | Ran | - | - | - | - | 5.81 | 2.00 | - | - | - | - | 2.35 | 2.26 |
|  | Ewe | - | - | - | - | 3.76 | 2.51 | - | - | - | - | 5.14 | 3.56 |
| Separated adipose tissue(x 107) : |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  | Ewe | 82.3 | 133.6 | 221.9 | 134.3 | 177.6 | 154.3 | 55.2 | 116.2 | 224.2 | 94.2 | 145.8 | 158.1 |
| Subcutaneous | Ram | - | 189.3 | 538.0 | 208.7 | 544.0 | 815.7 | - | 115.0 | 453.0 | 266.0 | 504.7 | 811.7 |
|  | Ewe | - | 145.0 | 439.7 | 363.7 | 441.0 | 448.7 | - | 331.0 | 533.3 | 240.0 | 426.0 | 685.3 |
| Intramuscular | Ram | - | - | - | - | 385.7 | 137.3 | - | - | - | - | 126.0 | 154.7 |
|  | Ewe | - | - | - | - | 206.0 | 160.7 | - | - | - | - | 189.3 | 221.0 |
| Adipocyte diameter ( $\mu \mathrm{m}$ ) : |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ras | 36.1 | 50.0 | 50.7 | 61.4 | 77.6 | 81.7 | 37.0 | 47.3 | 52.2 | 58.2 | 73.1 | 79.8 |
|  | Ewe | 37.4 | 53.2 | 54.2 | 59.1 | 72.9 | 94.7 | 37.7 | 50.3 | 53.6 | 58.6 | 68.5 | 93.8 |
| Subcutaneous | Ram | - | 50.6 | 51.3 | 69.0 | 78.9 | 93.1 | - | 50.6 | 52.7 | 68.2 | 90.0 | 87.6 |
|  | Ewe | - | 54.2 | 52.9 | 70.3 | 84.4 | 90.6 | - | 51.2 | 51.5 | 74.8 | 79.4 | 90.0 |
| Intramuscular | Ram | - | - | - | - | 50.8 | 63.3 | - | - | - | - | 51.1 | 65.3 |
| Adipocyte volume ( $\mathrm{mm}^{3} \times 10^{4}$ ) : Wwe - 50.463 .0 |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 2.5 | 6.5 | 6.8 | 12.3 | 25.3 | 28.7 | 2.6 | 5.7 | 7.9 | 11.2 | 20.8 | 27.6 |
|  | Ewe | 2.7 | 8.1 | 8.4 | 11.3 | 20.5 | 51.4 | 2.8 | 6.8 | 8.2 | 10.6 | 17.1 | 44.3 |
| Subcutaneous | Ram | - | 6.9 | 5.9 | 17.5 | 26.4 | 42.7 | - | 7.0 | 7.1 | 17.9 | 38.4 | 35.5 |
|  | Ewe | - | 8.5 | 7.4 | 18.7 | 32.3 | 39.1 | - | 7.0 | 7.3 | 22.0 | 26.5 | 38.2 |
| Intramuscular | Ram | - | - | - | - | 6.9 | 13.4 | - | - | - | - | 7.2 | 5.9 |
|  | Ewe | - | - | - | - | 7.1 | 14.1 | - | - | - | - | 14.6 | 14.4 |
| Lipid content per adipocyte(ng) : |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | Ram | 6.8 | 54.3 | 53.9 | 115.0 | 200.4 | 266.9 | 7.8 | 38.4 | 38.8 | 85.0 | 166.9 | 226.6 |
|  | Ewe | 9.5 | 60.3 | 66.4 | 90.2 | 160.9 | 432.3 | 9.3 | 46.9 | 68.2 | 122.9 | 136.1 | 409.6 |
| Subcutaneous | Ram | . | 41.3 | 38.1 | 106.5 | 230.9 | 275.2 | - | 33.9 | 37.5 | 105.8 | 266.0 | 256.4 |
|  | Ewe | - | 53.6 | 58.9 | 57.4 | 194.5 | 294.2 | - | 24.3 | 48.2 | 147.9 | 177.7 | 197.4 |
| Intramuscular | Ram | - | - | - | - | 4.73 | 13.60 | - | - | - | - | 10.03 | 21.20 |
|  | Ewe | - | - | - | - | 7.40 | 12.10 | - | - | - | - | 5.93 | 9.60 |

${ }^{\text {ameans are the average of } 3 \text { lambs. }}$

The lipid content per cell of intramuscular fat of rams was higher than that of ewes ( P <.05). In addition lipid content per cell of intramuscular depot was significantly ( $P<.05$ ) affected by the interaction of sex and growth rate (Appendix 11). A significant interaction ( $\mathrm{P}<.05$ ) between sex and age on the lipid content per cell of perirenal adipose tissue was also observed ( Appendix 13). None of the other interactions was significant ( $\mathrm{P}>.05$ ).

Cellularity of the Adipose Tissues During Growth

Adipocyte Number

Number of adipocytes was expressed either on a per gram of adipose tissue basis or as the total number for each adipose tissue depot (tables 11 and 12). As would be expected, the number of fat cells decreased with age when expressed on per gram of tissue (table 11). This is due to the increasing size of the cells with age, consequently fewer numbers of adipocytes were present per unit weight with advancing age. This observation is verified by the significant ( $p<.01$ ) negative correlation coefficients between adipocyte number (per gram basis) and volume of the fat cells in perirenal ( $r=-.69$ ), subcutaneous ( $r=-.79$ ) and intramuscular ( $r=-.58$ ) adipose tissues. Neither growth rate nor sex of the lambs significantly ( $p>.05$ ) affected the concentration of
adipocytes per gram of adipose tissues for any of the three depots. The interaction between growth rate and sex resulted in a significant $(\mathrm{P}<.01)$ effect on the number of adipocytes per gram of intramuscular adipose tissue (Appendix 11). No other interactions were significant ( $\mathrm{P}>.05$ ).

On the basis of total number of adipocytes per depot, the total number of adipocytes in perirenal and subcutaneous fat generally tended to increase with age (table 11). Although the total number of adipocytes in the perirenal adipose tissue increased from birth to 35 days, the increase was not significant ( $\mathrm{P}>.05$ ) . The largest increase ( $83 \%$ ) in this fat depot occurred between 35 and 70 days ( $P<.05$ ) which was followed by a $50 \%$ decrease $(P<.05)$ between 70 to 105 days. This latter decrease apparently is due to the events discussed earlier for the data at 105 days of age.

The total number of fat cells in the subcutaneous adipose tissue depot increased ( $150 \%, \mathrm{P}<.01$ ) between 35 and 70 days, but decreased between 70 and 105 days. The latter observation probably is attributed to the explanation presented earlier. The number of adipocytes in the subcutaneous connective tissue removed from the lambs at birth was very low and the few adipocytes present were extremely small in diameter as observed by microscopy. Thus, a considerable increase in subcutaneous adipocytes had to have occurred between birth and 35 days of age to account for the 195.1 x $10^{7}$ cells present at 35 days of age in this depot. In
contrast to perirenal fat, the number of fat cells in subcutaneous adipose tissue increased ( $\mathrm{P}<.05$ ) from 140 to 175 days. These data suggest that the perirenal fat hyperplasia has plateaued while subcutaneous adipose tissue still has an increase in cell number occurring between 140 and 175 days. This observation also indicates the earlier maturity of perirenal fat compared to that for subcutaneous fat.

The total number of adipocytes in intramuscular adipose tissue decreased (although not significantly) from 140 to 175 days. This latter decrease could be due to the change in aperture tube from $250 \mu \mathrm{~m}$ to $400 \mu \mathrm{~m}$ for intramuscular adipose tissue at age 175 days. Neither growth rate nor sex significantly affected the total number of adipocytes in any of the three fat depots. The interaction between growth rate and sex (Appendix 11) and between growth rate and age (Appendix 12) affected the total number of adipocytes in intramuscular adipose tissue ( $\mathrm{P}<.05$ ). No other interactions were significant ( $P>.05$ ).

Adipocyte Volume and Diameter

The mean diameter and volume of adipocytes in the three adipose tissue depots were significantly ( $P<.01$ ) affected by age (table 11 and 12). The diameter of the perirenal and subcutaneous adipocytes increased significantly with age, except between 35 and 70 days, until 175 days ( $P<.05$ ).

Between 35 and 70 days the diameter of perirenal and subcutaneous adipose tissue cells were similar, but at 105 and 140 days subcutaneous adipose tissue had larger adipocytes than perirenal fat. However, at 175 days fat cell size of the two depots was similar. Intramuscular adipose tissue fat cell diameter at 175 days was $30 \%$ less than that from subcutaneous and perirenal fat. These data indicate that intramuscular adipocytes hypertrophy occurs at later ages than for subcutaneous and perirenal fat. During the last two periods of the experiment, when fat cell diameter or volume of the three fat depots were increasing significantly, a significant ( $\mathrm{P}<.05$ ) increase in adipocyte number also occurred in subcutaneous fat but not in the perirenal or intramuscular adipose tissues. The results of fat cell number and diameter suggest that when the lambs weighed between 36 and 46 kg ( 140 and 175 days), hyperplasia apparently had been completed in perirenal adipose tissue and the increase in the fat depot during this time was primarily due to hypertrophy. However, when the lambs were younger, both hyperplasia and hypertrophy were responsible for the increase in weight of perirenal adipose tissue (table 11). On the other hand, both hyperplasia and hypertrophy continued to contribute to the increase in subcutaneous adipose tissue at 175 days. Hypertrophy contributed significantly to the increase in intramuscular fat, whereas hyperplasia data were beset with the events associated with change in aperture as discussed previously.

Perirenal adipose tissue of ewe lambs had larger diameter ( $\mathrm{P}=.07$ ) fat cells and volumes ( $\mathrm{P}<.05$ ) than rams. These data are consistent with the total mass of the perirenal adipose tissue since ewe lambs had significantly ( $\mathrm{P}<.05$ ) more fat in this depot. Diameter and volume of subcutaneous and intramuscular adipose tissue were not affected by sex ( $\mathrm{P}>.05$ ). Growth rate of the lambs did not affect the diameter or volume of the fat cells in any of the adipose tissues ( $\mathrm{P}>.05$ ). Fat cell diameter and volume of the perirenal adipose tissue was affected by the interaction between age and sex (Appendix 13). None of the other interactions was significant ( $\mathrm{P}>.05$ ).

Adipocyte Histograms

Figures 22 through 27 depict the frequency distributions of adipose cells isolated from the three adipose tissues. Each bar of the histogram represents the contribution in percentage of total adipocyte number made by the cells within a specified diameter range (abscissa). The histogram patterns for ram and ewe lambs were similar for each of the three adipose tissues. As shown in figures 22 and 23, approximately $95 \%$ of the adipose cells of the perirenal depot at birth had diameters of less than $40 \mu \mathrm{~m}$. With age up to 70 days, the percentage of small cells decreased while larger cells increased. This change caused a shift in bar height to the right at each age up to 70 days. At 105 days, the

## FASTGROWING

 Figure 22. Frequency distribution of perirenal adipocytes as affected by growth rate, age and sex.

## SLOWGROWING

RAM
EWE



AdiPocyte Diameter (Nmeler)
Figure 23. Frequency distribution of perirenal adipocytes as affected by growth rate, age and sex.
distribution pattern had changed because the percentage of small cells had increased compared to that at 70 days of age. This observation is consistent with the explanation discussed earlier for the perirenal fat data from the lambs at 105 days. An interesting observation is that the percentage of small cells (less than $30 \mu \mathrm{~m}$ ) that had increased at 105 days essentially maintained this level (between 30 and $40 \mu \mathrm{~m}$ ) throughout the remainder of the experiment. However, disregarding the very small cells, after 105 days, the remainder of the cells increased in diameter and the distribution gradually shifted to the right with age. The distribution of the cells at the last two ages (140 and 175 days) had a bimodal shape with the first mode being represented by the small cells (less than $30 \mu \mathrm{~m}$ ) and the second mode by the larger cells. The adipocyte distribution observed for perirenal was similar to that observed for subcutaneous adipose tissue (figures 24 and 25).

In intramuscular fat both small and large diameter fat cells were present. Compared to the data at 140 days, at 175 days the percentage of small cells had increased and the histogram bars had also shifted to the right (figures 26 and 27). Thus it appears that at 175 days both hyperplasia and hypertrophy probably contributed to the development of intramuscular adipose tissue.


Figure 24. Frequency distribution of subcutaneous adipocytes as affected by growth rate, age and sex.


Figure 25. Frequency distribution of subcutaneous adipocytes as affected by growth rate, age and sex.




# Changes in Body Weight and Muscle Weight and Composition During Growth 

Body Weight

Results of body weight gain as affected by growth rate, age and sex are presented in tables 13 and 14. At all ages fast growing rams and ewes (except for new born rams) were heavier than those of the slow growing group (table 14). As can be seen in figure 29 , the fast growing group was heavier at all ages from 35 days onward than the slow growing rams. In addition, the overall effect of growth rate resulted in significantly $(P<. O 1)$ heavier lambs in the fast growing group compared to the slow growing group (26.4 vs 22.8 kg , table 13). This difference in body weight was due to differences in daily gain ( 260 vs $230 \mathrm{~g} /$ day) rather than the differences in birth weights ( 4.2 vs 3.9 kg ). The greatest percentage increase in body weight occurred between birth and 35 days. The growth rate of rams was significantly ( $\mathrm{P}<.01$ ) greater than ewes (table 13). Average daily gain of rams and ewes was 260 and 224 g , respectively, for the entire experimental period. The growth rate of fast growing lambs and rams was greater than slow growing lambs and ewes. The highest (230\%) and the lowest ( $27 \%$ ) percentage increase was seen during the first ( 0 to 35 days) and the last (140 to 175 days) age periods, respectively. There was a significant ( $\mathrm{P}<.05$ )
interaction between sex and age (Appendix 16) on body weight. No other significant interaction was seen ( $\mathrm{P}>.05$ ) .

The growth curves presented in figure 28 to 30 indicate that the usual sigmoidal shaped curve was not observed in this experiment mainly because the experimental period was not long enough. This can be seen from the data (table 13) which show that even at the end of the experiment the lambs still had significant ( $\mathrm{P}<.05$ ) body weight and gastrocnemius (GT) and longissimus (LD) muscle weight increases.

Muscle Weight

The increase in GT and LD weights was very closely correlated with body weight ( $\mathrm{r}=.98, \mathrm{r}=.97$ for GT and LD respectively, $\mathrm{P}<.01$ ). This in agreement with the results reported by Hammond and Appleton (1932) in sheep and Orme et al. (1960) in cattle. Butterfield (1962) showed that age, weight and breed of the animal had no effect on the correlation between muscle weight and body weight. The regression equation for body weight ( X ) and GT weight of the 72 lambs in the present experiment is as follows: GT weight $(\mathrm{g})=3.40$ $\mathbf{x}$ body weight (kg) + 8.05. By applying the equation one can estimate the GT weight from live body weight of the lambs.

As was true for live body weight, GT and LD weights were significantly ( $\mathrm{P}<.01$ ) affected by growth rate, sex and age (table 13). During the early stages of growth when the lambs were tripling their birth weights, the greatest


Figure 2 8. Growth curves of body weights for fast vs slow growing lambs as affected by age.


Figure 29. Growth curves of body weight for rams vs ewes as affected by age.


Figure 30. Growth curves of body weight and gastocnemius (GT) and Longissimus (LD) muscles.
increase in rate of GT and LD weights also occurred. These data are similar to those reported by Butterfield (1976) and the emphasize that this is the period of maximum growth. There was no significant ( P >.05) interactions between the main affects, that is, growth rate, sex and age on muscle growth.

Another expression of muscle growth is the calculation of percentage of muscle relative to total body weight at various stages of growth (tables 13 and 14). On the latter basis, neither GT nor LD weights were affected ( $\mathrm{P}>.05$ ) by either growth rate or sex. However, age had a significant effect ( $\mathrm{P}<.01$ ) on the percentage $G T$ and LD weights (table 13). The percentage GT of body weight increased sharply from birth to 35 days. After 70 days the percentage GT of body weight plateaued, and the final percentage was significantly ( $P<.05$ ) lower than initially. The percentage LD of body weight increased between birth and 70 days followed by a nonsignificant ( $P>.05$ ) decrease between 70 and 105 days. The highest rate of increase in percentage LD was observed between 105 and 140 days, therafter the percentage plateaued (table 13). These data indicate that GT and LD were relatively constant proportions of live body weight after 70 and 105 days for GT and LD muscles, respectively.
table 13. EFFECTS OF GROWTH RATE, AGE AND SEX ON LIVE WEIGHT AND WEIGHT AND PERCENTAGE OF GASTROCNEMIUS (GT) AND LONGISSIMUS (LD) MUSCLES

| Measurements | Growth Rate ${ }^{\text {a }}$ |  | Pr. | Sex ${ }^{\text {a }}$ |  | Pr. | Age (days) ${ }^{\text {b }}$ |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | growing | growing |  | Ram | Ewe |  | 0 | 35 | 70 | 105 | 140 | 175 |  |
| $\begin{aligned} & \hline \text { Live weight } \\ & (\mathrm{kg}) \\ & \hline \end{aligned}$ | $26.43{ }^{\text {c }}$ | $22.82{ }^{\text {d }}$ | <. 001 | $26.62^{\text {c }}$ | 22.63 d | <. 001 | 3.95 C | $13.04{ }^{\text {d }}$ | 20.24 e | 28.09 f | 36.25 g | 46.7h | <. 001 |
| $\begin{aligned} & \hline \text { GT weight } \\ & \text { (g) } \end{aligned}$ | $101.2^{\text {c }}$ | $86.2{ }^{\text {d }}$ | <. 001 | $100.2^{\text {c }}$ | $87.8{ }^{\text {d }}$ | <. 001 | $15.6{ }^{\text {c }}$ | $63.3{ }^{\text {d }}$ | $82.4{ }^{\text {e }}$ | $107.4{ }^{\text {f }}$ | 134.08 | 159.7h | <. 001 |
| GT \% | . 40 | . 40 | . 97 | . 39 | . 40 | . 39 | .39d | .48e | . $40{ }^{\text {d }}$ | . 38 cd | . 37 c | . 35 c | <. 001 |
| LD weight <br> (g) | 357.5c | $310.4{ }^{\text {d }}$ | . 006 | 354.9C | 313d | . 01 | 44.7 c | 159.4 d | 273.9 e | 354.7 f | 533.6 g | 637.2h | <. 001 |
| LD \% | 1.29 | 1.31 | . 57 | 1.30 | 1.30 | . 97 | 1.12 C | 1.24 cd | 1.34 de | 1.27 d | 1.47 e | 1.38 e | <. 001 |

[^1]TABLE 14. INTERRELATIONSHIP OF GROWTH RATE, AGE AND SEX ON LIVE WEIGHT AND WEIGHT

| Measurement | Sex | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  |  | Age (days) |  |  |  |  |  | Age(days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Live weight(kg) | Ram | 3.88 | 14.81 | 24.72 | 32 | 43.24 | 53.24 | 4.46 | 11.94 | 19.81 | 27.42 | 38.10 | 45.66 |
|  | Ewe | 4.11 | 13.15 | 19.05 | 29.78 | 34.77 | 44.3 | 3.32 | 12.24 | 17.39 | 23.17 | 28.88 | 41.43 |
| GT weight (g) | Ram | 15 | 69 | 101 | 119 | 159 | 188 | 19 | 51 | 85 | 106 | 141 | 149 |
|  | Ewe | 15 | 72 | 75 | 113 | 128 | 160 | 13 | 60 | 69 | 90 | 108 | 141 |
| GT percentage | Ram | . 38 | . 46 | . 41 | . 37 | . 37 | . 35 | . 43 | . 43 | . 43 | . 39 | . 37 | . 33 |
|  | Ewe | . 38 | . 54 | . 39 | . 38 | . 37 | . 36 | . 39 | . 49 | . 40 | . 39 | . 37 | . 35 |
| LD weight (g) | Ram | 43 | 173 | 343 | 375 | 668 | 652 | 58 | 151 | 275 | 351 | 533 | 636 |
|  | Ewe | 40 | 164 | 267 | 394 | 526 | 644 | 38 | 149 | 211 | 298 | 408 | 616 |
| LD percentage | Ram | 1.09 | 1.21 | 1.38 | 1.18 | 1.55 | 1.21 | 1.28 | 1.13 | 1.39 | 1.28 | 1.42 | 1.39 |
|  | Ewe | . 99 | 1.26 | 1.40 | 1.32 | 1.50 | 1.45 | 1.13 | 1.22 | 1.21 | 1.29 | 1.41 | 1.49 |

${ }^{\text {a }}$ Means are the average of 3 lambs.

## Changes in Nucleic Acids During Growth

Nucleic Acid Concentrations

Tables 15 and 16 present the concentrations (milligrams per gram fresh muscle) of RNA and DNA in GT muscle of the lambs. Neither growth rate nor sex significantly affected the concentrations of the total nucleic acids in the GT ( $\mathrm{P}>.05$ ). Muscle RNA and DNA concentrations decreased ( $\mathrm{P}<.05$ ) by $62 \%$ and $50 \%$, respectively, between birth and 35 days (table 15, figure 31). Thereafter, concentration of RNA plateaued, while DNA showed a further, but small ( P <.05) decrease in nucleic acid concentrations. These decrease in muscle nucleic acid concentrations during postnatal growth agree with results reported previously (Enesco and Puddy, 1964; Robinson and Bradford, 1969; Powell and Aberle, 1975; Aberle and Doolittle, 1976; Johns and Bergen, 1976; Harbison et al., 1976). The high concentration of DNA in the baby lambs might be attributed, at least in part, to the presence of more muscle fibers per unit weight of muscle. In addition, skeletal muscle tissue of new born lambs is similar to that of embryonic muscle in which the nuclei and nucleoli constitute a high proportion of the muscle compared to that of later ages. During the period between birth and 35 days, the lambs showed the maximum muscle growth rate, and since
table 15. effect of growth rate, age and sex of lambs on the nucleic acid and nuclei

| Measurements | Growth Rate ${ }^{\text {a }}$ |  | Pr. | Sex ${ }^{\text {a }}$ |  | Pr . | Age(days) ${ }^{\text {b }}$ |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast growing | $\begin{gathered} \text { Slow } \\ \text { growing } \end{gathered}$ |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  | Ram | Ewe |  | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Concentration of RNA (mg/g) | 5.02 | 4.99 | . 91 | 4.96 | 5.05 | . 81 | $10.48^{\text {c }}$ | $4.02^{\text {d }}$ | $3.79{ }^{\text {d }}$ | $4.46^{\text {d }}$ | $4.04{ }^{\text {d }}$ | 3.25 d | <. 001 |
| Concentration of DNA (mg/g) | 1.83 | 1.76 | . 37 | 1.82 | 1.77 | . 54 | $3.43{ }^{\text {c }}$ | $1.71{ }^{\text {d }}$ | $1.68{ }^{\text {d }}$ | $1.36{ }^{\text {e }}$ | $1.36{ }^{\text {e }}$ | $1.23{ }^{\text {e }}$ | <. 001 |
| Total RNA(mg) | $408{ }^{\text {c }}$ | $348{ }^{\text {d }}$ | . 04 | 403 | 353 | . 09 | $167^{\text {c }}$ | $254{ }^{\text {cd }}$ | $311{ }^{\text {d }}$ | $470{ }^{\text {e }}$ | $544{ }^{\text {e }}$ | $522{ }^{\text {e }}$ | $<.001$ |
| Total $\mathrm{DNA}(\mathrm{mg})$ | $154{ }^{\text {c }}$ | $123{ }^{\text {d }}$ | <. 001 | $149^{\text {c }}$ | $127^{\text {d }}$ | . 004 | $53^{\text {c }}$ | $111{ }^{\text {d }}$ | $137{ }^{\text {e }}$ | $145{ }^{\text {e }}$ | $183{ }^{\text {f }}$ | 199 f | <. 001 |
| Protein/DNA | $120.2^{\text {c }}$ 2.77 | 130.4 2.84 | . 02 | 188.7 2.76 | 187.9 2.85 | $\begin{aligned} & .23 \\ & .70 \end{aligned}$ | 49.8 $3.10^{\text {c }}$ | $110.7{ }^{\text {d }}$ $2.40^{\text {d }}$ | $124.0{ }^{\text {e }}$ 2.35 | $150.0{ }^{\text {f }}$ $3.33^{\mathrm{C}}$ | 150.6 <br> 3.01 <br> c | 167.3 f 2.64 cd | <. 001 |
| RNA/DNA <br> Total number of nuclei ( $\times 10^{9}$ ) | ${ }_{24.655^{2}}$ | ${ }_{19.77}{ }^{2.84}{ }^{\text {d }}$ | .70 $<.001$ | 23.76 23.97 | 2.85 $20.47{ }^{\text {d }}$ | . 7006 | $3.10{ }^{\text {c }}$ $8.13^{\mathrm{C}}$ | $2.400^{\text {d }}$ 17.96 | ${ }_{22.144^{\text {de }}}$ | ${ }_{23.36{ }^{3} \mathrm{e}}$ | $3.011^{\text {c }}$ 29.55 | ${ }_{32.153^{\text {f }}}$ | .03 $<.001$ |
| Weight/nucleus $\left(\times 10^{-8} g\right)$ | 38.6 | 40.7 | . 16 | 39.2 | 40.0 | . 54 | $18.5{ }^{\text {c }}$ | $35.9{ }^{\text {d }}$ | $39.4{ }^{\text {e }}$ | $46.6{ }^{\text {f }}$ | $46.4{ }^{\text {f }}$ | $50.7{ }^{\text {f }}$ | <. 001 |

${ }^{a_{\text {Means }}}$ are the average of 36 lambs.
$\mathrm{b}_{\text {Means }}$ are the average of 12 lambs.
cdef Means within each main effect on the same row bearing the same superscripts are not statistically significant (p>.05).
Pr.=Probability for level of significance.
TABLE 16. INTERRELATIONSHIP OF GROWTH RATE, AGE AND SEX OF LAMBS ON THE NUCLEIC ACID

| Measurements | Sex | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  |  | Age(days) |  |  |  |  |  | Age (days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Concentration of | Ram | 9.8 | 4.3 | 4.5 | 4.1 | 4.4 | 3.5 | 10.4 | 4.4 | 3.2 | 4.1 | 3.7 | 3.1 |
| RNA (mg/g) | Ewe | 11.1 | 3.8 | 3.9 | 3.8 | 3.8 | 3.5 | 10.6 | 3.7 | 3.5 | 5.8 | 4.3 | 3 |
| Concentration of | Ram | 3.6 | 1.6 | 1.8 | 1.4 | 1.5 | 1.4 | 3.5 | 1.9 | 1.3 | 1.4 | 1.2 | 1.1 |
| DNA (mg/g) | Ewe | 3 | 1.7 | 2.1 | 1.2 | 1.3 | 1.3 | 3.6 | 1.6 | 1.6 | 1.4 | 1.3 | 1.2 |
| Total RNA(mg) | Ram | 148 | 293 | 440 | 471 | 713 | 651 | 206 | 231 | 275 | 428 | 523 | 457 |
|  | Ewe | 170 | 265 | 294 | 443 | 474 | 534 | 143 | 229 | 237 | 538 | 464 | 447 |
| Total DNA(mg) | Ram | 51 | 111 | 175 | 167 | 246 | 259 | 68 | 119 | 106 | 153 | 174 | 184 |
|  | Ewe | 46 | 119 | 160 | 134 | 171 | 205 | 47 | 97 | 107 | 126 | 141 | 169 |
| Protein/DNA | Ram | 46 | 113 | 107 | 139 | 129 | 147 | 49 | 87 | 163 | 140 | 164 | 187 |
|  | Ewe | 57 | 121 | 101 | 176 | 152 | 153 | 47 | 121 | 124 | 143 | 157 | 181 |
| RNA/DNA | Ram | 2.79 | 2.59 | 2.57 | 3.13 | 2.93 | 2.54 | 2.97 | 2.30 | 2.60 | 2.88 | 3.01 | 2.83 |
|  | Ewe | 3.76 | 2.62 | 2.0 | 3.12 | 2.87 | 2.62 | 2.89 | 2.44 | 2.71 | 4.19 | 3.22 | 2.58 |
| Total number of | Ram | 6.23 | 17.87 | 28.32 | 26.87 | 39.71 | 41.75 | 10.91 | 19.17 | 17.09 | 24.69 | 28.12 | 26.52 |
| nuclei (x10 ${ }^{9}$ ) | Ewe | 7.46 | 19.19 | 25.85 | 21.55 | 27.63 | 33 | 7.54 | 15.62 | 17.30 | 20.34 | 22.77 | 27.25 |
| Weight/nucleus | Ram | 17.5 | 38.1 | 36.8 | 45.3 | 41.3 | 45.7 | 17.7 | 28.2 | 49.7 | 43.0 | 50.5 | 56.2 |
| ( $\times 10^{-8}$ ) | Ewe | 21.0 | 38.0 | 31.1 | 53.6 | 46.5 | 48.6 | 17.7 | 39.5 | 40.3 | 44.4 | 47.3 | 52.4 |

apleans are the average of 3 lambs.


Figure 31. Changes in concentration (mg/g fresh muscle) of RNA and DNA as affected by age.

RNA is essential for protein synthesis, the high RNA concentration observed would be expected during this period. The sharp decrease in RNA concentration after birth is attributable to hypertrophy of the muscle fibers resulting in accretion of other constituents, thus diluting the nucleic acids which are accumulating at a much lower rate.

RNA to DNA Ratio

Aecording to Munro (1969) a high ratio of RNA to DNA concentration in muscle provides an indication of a high rate of protein synthesis. The pattern of changes of this ratio is presented in tables 15 and 16 . RNA to DNA ratios were not effected by growth rate or sex ( $P>.05$ ), however, age had a significant ( $P<.05$ ) effect on this ratio. The ratio decreased from birth to 35 days ( $\mathrm{P}<.05$ ) then remained unchanged until 70 days at which time it increased again ( $\mathrm{P}<.05$ ) and essentially regained and maintained the initial value thereafter.

The percentage increase of total DNA in the GT between birth and 35 days was approximately twice that of RNA, thus the ratio decreased. The highest ratio of RNA/DNA was observed at 105 days of age. This may be attributed to the following explanation. The lambs were weaned at 82 days of age which obviously reduced feed intake for the next few days. RNA concentration has been shown to decrease rapidly
following dietary restriction; whereas, DNA is much less sensitive to dietary intake (Howarth and Baldwin, 1971). Restriction of dietary intake to normal ad libitum levels has been reported to result and a marked increase in RNA synthesis (Howarth and Baldwin, 1971). Undoubtedly the RNA concentration at 105 days of age in the present study resulted from the compensatory increase in RNA synthesis to above normal levels when the lambs returned to normal feed intake levels during the week or days just prior to the 105 day sampling period. Other data also indicated that RNA/DNA ratio increased when stressed (Logan et al., 1952; Gluck et al., 1964). Goldspink (1964) reported that stress due to borderline protein intake caused a reduction in DNA and an increase in protein/DNA or RNA/DNA ratios. The present results were in agreement with the above observations (table 15).

The results of several experiments shows that total muscle growth is more closely related to total DNA than to rate of protein synthesis (Cheek et al., 1971; Buhlinger et al., 1978). The present experiment also confirms the above statement (Appendix 18) regarding the relationship of DNA to protein accretion. High ratios of protein/DNA and RNA/DNA both provide an indication of high rates of protein synthesis.

Total Amounts of Nucleic Acids

In addition to concentrations of DNA and RNA, the total amount of each in the GT was calculated. When compared to the slow growing group, fast growing lambs had more total RNA and DNA in their muscle ( P >.05). Rams had more total DNA in their GT muscles than ewes ( $\mathrm{P}>.01$ ). These observations are a reflection of the increased muscle weight among the fast growing lambs and rams. Although the total muscle RNA of rams was higher than ewes, the difference was not statistically significant ( $\mathrm{P}>.05$ ). Both total RNA and DNA contents of the GT were affected by age ( $\mathrm{P}<.01$ ). Total RNA increased $52 \%$ between birth and 35 days, and only slightly between 35 and 70 days which was followed by a gradual nonsignificant ( $\mathrm{P}>.05$ ) increase up to 175 days of age. The increase in DNA was more than twice (109\%) that of RNA during the first period (birth to 35 days). Between 35 and 70 days DNA showed a further significant ( P <.05) increase (23.4\%) but only a slight nonsignificant ( P >.05) increase (approximately 6\%) between 70 and 105 days of age. Between 105 and 140 days, DNA increased (27.6\%) significantly ( P <.05) followed by a nonsignificant (p>.05) increase (8.7\%) between 140 and 175 days of age. These nucleic acid data essentially parallel the increase in muscle weight with growth. The only significant ( $\mathrm{P}<.05$ ) interaction observed was between growth rate and age on the total DNA content of the GT (Appendix 15).

There was a high correlation ( $\mathrm{P}<.01$ ) between muscle weight and total DNA ( $\mathrm{r}=.89$ ) or total RNA ( $\mathrm{r}=.81$ ).

Changes in Number of Nuclei During Growth

In tissues that are composed of mononucleated cells, total DNA content is a direct indication of the total number of cells because their diploid nucleus contains a constant amount of DNA (Mirsky and Ris, 1949; Thomson et al., 1953; Vendrely, 1955). However, skeletal muscle is multinucleated and thus the relationship between the quantity of DNA and the number of muscle fibers is more complex. Cheek et al. (1971) introduced the "DNA Unit Concept" which considers the cytoplasm-to-nucleus ratio as a cell unit within the muscle cell. The number of nuclei in a given mass of muscle tissue can be calculated from the total DNA content. Thus, in multinucleated muscle cells the number of nuclei provides an indication of muscle growth potential. In order to study this relationship in the present experiment, the total number of nuclei was calculated by dividing the total DNA content of the GT muscle by 6.2 , since 6.2 is the amount of DNA in picograms in a single diploid nucleus (Enesco and Puddy, 1964). This index is used to estimate the cellularity (number of nuclei derived from total DNA) in the skeletal muscle tissue (Trenkle et al., 1978). The results of the total number of nuclei in the GT muscle are presented in table 15.

Fast growing lambs had more nuclei per GT ( $\mathrm{P}<.01$ ) than the slow growing group ( $24.65 \times 10^{9}$ vs $19.77 \times 10^{9}$ ). There was a significant interaction ( $\mathrm{P}<.05$ ) between age and growth rate for the total number of nuclei per GT (Appendix 15). Although the number of nuclei in the GT at birth was lower in fast growing lambs than the slow growing group, the higher rate of muscle growth in the fast growing lambs resulted in greater nuclei numbers during the remainder of experimental periods. The maximum increase in the number of nuclei occurred between birth and 35 days of age which was $163 \%$ and $89 \%$ for fast and slow growing lambs, respectively. As would be expected, rams had more nuclei in their muscles than ewes ( $\mathrm{P}<.01$ ) because they had heavier muscles. The number of nuclei increased ( P <.01) with increasing age, however, between 140 to 175 days the increase was nonsignificant ( $\mathrm{P}>.05$ ). High correlation coefficients were observed between number of nuclei and muscle weight ( $r=.89$ ) and with body weight ( $\mathrm{r}=.86$ ).

Some of the increase in muscle DNA may originate from the increase in nuclei associated with connective tissue and other cell types present in muscle tissue (Jablecki et al., 1973). However, Enesco and Puddy (1964) reported that a major proportion of the postnatal DNA increase was due to increases in nuclei within the muscle fibers. They also found that the proportion of DNA in muscle and connective tissue cells does not change during growth of skeletal
muscle. The results of the present experiment showed the percentage of connective tissue evaluated as stroma protein nitrogen in the GT was low (less than $1 \%$ ). The actual percentage of stroma protein nitrogen which was highest at birth (.6\%) decreased to approximately . $5 \%$ at 175 days of age. Since connective tissue contains relatively few cells, therefore few nuclei, it can be concluded that the contribution of nuclei from connective tissue in total muscle DNA was negligible.

With increasing age the cellularity (number of nuclei derived from total DNA) of the GT muscle increased ( $\mathrm{P}<.01$ ). The total number of nuclei increased threefold during muscle growth from birth to 175 days, however, the greatest increase occurred between birth and 35 days (120\%). After 35 days the total number of nuclei per muscle continued to increase ( $\mathrm{P}<.05$ ) up to 105 days and remained unchanged thereafter ( $\mathrm{P}>.05$, table 15). Buhlinger et al. (1978) concluded that the difference in number of muscle nuclei rather than differences in protein synthesis probably accounted for the differences in protein deposition of obese and lean pigs. In the present experiment it was found that measurements such as protein/DNA, muscle DNA or RNA concentrations and the ratio of RNA/DNA are not as highly correlated to muscle weight as total number of nuclei in the GT (Appendix 18). This observation is in agreement with the data reported by Ashmore and Robinson (1969), Ezekwe and Martin (1975) , Powell and Aberle (1975) and Buhlinger et al. (1978).

These conclusions support the DNA unit concept proposed by Cheek et al. (1971) that the total amount of muscle gained during growth is associated with the total DNA present. Muscle fibers do not undergo mitosis (Stromer et al., 1974) during postnatal growth, therefore the increase in total DNA during growth cannot originate from mitotic division of muscle nuclei. Recent data indicate that satellite cells are capable of postnatal mitosis and that one or both of their daughter cells resulting from this mitosis may be incorporated into the multinucleated muscle cell (Reger and Craig, 1968; Moss and LeBlond, 1970, 1971; Schultz, 1974). Thus, the DNA Unit Concept of Cheek et al. (1971) amplifies the importance of satellite cells for postnatal muscle growth.

Weight Per Nucleus

Since the ratio of cell protein/water is quite constant (Cheek et al., 1971), it can be deduced that protein/DNA ratio is an index of hypertrophy of muscle cells. Another parameter which also is an index for cell size is weight of the cellular constituents. This is an index of the amount of material associated with each nucleus, and therefore, it is influenced by size of the cell and the quantity of its intracellular materials. To calculate the weight per nucleus, weight of the muscle was divided by the total number of nuclei in the GT. The protein/DNA and weight/nucleus data
are presented in tables 15 and 16 . Weight per nucleus and protein/DNA ratios increased ( $P$ <.05) after birth until 70 days and remained unchanged thereafter ( $P>.05$ ). There was a high correlation ( $P<.01$ ) between muscle mass and protein/DNA ( $\mathrm{r}=.77$ ) or weight/nucleus $(\mathrm{r}=.76)$. High correlation coefficients ( $\mathrm{r}=.99$ ) between protein/DNA and weight/nucleus ratios and the similarity in the association of these two measurements with muscle mass indicates that they are indices of muscle fiber hypertrophy.

Although the difference in muscle weight as affected by growth rate was not detectable from the weight/nucleus ratio ( $\mathrm{P}>.05$ ), the significant difference ( $\mathrm{P}<.05$ ) in protein/DNA ratio suggests that there was more hypertrophy in slow growing lambs than the fast growing group. The effect of sex on both the protein/DNA and weight/nucleus ratio was nonsignificant ( P >.05). The weight/nucleus ratio was affected by the interaction between growth rate and age (Appendix 15). Other interactions were nonsignificant ( $P>.05$ ). In summary, it is concluded that the greater GT muscle mass of fast growing lambs and rams was due to more cellularity (DNA content) rather than size of the muscle fiber. The larger fibers of these lambs were associated with high concentrations of myofibrillar proteins and to a lesser extent to the sarcoplasmic proteins (table 19).

Chemical Composition of GT and LD Muscles

The percentage fat, protein and moisture of the GT and LD muscles are presented in tables 17 and 18. During muscle growth the percentage mositure decreased ( $\mathrm{P}<.01$ ) (from 79.4 to 72.9 in GT and from 79.1 to 73.6 in LD), while the percentage fat increased ( $\mathrm{P}<.01$ ) from .96 to 5.07 in GT and from . 62 to 3.66 in the LD. The percentage fat in the GT did not change significantly ( $\mathrm{P}>.05$ ) between 35 and 140 days, but then increased significantly ( $\mathrm{P}<.05$ ) between 140 and 175 days. In contrast to the GT muscle, the percentage fat in the LD showed a further increase ( $\mathrm{P}<.05$ ) between 35 to 70 days of age and then was followed by a decrease ( $\mathrm{P}<.05$ ) between 70 to 105 days. The decrease in fat content of LD muscle between 70 and 105 days may be attributed to weaning stresses suggesting that marbling in the LD muscle is more sensitive to these stresses than the GT. However, between 105 and 140 days the percentage fat in the LD returned to the value at 70 days and showed a further increase ( $\mathrm{P}<.05$ ) between 140 and 175 days.

The decrease in moisture content is a well established phenomenon (Callow, 1947; Dickerson and Widdonson, 1960; Reid et al., 1968; Hafez and Dyer, 1969) and can be explained by concomitant increase in fat and protein content as the muscle grows and develops. The negative correlations between moisture and fat in the GT or LD muscles are presented in
table 17. effect of growth rate, age and sex of the lambs on the chemical composition OF GASTROCNEMIUS (GT) AND LONGISSIMUS (LD) MUSCLES

| Measurements | Growth Rate ${ }^{\text {a }}$ |  | Sex ${ }^{\text {a }}$ |  |  | Age(days) ${ }^{\text {b }}$ |  |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast | Slow |  |  |  |  |  |  |  |  |  |  |  |
|  | growing | growing | Pr. | Ram | Ewe | Pr . | 0 | 35 | 70 | 105 | 140 | 170 |  |
| Percentage <br> fat in GT | $3.04{ }^{\text {c }}$ | $3.23{ }^{\text {d }}$ | . 05 | $3.68{ }^{\text {c }}$ | $3.18{ }^{\text {d }}$ | . 02 | .96C | $3.52^{\text {d }}$ | $3.51{ }^{\text {d }}$ | $3.56{ }^{\text {d }}$ | 3.97d | 5.07 e | <. 001 |
| Percentage protein in GT | $19.0{ }^{\text {c }}$ | $19.5{ }^{\text {d }}$ | . 003 | $19.1{ }^{\text {c }}$ | $19.5{ }^{\text {d }}$ | . 01 | $16.7{ }^{\text {c }}$ | $19.1{ }^{\text {d }}$ | $19.5{ }^{\text {d }}$ | 19.9 de | $20.1{ }^{\text {e }}$ | $20.4{ }^{\text {e }}$ | <. 001 |
| Percentage moisture in GT | 75.8 | 76.7 | . 52 | 75.9 | 75.7 | . 43 | $79.4{ }^{\text {c }}$ | $76.1{ }^{\text {d }}$ | $76.1{ }^{\text {d }}$ | $76.1{ }^{\text {d }}$ | $74.0{ }^{\text {e }}$ | $72.9{ }^{\text {f }}$ | <. 001 |
| Percentage <br> fat in LD | $1.90^{\text {c }}$ | $2.21{ }^{\text {d }}$ | . 03 | 2.05 | 2.06 | . 96 | $.62^{\text {c }}$ | $1.60{ }^{\text {d }}$ | $2.19{ }^{\text {e }}$ | $1.68{ }^{\text {d }}$ | $2.56{ }^{\text {e }}$ | $3.66{ }^{\text {f }}$ | <. 001 |
| Percentage protein in LD | 19.9 | 20.0 | . 57 | 19.8 | 20.1 | . 10 | $16.6{ }^{\text {c }}$ | $19.3{ }^{\text {d }}$ | $20.6{ }^{\text {e }}$ | $20.6{ }^{\text {e }}$ | $21.3{ }^{\text {f }}$ | $21.3{ }^{\text {f }}$ | <. 001 |
| Percentage moisture in LD | 76.6 | 76.3 | . 08 | $76.6{ }^{\text {c }}$ | $76.2^{\text {d }}$ | . 04 | $79.1{ }^{\text {c }}$ | $77.8{ }^{\text {d }}$ | $76.4{ }^{\text {e }}$ | $76.7^{\text {e }}$ | $75.1{ }^{\text {f }}$ | 73.68 | <. 001 |

${ }^{\text {a }}$ Means are the average of 36 lambs.
$\mathrm{b}_{\text {Means }}$ are the average of 12 lambs.
cdefg $_{\text {Means }}$ within each main effect significant( $p>.05$ ).
Pr.=Probability for level of significance.
TABLE 18. INTERRELATIONSHIP OF GROWTH RATE, AGE AND SEX OF LAMBS IN THE CHEMICAL COMPOSITION OF GASTROCNEMIUS (GT) AND LONGISSIMUS (LD) MUSCELS ${ }^{\text {a }}$

| Measurements | Sex | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  |  | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Percentage fat in GT | Ram | 1.1 | 3.5 | 4.4 | 4.2 | 4.4 | 6.4 | . 8 | 3.6 | 2.9 | 3.8 | 4.3 | 4.8 |
|  | Ewe | . 9 | 3.2 | 3.7 | 3.2 | 3.5 | 5.1 | 1.0 | 3.8 | 3.1 | 3.0 | 3.7 | 4.0 |
| Percentage protein in GT | Ram | 16.2 | 18.5 | 18.0 | 19.0 | 19.4 | 20.0 | 17.2 | 19.2 | 20.4 | 20.2 | 20.0 | 20.7 |
|  | Ewe | 16.9 | 19.6 | 20.4 | 20.3 | 20.3 | 19.6 | 16.5 | 19.0 | 19.2 | 20.0 | 20.6 | 21.4 |
| Percentage moisture in GT | Ram | 80.3 | 76.4 | 75.7 | 76.4 | 74.0 | 73.0 | 79.0 | 76.6 | 76.3 | 75.3 | 73.6 | 73.9 |
|  | Ewe | 79.2 | 76.1 | 75.9 | 76.0 | 74.0 | 73.0 | 79.0 | 75.5 | 76.4 | 76.5 | 74.0 | 71.9 |
| Percentage fat in LD | Ram | . 61 | 1.70 | 2.63 | 1.66 | 2.78 | 2.69 | . 60 | 1.43 | 2.21 | 1.63 | 2.27 | 4.37 |
|  | Ewe | . 63 | 1.73 | 1.57 | 1.61 | 2.14 | 3.01 | . 63 | 1.56 | 2.36 | 1.83 | 3.05 | 4.56 |
| Percentage protein in LD | Ram | 16.0 | 18.1 | 20.6 | 20.2 | 21.1 | 21.1 | 16.9 | 19.8 | 20.4 | 20.6 | 21.3 | 21.1 |
|  | Ewe | 16.5 | 20.0 | 20.9 | 20.9 | 21.5 | 21.7 | 16.9 | 19.4 | 20.3 | 20.6 | 21.2 | 21.2 |
| Percentage moisture in LD | Ram | 80.2 | 78.4 | 76.0 | 77.2 | 74.8 | 74.4 | 78.8 | 77.8 | 76.3 | 76.7 | 75.2 | 73.7 |
|  | Ewe | 78.7 | 77.1 | 76.6 | 76.6 | 75.7 | 73.6 | 78.8 | 77.7 | 76.6 | 76.1 | 74.6 | 72.6 |

$\mathrm{a}_{\text {Means }}$ are the average of 3 lambs.

Appendix 18. The amount of marbling of fast growing lambs in both GT and LD was lower than the slow growing group ( $\mathrm{P}<.05$ ). Rams had high ( $\mathrm{P}<.05$ ) percentages of fat and lower percentages of protein in their GT muscles, however, the percentage fat and protein in the LD of rams and ewes was similar ( $\mathrm{P}>.05$ ). Although the percentage protein in the GT of fast growing lambs was lower $(\mathrm{P}<.01)$ than the slow growing group the differences in the LD was nonsignificant ( $P>.05$ ). Percentage moisture of the GT and LD muscles was not affected by growth rate ( $P>05$ ). Rams had higher ( $P<.05$ ) percentages of moisture in the LD compared to ewes. The effect of sex on the percentage moisture of the GT was nonsignificant (P>.05). The maximum change in moisture and fat was observed during the first and the last 35 days of the experiment. However, the change during the first 35 days was greater than that of the last period.

The percentage protein in both GT and LD muscles increased sharply after birth until 35 days, but then the increase was more gradual thereafter. There was a significant ( $\mathrm{P}<.05$ ) interaction between growth rate and sex on the percentage protein in both GT and LD muscles. No other significant ( $\mathrm{P}>.05$ ) interactions were observed.

## Changes in Nitrogen Fractions During Growth

The nitrogen fraction data are expressed as: milligrams nitrogen per gram GT, milligrams nitrogen per GT and each fraction expressed as a percentage of total nitrogen (tables 19 and 20). Age had a significant effect ( $P<.01$ ) on all nitrogen fractions. As the GT growth progressed, the concentration of total, myofibrillar and sarcoplasmic nitrogen increased (table 19). The increase in concentration of myofibrillar nitrogen was essentially parallel to total nitrogen with a high correlation coefficient of .91 ( $\mathrm{P}<.01$ ). The concentration values are similar to those reported by Helander (1957) on gastrocnemius muscle of cattle. However, the percentage increase in sarcoplasmic nitrogen (46\%) from birth to 175 days was greater than for myofibrillar nitrogen (31\%). This is in agreement with data of Lawrie (1961) for the beef longissimus muscle. Stroma nitrogen per gram GT was the most variable when compared to the other nitrogen fractions (table 19). This might be explained by the fact that the stroma nitrogen is determined by difference which would reflect the combination of experimental errors in other fractions. However, the general trend of changes in stroma nitrogen concentration during GT growth were similar to those reported by Helander (1957) for this same muscle in cattle. Stroma nitrogen was negatively correlated ( $\mathrm{P}<.05$ ) with myofibrillar
table 19. effect of growth rate, age and sex of the lambs on the protein

\begin{tabular}{|c|c|c|c|c|c|c|c|c|c|c|c|c|c|}
\hline \multirow[t]{3}{*}{Measurements} \& \multicolumn{2}{|l|}{Growth Rate ${ }^{\text {a }}$} \& \multirow[t]{3}{*}{Pr .} \& \multicolumn{2}{|l|}{\multirow[t]{2}{*}{Sex ${ }^{\text {a }}$}} \& \multirow[t]{3}{*}{Pr.} \& \multicolumn{6}{|l|}{\multirow[t]{2}{*}{Age (days) ${ }^{\text {b }}$}} \& \multirow[t]{3}{*}{Pr.} <br>
\hline \& \multirow[t]{2}{*}{$$
\begin{aligned}
& \text { Fast } \\
& \text { growing }
\end{aligned}
$$} \& \multirow[t]{2}{*}{$$
\begin{aligned}
& \text { Slow } \\
& \text { growing }
\end{aligned}
$$} \& \& \& \& \& \& \& \& \& \& \& <br>
\hline \& \& \& \& Ram \& Ewe \& \& 0 \& 35 \& \& 105 \& 140 \& 175 \& <br>
\hline Concentration of nitrogen
$$
(\mathrm{mg} / \mathrm{g})
$$ \& $30.3{ }^{\text {c }}$ \& $31.0{ }^{\text {a }}$ \& . 007 \& $30.4{ }^{\text {c }}$ \& $30.9{ }^{\text {d }}$ \& . 02 \& $26.7^{\text {c }}$ \& $29.3{ }^{\text {d }}$ \& \& $31.8{ }^{\text {ef }}$ \& $32.0{ }^{\text {f }}$ \& 33.08 \& <. 001 <br>
\hline Concentration of mofibrillar nitrogen( $\mathrm{ng} / \mathrm{g}$ ) \& 15.1 \& 15.5 \& . 06 \& 15.2 \& 15.4 \& . 24 \& $12.7{ }^{\text {c }}$ \& $14.6{ }^{\text {d }}$ \& $15.4{ }^{\text {e }}$ \& $15.8{ }^{\text {f }}$ \& 16.98 \& $16.6{ }^{\text {h }}$ \& <. 001 <br>
\hline Concentration of sarcoplasmic nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) \& 5.64 \& 5.79 \& . 12 \& 5.63 \& 5.8 \& . 10 \& $4.40^{\text {c }}$ \& $5.81{ }^{\text {d }}$ \& $5.86{ }^{\text {d }}$ \& $5.67{ }^{\text {d }}$ \& $6.13^{\text {d }}$ \& $6.41^{\text {e }}$ \& <. 001 <br>
\hline Concentration of stroma nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) \& 5.60 \& 5.65 \& . 81 \& 5.58 \& 5.68 \& . 61 \& $6.00{ }^{\text {d }}$ \& $5.00^{\text {c }}$ \& $5.73{ }^{\text {d }}$ \& $6.84{ }^{\text {d }}$ \& $4.80{ }^{\text {c }}$ \& $6.00^{\text {d }}$ \& <. 001 <br>
\hline Concentration of nonprotein nitrogen (mg/g) \& $3.91{ }^{\text {c }}$ \& $4.08{ }^{\text {d }}$ \& . 01 \& 3.94 \& 3.99 \& . 80 \& $3.60{ }^{\text {c }}$ \& $3.90{ }^{\text {d }}$ \& $3.98{ }^{\text {d }}$ \& $4.10^{\text {de }}$ \& $4.88{ }^{\text {e }}$ \& $3.99{ }^{\text {d }}$ \& <. 001 <br>
\hline $\frac{\text { Total nitrogen(mg) }}{\text { Total myofibrillar }}$ \& 3160
$1600^{\text {c }}$ \& ${ }^{2760}{ }^{\text {d }}$ d ${ }^{\text {d }}$ \& . 003 \& $3140^{c}$
$1590^{c}$ \& 2770
1400 \& .009
.004 \& $$
\begin{aligned}
& 480^{c} \\
& 800^{c}
\end{aligned}
$$ \& $1850^{\text {c }}$
920 \& $$
\begin{aligned}
& 2560^{\mathrm{cd}} \\
& 1280^{\mathrm{de}}
\end{aligned}
$$ \& $$
\begin{aligned}
& 3410^{\mathrm{d}} \\
& 1680^{\mathrm{ef}}
\end{aligned}
$$ \& $$
\begin{array}{ll}
4270^{d} & 5 \\
2270^{f 8} & 2
\end{array}
$$ \& $$
\begin{aligned}
& 5250^{\mathrm{d}} \\
& 26508
\end{aligned}
$$ \& <. 001 <br>
\hline Total myofibrillar nitrogen (mg) Total sarcoplasmic nitrogen(mg) \& $1600^{\text {c }}$
600 \& 1390

530 \& .001
.005 \& 1590
590 \& $1400^{\text {d }}$
530 \& .004
.08 \& $800^{\text {c }}$
70 \& $920{ }^{\text {cd }}$

370 \& $12800^{\text {de }}$
480 \& $1680^{\text {ef }}$
$610^{\text {d }}$ \& 2270
888
8808 \& 2650
1020 \& <. 001 <br>
\hline Total stroma nitrogen(mg) \& $560{ }^{\text {c }}$ \& $490{ }^{\text {d }}$ \& . 02 \& $560^{\text {c }}$ \& $490{ }^{\text {d }}$ \& . 02 \& $90^{\text {c }}$ \& $320^{\text {d }}$ \& $470{ }^{\text {e }}$ \& $680{ }^{\text {f }}$ \& $640{ }^{\text {f }}$ \& 9488 \& <. 001 <br>
\hline Total non-protein nitrogen(mg) \& $400^{\text {c }}$ \& $350{ }^{\text {d }}$ \& . 001 \& $400^{\text {c }}$ \& $350{ }^{\text {d }}$ \& . 003 \& $60^{\text {c }}$ \& $240{ }^{\text {d }}$ \& $330{ }^{\text {e }}$ \& $440{ }^{\text {f }}$ \& 5608 \& $640^{\text {h }}$ \& <. 001 <br>
\hline . Myofibrillar nitrogen \& 49.8 \& 49.8 \& . 96 \& 49.9 \& 49.7 \& . 75 \& $48.1{ }^{\text {c }}$ \& $47.9{ }^{\text {c }}$ \& $50.8{ }^{\text {d }}$ \& $49.7{ }^{\text {cd }}$ \& $52.6{ }^{\text {e }}$ \& $50.6{ }^{\text {de }}$ \& <. 001 <br>
\hline \% Sarcoplasmic nitrogen \& 18.5 \& 18.6 \& . 87 \& 18.4 \& 18.6 \& . 50 \& $16.7{ }^{\text {c }}$ \& $19.0{ }^{\text {d }}$ \& $18.9{ }^{\text {d }}$ \& $17.9{ }^{\text {cd }}$ \& $19.1{ }^{\text {de }}$ \& $19.6{ }^{\text {e }}$ \& <. 001 <br>
\hline 7 Stroma nitrogen \& 18.8 \& 18.7 \& . 88 \& 18.8 \& 18.7 \& . 93 \& $21.7{ }^{\text {c }}$ \& $20.0{ }^{\text {cd }}$ \& $18.6{ }^{\text {cde }}$ \& $19.3{ }^{\text {cd }}$ \& $15.2{ }^{\text {e }}$ \& $17.7{ }^{\text {de }}$ \& . 006 <br>
\hline $\%$ Non-protein nitrogen \& 18.9 \& 13.0 \& . 71 \& 18.9 \& 18.9 \& . 90 \& $13.6{ }^{\text {c }}$ \& $12.8{ }^{\text {cd }}$ \& $12.7{ }^{\text {cd }}$ \& $13.0{ }^{\text {cd }}$ \& $13.2{ }^{\text {c }}$ \& $12.3{ }^{\text {d }}$ \& . 04 <br>
\hline Myofibrillar nitrogen/ Sarcoplasmic nitrogen \& 2.70 \& 2.71 \& . 93 \& 2.71 \& 2.70 \& . 84 \& $2.92{ }^{\text {c }}$ \& $2.53{ }^{\text {e }}$ \& $2.65{ }^{\text {d }}$ \& $2.79{ }^{\text {cd }}$ \& $2.75{ }^{\text {cd }}$ \& $2.59{ }^{\text {de }}$ \& <. 001 <br>
\hline
\end{tabular}

$$
{ }^{\text {a Means }} \text { are the average of } 36 \text { lambs. }
$$

cdefgh means within each main effect on the same row bearing the same superscripts are not statistically significant(p).05).
Pr.=Probability for level of significance.
TABLE 20. INTERRELATIONSHIP OF GROWTH RATE, SEX AND AGE OF LAMBS ON THE PROTEIN FRACTIONATION DATA OF GASTROCNEMIUS MUSCLE ${ }^{a}$

| Measurements | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast Growing |  |  |  |  |  |  | Slow Growing |  |  |  |  |  |
|  | Sex | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |
|  |  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |
| Concentration of non-protein | Ram | 3.5 | 3.8 | 3.4 | 3.9 | 4.2 | 3.9 | 3.7 | 3.9 | 4.2 | 4.1 | 4.2 | 4.0 |
| nitrogen(mg/g) | Ewe | 3.6 | 4.0 | 3.9 | 4.1 | 4.2 | 4.1 | 3.6 | 3.9 | 4.2 | 4.2 | 4.2 | 4.0 |
| Total nitrogen | Ram | 380 | 1970 | 3060 | 3610 | 5060 | 6090 | 530 | 1500 | 2670 | 3430 | 4500 | 4930 |
|  | Ewe | 420 | 2140 | 2360 | 3710 | 4070 | 5130 | 350 | 1820 | 2150 | 2890 | 3490 | 4860 |
| Total myofibrillar nitrogen(mg) | Ram | 180 | 970 | 1550 | 1760 | 2640 | 3110 | 860 | 740 | 1380 | 1670 | 2380 | 2460 |
|  | Ewe | 210 | 1070 | 1100 | 1860 | 2170 | 2630 | 160 | 910 | 1090 | 1460 | 1830 | 2390 |
| Total sarcoplasmic nitrogen(mg) | Ram | 60 | 390 | 590 | 630 | 940 | 1150 | 90 | 280 | 480 | 680 | 900 | 960 |
|  | Ewe | 70 | 410 | 470 | 650 | 770 | 1040 | 60 | 390 | 390 | 530 | 670 | 940 |
| Total stroma nitrogen(mg) | Ram | 90 | 350 | 540 | 760 | 810 | 1090 | 110 | 280 | 450 | 700 | 650 | 910 |
|  | Ewe | 90 | 370 | 500 | 730 | 590 | 810 | 80 | 280 | 390 | 520 | 540 | 970 |
| Total non-protein nitrogen(mg) | Ram | 50 | 260 | 380 | 460 | 670 | 740 | 70 | 200 | 360 | 440 | 600 | 600 |
|  | Ewe | 50 | 290 | 290 | 470 | 540 | 650 | 50 | 240 | 280 | 380 | 450 | 560 |
| Percentage myofibrillar nitrogen | Ram | 45.7 | 47.7 | 53.0 | 49.0 | 53.3 | 51.0 | 50 | 47.3 | 50 | 49.3 | 53.0 | 49.7 |
|  | Ewe | 50.0 | 47.7 | 45.3 | 50.3 | 52.3 | 58.7 | 46.7 | 4.9 | 51.7 | 50.3 | 51.7 | 49 |
| Percentage sarcoplasmic nitrogen | Ram | 16.7 | 19.0 | 20.3 | 17.3 | 18.7 | 18.7 | 17.0 | 18.0 | 17.7 | 18.3 | 20 | 12.3 |
|  | Ewe | 17.0 | 18.0 | 19.3 | 17.7 | 18.7 | 20.7 | 16.0 | 21.0 | 18.3 | 18.3 | 19.0 | 19.7 |
| Percentage stroma nitrogen | Ram | 24.0 | 20.7 | 14.7 | 20.7 | 14.3 | 18.0 | 19.0 | 22.0 | 19.7 | 19.7 | 14.0 | 18.7 |
|  | Ewe | 20.0 | 21.0 | 23.7 | 19.0 | 16.0 | 13.7 | 23.7 | 17.0 | 16.3 | 18.0 | 16.3 | 19.7 |
| Percentage non-protein nitrogen | Ram | 13.7 | 12.7 | 12.7 | 13.0 | 13.7 | 12.3 | 14.0 | 12.7 | 12.7 | 12.7 | 13.0 | 18.3 |
|  | Ewe | 13.0 | 13.0 | 11.7 | 13.0 | 13.0 | 13.0 | 13.7 | 13.0 | 13.7 | 13.3 | 13.0 | 11.7 |
| Concentration of nitrogen(mg/g) | Ram | 25.9 | 28.5 | 29.9 | 30.4 | 31.8 | 32.4 | 27.5 | 29.2 | 31.3 | 32.3 | 38.1 | 33.1 |
|  | Ewe | 27.0 | 29.6 | 31.5 | 32.5 | 31.9 | 32.1 | 26.4 | 29.9 | 31.3 | 32.0 | 38.3 | 34.3 |
| Concentration of myofibrillar nitrogen(mg/g) | Ram | 11.8 | 14.1 | 15.2 | 14.9 | 16.6 | 16.6 | 13.5 | 14.4 | 16.1 | 15.8 | 17.0 | 16.5 |
|  | Ewe | 13.2 | 14.9 | 14.6 | 16.4 | 16.9 | 16.4 | 12.3 | 14.9 | 15.8 | 16.1 | 16.9 | 16.8 |
| Concentration of sarcoplasmic nitrogen(mg/g) | Ram | 4.4 | 5.6 | 5.8 | 5.3 | 5.9 | 6.1 | 4.6 | 5.4 | 5.7 | 5.9 | 6.5 | 6.4 |
|  | Ewe | 4.4 | 5.7 | 6.3 | 5.6 | 6.0 | 6.5 | 4.2 | 6.4 | 5.6 | 5.9 | 6.2 | 6.6 |
| Concentration of stroma nitrogen (mg/g) | Ram | 6.2 | 4.9 | 5.2 | 6.3 | 5.1 | 5.8 | 5.7 | 5.5 | 5.3 | 6.4 | 4.4 | 6.2 |
|  | Ewe | 5.9 | 5.0 | 6.7 | 6.4 | 4.7 | 5.1 | 6.3 | 4.6 | 5.8 | 5.8 | 5.0 | 6.9 |
| Myofibrillar nitrogen/ <br> Sarcoplasmic nitrogen | Ram | 2.7 | 2.53 | 2.62 | 2.84 | 2.83 | 2.71 | 2.98 | 2.67 | 2.83 | 2.68 | 2.64 | 2.57 |
|  | Ewe | 3 | 2.61 | 2.34 | 2.91 | 2.80 | 2.54 | 3.05 | 2.32 | 2.81 | 2.74 | 2.75 | 2.53 |

${ }^{3}$ Means are the average of 3 lambs.
( $\mathrm{r}=-.29$ ), sarcoplasmic $(\mathrm{R}=-.31)$ and nonprotein nitrogen ( $\mathrm{r}=-.26$ ) fractions, but was not significantly ( $\mathrm{P}>.05$ ) correlated with total nitrogen ( $\mathrm{r}=.08$ ).

The ratio of myofibrillar to sarcoplasmic protein nitrogen varied during growth. The highest ratio was observed in the GT muscle of the new born lambs. Neither growth rate nor sex had an effect on these ratios ( $P>.05$ ).

Concentration of total and nonprotein nitrogen was higher in slow growing lambs than the fast growing group ( $P$ <.05). The other nitrogen fractions were not affected by growth rate ( $\mathrm{P}>.05$ ). Only total nitrogen (milligrams per gram) was significantly affected by sex ( $\mathrm{P}<.05$ ) which showed that rams had lower total nitrogen than ewes.

On the basis of the total milligrams of nitrogen in the GT muscle (table 19 and 20), all nitrogen fractions increased significantly ( $\mathrm{P}<.01$ ) during growth. Both sarcoplasmic and myofibrillar nitrogen fractions had a higher rate of increase in GT weight. This is in agreement with the results reported by Gorden et al. (1966) in rats. During the 175 days of the experiment the GT weight increased 9.2 times, while total sarcoplasmic and myofibrillar nitrogen increased 13.6 and 12.2 times, respectively. On the other hand, the increase in stroma ( 9.4 fold) and nonprotein nitrogen ( 9.7 fold) was similar to that of the GT increase of 9.2 fold. High correlation coefficients were obtained between the individual nitrogen fractions and they ranged from .92 to . 99 (Appendix
18). These data suggest that estimating the total amount of one nitrogen fraction may be a useful tool for predicting another nitrogen constituent. In addition, there was high correlation coefficients between GT and individual nitrogen fractions (r values ranged from . 94 to .99). Fast grwoing lambs had higher values for the nitrogen fraction compared to the slow growing group ( P <.05) . Rams had higher values for nitrogen fractions than ewes ( $\mathrm{P}<.05$ ) . The only significant interaction was observed between sex and age (Appendix 16) which affected ( $\mathrm{P}<.05$ ) total myofibrillar and nonprotein nitrogen.

When expressed on the basis of percentage of total nitrogen, the percentage of both myofibrillar and sarcoplasmic fractions increased significantly between birth and 35 days and maintained a relatively constant percentage thereafter (table 19). The percentage stroma and nonprotein nitrogen tended to decrease during GT growth, however, the changes were not consistent. Neither growth rate nor sex significantly ( P >.05) affected the percentages of the nitrogen fractions. No significant ( $P>.05$ ) interactions were observed for these data.

## SUMMARY

This study was designed to determine the effects of growth rate, sex and age on muscle growth and fattening of lambs from birth to 175 days of age. Sixty ewes with the fastest and 60 ewes with the slowest growing lambs were mated to Suffolk and Dorset rams respectively. Three ram and three ewe lambs of each growth rate were slaughtered at each age (birth, 35, 70, 105, 140 and 175 days). The lambs were weaned at 82 days of age and then divided into fast growing rams and ewes, and slow growing rams and ewes. Each group was penned and fed separately until slaughter time.

The new born lambs were slaughtered within 10 to 12 hr after birth, while the lambs of the other age groups were fasted approximately 15 hr prior to slaughter. All muscle and fat were rapidly removed, weighed, frozen, powdered and stored at -85 C for subsequent analyses.

Perirenal, subcutaneous and intramuscular adipose tissues were assayed for glyceride synthetase, cellularity (size and number of fat cells) and fat, protein and moisture. The GT muscle was analyzed for nucleic acid and protein fractions. Both GT and LD muscles were analyzed for fat, protein and moisture contents.

Fast growing lambs and rams had higher average daily gains than the slow growing group and ewes, respectively. Ewe lambs were superior to rams in feed conversion between 140 and 175 days of age. Body weight, muscle and adipose tissue weights increased with increasing age. Although fast growing lambs deposited more total protein than the slow growing group,the increase in total adipose tissue of the fast and slow growing lambs was similar. However, higher percentages of subcutaneous fat were detected in the slow growing group compared to fast growing lambs. Rams had more subcutaneous but less perirenal fat than ewes. Rams also had greater muscle weights than ewes.

When expressed per milligram protein or cell basis, glyceride synthetase activity increased with increasing age, but on a gram of adipose tissue basis the activity decreased. The effect of growth rate and sex differed between depots and among the expression of the enzyme activities.

The data for fat cell number and diameter suggest that when the lambs weighed between 36 to 46 kg ( 140 to 175 days) hyperplasia apparently had been completed in perirenal adipose tissue and the increase in this depot during this period was primarily due to hypertrophy. On the other hand, hyperplasia and hypertrophy contributed to the increase in subcutaneous adipose tissue at age 175 days.

However, cellularity in both perirenal and subcutaneous adipose tissues was affected by the stresses associated with weaning of the lambs. Hypertrophy contributed to the increase in intracellular fat. Results of different adipose tissue measurements indicated that the growth sequence is perirenal> subcutaneous > intramuscular.

Muscle RNA and DNA concentrations decreased while the total RNA and DNA increased in the GT muscle with increasing age. Although, growth rate and sex had no effect on the concentrations of nucleic acids in the GT muscle, fast growing lambs and rams had more total RNA and DNA and also more total nuclei in the GT muscle than slow growing lambs and rams, respectively.

Although weight/nucleus and protein/DNA increased with age, these ratios were not affected by sex of the lambs. On the other hand protein/DNA (hypertrophy) of the slow growing lambs was higher than the fast growing group.

As GT growth progressed, the concentration of total myofibrillar and sarcoplasmic nitrogen and the total of each fraction in the GT increased. The highest value of myofibrillar/sarcoplasmic ratio was observed in the GT muscle of the newborn lambs. Concentration of total and non-protein nitrogen was higher in slow growing lambs than the fast growing group. Rams had lower nitrogen concentrations but higher values for total nitrogen fraction than ewes.

From the results of the present study it is concluded that although Suffolk-sired lambs grew faster and had heavier GT and LD muscles than the Dorset-sired lambs, weight of the adipose tissue depots of the two groups of the lambs were similar. However, because the slow growing lambs were lighter in weight and had similar quantities of adipose tissue, compared to fast growing lambs, the fat depots expressed as a percentage of body weight were greater for the slow growing group. Even so, the unexpected results for the adipose tissue mass in the present study suggests that the two groups did not differ greatly in their genetical propensity toward fatness.

APPENDICES

## APPENDIX 1

TRIS-SUCROSE BUFFER PREPARATION, pH 7.2

| Ingredient | g/liter |
| :--- | :---: |
| 30 mM tris | 3.63 |
| .3 M sucrose | 102.69 |
| 1 mM glutathione (GSH) | .3073 |
| 1 mM EDTA | .3722 |
| Dissolve and dilute to 1 liter with distilled |  |
| water. Adjust pH to 7.2 and store at 2 to 3 C. |  |

## APPENDIX 2

COMPOSITION OF FATTY ACID MIXTURE ${ }^{\text {a }}$

| Fatty acid | Molecular description | $\%$ | $\mathrm{~g} / 100 \mathrm{ml}$ |
| :--- | :---: | :---: | :---: |
| Myristic | 14 | 4.38 | .0216 |
| Palmitic | 16 | 25.58 | .1402 |
| Stearic | 18 | 17.74 | .2372 |
| 0leic | $18: 1$ | 44.57 | .0971 |
| Linoleic | $18: 2$ | 5.86 | .0296 |
| Linolenic | $18: 3$ | 1.84 | .0081 |

$\mathrm{a}_{\text {These }}$ data are for medium weight lambs and has been calculated from the tables reported by Tichenor et al., (1970)

## APPENDIX 3

PREPARATION OF50 mM ISOTONIC COLLIDINE SOLUTION, pH 7.4

| Ingredient | ml |
| :---: | :---: |
| (a) .2 M Collidine ( $2,4,6$-trimethyl pyridine) (. 609 g per 100 ml distilled water) | 37.5 |
| (b) .3 M NaCl <br> (1.753 g per 100 ml distilled water) | 39.4 |
| (c) $.1 \mathrm{M} \underset{(.833 \mathrm{ml}}{\mathrm{HCl}} 12 \mathrm{~N} \mathrm{HCl}$ per 100 ml distilled water) ${ }^{25}$ | $25.0$ |
| (d) Distilled water 4 | 48.1 |
| Adjust to pH 7.4 |  |
| To obtain a $3 \%$ osmium tetroxide solution, dissolve 1 g of $\mathrm{OsO}_{4}$ in 33.3 ml of the above collidine buffer. |  |

CALCULATIONS FOR DETERMINING THE NUMBER AND VOLUME OF FAT CELLS IN COULTER COUNTER
(a) Calculate mean radius ( $r$ ) of the cells in each range by changing $I$ and $A$ in the following equation:
$\mathrm{k}=\frac{\mathrm{V}}{\mathrm{I} \cdot \mathrm{A} \cdot \mathrm{T}} . \quad=\frac{4 / 3 \pi r^{3}}{\mathrm{I} \cdot \mathrm{A} \cdot \mathrm{T}}$.
$\mathrm{V}=\mathrm{volume}$ of the cell ( $\mu^{3}$ )
$\mathrm{r}=$ radius of the cell
I = aperture current setting
A = amplifier setting
$t=$ lower threshold at $50 \%$ count
(b) Subtotal no. of the cells per range $=\mathrm{F} \times$ height of the peak
$F$, the average no. of cells per line $=$ no. of cells in a particular window (this window usually has an average peak) : distance of the peak from the base line.
(c) Sum all the subtotals for part (b)
(d) \% of cells per range $=(b) \times 100$ (c)
(e) Total cells per range $=$ Total no. of cells in total volume $x \frac{(d)}{100}$.

Total no. of fat cells $=$ no. of cell in 2 ml suspension $x$ total volume of suspension/2
(f) Weight in mg of total cells per range $=\{2(a)\}^{3}$. (e). $.4719 \times 10-9$
(g) \% recovery $=$ Sum of (f) Sample weight in mg 100
(h) Adjusted total no. of cells per range $=\frac{(e)}{(g)}$
(i) Volume of cells per range $=(h) \cdot\{(a)\}^{3} \cdot(4 / 3) \cdot \pi$

PREPARATION OF RNA STANDARDS
(a) Dissolve 12.5 mg RNA in $250 \mathrm{ml} 5 \%$ (w/v) PCA This solution contains 50 mg RNA per ml.
(b) Add $12.5 \mathrm{ml} 5 \%$ (w/v) PCA to 37.5 ml of (a).

This solution contains 37.5 mg RNA per ml.
(c) Add $25 \mathrm{ml} \mathrm{5} \mathrm{\%} \mathrm{(w/v)} \mathrm{PCA} \mathrm{to} 25 \mathrm{ml}$ of (a).

This solution contains 25 mg RNA per ml.
(d) Add $37.5 \mathrm{ml} \mathrm{5} \mathrm{\%} \mathrm{(w/v)} \mathrm{PCA} \mathrm{to} 12.5 \mathrm{ml}$ of (a).

This solution contains 12.5 mg RNA per ml .
(e) Store all the above solutions at 2 to 3 C .

## APPENDIX 6 <br> PREPARATION OF DNA STANDARDS

(a) Dissolve 12.5 mg DNA in 250 ml of $10 \%$ ( $\mathrm{w} / \mathrm{v}$ ) PCA. This solution contains 50 mg DNA/ml.
(b) Add $12.5 \mathrm{ml} 10 \%$ (w/v) PCA to 37.5 ml of (a). This solution contains 37.5 mg DNA per ml.
(c) Add 25 ml of $10 \%$ (w/v) PCA to 25 ml of (a). This solution contains 25 mg DNA per ml.
(d) Add 37.5 ml of $10 \%$ (w/v) PCA to 12.5 ml of (a). This solution contains 12.5 mg DNA per ml .
(e) Store all the above solutions at 2 to 3 C .

## APPENDIX 7

PREPARATION OF $1 \%$ (w/v) ORCINOL REAGENT
(a) Make $10 \%$ (w/v) of $\mathrm{FeCl}_{3}$ in concentrated HC1.
(b) Make $.05 \% \mathrm{FeCl}_{3}$ solution by taking 5 ml of (a). and diluting to 1 liter with concentrated HC1 in volumetric flask. This will be stock solution.
(c) Make $1 \%$ orcinol solution by adding 100 ml of (b) to 1 gm orcinol in a volumetric flask and stirring vigorously with a magnetic bar for about 20 min . This solution must be made fresh just prior to use.

## APPENDIX 8

PREPARATION OF 4\% (w/v) DIPHENYLAMINE REAGENT

Make 4\% (w/v) diphenylamine solution by adding 100 ml glacial acetic acid to 4 g of diphenylamine and store at 2 to 3 C .

## APPENDIX 9

PREPARATION OF ACETALDEHYDE SOLUTION
(a) Add . 4 ml of acetaldehyde to a 250 ml volumetric flask.
(b) Bring to 250 ml and store at 2 to 3 C .

## APPENDIX 10

REAGENTS USED IN PROTEIN FRACTIONATION

1. . 015 M Potassium phosphate buffer pH 7.5
$\mathrm{K}_{2} \mathrm{HPO}_{4} \quad 2.16 \mathrm{~g}$
$\mathrm{KH}_{2} \mathrm{PO}_{4} \quad .326 \mathrm{~g}$
Dissolve and dilute to 1 liter with distilled water. Adjust pH to 7.5 and store at 2 to 3 C .
2. 1.1 M KI, . 1 M phosphate buffer pH 7.5

| $\mathrm{K}_{2} \mathrm{HPO}_{4}$ | 14.631 | g |
| :--- | ---: | :--- |
| $\mathrm{KH}_{2} \mathrm{PO}_{4}$ | 2.178 | g |
| KI | 182.6 | g |

Dissolve and dilute to 1 liter with distilled water. Adjust pH to 7.5 and store at 2 to 3 C .
APPENDIX 11. RESULTS OF INTERACTION BETWEEN GROWTH RATE AND SEX ON SOME CHARACTERISTICS OF PERIRENAL,

| Measurements | PERIRENAL FAT |  |  |  | SUBCUTANEOUS FAT |  |  |  |  | INTRAMUSCULAR FAT |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast Growing |  | Slow Growing |  | Pr. | Fast Growing |  | Slow Growing |  | Pr. | Fast Growing |  | Slow Growing |  |  |
|  | Ram | Ewe | Ram | Ewe |  | Ram | Ewe | Ram | Ewe |  | Ram | Ewe | Ram | Ewe |  |
| Adipose tissue weight (g) | 193 | 239 | 195 | 221 | . 64 | 1029 | 773 | 1017 | 854 | . 66 | 9.20 | 7.32 | 9.26 | 9.07 | . 45 |
| Adipose tissue percentage | . 57 | . 84 | . 64 | . 85 | . 63 | 2.39 | 2.27 | 2.72 | 2.78 | . 66 | . 029 | . 024 | . 029 | . 032 | . 21 |
| Chemical composition of adipose tissue: |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Percentage lipid | 74.2 | 78.7 | 75.7 | 77.5 | . 21 | 61.6 | 61.5 | 58.8 | 60.4 | . 65 | 53.8 | 52.6 | 49.0 | 59.0 | . 21 |
| Percentage protein | 3.78 | 3.33 | 3.67 | 3.58 | . 32 | 6.57 | 6.14 | 6.21 | 7.15 | . 13 | 16.24 | 46.72 | 15.15 | 17.66 | . 34 |
| Percentage moisture | 21.7 | 17.7 | 20.8 | 18.8 | . 30 | 32.1 | 32.5 | 35.4 | 32.9 | . 39 | 41.7 | 48.8 | 37.6 | 33.3 | . 09 |
| Nmoles substrate utilized per minute per: |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| mg protein | 2.64 | 2.79 | 2.54 | 2.20 | . 03 | 1.35 | 1.66 | . 99 | 1.06 | . 10 | . 014 | . 004 | . 010 | . 009 | . 14 |
| 8 adipose tissue | 34.6 | 31.0 | 34.8 | 30.0 | . 74 | 11.23 | 9.35 | 8.62 | 8.96 | . 16 | . 103 | . 035 | . 065 | . 055 | . 07 |
| $10^{7}$ adipocytes | 38.4 | 33.7 | 27.7 | 31.2 | . 15 | 28.1 | 19.83 | 10.9 | 22.6 | . 04 | . 303 | . 145 | . 263 | . 135 | . 40 |
| Number of adipocytes per: $g$ tissue ( $\times 10^{6}$ ) | 15.25 | 14.40 | 16.83 | 14.96 | . 56 | 10.20 | 8.35 | 9.81 | 10.83 | . 22 | 3.90 | 3.13 | 2.31 | 4.35 | . 005 |
| Seperated adipose tissue $\left(\times 10^{7}\right)$ | 126.6 | 150.7 | 151.0 | 135.6 | . 14 | 459.1 | 367.6 | 429.5 | 443.1 | . 30 | 261.5 | 183.3 | 140.3 | 205.2 | . 05 |
| Adipocyte diameter ( $\mu \mathrm{m}$ ) | 59.6 | 62.6 | 58.9 | 60.4 | . 86 | 68.6 | 70.5 | 69.8 | 69.4 | . 48 | 57.0 | 56.7 | 58.2 | 56.5 | . 73 |
| Adipocyte diameter ( $\mu^{\mathrm{m}} \times{ }^{3} 0^{4}$ ) | 13.7 | 17.1 | 12.6 | 15.0 | . 73 | 19.9 | 21.2 | 21.3 | 20.2 | . 40 | 10.2 | 10.6 | 10.9 | 10.1 | . 48 |
| Lipid content per adipocyte (ng) | 116.2 | 136.6 | 97.2 | 132.2 | . 56 | 138.4 | 131.7 | 139.9 | 119.1 | . 59 | 9.17 | 9.75 | 15.62 | 7.77 | . 02 |

[^2]

| Measurements | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  | Pre |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |  |
|  | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |  |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| $\begin{array}{lllllllll}\text { Live weight (kg) } & 4.00 & 13.98 & 21.88 & 30.89 & 39.01 & 48.83 & 3.89 & 12.09\end{array}$ |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 20 | 89 | 129 | 122 | 339 | 596 | 16 | 56 | 167 | 113 | 314 | 583 | . 95 |
| Subcutaneous | - | 149 | 280 | 442 | 1276 | 2385 | - | 105 | 311 | 417 | 1315 | 2493 | . 98 |
| Intramuscular | . 26 | 2.95 | 6.33 | 6.34 | 14.98 | 18.78 | . 29 | 2.24 | 5.58 | 5.64 | 12.38 | 28.86 | . 01 |
| Adipose tissue percentage: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | . 49 | . 62 | . 62 | . 38 | . 87 | 1.25 | . 42 | . 46 | . 91 | . 43 | . 90 | 1.34 | . 38 |
| Subcutaneous | - | 1.09 | 1.27 | 1.35 | 3.20 | 4.75 | - | . 85 | 1.65 | 1.65 | 3.94 | 5.65 | . 47 |
| Intramuscular | . 016 | . 028 | . 028 | . 020 | . 037 | . 039 | . 007 | . 018 | . 030 | . 024 | . 039 | . 066 | - |
| Chemical Composition of |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Adipose Tissue: <br> Percentage lipid: |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 33.4 | 83.2 | 84.3 | 79.6 | 87.0 | 91.1 | 34.7 | 76.0 | 86.0 | 83.6 | 87.9 | 91.2 | . 10 |
| Subcutaneous | 2.6 | 67.6 | 75.4 | 68.0 | 77.9 | 77.9 | 4.4 | 50.3 | 72.8 | 70.4 | 76.2 | 83.5 | . 02 |
| Intramuscular | - | - | - | - | 49.5 | 56.9 | - | - | - | - | 46.7 | 61.3 | . 42 |
| Percentage protein: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 10.99 | 2.32 | 1.82 | 2.77 | 1.89 | 1.55 | 11.34 | 2.63 | 1.79 | 2.56 | 1.74 | 1.67 | . 91 |
| Subcutaneous | 17.90 | 4.47 | 4.17 | 4.68 | 3.17 | 3.75 | 17.25 | 6.57 | 4.51 | 4.81 | 3.86 | 3.06 | . 52 |
| Intranuscular | - | - | - | - | 17.22 | 17.43 | - | - | - | - | 16.02 | 16.79 | . 42 |
| Percentage moisture: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 53.4 | 15.5 | 12.9 | 18.0 | 11.3 | 7.1 | 52.8 | 20.7 | 12.2 | 14.4 | 11.2 | 7.3 | . 22 |
| Subcutaneous | 79.9 | 27.9 | 20.8 | 27.6 | 19.3 | 18.2 | 80.1 | 42.6 | 22.6 | 25.6 | 20.5 | 13.7 | . 03 |
| Intramuscular | - | - | - | - | 45.3 | 35.5 | - | - | - | - | 44.4 | 30.7 | . 55 |
| Nmoles Substrate Utilized |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Per Minute Per: mg protein: |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 1.34 | 3.10 | 3.39 | 2.65 | 3.05 | 2.78 | 1.66 | 2.21 | 2.10 | 2.93 | 2.83 | 2.84 | . 001 |
| Subcutaneous | . 01 | 1.55 | 1.42 | 1.37 | 1.50 | 1.67 | . 01 | . 78 | . 79 | 1.49 | 1.64 | 1.47 | . 002 |
| Intramuscular | - | - | - | - | . 009 | . 010 | - | - | - | - | . 013 | . 006 | . 16 |
| g adipose tissue: 66.3 l 27.0 |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 66.3 | 27.0 | 28.0 | 28.2 | 25.5 | 21.8 | 81.2 | 28.2 | 16.2 | 28.8 | 22.5 | 17.7 | . 001 |
| Subcutaneous | . 13 | 12.55 | 9.83 | 11.38 | 12.15 | 15.70 | . 10 | 5.65 | 5.95 | 15.18 | 13.43 | 12.45 | . 003 |
| Intramuscular | - | - | - | - | . 065 | . 073 | - | - | - | - | . 080 | . 040 | . 13 |
| $\frac{10^{\prime} \text { adipocytes: }}{\text { Perirenal }}$ | 15.8 | 17.5 | 19.90 | 38.2 | 52.5 | 72.3 | 19.9 | 14.5 | 12.3 | 32.1 | 37.1 | 60.9 | . 45 |
| Subcutaneous | - | 8.4 | 6.0 | 18.2 | 30.4 | 56.7 | - | 3.2 | 3.7 | 21.7 | 39.0 | 41.1 | . 03 |
| Intramuscular | - | - | - | - | . 145 | . 353 | - | - | - | - | . 270 | . 128 | . 02 |

APPENDIX 12. ( con't $^{\prime}$ )

| Measurements | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  | Pr . |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |  |
|  | Age (days) |  |  |  |  |  | Age(days) |  |  |  |  |  |  |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Number of Adipocytes Per: g tissue ( $\times 10^{6}$ ): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 42.02 | 15.44 | 14.31 | 9.08 | 5.16 | 3.24 | 41.49 | 19.25 | 14.49 | 10.56 | 6.25 | 3.36 | . 82 |
| Subcutaneous | - | 15.75 | 17.47 | 6.43 | 3.96 | 2.77 | - | 22.48 | 15.86 | 6.54 | 3.64 | 3.08 | . 90 |
| Intramuscular | - | - | - | - | 4.78 | 2.25 | - | - | - | - | 3.75 | 2.91 | . 06 |
| Separated adipose tissue (x107): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 81.5 | 126.9 | 182.7 | 106.7 | 169.2 | 164.8 | 66.8 | 101.8 | 236.5 | 104.0 | 173.6 | 176.9 | . 62 |
| Subcutaneous | - | 167.1 | 488.8 | 286.2 | 492.5 | 632.2 | - | 223.0 | 493.2 | 253.0 | 463.8 | 748.5 | . 86 |
| Intramuscular | - | - | - | - | 259.8 | 149.0 | - | - | - | - | 159.7 | 187.8 | . 02 |
| Adipocyte Diameter ( $\mu \mathrm{m}$ ) : |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 36.7 | 51.6 | 52.4 | 60.2 | 75.2 | 90.2 | 37.4 | 48.8 | 52.9 | 58.4 | 70.8 | 86.8 | . 90 |
| Subcutaneous | - | 52.4 | 52.1 | 69.6 | 81.7 | 91.9 | - | 50.9 | 52.1 | 71.5 | 84.7 | 88.8 | . 78 |
| Intramuscular | ) | - | - | - | 50.6 | 63.2 | - | - | - | - | 49.6 | 65.1 | . 47 |
| Adipocyte Volume ( $\mu \mathrm{m}^{3} \times 10^{4}$ ) : |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 2.6 | 7.3 | 7.6 | 11.8 | 22.9 | 40.1 | 2.7 | 6.2 | 7.9 | 10.9 | 19.0 | 36.0 | . 90 |
| Subcutaneous | - | 7.7 | 6.6 | 18.1 | 29.4 | 40.9 | - | 7.0 | 7.6 | 20.0 | 32.4 | 36.9 | . 57 |
| Intramuscular | - | - | - | - | 7.02 | 13.8 | - | - | - | - | 6.5 | 14.5 | . 51 |
| Lipid Content Per Adipocyte (ng): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 8.1 | 57.3 | 60.1 | 102.6 | 180.6 | 349.6 | 8.5 | 42.6 | 63.5 | 103.9 | 151.5 | 318.1 | . 92 |
| Subcutaneous | - | 47.4 | 48.5 | 81.9 | 212.7 | 284.7 | - | 29.1 | 42.8 | 126.8 | 221.8 | 226.9 | . 85 |
| Intramuscular | - | - | - | - | 6.07 | 12.85 | - | - | - | - | 7.98 | 15.40 | . 85 |

[^3]appendix 13. results of interaction between age and sex on some characteristics of perirenal

| Measurements | Ram |  |  |  |  |  | Ewe |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Age(days) |  |  |  |  |  | Age (days) |  |  |  |  |  | Pr. |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Live Weight (kg) | 4.17 | 13.38 | 22.26 | 29.71 | 40.67 | 49.52 | 3.72 | 12.70 | 18.22 | 26.48 | 31.83 | 42.86 | . 04 |
| Adipose tissue weight (8) :PerirenalSubcutaneousIntramuscular |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  | 18 | 64 | 119 | 103 | 375 | 484 | 17 | 81 | 177 | 132 | 278 | 695 | . 005 |
|  | - | 108 | 302 | 368 | 1604 | 2732 | - | 146 | 289 | 491 | 1023 | 2120 | . 07 |
|  | . 30 | 2.61 | 7.32 | 6.01 | 15.46 | 23.68 | . 25 | 2.57 | 4.60 | 5.97 | 11.81 | 23.97 | . 57 |
| Adipose tissue percentage:PerirenalSubcutaneousIntramuscular |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  | . 45 | . 45 | . 55 | . 33 | . 91 | . 96 | . 46 | . 63 | . 99 | . 48 | . 86 | 1.63 | . 01 |
|  | - | . 80 | 1.34 | 1.23 | 3.95 | 5.45 | - | 1.15 | 1.57 | 1.76 | 3.19 | 4.94 | . 23 |
|  | . 007 | . 026 | . 033 | . 022 | . 037 | . 048 | . 007 | . 020 | . 025 | . 022 | . 038 | . 056 | . 57 |
| Chemical composition of adipose tissue: <br> Percentage lipid: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 31.2 | 77.7 | 83.2 | 79.7 | 87.9 | 89.7 | 36.8 | 81.5 | 87.2 | 83.5 | 87.0 | 92.6 | . 66 |
| Subcutaneous | 3.8 | 60.8 | 71.9 | 66.7 | 78.2 | 79.9 | 3.3 | 57.1 | 76.3 | 71.7 | 75.9 | 81.5 | . 74 |
| Intramuscular | - | - | - | - | 46.3 | 56.5 | - | - | - | - | 49.9 | 617 | . 86 |
| Percentage protein |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 11.35 | 2.68 | 1.97 | 2.86 | 1.72 | 1.76 | 10.98 | 2.27 | 1.64 | 2.47 | 1.91 | 1.47 | . 92 |
| Subcutaneous | 16.95 | 5.04 | 4.60 | 4.96 | 3.26 | 3.53 | 18.21 | 6.00 | 4.08 | 4.53 | 3.77 | 3.28 | . 80 |
| Intramuscular | - | - | - | - | 15.58 | 15.81 | - | - | - | - | 17.65 | 46.73 | . 32 |
| Percentage moisture: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 56.1 | 20.0 | 14.2 | 18.0 | 10.7 | 8.3 | 50.0 | 16.2 | 10.9 | 14.4 | 11.8 | 6.1 | . 43 |
| Subcutaneous | 80.6 | 34.3 | 23.7 | 28.5 | 18.7 | 16.8 | 79.5 | 36.2 | 19.7 | 24.7 | 21.1 | 15.1 | . 81 |
| Intramuscular | - | - | - | - | 45.1 | 37.3 | - | - | - | - | 44.7 | 28.9 | . 23 |
| Nwoles substrate utilized per minute per: mp protein: |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  | 1.46 | 3.16 | 2.64 | 3.05 | 2.85 | 2.38 | 1.54 | 2.15 | 2.85 | 2.54 | 3.03 | 2.89 | . 002 |
| Subcutaneous | . 01 | 1.09 | 1.04 | 1.51 | 1.72 | 1.63 | . 01 | 1.20 | 1.72 | 1.35 | 1.43 | 1.51 | . 60 |
| Int ramuscular | - | - | - | - | . 016 | . 009 | - | - | - | - | . 007 | . 006 | . 32 |
| g adipose tissue: Perirenal | 71.2 | 35.2 | 24.3 | 32.5 | 22.8 | 22.2 | 76.3 | 20.0 | 19.8 | 24.5 | 25.2 | 17.3 | . 01 |
| Subcutaneous | . 15 | 8.5 | 7.8 | 14.5 | 12.4 | 16.1 | . 07 | 9.7 | 8.0 | 12.0 | 13.2 | 12.0 | . 32 |
| Intramuscular | - | - | - | - | . 098 | . 070 | - | - | - | - | . 047 | . 043 | . 42 |

APPENDIX 13. ( con't $^{\prime}$ )

| Measurements | Ram |  |  |  |  |  | Ewe |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Age (days) |  |  |  |  |  | Age(days) |  |  |  |  |  |  |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Nmole substrate utilized per minute per: $10^{7}$ adipocytes: |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 16.3 | 19.7 | 16.4 | 43.0 | 46.8 | 56.2 | 19.4 | 12.2 | 15.8 | 27.4 | 42.8 | 77.1 | . 01 |
| Subcutaneous | - | 5.3 | 4.4 | 18.5 | 39.6 | 54.5 | - | 6.2 | 5.3 | 21.4 | 29.8 | 43.3 | . 18 |
| Intramuscular | - | - | - | - | . 300 | . 320 | - | - | - | - | . 118 | . 162 | . 83 |
| Number of adipocytes per: <br> tissue (x106): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 43.58 | 18.33 | 15.18 | 9.92 | 5.21 | 4.01 | 39.93 | 16.36 | 13.62 | 9.72 | 6.19 | 2.59 | . 81 |
| Subcutaneous | - | 18.22 | 18.18 | 7.34 | 3.2 | 3.08 | - | 20.00 | 15.15 | 5.62 | 4.40 | 2.77 | . 66 |
| Intramuscular | - | - | - | - | 4.08 | 2.13 | - | - | - | - | 4.45 | 3.03 | . 55 |
| Separated adipose tissue ( $\times 10^{7}$ ): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 79.6 | 103.9 | 186.1 | 96.5 | 181.1 | 185.6 | 68.7 | 124.9 | 233.1 | 114.2 | 161.7 | 156.2 | . 56 |
| Subcutaneous | - | 152.2 | 495.5 | 237.3 | 522.8 | 813.7 | - | 238.0 | 486.5 | 301.8 | 433.5 | 467.0 | . 23 |
| Intramuscular | - | - | - | - | 255.8 | 146.0 | - | - | - | - | 197.7 | 190.8 | . 15 |
| Adipocyte diameter ( $\mu \mathrm{m}$ ) |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 35.6 | 48.7 | 51.5 | 59.8 | 75.4 | 80.7 | 37.5 | 51.8 | 53.9 | 58.9 | 70.7 | 96.2 | . 01 |
| Subcutaneous | - | 50.6 | 52.0 | 68.6 | 84.4 | 90.3 | - | 52.7 | 52.2 | 72.5 | 81.9 | 90.9 | . 78 |
| Intramuscular | - | S | - | - | 50.9 | 64.3 | - | - | - | - | 49.2 | 63.9 | . 75 |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal . | 2.6 | 6.1 | 7.2 | 11.8 | 23.1 | 28.1 | 2.8 | 7.4 | 8.3 | 10.9 | 18.8 | 47.9 | . 001 |
| Subcutaneous | - | 6.9 | 6.9 | 17.7 | 32.4 | 39.1 | - | 7.8 | 7.3 | 20.3 | 29.4 | 38.6 | . 79 |
| Intramuscular | - | - | - | - | 7.1 | 14.0 | - | - | - | - | 6.5 | 14.2 | . 64 |
| Lipid content per adipocytes (ng): |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Perirenal | 7.3 | 46.3 | 56.3 | 180.0 | 183.6 | 246.7 | 9.4 | 53.6 | 67.3 | 106.5 | 148.5 | 420.9 | . 001 |
| Subcutaneous | - | 37.6 | 37.8 | 106.1 | 248.4 | 265.8 | - | 38.9 | 53.5 | 102.7 | 186.1 | 245.8 | . 39 |
| Intramuscular | - | - | - | - | 7.38 | 17.40 | - | - | - | - | 6.67 | 10.85 | .10 |

[^4]APPENDIX 14. RESULTS OF INTERACTION BETWEEN GROWTH RATE AND SEX ON SOME CHARACTERISTICS OF GASTROCNEMIUS (GT) AND LONGISSIMUS MUSCLES ${ }^{\text {a }}$

| Gastrocnemius (GT) muscle data | GROWTH RATE |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast Growing |  | Slow Growing |  |  |
|  | Sex |  | Sex |  |  |
|  | Ram | Ewe | Ram | Ewe |  |
| Live weight (kg) | 28.67 | 24.20 | 24.56 | 21.07 | . 56 |
| GT weight (g) | 108.5 | 94.0 | 91.9 | 80.4 | . 69 |
| GT Percentage | . 39 | . 40 | . 39 | . 40 | . 47 |
| Percentage fat | 4.01 | 3.27 | 3.35 | 3.10 | 26 |
| Percentage protein | 18.54 | 19.53 | 19.61 | 19.46 | . 001 |
| Percentage moisture | 75.93 | 75.77 | 75.80 | 75.55 | . 87 |
| Concentration of nitrogen(mg/g) | 29.8 | 30.8 | 30.9 | 31.1 | . 10 |
| Concentration of myofibrillar nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 14.9 | 15.4 | 15.5 | 15.5 | . 13 |
| Concentration of sarcoplasmic nitrogen (mg/g) | 5.57 | 5.77 | 5.76 | 5.82 | . 33 |
| Concentration of stroma nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 5.58 | 5.62 | 5.58 | 5.73 | . 79 |
| Concentration of non-protein nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 3.84 | 3.97 | 4.03 | 4.01 | . 09 |
| Total nitrogen (mg) | 3360 | 2960 | 2930 | 2600 | . 87 |
| Total myofibrillar nitrogen(mg) | 1700 | 1500 | 1480 | 1310 | . 89 |
| Total sarcoplasmic nitrogen(mg) | 630 | 570 | 560 | 500 | . 95 |
| Total stroma nitrogen (mg) | 600 | 510 | 510 | 460 | . 50 |
| Total non-protein nitrogen (mg) | 430 | 380 | 380 | 330 | . 87 |
| \% myofibrillar nitrogen | 49.9 | 49.7 | 49.9 | 49.7 | . 96 |
| \% sarcoplasmic nitrogen | 18.4 | 18.5 | 18.4 | 18.7 | . 74 |
| \% stroma nitrogen | 18.7 | 18.9 | 18.8 | 18.5 | . 79 |
| \% non-protein nitrogen | 13.0 | 12.8 | 12.9 | 13 | . 39 |
| Myofibrillar nitrogen/ sarcoplasmic nitrogen | 2.70 | 2.70 | 2.71 | 2.70 | . 90 |
| Concentration of RNA ( $\mathrm{mg} / \mathrm{g}$ ) | 5.10 | 4.95 | 4.83 | 5.14 | . 51 |
| Concentration of DNA ( $\mathrm{mg} / \mathrm{g}$ ) | 1.89 | 1.77 | 1.75 | 1.78 | . 30 |
| Total RNA (mg) | 453 | 363 | 353 | 343 | . 48 |
| Total DNA (mg) | 168 | 139 | 131 | 114 | . 40 |
| Protein/DNA | 113.7 | 128.7 | 131.8 | 129.1 | . 07 |
| RNA/DNA | 2.76 | 2.77 | 2.76 | 2.92 | . 72 |
| Total number of nuclei ( $\times 10{ }^{9}$ ) | 26.85 | 22.45 | 21.08 | 18.47 | . 46 |
| Weight/nucleus ( $\times 10^{-8}$ g) | 37.4 | 39.8 | 40.9 | 40.2 | . 29 |

APPENDIX 14. (con't)

${ }^{\text {a Means }}$ are average of 18 lambs.
Pr. $=$ Probability for level of significance.
APPENDIX 15. RESULTS OF INTERACTION BETWEEN GROWTH RATE AND AGE ON SOME CHARACTERISTICS

| Gastrocnemius (GT) muscle data | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |  |
|  | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |  |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Live weight (kg) | 4.00 | 13.98 | 21.88 | 30.89 | 39.01 | 48.83 | 3.89 | 12.09 | 18.60 | 25.29 | 33.49 | 43.54 | . 32 |
| GT weight (g) | 15.0 | 70.5 | 88.1 | 116.3 | 143.5 | 173.9 | 16.2 | 56 | 76.6 | 98.4 | 124.3 | 145.5 | . 35 |
| GT percentage | . 38 | . 50 | . 40 | . 37 | . 36 | . 35 | . 41 | . 46 | . 41 | . 39 | . 37 | . 34 | . 11 |
| Percentage fat | 1.02 | 3.34 | 4.04 | 3.72 | 3.96 | 5.76 | . 88 | 3.71 | 2.98 | 3.39 | 3.99 | 4.38 | . 18 |
| Percentage protein | 16.6 | 19.1 | 19.2 | 19.7 | 19.9 | 19.8 | 16.9 | 19.1 | 19.8 | 20.1 | 20.3 | 21 | . 37 |
| Percentage moisture | 79.7 | 76.2 | 75.6 | 76.2 | 74.3 | 72.8 | 79.1 | 76 | 76.3 | 75.9 | 73.8 | 72.9 | . 81 |
| Concentration of nitrogen (mg/g) | 26.5 | 29.1 | 30.7 | 31.5 | 31.8 | 32.3 | 27 | 29.5 | 31.3 | 32.1 | 32.2 | 33.7 | . 83 |
| Concentration of myofibrillar nitrogen (mg/g) | 12.5 | 14.5 | 14.9 | 15.7 | 16.8 | 16.5 | 12.9 | 14.7 | 16 | 15.9 | 17 | 16.6 | . 77 |
| Concentration of sarcoplasmic nitrogen (mg/g) | 4.39 | 5.68 | 6.05 | 5.45 | 5.96 | 6.30 | 4.41 | 5.94 | 5.67 | 5.90 | 6.31 | 5.53 | .19 |
| Concentration of stroma nitrogen (mg/g) | 6.01 | 4.97 | 5.93 | 6.38 | 4.90 | 5.46 | 5.98 | 5.03 | 5.54 | 6.12 | 4.71 | 6.54 | . 28 |
| Concentration of non-protein nitrogen (mg/g) | 3.53 | 3.89 | 3.73 | 4.02 | 4.21 | 4.0 | 3.65 | 3.92 | 4.18 | 4.19 | 6.22 | 3.98 | . 10 |
| Total nitrogen (mg) | 400 | 2050 | 2700 | 3660 | 4550 | 5600 | 430 | 1670 | 2400 | 3160 | 4000 | 4890 | .67 |
| Total myofibrillar nitrogen(mg) | 190 | 1020 | 1320 | 1810 | 2400 | 2870 | 210 | 830 | 1230 | 1560 | 2110 | 2420 | . 33 |
| Total sarcoplasmic nitrogen(mg) | 70 | 400 | 530 | 640 | 850 | 1090 | 70 | 340 | 430 | 580 | 790 | 950 | . 64 |
| Total stroma nitrogen (mg) | 90 | 360 | 520 | 740 | 700 | 950 | 90 | 280 | 420 | 610 | 580 | 940 | . 68 |

APPENDIX 15. ( $\operatorname{con}^{\prime} \mathrm{t}$ )

| Gastrocnemius muscle data | GROWTH RATE |  |  |  |  |  |  |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Fast Growing |  |  |  |  |  | Slow Growing |  |  |  |  |  |  |
|  | Age(days) |  |  |  |  |  | Age(days) |  |  |  |  |  |  |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Total non-protein nitrogen (mg) | 50 | 270 | 330 | 470 | 600 | 690 | 60 | 220 | 320 | 410 | 520 | 580 | . 22 |
| Percentage myofibillar nitrogen | 47.8 | 47.7 | 49.2 | 49.7 | 52.8 | 51.8 | 48.3 | 48.2 | 50.8 | 49.8 | 52.3 | 49.3 | . 50 |
| Percentage sarcoplasmic nitrogen | 16.8 | 18.5 | 19.8 | 17.5 | 18.7 | 19.7 | 16.5 | 19.5 | 18 | 18.3 | 19.5 | 19.5 | . 13 |
| Percentage stroma nitrogen | 22 | 20.8 | 19.2 | 19.8 | 15.2 | 15.8 | 21.3 | 19.0 | 18.0 | 18.8 | 15.2 | 19.2 | . 72 |
| Percentage non-protein nitrogen | 13.3 | 12.8 | 12.2 | 13 | 13.3 | 12.7 | 13.8 | 12.8 | 13.2 | 13 | 13 | 12 | . 34 |
| Myofibillar nitrogen/Sarcoplasmic nitrogen | 2.85 | 2.57 | 2.48 | 2.88 | 2.81 | 2.62 | 2.98 | 2.49 | 2.82 | 2.71 | 2.70 | 2.55 | . 08 |
| Concentration of RNA (mg/g) | 10.45 | 4.03 | 4.20 | 3.94 | 4.11 | 3.41 | 10.51 | 4.01 | 3.37 | 4.97 | 3.96 | 3.09 | . 73 |
| Concentration of DNA (mg/g) | 3.30 | 1.67 | 1.94 | 1.30 | 1.43 | 1.32 | 3.56 | 1.74 | 1.42 | 1.42 | 1.28 | 1.14 | . 06 |
| Total RNA (mg) | 159 | 279 | 367 | 457 | 594 | 593 | 174 | 230 | 256 | 483 | 492 | 452 | . 48 |
| Total DNA (mg) | 49 | 115 | 168 | 150 | 208 | 231 | 57 | 108 | 107 | 139 | 158 | 167 | . 02 |
| Protein/DNA | 51.1 | 117.1 | 104.0 | 157.6 | 140.6 | 150.4 | 48.1 | 104.2 | 143.9 | 141.7 | 160.5 | 184.2 | . 001 |
| RNA/DNA | 3.27 | 2.42 | 2.29 | 3.13 | 2.9 | 2.58 | 2.93 | 2.37 | 2.44 | 3.53 | 3.11 | 2.70 | . 92 |
| Total number of nuclei ( $\times 10^{9}$ ) | 7.04 | 18.52 | 27.08 | 24.21 | 33.69 | 37.37 | 9.22 | 17.39 | 17.19 | 22.51 | 25.44 | 26.88 | . 01 |
| Weight/nucleus ( $\times 10^{-8} \mathrm{~g}$ ) | 22 | 20.8 | 19.2 | 19.8 | 15.2 | 15.8 | 21.3 | 19 | 18 | 18.8 | 15.2 | 19.2 | . 72 |
| Longissimus (LD) muscle data |  |  |  |  |  |  |  |  |  |  |  |  |  |
| LD weight (g) | 50 | 162 | 309 | 363 | 600 | 644 | 39 | 157 | 239 | 346 | 467 | 630 | . 25 |
| LD percentage | 1.04 | 1.23 | 1.39 | 1.25 | 1.52 | 1.33 | 1.20 | 1.25 | 1.30 | 1.28 | 1.41 | 1.44 | . 23 |
| Percentage fat | . 62 | 1.71 | 2.1 | 1.64 | 2.46 | 2.83 | . 62 | 1.49 | 2.28 | 1.73 | 2.66 | 4.47 | . 009 |
| Percentage protein | 16.3 | 19.1 | 20.8 | 20.5 | 21.3 | 21.4 | 16.9 | 19.6 | 20.4 | 20.6 | 21.3 | 21.1 | . 42 |
| Percentage moisture | 79.4 | 77.8 | 76.3 | 76.9 | 75.2 | 74.0 | 78.8 | 77.8 | 76.5 | 76.4 | 74.9 | 73.2 | . 69 |

[^5]appendix 16. RESULTS OF interaction between age and sex on some characteristics of

| Gastrocnemius (GT) muscle data | Ram |  |  |  |  |  | Ewe |  |  |  |  |  | Pr. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Age (days) |  |  |  |  |  | Age(days) |  |  |  |  |  |  |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Live weight (kg) | 4.17 | 13.88 | 22.26 | 29.71 | 40.67 | 49.52 | 3.72 | 12.7 | 18.22 | 26.48 | 31.83 | 42.86 | . 04 |
| GT weight | 17.0 | 60.2 | 93.1 | 112.8 | 150.0 | 168.4 | 14.2 | 66.3 | 71.6 | 102.0 | 118.0 | 151.0 | . 07 |
| GT percentage | . 40 | . 45 | . 42 | . 38 | . 37 | . 34 | . 38 | . 51 | . 39 | . 38 | . 37 | . 35 | . 02 |
| Percentage fat | . 96 | 3.54 | 3.63 | 4.03 | 4.36 | 5.59 | . 95 | 3.51 | 3.39 | 3.08 | 3.59 | 4.55 | . 57 |
| Percentage protein | 16.7 | 18.9 | 19.2 | 19.6 | 19.7 | 20.4 | 16.7 | 19.3 | 19.8 | 20.2 | 20.4 | 20.5 | . 75 |
| Percentage moisture | 79.7 | 76.5 | 76 | 75.8 | 73.8 | 73.3 | 95.1 | 75.8 | 76.1 | 76.3 | 74.2 | 72.4 | . 48 |
| Concentration of nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 26.7 | 28.9 | 30.6 | 31.3 | 31.9 | 32.7 | 26.7 | 29.7 | 31.4 | 32.3 | 32.1 | 33.2 | . 82 |
| Concentration of myofibrillar nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 12.7 | 14.3 | 15.7 | 15.4 | 16.8 | 16.5 | 12.8 | 14.9 | 15.2 | 16.2 | 16.9 | 16.6 | . 44 |
| Concentration of sarcoplasmic nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 4.49 | 5.53 | 5.76 | 5.58 | 6.17 | 6.27 | 4.31 | 6.08 | 5.96 | 5.76 | 6.10 | 6.56 | . 32 |
| Concentration of stroma nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 5.91 | 5.21 | 5.24 | 6.38 | 4.76 | 5.97 | 6.08 | 4.79 | 6.22 | 6.10 | 4.85 | 6.03 | . 37 |
| Concentration of non-protein nitrogen ( $\mathrm{mg} / \mathrm{g}$ ) | 3.6 | 3.85 | 3.95 | 4.02 | 4.22 | 3.97 | 3.59 | 3.95 | 4.01 | 4.19 | 4.21 | 4.01 | . 84 |
| Total nitrogen (mg) | 460 | 1730 | 2860 | 3520 | 4780 | 5510 | 370 | 1970 | 2250 | 3300 | 3790 | 5000 | . 13 |
| Total myofibrillar nitrogen(mg) | 220 | 850 | 1460 | 1720 | 2510 | 2790 | 180 | 990 | 1090 | 1660 | 2000 | 2510 | . 04 |
| Total sarcoplasmic nitrogen(mg) | 80 | 340 | 540 | 620 | 920 | 1050 | 60 | 400 | 430 | 590 | 720 | 990 | . 07 |
| Total stroma nitrogen (mg) | 100 | 310 | 490 | 730 | 720 | 1000 | 80 | 320 | 440 | 620 | 570 | 890 | . 62 |
| Total non-protein nitrogen (mg) | 60 | 230 | 370 | 450 | 630 | 670 | 50 | 260 | 270 | 430 | 500 | 610 | . 04 |

APPENDIX 16. ( $\operatorname{con}^{\prime} \mathrm{t}$ )

| Gastrocnemius (GT) muscle data | Ram |  |  |  |  |  | Ewe |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Age(days) |  |  |  |  |  | Age (days) |  |  |  |  |  | Pr. |
|  | 0 | 35 | 70 | 105 | 140 | 175 | 0 | 35 | 70 | 105 | 140 | 175 |  |
| Percentage myofibrillar nitrogen | 47.8 | 47.5 | 51.5 | 49.2 | 53.2 | 50.3 | 48.3 | 48.3 | 48.5 | 50.3 | 52.0 | 50.8 | . 35 |
| Percentage sarcoplasmic nitrogen | 16.8 | 18.5 | 19.0 | 17.8 | 19.3 | 19 | 16.5 | 19.5 | 18.8 | 18 | 18.8 | 20.2 | . 58 |
| Percentage stroma nitrogen | 21.5 | 21.3 | 17.2 | 20.2 | 14.2 | 18.3 | 21.8 | 19 | 20 | 18.5 | 16.2 | 16.7 | . 52 |
| Percentage non-protein nitrogen | 13.8 | 12.7 | 12.7 | 12.8 | 13.3 | 12.3 | 13.3 | 13 | 12.7 | 13.2 | 12 | 12.3 | . 86 |
| Myofibrillar nitrogen/sarcoplasmic nitrogen | 2.81 | 2.60 | 2.72 | 2.76 | 2.73 | 2.64 | 3.03 | 2.46 | 2.57 | 2.82 | 2.78 | 2.53 | . 32 |
| Concentration of RNA (mg/g) | 10.13 | 4.32 | 3.86 | 4.13 | 4.06 | 3.29 | 10.83 | 3.73 | 3.72 | 4.78 | 4.02 | 3.8 | . 87 |
| Concentration of DNA (mg/g) | 3.55 | 1.78 | 1.52 | 1.43 | 1.39 | 1.24 | 3.31 | 1.63 | 1.85 | 1.28 | 1.33 | 1.23 | . 29 |
| Total RNA (mg) | 177 | 262 | 358 | 450 | 618 | 554 | 157 | 246 | 265 | 490 | 469 | 490 | . 53 |
| Total DNA (mg) | 60 | 45 | 141 | 160 | 210 | 212 | 46 | 108 | 134 | 130 | 156 | 187 | . 45 |
| Protein/DNA | 47.3 | 100.3 | 135.3 | 139.8 | 146.4 | 167.4 | 52.2 | 121.0 | 112.7 | 159.6 | 154.8 | 167.1 | . 06 |
| RNA/DNA | 2.88 | 2.45 | 2.59 | 3.01 | 2.97 | 2.68 | 3.33 | 2.35 | 2.10 | 3.65 | 2.6 | 2.85 | . 63 |
| Total number of nuclei (x10 ${ }^{9}$ ) | 8.77 | 18.52 | 22.7 | 25.78 | 33.91 | 34.13 | 7.5 | 17.4 | 21.57 | 20.94 | 25.2 | 30.12 | . 41 |
| Weight/nucleus ( $\times 10^{-8} \mathrm{~g}$ ) | 17.6 | 33.1 | 43.2 | 44.1 | 45.9 | 50.9 | 19.3 | 38.7 | 35.7 | 49 | 46.9 | 46.9 | . 10 |
| Longissimus (LD) muscle data |  |  |  |  |  |  |  |  |  |  |  |  |  |
| LD weight (g) | 50 | 162 | 309 | 363 | 600 | 644 | 39 | 157 | 239 | 346 | 467 | 630 | . 18 |
| LD percentage | 1.18 | 1.24 | 1.38 | 1.23 | 1.48 | 1.30 | 1.06 | 1.24 | 1.31 | 1.30 | 1.45 | 1.47 | . 22 |
| Percentage fat | . 61 | 1.56 | 2.42 | 1.64 | 2.53 | 3.53 | . 63 | 1.64 | 1.96 | 1.72 | 2.59 | 3.79 | . 80 |
| Percentage protein | 16.5 | 19.0 | 20.5 | 20.4 | 21.2 | 21.1 | 16.7 | 19.7 | 20.6 | 20.7 | 21.3 | 21.4 | . 90 |
| Percentage moisture | 79.5 | 78.1 | 76.2 | 77.0 | 75.0 | 74.0 | 78.7 | 77.4 | 76.6 | 76.4 | 75.1 | 73.1 | . 23 |

[^6]Pr.-Probability for level of significance.

## APPENDIX 17 NUMBER AND DEFINITION OF VARIABLES USED IN RAW

 data and correlation coefficientsVariable
Number Definition

MUSCLE DATA
Live weight of lamb at slaughter (kg)
38 Weight of gastrocnemius (GT) muscle (g)

Weight of longissimus (L.D.) muscle (g)
Percentage L.D. of live weight
Percentage fat in GT
Percentage protein in GT
Percentage moisture in GT
Percentage fat in L.D.
Percentage protein in L.D.
Percentage moisture in L.D.
Total protein in L.D. (g)
Total protein in GT (g)
Total nitrogen in GT (mg)
Total myofibrillar nitrogen in GT (mg)
Total sarcoplasmic nitrogen in GT (mg)
Total stroma nitrogen in GT (mg)
Total non protein nitrogen in GT (mg)
Concentration of nitrogen in GT (mg/g)
Concentration of myofibrillar nitrogen in GT (mg/g)
Concentration of sarcoplasmic nitrogen in GT ( $\mathrm{mg} / \mathrm{g}$ )
Concentration of non protein nitrogen in GT (mg/g)
Concentration of stroma nitrogen in GT ( $\mathrm{mg} / \mathrm{g}$ )
Precentage myofibrillar nitrogen of total nitrogen
in GT
Percentage sarcoplasmic nitrogen of total nitrogen
in $G T$
Percentage non protein nitrogen of total nitrogen
in GT
Percentage stroma nitrogen of total nitrogen in GT
Myofibrillar/sarcoplasmic ratio
Concentration of RNA in GT (mg/g)
Concentration of DNA in GT (mg/g)
Total RNA in GT (mg)
Total DNA in GT (mg)
Protein/RNA ratio in GT
Protein/DNA ratio in GT
RNAYBIA ratio in $\mathrm{GT}^{-}$
Total number of nuclei in GT ( $\mathrm{x} 10^{9}$ )
GT weight/nucleus (ng)

APPENDIX 17 NUMBER AND DEFINITION OF VARIABLES USED IN RAW (cont.) DATA AND CORRELATION COEFFICIENTS

Variable Number

## LIPID DATA

Weight of perirenal adipose tissue (PAT) (g)
Weight of subcutaneous adipose tissue (SAT) (g) Weight of intramuscular adipose tissue (IAT) (g)
Percentage PAT of live weight
Percentage SAT of live weight
Percentage IAT of live weight
Percentage lipid in PAT
Percentage protein in PAT
Percentage moisture in PAT
Percentage lipid in SAT
Percentage protein in SAT
Percentage moisture in SAT
Percentage lipid IAT
Percentage protein in IAT
Percentage moisture in IAT
Lipid content per PAT adipocyte (ng)
Lipid content per SAT adipocyte (ng)
Lipid content per IAT adipocyte (ng)
mg soluble protein per ml PAT hemogenate
mg soluble protein per ml SAT hemogenate
mg soluble protein per ml IAT hemogenate
nmoles substrate utilized per min per mg protein in PAT
nmoles substrate utilized per min per mg protein in SAT
nmoles substrate utilized per min per mg protein in IAT
nmoles substrate utilized per min per $g$ wet PAT mnoles substrate utilized per min per $g$ wet SAT nmoles substrate utilized per min per g wet IAT nmoles substrate utilized per min per 107 adipocytes in PAT nmoles substrate utilized per min per $10^{7}$ adipocytes in SAT nmoles substrate utilized per min per $10^{7}$ adipocytes in IAT Concentration of adipocytes Der $g$ wet PAT ( $x 100_{6}^{6}$ ) Concentration of adipocytes per $g$ wet SAT ( $\begin{array}{ll} & 106 \text { ) }\end{array}$
Concentration of adipocytes per g wet IAT,
Total number of adipocytes per PAT ( x 10 )
Total number of adipocytes per SAT ( $x$ 107)
Total number of adipocytes per IAT ( $x$ 107)
Diameter of adipocytes in PAT ( $\mu \mathrm{m}$ )
Diameter of adipocytes in SAT ( $\mu \mathrm{m}$ )
Diameter of adipocytes in IAT ( $\mu \mathrm{m}$ )
Volume of adipocytes in PAT ( $\mu \mathrm{m} \mathrm{m}^{3} \times 10^{4}$ )
Volume of adipocytes in SAT $\left(\mu \mathrm{m}^{3} \times 10^{4}\right)$
Volume of adipocytes in IAT ( $\mu \mathrm{m}^{3} \times 10^{4}$ )

|  | 174 |
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| a |  <br>  <br> 11 |
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| $m$ |  |
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APPENDIX 18 (cont.

| Var <br> No. | able 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 |
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| 71 | -. 20 | -. 24 | .00 | $-.34$ | $-.29$ | .21 | . 20 | -. 23 | -. 30 | -. 25 | -. 29 | .34 | . 38 | -. 28 | -. 13 |
| 72 | -. 24 | .13 | .06 | -. 18 | -. 34 | .29 | . 30 | . . 41 | -. 31 | -. 38 | -. 34 | .29 | . 25 | -. 25 | -. 26 |
| 73 | . 55 | .01 | .07 | . 65 | . 78 | .46 | .50 | . 75 | . 68 | . 74 | .76 | -. 74 | -. 67 | . 52 | . 61 |
| 74 | . 34 | . 22 | .41 | .24 | .40 | .17 | . 20 | . 20 | -. 03 | .45 | .40 | -. 04 | -. 18 | $-.09$ | . 39 |
| 75 | .17 | $-.07$ | . 18 | .10 | .21 | .02 | -. 06 | .26 | -. 18 | .21 | . 21 | -. 16 | $-.12$ | -. 22 | .15 |
| 76 | -. 58 | -. 20 | $-.04$ | -. 38 | -. 35 | $-.47$ | -. 14 | -. 48 | $-.40$ | -. 49 | -. 35 | -. 04 | .05 | -. 42 | -. 61 |
| 77 | . 05 | . 04 | . 03 | $-.03$ | .01 | -. 13 | . 05 | .11 | -. 21 | . 03 | . 01 | $-.12$ | $-.14$ | -. 22 | .05 |
| 78 | . 88 | .12 | .01 | .77 | .80 | . 66 | . 34 | . 86 | .82 | . 81 | . 80 | $-.44$ | -. 44 | .78 | .74 |
| 79 | . 65 | . 08 | .01 | .62 | .89 | .57 | . 38 | . 73 | .71 | .66 | .69 | -. 55 | -. 51 | -. 41 | . 52 |









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APPENDIX

| APPENDIX | 18 | t |  |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Variable |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| No. 16 | 17 | 18 | 19 | 20 | 21 | 22 | 23 | 24 | 25 | 26 | 27 | 28 | 29 | 30 |




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APPENDIX 18 (cont.)

| Variable |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
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| No. 31 | 32 | 33 | 34 | 35 | 36 | 37 | 38 | 39 | 40 | 41 | 42 | 43 | 44 | 45 |







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APPENDIX 18 (cont.)

| Var No. | able <br> 31 | 32 | 33 | 34 | 35 | 36 | 37 | 38 | 39 | 40 | 41 | 42 | 43 | 44 | 45 |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 66 | -. 03 | . 37 | -. 25 | -. 16 | . 66 | . 46 | -. 04 | . 96 | . 73 | -. 66 | -. 73 | . 70 | -. 64 | -. 71 | . 28 |
| 67 | -. 23 | . 12 | -. 48 | -. 31 | . 66 | . 44 | -. 09 | . 65 | . 76 | -. 73 | -. 76 | . 76 | -. 74 | -. 76 | . 19 |
| 68 | -. 02 | -. 06 | . 55 | . 39 | -. 76 | -. 55 | . 18 | -. 70 | -. 90 | . 89 | . 89 | -. 92 | . 91 | . 92 | -. 55 |
| 69 | . 25 | -. 04 | -. 08 | . 25 | . 32 | . 47 | -. 49 | . 39 | . 26 | -. 31 | -. 25 | . 29 | -. 29 | -. 30 | -. 34 |
| 70 | -. 22 | -. 29 | -. 19 | -. 74 | , 83 | , 31 | -. 45 | . 41 | . 45 | -. 45 | -. 44 | . 52 | -. 53 | -. 54 | . 00 |
| 71 | . 19 | -. 15 | . 23 | . 21 | -. 26 | . 27 | -. 32 | -. 27 | -. 34 | . 34 | . 35 | -. 31 | . 28 | . 33 | -. 19 |
| 72 | -. 14 | . 15 | . 08 | . 05 | -. 49 | -. 52 | . 58 | -. 37 | -. 28 | . 31 | . 27 | -. 34 | . 34 | . 34 | . 27 |
| 73 | . 24 | -. 02 | -. 34 | -. 05 | . 77 | . 76 | . 29 | . 76 | . 77 | -. 77 | $-.75$ | . 79 | -. 76 | -. 79 | -. 23 |
| 74 | -. 45 | -. 17 | . 29 | -. 05 | . 02 | -. 24 | -. 11 | . 44 | -. 03 | . 22 | . 02 | -. 02 | -. 20 | -. 03 | -. 16 |
| 75 | . 09 | -. 24 | . 10 | . 19 | -. 12 | -. 15 | -. 18 | . 25 | -. 07 | . 23 | . 06 | -. 26 | -. 15 | . 24 | -. 05 |
| 76 | . 31 | . 40 | -. 03 | -. 17 | -. 09 | -. 12 | -. 27 | -. 35 | -. 11 | . 04 | . 12 | -. 04 | -. 02 | . 06 | -. 02 |
| 77 | . 08 | -. 51 | . 27 | . 08 | . 03 | -. 06 | -. 16 | . 05 | -. 17 | . 27 | . 16 | -. 27 | -. 12 | . 28 | . . 16 |
| 78 | . 31 | . 33 | -. 19 | -. 18 | . 58 | . 25 | . 00 | . 78 | . 57 | -. 48 | -. 57 | . 54 | -. 47 | -. 56 | . 38 |
| 79 | . 25 | . 27 | -. 24 | -. 27 | . 65 | . 27 | -. 09 | . 66 | . 64 | -. 56 | -. 64 | . 63 | -. 60 | -. 64 | . 32 |

APPENDIX 18 (cont.)

|  | $\begin{gathered} \text { iable } \\ 61 \\ \hline \end{gathered}$ | 62 | 63 | 64 | 65 | 66 | 67 | 68 | 69 | 70 | 71 | 72 | 73 | 74 | 75 |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 61 | 1.00 |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| 62 | -. 40 | 1.00 |  |  |  |  |  |  |  |  |  |  |  |  |  |
| 63 | . 79 | . 20 | 1.00 |  |  |  |  |  |  |  |  |  |  |  |  |
| 64 | . 46 | . 06 | . 62 | 1.00 |  |  |  |  |  |  |  |  |  |  |  |
| 65 | . 57 | . 05 | . 74 | . 70 | 1.00 |  |  |  |  |  |  |  |  |  |  |
| 66 | . 73 | . 47 | . 96 | . 82 | . 79 | 1.00 |  |  |  |  |  |  |  |  |  |
| 67 | . 76 | -. 12 | . 64 | . 20 | . 51 | . 61 | 1.00 |  |  |  |  |  |  |  |  |
| 68 | -. 79 | -. 19 | -. 69 | -. 21 | -. 49 | -. 62 | -. 75 | 1.00 |  |  |  |  |  |  |  |
| 69 | . 35 | . 17 | . 43 | . 43 | . 24 | . 43 | . 17 | -. 25 | 1.00 |  |  |  |  |  |  |
| 70 | . 38 | -. 16 | . 42 | . 20 | . 34 | . 41 | . 39 | -. 50 | . 40 | 1.00 |  |  |  |  |  |
| 71 | -. 23 | . 05 | -. 21 | . 01 | -. 30 | -. 23 | -. 31 | . 30 | . 37 | -. 03 | 1.00 |  |  |  |  |
| 72 | -. 35 | -. 07 | -. 41 | -. 39 | -. 23 | . 40 | -. 21 | . 29 | -. 93 | -. 65 | -. 48 | 1.00 |  |  |  |
| 73 | . 80 | . 16 | . 79 | . 64 | . 54 | . 75 | . 62 | -. 71 | . 68 | . 49 | -. 10 | -. 64 | 1.00 |  |  |
| 74 | . 00 | -. 31 | . 39 | . 28 | -. 05 | . 19 | . 11 | . 17 | . 03 | -. 01 | -. 14 | . 02 | -. 03 | 1.00 |  |
| 75 | -. 17 | . 18 | . 21 | . 25 | -. 12 | . 26 | . 03 | -. 03 | . 13 | -. 13 | . 00 | -. 05 | . 03 | . 76 | 1.00 |
| 76 | -. 25 | . 35 | -. 36 | -. 55 | -. 41 | -. 47 | -. 07 | -. 03 | -. 03 | . 31 | -. 49 | -. 51 | -. 26 | -. 06 | . 08 |
| 77 | -. 09 | . 09 | . 01 | . 17 | -. 14 | . 10 | . 04 | . 03 | . 17 | . 04 | . 03 | -. 14 | . 11 | . 69 | . 93 |
| 78 | . 60 | . 40 | . 76 | . 75 | . 82 | . 87 | . 53 | -. 50 | -. 30 | . 36 | . 31 | . 27 | . 61 | -. 09 | . 03 |
| 79 | . 61 | . 43 | . 64 | . 55 | . 70 | . 74 | . 54 | -. 60 | . 25 | . 44 | -. 40 | -. 25 | . 62 | -. 28 | -. 07 |

APPENDIX 18 (cont.)

|  | 182 |
| :---: | :---: |
| $0$ |  <br>  $\qquad$ I <br> 11 <br> I |
| $\begin{aligned} & \infty \\ & n \end{aligned}$ |  <br>  $\qquad$ |
| $\cdots$ |  <br>  - $\qquad$ <br> 1 <br> 11 <br> 111 |
| N |  <br>  <br> $\stackrel{\square}{\bullet}$ |
| o |  <br>  - i i i i i <br> 11111 <br> 11 |
| $\sim$ |  <br>  $\infty$ - |
| $\stackrel{\downarrow}{\sim}$ |  <br>  <br> $\rightarrow 11$ |
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| $N$ |  <br>  <br> - $\quad i$ |
| $\stackrel{\sim}{n}$ |  ON. <br> $\rightarrow$ |
| $\bigcirc$ |  <br>  <br> $-$ |
| on $\pm$ |  <br>  $\rightarrow$ $\qquad$ 11 <br> I <br> 1 I |
| $\cdots$ |  <br>  $\mapsto$ |
| $\pm$ |  <br>  |
| $$ |  <br>  |

APPENDIX 18 (cont.)

| Variable |  |  |  |
| :--- | ---: | ---: | ---: |
| No. | 76 | 77 | 78 |

APPENDIX 19-A ALLOTMENT OF LAMBS BY NUMBER TO GROWTH RATE GROUP, SEX AND AGE

| $\text { Age (days) } 0$ |  | 35 |  | 70 |  | 105 |  | 140 |  | 175 |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fast | Growing |  |  |  |  |  |  |  |
| No. Ram | Ewe |  |  | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe |  |
| 1 | 9 | 17 | 14 | 28 | 26 | 39 | 38 | 50 | 53 | 65 | 62 |  |
| 3 | 11 | 21 | 15 | 29 | 27 | 46 | 43 | 52 | 56 | 67 | 69 |  |
| 4 | 12 | 22 | 19 | 30 | 35 | 48 | 44 | 55 | 57 | 68 | 71 |  |
|  |  |  |  | Slow | Growing |  |  |  |  |  |  |  |
| 2 | 6 | 16 | 20 | 33 | 31 | 40 | 42 | 54 | 51 | 63 | 64 | $\stackrel{\infty}{+}$ |
| 7 | 10 | 18 | 24 | 34 | 32 | 41 | 45 | 59 | 58 | 66 | 70 |  |
| 8 | 13 | 23 | 25 | 36 | 37 | 49 | 47 | 61 | 60 | 73 | 72 |  |

APPENDIX 19-B

RAW DATA



RAW DATA


RAW DATA

APPENDIX 19－B（cont．）RAW DATA

|  | ¢ |  |  | $\begin{aligned} & \text { जNn } \\ & \text { Nさ~ } \\ & \text { onn m } \\ & \text { min } \end{aligned}$ |
| :---: | :---: | :---: | :---: | :---: |
| $\stackrel{n}{n}$ | $\begin{gathered} \text { 芯 } \\ \end{gathered}$ | $\begin{aligned} & N \sim \infty \\ & i N o \\ & -i N M \end{aligned}$ |  | ヘナに ○ルO N～ن mmm |
|  | $\begin{gathered} 0 \\ 3 \\ \mathbf{3} \end{gathered}$ | $\begin{aligned} & \text { Nito } \\ & \text { Nin } \\ & \text { Nin } \end{aligned}$ |  |  |
| $\begin{aligned} & 0 \\ & \underset{-}{\circ} \end{aligned}$ | $\begin{aligned} & \text { EI } \\ & \text { ® } \end{aligned}$ | $\begin{aligned} & \text { Hon } \\ & \text { Fio } \\ & \text { N } \\ & \text { m } \end{aligned}$ |  | $\begin{aligned} & \text { an m } \\ & \text { mon } \\ & \text { rin } \\ & \text { n m } \end{aligned}$ |
|  | $\begin{gathered} 0 \\ 3 \\ \mathbf{3} \end{gathered}$ | $\begin{aligned} & \text { Mホの } \\ & \text { ñ~ } \\ & \text { mलn } \end{aligned}$ |  |  |
| $\stackrel{n}{0}$ | 范 |  |  | $\begin{aligned} & \infty \text { nN } \\ & \sim N \\ & \text {-iN } \end{aligned}$ |
|  |  | n $0 \infty$ nnin लंलंल <br> moN $0 \infty$ $\infty-\dot{0}$ NMN | Slow Growing | $\begin{aligned} & \text { oon } \\ & 00 \text { n } \\ & \text { non } \\ & \text { no } \\ & \text { in } \\ & \text { लंल } \end{aligned}$ |




$\begin{array}{ll}29.00 & 21.86 \\ 25.82 & 17.83 \\ 19.26 & 21.32\end{array}$


19
20
－
$\begin{array}{ll}15.90 & 16.22 \\ 18.03 & 19.96 \\ 17.34 & 15.71\end{array}$
電







Age（days）
Variable
no．
1925


| Age（days） |  |  | 35 |  |
| :---: | :---: | :---: | :---: | :---: |
| Variable |  |  |  |  |
| no． | Ram | Ewe | Ram | Ewe |
| 19 | 25.97 | 27.84 | 28.42 | 28.54 |
|  | 24.26 | 26.99 | 28.67 | 30.96 |
|  | 27.62 | 26.33 | 28.46 | 29.34 |

RAW DATA


APPENDIX 19－B（cont．）RAW DATA

| $\stackrel{0}{\substack{3}}$ | $\begin{aligned} & 0 \text { ợ } \\ & \text { 于 } \\ & \text { inici } \end{aligned}$ |  | ヘ̂กn $\rightarrow \boldsymbol{r}$ |  | $\begin{aligned} & \text { NNM } \\ & \text { iNiri } \end{aligned}$ |  | $\begin{aligned} & \text { Nonin } \\ & \text { ninini } \end{aligned}$ |  | $\overbrace{0}^{\infty} 0$ <br> ナポ |  | NOㅇN nin |
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| $\begin{array}{cc} \text { O} \\ \text { 等 } & \text { 慁 } \\ & \end{array}$ | $\begin{aligned} & \text { Nin } n_{n} \\ & \text { Ninini } \end{aligned}$ |  | $\begin{aligned} & \text { Nino } \\ & \text { Ninin } \end{aligned}$ |  | बूñ |  |  |  |  |  | $\begin{aligned} & \text { Nón } \\ & \text { inive } \end{aligned}$ |
| \％ | min <br> ーーか |  | mNN ーー～ |  | 오․․ |  | 응앙 |  | $\begin{aligned} & \text { ging } \\ & \text { gís } \\ & \dot{n} \dot{y} \text { in } \end{aligned}$ |  | ががo <br> ナナホ |
|  |  |  |  |  | ${ }_{n}^{n}{ }_{n}^{\infty}$ |  | №Non |  | $\begin{aligned} & \text { rive } \\ & \dot{A} \text { min } \end{aligned}$ |  | $\begin{aligned} & \text { FMo } \\ & \dot{\forall} \dot{+} \end{aligned}$ |
|  |  | $\begin{aligned} & 00 \\ & .0 \\ & .7 \\ & 0 \\ & 0 \\ & 0 \\ & 0 \\ & 3 \\ & 0 \\ & \cdots \\ & \omega \end{aligned}$ |  | $\begin{gathered} a g \\ . \\ .0 \\ 0 \\ 0 \\ 0 \\ 0 \\ + \\ 0 \\ 0 \\ 0 \\ 0 \end{gathered}$ | $\infty \times \text { on }$ <br> $\underset{\sim}{\infty}{ }^{\infty}$ ナーツ |  |  | 0 <br>  <br> 0 <br> 0 <br> 0 <br> 0 <br> 0 <br> 0 <br> 0 <br> 0 <br> 4 <br> 4 |  mNm <br> $\stackrel{\sim}{\infty}$ <br> Non <br> NナM |  |  |
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APPENDIX 19-B (contan_ RAW DATA



APPENDIX 19-B (cont.) RAW DATA

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| APPENDIX 19-B (cont.) |  |  |  |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Age (days) Variable | 0 |  | 35 |  | 70 |  | 105 |  | 140 |  | 175 |  |
|  |  |  |  |  | Fast | owing |  |  |  |  |  |  |
| no. | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe |
| 52 | .39 .14 .07 | .08 .04 .04 | $\begin{array}{r} 14.1 \\ 10.8 \\ 9.6 \end{array}$ | $\begin{array}{r} 9.3 \\ 13.5 \\ 18.0 \end{array}$ | $\begin{aligned} & 10.5 \\ & 11.1 \\ & 12.2 \end{aligned}$ | $\begin{aligned} & 7.1 \\ & 8.3 \\ & 9.8 \end{aligned}$ | $\begin{array}{r} 13.1 \\ 5.8 \\ 15.2 \end{array}$ | $\begin{array}{r} 5.4 \\ 9.6 \\ 19.2 \end{array}$ | $\begin{array}{r} 16.7 \\ 9.8 \\ 8.6 \end{array}$ | $\begin{array}{r} 11.8 \\ 7.2 \\ 18.8 \end{array}$ | $\begin{aligned} & 18.6 \\ & 22.9 \\ & 22.5 \end{aligned}$ | $\begin{array}{r} 10.2 \\ 8.5 \\ 11.5 \end{array}$ |
|  |  |  |  |  | Slow | owing |  |  |  |  |  |  |
| 52 | .03 .21 .09 | .19 .01 .06 | $\begin{aligned} & 6.7 \\ & 7.8 \\ & 2.2 \end{aligned}$ | $\begin{aligned} & 5.3 \\ & 6.0 \\ & 5.9 \end{aligned}$ | 3.3 3.7 5.9 | $\begin{array}{r} 6.0 \\ 4.2 \\ 12.6 \end{array}$ | $\begin{aligned} & 23.0 \\ & 16.0 \\ & 14.0 \end{aligned}$ | $\begin{aligned} & 13.6 \\ & 12.7 \\ & 11.8 \end{aligned}$ | $\begin{aligned} & 10.9 \\ & 14.7 \\ & 13.8 \end{aligned}$ | $\begin{aligned} & 17 \cdot 4 \\ & 10.5 \\ & 13 \cdot 3 \end{aligned}$ | $\begin{array}{r} 14.0 \\ 8.4 \\ 10.5 \end{array}$ | $\begin{array}{r} 18.6 \\ 9.5 \\ 13.7 \end{array}$ |
|  |  |  |  |  | Fast | owing |  |  |  |  |  |  |
| 53 |  |  |  |  |  |  |  |  | $\begin{aligned} & 5.4 \\ & 5.4 \\ & 3.4 \end{aligned}$ | $\begin{array}{r} 13.0 \\ 5.3 \\ 3.9 \end{array}$ | $\begin{aligned} & 13 \cdot 4 \\ & 14.1 \\ & 13.3 \end{aligned}$ | $\begin{aligned} & 11.6 \\ & 11.0 \\ & 13.7 \end{aligned}$ |
|  |  |  |  |  | Slow | owing |  |  |  |  |  |  |
| 53 |  |  |  |  |  |  |  |  | $\begin{array}{r} 13.3 \\ 9.9 \\ 6.9 \end{array}$ | $\begin{aligned} & 7.0 \\ & 5.9 \\ & 4.9 \end{aligned}$ | $\begin{aligned} & 26.8 \\ & 15.9 \\ & 20.9 \end{aligned}$ | $\begin{aligned} & 11.4 \\ & 16.9 \\ & 11.2 \end{aligned}$ |
|  |  |  |  |  | Fast | owing |  |  |  |  |  |  |
| 54 | 15.7 15.7 16.7 | 15.3 16.9 17.2 | $19 \cdot 3$ $19 \cdot 3$ $15 \cdot 7$ | $\begin{aligned} & 20.2 \\ & 20.7 \\ & 19.1 \end{aligned}$ | $\begin{aligned} & 20.5 \\ & 20.7 \\ & 20.6 \end{aligned}$ | $\begin{aligned} & 20.7 \\ & 21.3 \\ & 20.7 \end{aligned}$ | $\begin{aligned} & 20.3 \\ & 20.6 \\ & 19.7 \end{aligned}$ | $\begin{aligned} & 21.2 \\ & 21.5 \\ & 19.9 \end{aligned}$ | $\begin{aligned} & 20.6 \\ & 21.2 \\ & 21.5 \end{aligned}$ | $\begin{aligned} & 21.2 \\ & 21.8 \\ & 21.5 \end{aligned}$ | $\begin{aligned} & 21.1 \\ & 21.1 \\ & 21.1 \end{aligned}$ | $\begin{aligned} & 22.1 \\ & 21.1 \\ & 21.8 \end{aligned}$ |
| 54 | Slow Growing |  |  |  |  |  |  |  |  |  |  |  |
|  | 16.7 | 16.0 | 19.9 | 18.9 | 19.9 | 21.0 | 20.7 | 20.7 | 21.2 | 21.3 | 21.1 | 21.7 |
|  | 17.5 | 17.7 | 19.9 | 19.8 | 22.5 | 18.3 | 20.7 | 20.7 | 21.8 | 21.4 | 21.2 | 20.8 |
|  | 16.7 | 16.9 | 19.7 | 19.4 | 20.9 | 21.5 | 20.5 | 20.4 | 21.0 | 21.0 | 21.0 | 21.1 |


APPENDIX 19-B (cont.) RAW DATA
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APPENDIX 19-B (cont.) RAW DATA


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| APPENDIX |  | 19-B (cont.) |  |  | RAW DATA |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Age ( | ) 0 |  | 35 |  | 70 |  | 105 |  | 140 |  | 175 |  |
| Variable |  |  |  |  | Fast | owin |  |  |  |  |  |  |
| no. | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe |
| 70 | 15 | 18 | 20 | 16 | 20 | 19 | 17 | 18 | 19 | 19 | 18 | 22 |
|  | 18 | 15 | 20 | 18 | 21 | 20 | 17 | 18 | 18 | 18 | 20 | 19 |
|  | 17 | 18 | 17 | 20 | 20 | 19 | 18 | 17 | 19 | 19 | 18 | 21 |
|  |  |  |  |  | Slow | owin |  |  |  |  |  |  |
| 70 | 17 | 17 | 15 | 21 | 18 | 19 | 18 | 18 | 18 | 19 | 20 | 20 |
|  | 16 | 19 | 20 | 21 | 17 | 17 | 19 | 18 | 22 | 19 | 20 | 20 |
|  | 18 | 12 | 19 | 21 | 18 | 19 | 18 | 19 | 20 | 19 | 18 | 19 |
|  |  |  |  |  | Fast | owin |  |  |  |  |  |  |
| 71 | 11 | 13 | 12 | 12 | 11 | 13 | 13 | 13 | 13 | 13 | 12 | 14 |
|  | 15 | 13 | 12 | 13 | 14 | 11 | 13 | 13 | 13 | 13 | 13 | 12 |
|  | 15 | 13 | 14 | 14 | 13 | 11 | 13 | 13 | 15 | 13 | 12 | 13 |
|  |  |  |  |  | Slow | owin |  |  |  |  |  |  |
| 71 | 15 | 15 | 13 | 13 | 13 | 13 | 12 | 13 | 13 | 12 | 13 | 12 |
|  | 13 | 14 | 12 | 13 | 12 | 14 | 13 | 13 | 12 | 13 | 12 | 12 |
|  | 14 | 12 | 13 | 13 | 13 | 14 | 13 | 14 | 14 | 14 | 12 | 11 |
|  |  |  |  |  | Fast | owin |  |  |  |  |  |  |
| 72 |  | 20 | 21 | 29 | 22 |  |  | 21 |  | 17 |  | 9 |
|  | 19 | 22 | 22 | 18 | 8 | 28 | 19 | 18 | 17 | 15 | 12 | 19 |
|  | 20 | 18 | 19 | 16 | 14 | 20 | 20 | 18 | 11 | 16 | 21 | 13 |
|  |  |  |  |  | Slow | owin |  |  |  |  |  |  |
| 72 | 16 | 24 | 27 | 19 | 17 | 16 | 22 | 20 | 19 | 19 | 19 | 17 |
|  | 23 | 18 | 20 | 15 | 21 | 20 | 19 | 18 | 12 | 16 | 16 | 18 |
|  | 18 | 29 | 19 | 17 | 21 | 13 | 18 | 16 | 11 | 14 | 21 | 24 |

RAW DATA

| 105 |  |  | 140 |  | 175 |  |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :---: | :---: |
| Ram | Ewe | Ram | Ewe | Ram | Ewe |  |  |
| 13.7 | 16.1 | 16.8 | 16.7 | 15.4 | 17.0 |  |  |
| 15.6 | 16.7 | 16.4 | 17.1 | 17.6 | 15.6 |  |  |
| 15.6 | 16.4 | 16.5 | 17.0 | 16.7 | 16.6 |  |  |
|  |  |  |  |  |  |  |  |
| 15.1 | 16.6 | 16.4 | 16.4 | 15.7 | 18.4 |  |  |
| 16.0 | 15.9 | 17.6 | 17.3 | 16.9 | 16.7 |  |  |
| 16.3 | 15.9 | 17.0 | 17.1 | 16.8 | 15.3 |  |  |


| APPENDIX | 19－B（cont．） |  |  |  | RAW DATA |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Age（days） | ） 0 |  | 35 |  | 70 |  |
| Variable |  |  |  |  | Fast | rowing |
| no． | Ram | Ewe | Ram | Ewe | Ram | Ewe |
| 73 | 10.2 | 13.3 | 13.7 | 13.7 | 14.7 | 14.2 |
|  | 12.0 | 13.2 | 14.1 | 16.2 | 15.7 | 14.4 |
|  | 13.2 | 13.1 | 14.7 | 14.8 | 15.2 | 15.4 |
|  |  |  |  |  | Slow Growing |  |
| 73 | 14.1 | 11.0 | 13.3 | 13.9 | 16.3 | 16.2 |
|  | 13.1 | 13.4 | 14.9 | 16.0 | 16.6 | 15.0 |
|  | 13.3 | 12.6 | 14.9 | 14.9 | 15.5 | 16.2 |

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APPENDIX 19-B (cont.) RAW DATA

| Age (days) Variable no. | 0 |  | 35 |  | 70 |  | 105 |  | 140 |  | 175 |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  |  | Fast | Growing |  |  |  |  |  |  |
|  | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe | Ram | Ewe |
| 79 | . 007 | . 008 | . 022 | . 018 | . 045 | . 022 | . 022 | . 026 | . 047 | . 028 | . 040 | . 041 |
|  | . 004 | . 008 | . 064 | . 027 | . 025 | . 019 | . 018 | . 017 | . 050 | . 029 | . 016 | . 052 |
|  | . 008 | . 004 | . 018 | . 020 | . 036 | . 024 | . 018 | . 020 | . 029 | . 040 | . 046 | . 037 |
|  | Slow Growing |  |  |  |  |  |  |  |  |  |  |  |
| 79 | . 008 | . 006 | . 013 | . 019 | . 030 | . 036 | . 030 | . 023 | . 029 | . 053 | . 047 | . 093 |
|  | . 008 | . 008 | . 023 | . 018 | . 042 | . 021 | . 020 | . 017 | . 045 | . 040 | . 091 | . 077 |
|  | . 007 | . 007 | . 017 | . 018 | . 020 | . 029 | . 022 | . 031 | . 024 | . 036 | . 047 | . 039 |

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[^0]:    F $\mathcal{P} \geq$ ure 2. Pathways and ensymes in triglyceride biosynthesis Fatty acid CoA ligase, (2) acyl-CoA-L-glycerol 3 phoste o-acyl transferase, (3) L-a-phosphatidate phosphohyIyase, (4) acyl-CoA-1,2-diglyceride o-acyl transferase, acyl-CoA-2-monoglyceride o-acyl transferase.

[^1]:    ${ }^{\mathrm{a}}$ Means are the average of 36 lambs.
    $\mathrm{b}_{\text {Means }}$ are the average of 12 lambs.
    cdefgh Means within each main effect on the same row bearing the same superscripts are not statistically significant( $p>$.05).

[^2]:    $a_{\text {Means }}$ of 18 lambs for perirenal or subcutaneous and 6 lambs for intramuscular adipose tissues
    Pr=Probability for level of significance.

[^3]:    ${ }^{\text {a }}$ Means are the average of 6 lambs.
    Pr. =Probability for level of significance.

[^4]:    ameans are the average of 6 lambs.
    Pr=Probability for level of significance.

[^5]:    Means are the average of 6 lambs.
    Pr.-Probability for level of significance.

[^6]:    aMeans are the average of 6 lambs.

