INFLUENCE OF PROTEIN INTAKE, ENERGY INTAKE AND STAGE OF GESTATION ON GROWTH, REPRODUCTIVE PERFORMANCE NITROGEN BALANCE, CREATININE EXCRETION AND CARCASS COMPOSITION OF THE GRAVID GILT

By

GAWAIN MERLIN WILLIS

Bachelor of Science
Oklahoma State University
Stillwater, Oklahoma
1974

Submitted to the Faculty of the
Graduate College of the
Oklahoma State University
in partial fulfillment of
the requirements for
the Degree of
MASTER OF SCIENCE
July, 1978

INFLUENCE OF PROTEIN INTAKE, ENERGY INTAKE AND STAGE OF GESTATION ON GROWTH, REPRODUCTIVE PERFORMANCE NITROGEN BALANCE, CREATININE EXCRETION AND CARCASS COMPOSITION OF THE GRAVID GILT

Thesis Approved:

Charles V. Maxwell
Thesis Adviser

Rodger K. Johnson

F. N. Owens

Norman N Durham
Dean of Graduate College

ACKNOWLEDGMENTS

The author wishes to take this opportunity to thank Dr. C. V. Maxwell, major adviser, for the guidance and support in the course of this thesis project.

Special thanks are extended to Dr. R. K. Johnson for assistance in statistical analysis of data and critical evaluation of the thesis. Appreciation is also given to Dr. F. N. Owens for his suggestions and professional guidance.

Special recognition is extended to Jim Schooley, herdsman, who not only cared for the experimental animals, but also devoted particular attention to the details of the study. Others who deserve thanks are Leroy Kimbrell, Sandy Parr and numerous fellow graduate students.

This thesis is dedicated to my wife, Karmen, for her support, encouragement and continued hard work through these studies.

TABLE OF CONTENTS

LIST OF TABLES

CHAPTER I

INTRODUCTION

The gestating gilt has the capacity to retain more nitrogen in the body then can be accounted for by increases in direct products of conception. This retention increases with protein intake and has been attributed to storage of protein in extrauterine tissues. The extent, nature and physiological importance of such protein reserves has not been documented in the gilt.

Whether such stored protein can be translocated from the dam's tissues to fetal tissues at advanced stages of gestation is uncertain.

The aims of this study were to determine the effects of protein intake, energy intake and stage of gestation of (1) growth, reproductive performance and nitrogen balance in gilts and (2) evaluate tissue growth and protein content of skeletal muscle and of the gravid uterus. These investigations should lead to a more detailed definition of the protein requirements of the gestating gilt.

CHAPTER II

LITERATURE REVIEW

The effects of protein, energy, and stage of gestation on growth of replacement gilts and subsequent reproductive performance have been reviewed extensively by Duncan and Lodge (1960), Pond (1973), Jones (1973) and Jones (1975). Further, the effects of protein and stage of gestation on nitrogen balance have been reviewed by Rippel (1967), Jones (1973) and Jones (1975).

This review will concern more recent data of the effects of 1) energy intake on nitrogen balance and 2) protein or energy nutrition and/or stage of gestation on creatinine excretion and 3) usefulness of nitrogen balance and creatinine excretion as indices of nitrogen retention associated with gestation.

Nitrogen Balance

Energy Intake Levels

Munro and Naismith (1953) studied rats receiving diets either rich in protein or deficient in protein, in combination with various levels of energy intake (from about 850 to 1700 kcal/sq. m. of body surface area) obtained by adding carbohydrate of fat to standard basal diets. Their results showed that when diets provided adequate amounts of protein, increments in energy intake produced by adding either carbohydrate or fat

to a submaintenance diet caused a linear improvement in nitrogen balance. Munro (1964) reviewed the influence of dietary energy on nitrogen balance and summarized the available data by stating that under normal nutritional conditions, nitrogen retention is increased by an increase in energy intake as long as protein intake is adequate. Conversely, an increase in protein intake may not result in increased nitrogen retention if energy intake is inadequate. Studies with adult rats, dogs and humans showed that within limits, additional energy intake resulted in linear increases in nitrogen retention. Calloway and Spector (1954), in a survey of data on human subjects, reached the same basic conclusion. They stated that on fixed adequate protein intake, energy intake is the deciding factor in nitrogen balance and with a fixed adequate caloric intake, protein level is the determinant. That is, at each fixed inadequate protein intake there is an individual limiting energy level beyond which increasing calories without protein or protein without calories is without benefit.

More recently, in an experiment with men, Calloway (1975) fed marginal intakes of two levels of protein with energy constant or three levels of energy with protein constant. Energy intake appeared to have a much greater effect on nitrogen balance than did protein intake in the marginally adequate ranges of intake.

Similarly, ewes fed one of four diets with roughage to concentrate ratios of 20:80, 40:60, 60:40 and 80:20 were observed by Guada et al. (1975) from sixty-two day of gestation to parturition. In these studies, nitrogen retention and the efficiency with which the apparently digested nitrogen intake was retained increased as the dietary energy

concentration was increased. The mean daily increases in nitrogen retention were 0.0135 and 0.0039 g for each percentage unit increase in the concentrate portion of the diet at mid and late gestation respectively.

In experiments with gestating sows Pike (1970), found that doubling the energy intake caused a greater increase in nitrogen retention than did doubling protein intake. These findings suggest that energy level of the diet influences nitrogen retention.

Miller and Payne (1961) have proposed the value 150 kcal/gN as the energy required for retention of nitrogen. This includes not only the stored energy of the protein but also 'the work aspect of growth' analogus to 'that expended in rearranging the chairs in a room in which case the potential energy of the chairs, or the room, is not increased.' When energy is restricted, this assumption leads to an expression for net protein utilization, i.e., the proportion of the intake of N that is retained (Miller and Payne, 1964). Thus: $\theta = \frac{6.8\ (C-C_B)}{I}$, where C is the caloric intake, C_B is the energy required for basal metabolism (70 kcal/day $kg^{0.73}$) and I is the nitrogen intake (mg/day/$W^{0.73}$). Munro (1964) observed that an acceptable explanation of the action of energy intake on protein metabolism must provide a reason why the effects of protein and energy intake on N balance are sometimes interdependent. Miller and Payne (1964) attempted to explain this by combining equations they had presented into two expressions for nitrogen balance (mg/day/$W^{0.73}$)

$$\Delta B = I - M, \qquad (1)$$

$$\theta = S(1 - kP), \qquad (2)$$

$$\theta = \frac{6.8(C-C_B)}{I} \qquad (3)$$

From equation 1 and 2 combined:

$$\Delta B = 0.4\ SCP - 0.0076\ SCP^2 - 250.$$

Alternatively from equations 1 and 3:

$$\Delta B = 6.8\ C - 726.$$

where

ΔB = nitrogen balance in mg/day $kg^{0.73}$ body - weight

I = nitrogen intake in mg/day $kg^{0.73}$ body - weight = 0.4 PC

θ = net protein utilization (NPU) expressed as a fraction

M = nitrogen used for maintenance = 250 mg N/day $kg^{0.73}$

S = protein score expressed as a fraction

k = constant = 0.019

P = protein calories as percent of total calories

C = calorie intake in kcal/day $kg^{0.73}$ body - weight

C_B = energy for basal metabolism = 70 kcal/day/$kg^{0.73}$ body - weight.

The lesser of the two values for N balance is the one that applies (Miller and Payne, 1963). Using these equations Miller and Payne (1964) reported substantial agreement between predicted values for nitrogen balance and those found in the literature. If that is the case, then it becomes apparent that the value obtained for nitrogen balance is a combination of nitrogen intake, amino acid composition of that intake and caloric intake of the subject being studied.

Further study is needed to evaluate the interplay of these factors on nitrogen balance. This is especially true for nitrogen balance of the gestating gilt, where the stress of pregancy and subsequent lactation is added to the demands for growth.

Urinary Creatinine Excretion

Diet, Stage of Gestation, Sampling Method and Uses

Folin (1905) first expressed the view that creatinine excretion in urine was constant for a given individual on a meat free diet. The results of Camera et al. (1951) reemphasized the effect of meat content of the diet on creatinine excretion. The effect of diet on creatinine excretion in one individual who interposed periods of no meat-low protein intake (13 g per day), no meat-high protein intake (140 g per day) and high meat intake (100 g total protein per day of which 60 g was meat) into periods in which his normal diet was consumed was studied by Chattaway et al. (1969). The whole experiment extended over 60 days. Analysis of the mean values confirmed that the creatinine excretion of an individual subject decreases on a meat free diet and increases on a high meat intake. However, Vestergaard and Leverett (1958) in an experiment involving 18 adults could find no correlation between meat intake and increases in creatinine excretion. Conflicting reports such as these point out one of the major problems with urinary creatinine data presented in literature. In their study Vestergaard and Leverett (1958) found the constancy of urinary excretion of creatinine in 24-hour collection periods was highly dependent on the subject under investigation. Similarly Miller and Blyth (1952) found that a single urine collection may yield grossly erroneous results and recommended at least three consecutive collections to obtain more consistent results. In a study of phenylketonuric, galatosemic and normal children, Lewis et al. (1975) concluded that creatinine excretion is not constant for a subject, nor is it consistently elevated or depressed at a given time of

day. Therefore, there is no period during the day that a urine specimen can be assumed to be representative of the 24-hour creatinine excretion rate. It was, however, suggested that results could be improved using 24-hour excretion rates. Despite these problems, urinary creatinine has been widely used as an index compound for other constituents excreted in the urine. In such cases De Groot and Aafjes (1960) have proposed that the concentration of urinary constituents be expressed in terms of a creatinine quotient $\left(\frac{\text{content of constituent}}{\text{content of creatinine}}\right)$ rather than in absolute units when 24-hour urine samples cannot be taken.

More recently (Plotka and Erb, 1969), urinary creatinine of the ewe has been used successfully as an index compound to express estrogen excretion as the ratio, ng.steriod/mg.creatinine. The correlation between the ratio and µg./24 hr based on total volume of urine were 0.97, 0.98 and 0.98 for estrone, estradiol and estrogen respectively. A corresponding correlation of 0.96 has been reported for sows (Erb et al., 1970). These results show that measuring total 24-hour urine volume is unnecessary for estimating relative excretion rates of estrogen in urine.

Studies have also been conducted to evaluate the use of urinary creatinine to determine nutrient status in animals. Fisher (1965) in a series of experiments with adult male rats, studied the effect of varying dietary protein and amino acid content of rations in relation to the daily creatinine excretion. It was observed that creatinine excretion was not constant and varied with protein intake and amino acid content of the diet. The authors could find no pattern which would permit prediction of creatinine excretion as it relates to the

level of dietary N or of amino acids. With an amino acid free diet, which provided the least amount of dietary N offered in these experiments, daily creatinine excretion was highest. That excretion was approximately 3 times as high as with a diet which provided 15% crude protein. Considerable variation in creatinine excretion of individual rats fed any given diet was observed, particularly following a change from one diet to another.

Butcher and Harris (1957) conducted a series of trials with sheep and cattle to investigate the use of creatinine as an index material in the urine. They found creatinine excretion to be independent of protein intake. The diurnal variation in the excretion of urinary nitrogen and creatinine, and the creatinine/nitrogen ratio (grams creatine/grams nitrogen) was found to be significantly different for different times of day. Small samples of sheep urine collected morning and evening and composited were found to have essentially the same creatinine-urinary nitrogen ratios as samples from total collections. It was stated, creatinine may have merit as an index material for urine, with the limitation that average creatinine excretion must be determined for experimental animals prior to balance-trials by index materials. Total daily excretion of urinary nitrogen and creatinine/urinary nitrogen ratio were found to be closely associated with level of protein intake, when energy intake was relatively constant. Lee and Lucia (1963) studying rats fed 29.05 kcal/day or 64.30 kcal/day found creatinine excretion increased with time on the high energy diet, while remaining relatively constant in the low energy diets.

Thirty-nine hereford cows and steers were involved in three experiments to determine the factors contributing to the variation in urinary creatinine and creatinine/nitrogen ratios in beef cattle (Albin and Clanton, 1966). Four rations varying in protein and energy were fed. The high protein (HP) and high energy (HE) diets fed to cows were approximately 70% and 65% respectively, of the N.R.C. levels, whereas the low protein (LP) and low energy (LE) level were approximately 33% and 50% respectively. The HP and HE level fed to steer calves were calculated to supply the amounts recommended by N.R.C. for calves to gain 0.45 kg per day. The LP and LE levels were calculated to supply 60% and 80% of recommended levels respectively. In experiment 2, cows received 6.8 kg per head daily of a different ration (8.1 - 10.5% crude protein and 1.86 - 1.97 meg cal/kg D.E.) in each of three metabolism trials. There was no significant variation in urinary creatinine due to animals except in experiment three involving four cows studied at different stages of reproduction. Folin (1905) and Butcher and Harris (1957) have pointed out large variability of creatinine excretion among animals. Ration differences affected ($P < .05$) urinary creatinine in one experiment but not in the other experiments. The urinary creatinine/N ratio was significantly affected by rations. When nitrogen intake increased, urinary nitrogen increased; however, the creatinine/nitrogen ratio decreased, because creatinine did not fluctuate for a given individual with respect to rations. The converse was true when urinary nitrogen decreased. The authors suggest two major applications for the use of urinary creatinine values. First creatinine may be used as an index material in urine for balance studies. Second,

creatinine/N ratio may be used to indicate the balance of protein and energy. They stated that a ratio of 0.40 suggests that balance of protein and energy in the ration has been approached, but not necessarily whether both are low or high in the ration. Chetal et al. (1975) fed male and female buffalo calves from 3 to 6 months of age equal amounts of 13, 20 or 24% crude protein diets. Twenty-four hour urine samples were collected into toluene. The creatinine excretion did not differ with the level of protein in the diet; but differed from day to day. Total creatinine excretion in urine also differed significantly between blocks (made on basis of body weight) indicating that creatinine excretion was related to body weight. The creatinine nitrogen ratios for both male and female calves were significantly affected by protein level of the ration, but day to day variation and the interaction of day by protein were large ($P < .01$). This indicated that the effect of protein differed on different days.

It was concluded that urinary creatinine (percent and total) and the creatinine coefficient (mg/24 hr/kg) cannot be used as an index either in nutrient balance studies or for evaluating general nutritional status of ruminents. Further, due to significant day x protein interaction, creatinine/nitrogen ratios could not provide a suitable evaluation of protein status of the animals. In similar experiments with cattle Kertz et al. (1970) fed 8.9, 11.3 and 14.2% crude protein rations according to N.R.C. energy requirements with constant amounts of alfalfa hay and corn silage. Creatinine and nitrogen concentrations and their ratio were significantly affected by both level of nitrogen intake and time of day. Total urinary creatinine and nitrogen and

creatinine coefficient were not significantly affected by treatment.

Pike (1970) fed gestating (10-112 days post-breeding) sows two levels of protein (10.5 or 19.5% of crude protein) at one of three levels of intake (1.8, 2.7 or 3.6 kg/day). It was assumed that lean body mass at service was the same in the groups of sows studied and that creatinine output of the sow was related to lean body mass. Differences in protein deposition during gestation were used to explain the differences in creatinine output between high protein and low protein sows. He found a close relationship between urinary creatinine output and extra-uterine nitrogen deposition as determined by difference between nitrogen retention over the whole pregnancy less nitrogen deposited in uterus.

Albin and Clanton (1966) in their work with cattle, found urinary creatinine excretion was significantly decreased at the third month of gestation as compared to the eighth month of gestation and 2 months post-partum. In studies of sex hormones in women, Smith (1942) showed the only consistent change in urinary creatinine excretion during normal pregnancy was during the course of labor, when a striking drop in rate of creatinine excretion occurred. In an experiment involving 36 sows fed 2.3 kg per day of a 13% crude protein ration, Erb et al. (1970) found no difference in creatinine excretion (mg/hr) between nonpregnant and pregnant gilts. He listed mean excretion rates of 1.35 to 1.39 mg/hr/kg as compared to 0.79 mg/hr/kg for ewes (Hodgen et al., 1967).

Creatinine excretion has also been used to predict the lean body mass of an individual. Miller and Blyth (1952) used 43 students to predict lean body mass (calculated by subtracting densiometrically

determined body fat from total body weight). The correlation between creatinine excretion and lean body mass was 0.826. In 90% of the cases, the lean body mass for an individual predicted from creatinine excretion agrees within ±13.1% with the densiometrically determined value. Lofgreen and Garrett (1954) used 18 steers to correlate creatinine excretion (average of 5 day collection) and percent separable lean in soft tissue of 9, 10 and 11th rib. The correlation was 0.67. Urinary creatinine as an index of body composition has also been studied by Van Nieberk et al. (1963a). In these studies, 65 sheep ranging from 4 to 27 months of age were divided into three slaughter groups. These animals had been exposed to various nutritional treatments to effect a wide range in body composition. Urine was collected for 7 to 10 days to determine urinary creatinine. The regression of urinary creatinine (mg/24-hrs) on total empty body protein (gm), empty body water (kg) yielded a correlation coefficient of 0.97 in all three cases. Creatinine output was superior to body weight as an index of protein content in sheep containing 28 to 47% fat. Over the ranges studied and under the conditions studied, one milligram of creatinine was excreted per 24 hours for each 5.13±0.43 gm of body protein. Similar studies have been conducted in rats (Kumar et al., 1959).

It should be apparent that the use of creatinine excretion as an index material to determine concentration of urine constituents or as a predictor of carcass composition can be severely limited by the day to day inconsistency of urinary creatinine excretion within individuals. Van Niekerk et al. (1963b) found creatinine was unstable and decayed

rapidly at the normal pH (8.4 to 8.7) in sheep urine under temperatures ranging from 15 to 39°C. However, no loss of creatinine occurred during a 5-month period when urine was stored at its normal pH but at 4°C. Urine acidified to pH 2.5 to 3.5 and held at a temperature of 28 to 39°C progressively increased in creatinine concentration with increasing time of storage. Such effects plus differences in creatinine assay methods can account for a part of the variation in results using creatinine. Yet, more research is needed to determine the biological or technical reasons for inconsistency in individual creatinine excretion.

CHAPTER III

INFLUENCE OF PROTEIN INTAKE, ENERGY INTAKE AND STAGE OF GESTATION ON GROWTH, REPRODUCTIVE PERFORMANCE NITROGEN BALANCE, CREATININE EXCRETION AND CARCASS COMPOSITION OF THE GRAVID GILT

Summary

Thirty-six crossbred gilts were fed 3 levels of protein (146, 255 and 364 g/day) and 2 levels of energy (approximately 6200 kcal DE/day and 6200 kcal DE/day + 20%) throughout gestation. Five day nitrogen balance and creatinine excretion studies were conducted at early (0-30 days), mid (30-60 days) and late (60-90 days) gestation. At slaughter (90 days gestation), reproductive tracts were evaluated for reproductive performance and samples of the reproductive tract, liver and semimembranosus muscle were analyzed for crude protein.

The results of this experiment suggest that 146 g of crude protein per day during gestation is just as effective as the higher levels of crude protein intake for litter size or storage of nitrogen in the reproductive tissue. However, storage of protein in muscle tissue increased to intakes between 255 and 364 g of crude protein per day. No advantage for the higher energy diets for these traits was noted.

The fate of this stored muscle nitrogen at parturition and its role in subsequent lactation and rebreeding performance is an unanswered question.

Due to significant interactions and decreasing urinary creatinine excretion with increasing gilt weight, creatinine excretion could not be used as a predictor of lean body mass to follow changes in lean body mass throughout gestation.

Introduction

The literature yields no consistent relationship between protein and energy intake and subsequent litter size or pig birth weight (Clawson et al., 1963; Rippel et al., 1965a; Frobish et al., 1966; Baker et al., 1970; Hawton and Meade, 1971; Degeeter et al., 1972). A clear understanding of such a relationship is necessary to establish protein allowances for gestating gilts.

Nitrogen balance of gilts has been documented by Rippel et al. (1965b), Miller et al. (1969), and Hesby et al. (1971) with little evidence of a relationship between nitrogen retention and parturition performance.

The use of urinary creatinine excretion in the prediction of lean body mass has been demonstrated by Miller and Blyth (1952), Van Niekerk et al. (1963a) and Pike (1970). It appears that urinary creatinine could be of value in monitoring changes in lean body mass throughout gestation.

The objectives of this study were to examine nitrogen retention and urinary creatinine excretion in the pregnant gilt as affected by

protein intake, energy intake and stage of gestation. Concurrent evaluation of reproductive performance, growth rate, and tissue protein content were conducted.

Materials and Methods

Thirty-six crossbred (two and three breed crosses of Hampshire, Yorkshire and Duroc) gilts were fed three levels of protein (8, 14 or 20% crude protein) and two levels of energy (moderate [M] and high [H]). The M20% crude protein corn-soybean meal diet was extended with corn starch to give the M8% and M14% crude protein rations. The moderate energy rations were further extended with corn starch to increase the digestible energy content of the moderate energy rations by twenty percent to yield the high energy rations (Table I). Thus, amino acid ratios across protein levels were maintained.

Gilts were approximately 270 days of age at the start of the experiment. From cursory observation, groups of gilts were observed exhibiting estrus before the experiment began. Gilts were randomly assigned to one of four dirt lots. Gilts were fed once daily in individual feeding stalls and had access to drinking water and shelter. Gilts were observed daily for signs of estrus by introducing a teaser boar into the pens. Thirteen days after the first estrous on trial, the animals were placed in metal metabolism stalls. After a one day adjustment to the crate, a four day total collection of urine was conducted. Each day's urine output was collected into concentrated hydrochloric acid (diluted 1:2 with water) and diluted to twenty liters with water. A 200 ml aliquot was saved each day and the four daily

samples composited after each collection. The gilts were weighed before and after each collection. Subsequently, the animals were returned to the dirt lots and mated at the next estrus to two different boars on consecutive days and changed from 2.27 kg of a 16% crude protein diet to 1.81 kg (moderate energy rations) or 2.15 kg (high energy rations) of the experimental diets. This provided daily crude protein and digestible energy intakes shown in Table II. One days 25, 55 and 85 of gestation urine and feces were collected for four days. A 20% aliquot of the daily feces output was frozen and later composited in a Hobart mixer. Urine samples were frozen and later analyzed for Kjeldahl nitrogen (A.O.A.C., 1960) and creatinine (Hawk et al., 1954). Fecal samples were analyzed for Kjeldahl nitrogen and dry matter content.

Gilts were slaughtered on day 90 of gestation. Reproductive tract were recovered as quickly as possible after slaughter and uniformly trimmed by severing the uterus approximately 7.5 cm posterior to the junction of the uterine horns. The reproductive tracts were weighed and evaluated immediately for reproductive status. Corpora lutea counts were recorded for each ovary and each uterine horn was dissected from the cervical end to obtain embryo numbers. All solid tissues including the uterus, ovaries, placenta, and fetuses were ground twice through a 4.23 mm screen in a Hobard Model 4332 grinder and a 100 g ground sample was quickly frozen in a dry ice-ethanol bath. Volumes were recorded for freely draining uterine fluids and a 100 ml sample was frozen for Kjeldahl nitrogen analysis (A.O.A.C., 1960). The semimembranosus muscle of each left ham was removed as soon after slaughter as possible. The muscle was weighed, quickly ground and 100 g samples frozen in a dry

ice-ethanol bath. Muscle samples were subsequently subjected the ether extraction and Kjeldahl nitrogen analysis (A.O.A.C., 1960). Liver weights were recorded and 20 g samples were frozen for Kjeldahl nitrogen analysis (A.O.A.C., 1960).

Carcass weights were obtained and the right side of the carcass was subject to a physical separation of fat, lean and bone.

Body weight and growth data, nitrogen balance data and creatinine excretion data were analyzed as a 3x2x3 factorial arrangement of treatments in a split plot design. Each animal was a main plot with main plot treatments being level of protein in the diet (8, 14 and 20%) and dietary energy level (moderate and high). The subplot treatment corresponded to stages of gestation (30, 60 and 90 days). Remaining data, variables measured in just one period, were analyzed as a 3x2 factorial arrangement of treatments in a completely randomized design. The two treatment factors were protein level in the diet (8, 14 and 20%) and energy level in the diet (moderate and high).

Results and Discussion

Growth

The initial weight and slaughter weight of gilts is presented in Table III. There was no significant trends in slaughter weight of gilts due to protein level or energy level of the diet. The mean slaughter weights (kg) were 170.9, 177.5 and 176.2 for the 8, 14 and 20% protein diets and 173.9 and 175.6 for the M (moderate) and H (high) energy levels respectively.

Due to significant protein linear by gestation quadratic and energy linear by gestation quadratic interaction weight gain and average daily gain means are presented within each gestation period.

From 0-30 days gestation, weight gain (Table IV) increased as the protein intake increased. However, from 30-60 and 60-90 days of gestation the increase in weight gain with protein level did not carry through to the 20% level. Further, gaines in late gestation (60-90 days) were lower than in the early (0-30 days) and mid (30-60 days) gestation.

Gilt gains increased as energy increased in early and mid gestation. In late gestation however, gains were very similar on the H (High) energy diet and the M (Moderate) energy diet. Elsley et al. (1968) and Frobish (1970) have shown that total sow weight gain during gestation increased as energy level increased. Weight gain in the late gestation period, at the two energy intakes, was depressed compared to early and mid gestation, with weight gain being highest at mid gestation. Elsley et al. (1968) showed that sow weight gain increased steadily from 0 to 12 weeks gestation, with no depression in weight gain at 8 to 12 weeks of gestation, as indicated by these data. It is possible that the depression in weight gain of gilts at late gestation, in this experiment, was due to low ambient temperatures during the late gestation period. Thus, more energy would be used for maintenance of body temperature at the expense of weight gain.

Average daily gain (Table V) increased quadratically, at the three stages of gestation studied, with increasing protein level, although, the magnitude of the response was largest in mid gestation. Average

daily gain also increased quadratically with stage of gestation at each level of protein intake and increased on the H energy diets as compared to the M energy diets at the three stages of gestation. Average daily gain displayed a quadratic response to stage of gestation at the two energy levels. Average daily gains were reduced in late gestation, for the three levels of protein intake and the two levels of energy intake.

Reproductive Performance

Numbers of corpora lutea were 13.3, 13.9 and 12.6 for protein levels of 8%, 14% and 20% respectively (Table VI). For the M and H energy levels the mean numbers of corpora lutea were 13.4 and 13.2 respectively. None of these values was significantly affected by level of protein intake or level of energy intake. Mean ovulation rate (as indicated by corpora lutea) was 13.3 for all gilts. A similar value of 13.2 was reported by Jones (1975).

The mean embryo numbers were 10.9, 9.8 and 10.4 for the 8%, 14% and 20% crude protein diets respectively. The embryo numbers for both the M and H energy diets were 10.4. There was no significant trends in these values caused by level of protein intake or energy intake. Holden et al. (1968), Baker et al. (1970) and Hawton and Meade (1971) have also shown that crude protein intakes within this range did not significantly affect litter size. Frobish et al. (1966) and O'Bannon et al. (1966) have reported sows on lower dietary energy intakes farrow slightly more pigs per litter than sows on high energy intakes.

Nitrogen Balance

As protein level increased, nitrogen retention increased linearly ($P < .005$) with mean values of 10.3, 16.6 and 22.6 g/day for protein levels 8, 14, and 20% respectively (Table VII). This linear relationship is similar to that reported by Miller et al. (1969) during early gestation, Jones and Maxwell (1974) in early gestation and Jones (1975) throughout gestation.

As energy level increased, nitrogen retention increased linearly ($P < .005$) with mean values of 14.6 and 18.0 g/day for M and H energy levels respectively. Pine (1970) also showed increasing nitrogen retention with increasing energy levels.

Nitrogen retention increased linearly ($P < .005$) as stage of gestation progressed with mean values of 13.9, 14.6 and 20.5 for 30, 60 and 90 days gestation respectively. These data agree with earlier work by Elsley et al. (1966) and Kline et al. (1972) which showed increasing retention from a low value early in gestation to maximum retentions in late gestation.

There was a significant ($P < .05$) energy linear by protein linear interaction for urinary nitrogen. However, urinary nitrogen increased linearly as protein level increased at each energy level and displayed a significant ($P < .05$) quadratic effect as stage of gestation progressed.

Fecal nitrogen responded in a linear ($P < .005$) manner to protein intake. Fecal nitrogen increased ($P < .005$) with stage of gestation but decreased linearly ($P < .10$) with increasing energy intake.

Protein retention efficiency decreased linearly ($P < .10$) with increasing protein level. Since amino acid ratios are similar across protein levels, this suggests that protein was being supplied in excess of that needed for growth and reproduction, with catabolism and excretion of the excess protein. Retention efficiency increased linearly ($P < .025$) as energy level increased. Further, there was a significant ($P < .05$) quadratic increase in retention efficiency as stage of gestation progressed, with mean values of 42.0, 46.7 and 67.6 for 30, 60 and 90 days gestation respectively. Changes in retention efficiency suggest changes in protein synthesis. Bergen (1974) states that total protein synthesis can only be increased by increasing the synthetic capacity (elevated rRNA) through improved substrate availability and hormonal influence or by increasing the efficiency of synthetic machinery through hormonal factors. If stage of gestation affects protein synthesis, the control mechanism would probably be through hormonal modification of RNA synthesis (Manchester, 1970) or through an increased rate of ribosome movement along mRNA (Manchester, 1972). The exact control is not yet understood.

Dry matter digestibility decreased linearly ($P < .01$) as protein level increased with mean values of 84.3, 80.7 and 78.1 for the 8%, 14%, and 20% protein rations. There was no significant trend in dry matter digestibility as stage of gestation progressed. However, dry matter digestibility showed a significant ($P < .01$) linear increase as level of energy intake increased. It appears that energy levels at least up to the H level of this experiment can increase DMD.

Creatinine Excretion

Urinary creatinine excretion values are presented in Table VIII. There were significant ($P < .05$) protein linear by gestation linear, protein quadratic by gestation linear, protein quadratic by gestation quadratic and energy linear by gestation linear interactions in urinary creatinine excretion expressed as mg/day, mg/kg/day or $mg/kg^{0.75}/day$. Creatinine excreted showed a quadratic trend with the highest value on the 14% protein level intakes in early and late gestation. However, creatinine excretion responded in a linear fashion, by increasing as protein intake increased, at mid gestation. Creatine excretion decreased with increasing energy levels at mid and late gestation, however, the excretion increased with increasing energy intake in early gestation. Creatinine excretion responded quadratically to stage of gestation at each level of protein intake, with the highest creatinine excretions in mid gestation. The same affect of stage of gestation is seen at each energy level.

Since creatinine is the breakdown product of creatine, and 98% of the creatinine occurs in muscle tissue (West and Todd, 1961), it has been assumed that creatinine excretion in the urine should be closely related to lean body mass. This is based on several assumptions. First, urinary creatinine excretion values used to predict lean body mass are not greatly affected by dietary creatine or creatinine. This is probably true for diets low in creatine or creatinine (Folin, 1905; Camara et al., 1951; Vestergaard and Leverett, 1958; Chattaway et al., 1969) or when diets fed contain relatively similar amounts of creatine

and creatinine across animals. Second, there should be little or no reconversion of creatinine to creatine. Third, the conversion rate of creatine to creatinine should be relatively constant within an animal. Borsook and Dubnoff (1947) observed that 2% of the creatine of the body is converted each day into creatinine which is excreted in the urine.

In this experiment, urinary creatinine values were to be used to develop prediction equations for dry ether extracted lean (DEEL) in the carcass of gilts slaughtered at 90 days gestation. Using these prediction equations, it was hoped that the change in DEEL could be monitored throughout gestation. This was not possible. The mean pre-breeding value for urinary creatinine (mg/day) was 4847 (Table IX), whereas, the values for early, mid and late gestation were 3463, 5131 and 2988 respectively. Gilts weighed approximately 133 kilograms at the time pre-breeding values were determined and 175 kilograms when late gestation values were determined. Thus any prediction equation developed for DEEL at late gestation (90 days) using creatinine excretion in the urine could not accurately predict the DEEL at prebreeding or any other stage of gestation in this experiment. The urinary creatinine excretion decreased as weight increased. The reason for this trend in urinary creatinine excretion is not known. It seems plausible for the problem to be of a technical rather than a biological origin. Van Niekert (1963b) has indicated that creatinine varies in stability, depending on method of storage. Perhaps, the atypical trends for creatinine excretion in this experiment are due to method and time of storage. Daily urine samples were collected into concentrated hydrochloric acid (diluted 1:1 with water). Each day's collection was

diluted to 20 liters and a 200 ml sample was stored at 4°C for 1 to 7 days before being frozen. Samples were frozen from 4 to 11 months before analysis. Erb et al. (1970) has reported no difference in creatinine excretion of pregnant and non-pregnant sow with urinary creatinine excretion values of 1.35 - 1.39 mg/kg/hr. In this experiment, mean values of 0.71 - 1.51 mg/kg/hr were obtained over collection periods with an overall mean of 1.12 mg/kg/hr.

In view of the results obtained for urinary creatinine excretion over time in this experiment, it appears that creatinine excretion is of little value in monitoring changes in lean body mass over time. However, if the nature of the discrepancy in creatinine excretion over time can be elucitated, this conclusion could change.

Carcass Composition

Liver weight increased linearly ($P < .01$) as protein intake increased, with mean values of 1540, 1614 and 1848 grams for the 8, 14 and 20% diets respectively (Table X). There was no significant trend in liver weight due to energy, with mean values of 1603 and 1717 grams for the M and H energy levels respectively. Liver dry matter percent was very similar for the three levels of intake with mean values of 26.4, 26.9 and 26.8% for the 8, 14 and 20% protein levels. An energy increased, the liver dry matter percent increased. Mean dry matter percents were 26.1 and 27.3 for the M and H energy levels respectively. Percent nitrogen in the liver (dry tissue basis) showed no significant trends for the 8, 14 and 20% protein levels with mean values of 11.3, 11.6 and 11.9 respectively. Percent nitrogen in the liver decreased

linearly ($P < .05$) with increasing energy intake. The total grams of nitrogen contained in the liver (dry tissue basis) is presented in Table XIII. There was a significant ($P < .05$) energy linear x protein linear interaction. This interaction is the result of an inconsistently low amount of nitrogen in the livers of gilts fed the H energy 8% protein diet. Total grams of nitrogen in the liver increased with increasing nitrogen intake at each level of energy, whereas, total liver nitrogen decreased with increasing energy intake at the 8% protein level, but increased with increasing energy at the 14 and 20% protein levels.

The increase in total liver nitrogen with increasing protein intake is due largely to the increasing liver weights with increasing protein intake, as noted previously. The decrease in total liver nitrogen with increasing energy intake at the 8% protein level, is primarily due to lowered liver dry matter percent and lowered percent liver nitrogen of gilts fed the H energy 8% protein diet.

Semimembranosus (SM) muscle weight, percent dry matter, percent nitrogen and percent lean (dry tissue basis) is presented in Table X. SM muscle weight responded quadratically ($P < .05$) to increasing level of protein intake. Mean values were 1856, 2214 and 2089 grams for the 8, 14 and 20% protein levels. No significant trend in SM muscle weight was noted due to increasing energy intake. Mean values were 2108 and 1990 grams for the M and H energy levels respectively. No significant trends were noted in SM muscle percent dry matter due to protein or energy intake. SM muscle mean percent dry matter values were 23.1 percent for the M and H energy intakes and 23.1, 23.5 and 22.7% for the

8, 14 and 20% protein levels respectively. The increase in SM muscle percent nitrogen with increased protein intake approach linearity ($P < .10$). Increasing energy intake did not significantly affect percent nitrogen of the SM muscle. Mean percent nitrogen (dry ether extracted tissue basis) were 14.3 and 13.9% for the M and H energy diets respectively and 13.6, 14.3 and 14.6% for the 8, 14 and 20% protein diets respectively. Percent lean (dry tissue basis), of the SM muscle, showed no significant trends for level of protein or energy intake. The quadratic trend for total grams of nitrogen in the SM muscle with increasing protein level approached significance ($P < .10$). No significant trends were noted due to energy intake. Mean total nitrogen values of 46.5, 58.9 and 57.3 grams were obtained for 8, 14 and 20% protein levels respectively and 56.1 and 51.8 grams for the M and H energy levels respectively.

Carcass weight and carcass lean (physically separated) means are presented in Table XII. There were no significant trends apparent in carcass weight due to protein or energy treatment. The mean carcass lean values for the 8, 14 and 20% protein levels were 57.1, 69.7 and 66.6 kg respectively. Carcass lean (kg) showed a significant ($P < .001$) quadratic trend with increasing protein intake. No significant trends in carcass lean were noted due to energy intake.

By assuming that the percent nitrogen (dry tissue basis), percent dry matter and percent lean (dry tissue basis) were the same for the physically separated carcass lean as for the semimenbranosus muscle, the total nitrogen content of the carcass was calculated. These values are presented in Table XIII. The mean carcass nitrogen values were

1425, 1863 and 1768 grams for the 8, 14 and 20% protein levels. Mean values for the M and H energy levels were 1694 and 1654 grams, respectively. Grams of carcass nitrogen responded quadratically ($P < .05$) to increasing protein intake. However, no significant trend was noted due to energy intake. The quadratic response of grams of carcass nitrogen to protein intake is largely due to the quadratic response of physically separated carcass lean to protein intake. The quadratic response noted in grams of nitrogen in the carcass, grams of nitrogen in the semimenbranosus muscle, kilograms of physically spearated lean in the carcass and weight of the semimembranosus muscle to increasing protein level, all suggest, from a pure weight standpoint, the optimum level of nitrogen intake for protein storage in the muscle is somewhat less than that supplied by the 20% crude protein diets.

Uterine fluid volume decreased linearly ($P < .05$) with increasing protein level (Table XI). No significant trend was evident due to energy intake. Uterine fluid percent nitrogen increased linearly ($P < .001$) with increasing nitrogen intake. Energy intake did not significantly effect the percent nitrogen in the uterine fluid. A significant ($P < .05$) energy linear by protein linear interaction was noted for uterine fluid dry matter but no other affects of protein intake or energy intake were apparent.

The total grams of nitrogen in the uterine fluid were 5.9, 6.1 and 5.5 grams for the 8, 14 and 20% protein levels respectively. Values for the M and H energy intakes were 5.8 and 6.0 grams respectively. No significant trends were noted in grams of nitrogen in the uterine fluid due to level of protein or energy intake. It should be noted, no

overall affect of protein intake on grams of nitrogen in the uterine fluid could be detected even though significant trends were noted in percent nitrogen of the uterine fluid and volume of the uterine fluid. It seems that the nitrogen concentration is increased as the uterine fluid volume is decreased. Thus, total nitrogen content is not affected by protein intake.

Mean uterine weights, percent dry matter and percent nitrogen are presented in Table XI. The linear decrease in uterine weight due to increasing protein intake approached significance ($P < .10$) with mean values of 17.7, 16.3 and 14.7 kg for the 8, 14 and 20% protein levels respectively. Uterine weight decreased from 16.6 to 16.0 kg as energy intake increased, but this trend was not significant. Uterine percent dry matter contained a significant ($P < .01$) energy linear by protein quadratic interaction. Uterine percent nitrogen contained significant ($P < .05$) energy linear by protein linear and significant ($P < .05$) energy linear x protein quadratic interactions. No other trends were noted in uterine percent dry matter and percent nitrogen due to level of protein and energy intake.

The grams of nitrogen (dry tissue basis) in the uterine tissue (not including nitrogen in the uterine fluid) for the 8, 14 and 20% protein levels were 174.9, 177.6 and 170.1 g respectively. For the M and H energy levels, the values were 170.2 and 179.0 g respectively. No significant trends were found due to level of protein or energy intake. This suggests that the level of nitrogen intake supplied by the 8% crude protein diets was as effective as that supplied by the higher levels of nitrogen intake for nitrogen deposition in the uterine tissue.

TABLE I

CALCULATED COMPOSITION OF EXPERIMENTAL DIETS

Ingredients	International Reference No.	M 8%	H 8%	M 14%	H 14%	M 20%	H 20%
Corn (9%)[a] (3610)[b]	4-02-931	24.75	20.81	44.76	37.70	64.77	54.64
Soybean meal (44%)[a] (3300)[b]	5-04-604	12.31	10.35	22.25	18.74	32.20	27.17
Corn Starch (0.6%)[a] (3670)[b]	4-02-889	59.24	65.73	29.62	40.72	-----	15.63
Dicalcium phosphate	6-01-080	1.94	1.63	1.33	1.12	0.71	0.60
Calcium carbonate	6-01-069	0.76	0.64	1.04	0.88	1.32	1.11
Vitamin T. M. Premix[c]		0.50	0.42	0.50	0.42	0.50	0.42
Salt		0.50	0.42	0.50	0.42	0.50	0.42

[a]Estimated percent protein given in parentheses.

[b]Estimated Kcal/Kg of digestible energy.

[c]Vitamin-trace mineral premix supplied 4409 IU Vitamin A, 330.7 IU Vitamin D_3, 11 IU Vitamin E, 4.4 mg riboflavin, 22 mg d-pantothenic acid, 33 mg niacin, 882 mg choline chloride, .0165 mg Vitamin B_{12}, 2.2 mg menadione sodium bisulfite per kg of diet and 22 PPM Mn, 100 PPM Zn, .22 PPMI, Fe and 11 PPM Cu. (High energy rations supply 84% of these values.)

TABLE II

DAILY NITROGEN, CRUDE PROTEIN AND DIGESTIBLE ENERGY INTAKES OF GILTS

Item	Ration					
	M 8%	M 14%	M 20%	H 8%	H 14%	H 20%
Calculated N[a]	23.4	40.8	58.2	23.4	40.8	58.2
Actual N[a]	23.3	39.0	53.4	22.1	35.5	56.1
Calculated Crude Protein[a]	146	255	364	146	255	364
Actual Crude Protein[a]	146	244	334	138	222	351
Calculated Digestible Energy (Kcal/day)	6303	6236	6170	7563	7483	7404

[a]Values given in g/day.

TABLE III

BODY WEIGHT AND GROWTH DATA OF GILTS[a]

Item	Protein level				Energy level		
	8%	14%	20%	±SE	M	H	±SE
No. of Gilts[d]	12	11	10		17	16	
Initial weight (Kg)	133.4	132.8	133.3	3.9	135.0	131.3	3.1
Slaughter weight (Kg)	170.9	177.5	176.2	3.2	173.9	175.6	2.5

TABLE IV

BODY WEIGHT AND GROWTH DATA OF GILTS[a]

	Weight Gain (Kg)						
	Protein level[b]				Energy level[c]		
Gestation	8%	14%	20%	±SE	M	H	±SE
0-30	11.5	14.8	15.8	1.3	13.3	14.3	1.1
30-60	17.3	18.6	15.9	1.3	15.3	19.2	1.1
60-90	8.8	11.4	10.5	1.3	10.3	10.0	1.1

[a]Means

[b]Significant ($P < .05$) Protein linear X Gestation quadratic interaction.

[c]Significant ($P < .05$) Energy linear X Gestation quadratic interaction.

[d]Three gilts were removed from the trial for reasons not related to the experiment.

TABLE V

BODY WEIGHT AND GROWTH DATA OF GILTS[a]

Days Gestation	Average Daily Gain						
	Protein level[b]				Energy level[c]		
	8%	14%	20%	±SE	M	H	±SE
0-30	.33	.46	.45	.04	.39	.43	.03
30-60	.58	.62	.53	.04	.51	.64	.03
60-90	.29	.38	.35	.04	.34	.35	.03

[a]Means

[b]Significant ($P < .05$) Protein linear x Gestation quadratic interaction.

[c]Significant ($P < .05$) Energy linear x Gestation quadratic interaction.

TABLE VI

REPRODUCTIVE PERFORMANCE OF GILTS[a]

Item	Protein level				Energy level		
	8%	14%	20%	±SE	M	H	±SE
No. of Corpora lutea	13.3	13.9	12.6	.9	13.4	13.2	.7
No. of Embryos	10.9	9.8	10.4	.8	10.4	10.4	.6

[a]Means

TABLE VII

NITROGEN BALANCE OF GILTS[a]

Item	Protein level				Days Gestation				Energy level		
	8%	14%	20%	SE	30	60	90	SE	M	H	SE
Nitrogen retention (g/day)	10.3	16.6	22.6[b]	1.2	13.9	14.6	20.5[b]	1.1	14.6	18.0[b]	.9
Fecal N (g/day)	4.0	5.1	7.5[b]	.3	4.6	5.3	6.5[b]	.3	5.7	5.1	.2
Retained N (% of digested)	55.3	52.1	48.1[c]	3.2	42.0	46.7	67.6[d]	3.3	47.1	56.3[e]	2.6
Dry Matter digestibility (%)	84.3	80.7	78.1[f]	1.6	81.6	82.3	79.5	1.5	78.5	83.8[f]	1.3

	Energy level[3]								Days Gestation			
	M Protein level				H Protein level							
	8%	14%	20%	±SE	8%	14%	20%	±SE	30	60	90	±SE
Urinary N (g/day)	8.9	18.6	25.8	1.6	8.0	12.2	23.5	1.6	19.4	17.9	10.3[d]	1.1

[a]Means

[b]Linear effect significant ($P < .005$).

[c]Linear effect approached significance ($P < .10$).

[d]Quadratic effect significant ($P < .05$).

[e]Linear effect significant ($P < .025$).

[f]Linear effect significant ($P < .01$).

[g]Significant ($P < .05$) Energy Linear X Protein Linear interaction.

TABLE VIII

CREATININE EXCRETION OF GILTS[a]

Item	Days gestation[b] 30 Protein level 8%	30 Protein level 14%	30 Protein level 20%	60 Protein level 8%	60 Protein level 14%	60 Protein level 20%	90 Protein level 8%	90 Protein level 14%	90 Protein level 20%	±SE
Urinary creatinine (mg/day)	2072	4659	3784	4694	5192	5545	2568	3318	3129	348
Urinary creatinine (mg/Kg/day)	14.3	31.6	25.3	19.0	31.3	33.6	14.9	18.6	18.0	2.1
Urinary creatinine (mg/Kg·^{75}/day)	49.5	110.0	88.5	103.4	112.3	120.4	53.9	68.1	65.3	7.6

Item	Days gestation[c] 30 Energy level M	30 Energy level H	60 Energy level M	60 Energy level H	90 Energy level M	90 Energy level H	±SE
Urinary creatinine (mg/day)	3213	3714	5334	4927	3289	2668	288
Urinary creatinine (mg/Kg/day)	21.7	25.2	32.6	29.8	18.9	15.1	1.8
Urinary creatinine (mg/Kg·^{75}/day)	75.7	87.7	116.7	106.9	68.7	55.1	7.3

[a]Means

[b]Significant ($P < .05$) Protein linear X Gestation linear, Protein quadratic X Gestation linear and Protein quadratic X Gestation quadratic interactions.

[c]Significant ($P < .05$) Energy linear X Gestation linear interaction.

TABLE IX

PRE-BREEDING CREATININE EXCRETION OF GILTS[a]

Item	Protein level				Energy level		
	8%	14%	20%	±SE	M	H	±SE
Urinary creatinine (mg/day)	4579	5108	4853	355	5013	4655	272
Urinary creatinine (mg/kg/day)	34.0	38.7	36.5	2.5	37.3	35.4	1.9
Urinary creatinine ($mg/kg^{.75}$/day)	115.8	131.0	123.9	8.5	126.9	119.7	6.5

[a]Means

TABLE X

CARCASS COMPOSITION OF GILTS[a]

Item	Protein level				Energy level		
	8%	14%	20%	±SE	M	H	±SE
Liver							
Weight (g)	1540	1614	1848[b]	78	1603	1717	62
Dry matter (%)	26.4	26.9	26.8	.6	26.1	27.3	.5
Nitrogen (%)	11.3	11.6	11.9	.3	12.0	11.2	.3
Semimembranosus muscle							
Weight (g)	1856	2214	2089[e]	89	2108	1990	69
Dry matter (%)	23.1	23.5	22.7	.6	23.1	23.1	.5
Nitrogen (%)	13.6	14.3	14.6	.4	14.3	13.9	.3
Lean (% of dry weight)	79.6	80.4	80.0	2.6	79.4	80.6	2.0

[a]Means

[b]Linear effect significant ($P < .01$).

[c]Linear effect approached significance ($P < .10$).

[d]Linear effect significant ($P < .05$).

[e]Quadratic effect significant ($P < .05$).

TABLE XI

CARCASS COMPOSITION OF GILTS[a]

Item	Protein level 8%	14%	20%	±SE	Energy level M	H	±SE
Uterine fluid							
Volume (ml)	2464	2212	1742[b]	±240	1980	2141	±190
Nitrogen (%)	8.2	9.0	9.7	± .3	8.8	9.0	± .2
Uterine							
Weight (Kg)	17.7	16.3	14.7[d]	±2.5	16.6	16.0	±2.0

Item	Energy level M Protein level 8%	14%	20%	H Protein level 8%	14%	20%	±SE
Uterine fluid							
Dry matter (%)[e]	3.2	2.9	3.0	2.9	3.3	3.5	± .2
Uterine							
Dry matter (%)[f]	15.0	13.1	13.3	11.7	16.0	13.4	±1.1
Nitrogen (%)[eg]	7.7	9.4	9.7	10.2	8.5	9.8	± .7

[a]Means
[b]Linear effect significant ($P < .05$).
[c]Linear effect significant ($P < .001$).
[d]Linear effect approached significance ($P < .10$).
[e]Significant ($P < .05$) Energy linear X Protein linear interaction.
[f]Significant ($P < .01$) Energy linear X Protein quadratic interaction.
[g]Significant ($P < .05$) Energy linear X Protein quadratic interaction.

TABLE XII

CARCASS COMPOSITION OF GILTS[a]

Item	Protein level				Energy level		
	8%	14%	20%	±SE	M	H	±SE
Carcass							
Weight (Kg)	109.2	115.3	113.7	±2.4	110.9	114.4	±1.9
Lean (Kg)	57.1	69.7	66.6[b]	±1.7	64.6	63.6	±1.4

[a]Mean

[b]Quadratic effect significant ($P < .001$).

TABLE XIII

CARCASS COMPOSITION OF GILTS[a]

	Energy level						
	M			H			
	Protein level			Protein level			
Item	8%	14%	20%	8%	14%	20%	±SE
Liver N (g)[bc]	47.2	49.1	53.2	43.8	51.1	63.7	2.8

	Protein level				Energy level		
Item	8%	14%	20%	±SE	M	H	±SE
Semimembranosus muscle N (g)[d]	46.5	58.9	57.3[e]	3.4	56.1	51.8	2.6
Uterine Fluid N (g)[b]	5.9	6.1	5.5	.6	5.8	6.0	.5
Uterine N (g)[b]	174.9	177.6	170.1	14.1	170.2	179.0	10.9
Carcass Lean N (g)[d]	1425	1863	1768[f]	93	1694	1654	73

[a]Means

[b]Dry tissue basis.

[c]Significant ($P < .05$) Energy linear X Protein linear interaction.

[d]Dry ether extracted tissue basis.

[e]Quadratic effect approached significance ($P < .10$).

[f]Quadratic effect significant ($P < .05$).

TABLE XIV

TYPICAL ANALYSIS OF VARIANCE TABLE

Source	Degrees of Freedom	
Total	100	
Between animals	33	
Protein	2	
Linear		1
Quadratic		1
Energy	1	
Linear		1
Protein X Energy	2	
Protein Linear X Energy Linear		1
Protein Quadratic X Energy Linear		1
Animals (Protein X Energy)	28	
Within animals	67	
Gestation	2	
Linear		1
Quadratic		1
Protein X Gestation	4	
Protein Linear X Gestation Linear		1
Protein Quadratic X Gestation Linear		1
Protein Linear X Gestation Quadratic		1
Protein Quadratic X Gestation Quadratic		1
Energy X Gestation	2	
Energy Linear X Gestation Linear		1
Energy Linear X Gestation Quadratic		1
Error	59	

SELECTED BIBLIOGRAPHY

Albin, R. C. and D.C. Clanton. 1966. Factors contributing to the variation in urinary creatinine and creatinine-nitrogen ratios in beef cattle. J. Anim. Sci. 25:107.

Baker, D. H., D. E. Becker, A. H. Jensen and B. G. Harmon. 1970. Reproductive performance and progeny development in swine as influenced by protein restriction during various portions of gestation. J. Anim. Sci. 31:526.

Bergen, W. G. 1974. Protein synthesis in animal models. J. Anim Sci. 38:1079.

Borsook, H. and J. W. Dubnoff. 1947. The hydrolysis of phosphocreatine and the origin of urinary creatinine. J. Biol. Chem. 168:493

Butcher, J. E. and L. E. Harris. 1957. Creatinine as an index material for evaluating ruminant nutrition. J. Anim. Sci. 16:1020.

Calloway, D. H. 1975. Nitrogen balance of men with marginal intakes of protein and energy. J. Nutr. 105:914.

Calloway, D. H. and H. Spector. 1954. Nitrogen balance as related to caloric and protein intake in active young men. J. Clin. Nutr. 2:405.

Camara, A. A., K. D. Arn, A. Reimer, and L. H. Mewburgh. 1951. The twenty-four hourly endogenous creatinine clearance as a clinical measure of the functional state of the kidneys. J. Lab. Clin. Med. 37:743.

Chattaway, F. W., R. P. Hullin, and F. C. Odds. 1969. The variability of creatinine excretion in normal subjects, mental patients and pregnant women. Clin. Chim. Acta. 26:567.

Chetal, U., U. R. Mehra, K. Nath, and S. K. Ranjhan. 1975. On the variation of urinary creatinine in buffalo calves and the effect of dietary protein intake on urinary creatinine, creatinine-nitrogen ratio and creatinine coefficient. J. Agr. Sci. 84:1.

Clawson, A. J., H. L. Rickards, G. Matrone, and R. E. Barrick. 1963. Influence of level of total nutrient and protein intake on reproductive performance in swine. J. Anim. Sci. 22:662.

Degeeter, M. J., V. M. Hays, D. D. Kratzer, and G. L. Cromwell. 1972. Reproductive performance of gilts fed diets low in protein during gestation and lactation. J. Anim. Sci. 35:772.

DeGroot, T. and J. H. Aafjes. 1960. On the constancy of creatinine excretion in the urine of the dairy cow. Brit. Vet. J. 116:409.

Duncan, D. L. and G. A. Lodge. 1960. Diet in relation to reproduction and the viability of the young. Part III. Pigs. Technical Communication No. 21. Commonwealth Bureau of Animal Nutrition, Aberdeen, Scotland.

Elsley, F. W. H., R. M. MacPherson, and I. McDonald. 1968. The influence of intake of dietary energy in pregnancy and lactation upon sow productivity. J. Agric. Sci. 71:215.

Elsley, F. W. H., D. M. Anderson, I. McDonald, R. M. MacPherson, and R. Smart. 1966. A comparison of the liveweight changes, nitrogen retention and carcass composition of pregnant and non-pregnant gilts. Anim. Prod. 8:391.

Erb, R. E., S. A. Tillson, G. D. Hodgen, and E. D. Plotka. 1970. Urinary creatinine as an index compound for estimating rate of excretion of steriods in the domestic sow. J. Anim. Sci. 30:79.

Fisher, H. 1965. Variations in the urinary creatinine excretion of rats fed diets with different protein and amino acid content. J. Nutr. 85:181.

Folin, O. 1905. Laws governing the chemical composition of urine. Am. J. Physiol. 13:66.

Frobish, L. T. 1970. Effect of energy intake on reproductive performance and estrous synchronization of gilts. J. Anim. Sci. 31:486.

Frobish, L. T., V. C. Speer and V. W. Hays. 1966. Effect of protein and energy intake on reproductive performance in swine. J. Anim. Sci. 25:729.

Guada, J. A., J. J. Robinson, and C. Fraser. 1975. The effect of dietary energy concentration on protein utilization during late pregnancy in ewes. J. Agr. Sci. 85:175.

Hawk, P. B., B. L. Oser, and W. H. Summerson. 1954. In Practical Physiological Chemistry. (13th Ed.). Blakiston Co. Inc. New York, New York.

Hawton, J. D. and R. J. Meade. 1971. Influence of quantity and quality of protein fed the gravid female on reproductive performance and development of offspring in swine. J. Anim. Sci. 32:88.

Hesby, J. H., J. H. Conrad, M. P. Plumlee, and R. B. Harrington. 1971. Nitrogen balance and serum protein response of gestating swine fed opaque-2 corn, normal corn and corn-soybean diets. J. Anim. Sci. 31:481.

Hodgen, G. D., R. E. Erb, and E. D. Plotka. 1967. Estimating creatinine excretion in sheep. J. Anim. Sci. 26:586.

Holden, P. J., E. W. Lucas, V. C. Speer and V. W. Hays. 1968. Effect of protein level during pregnancy and lactation on reproductive performance in swine. J. Anim. Sci. 27:1587.

Jones, R. D. 1975. Influence of protein intake and stage of gestation on growth, reproductive performance, nitrogen balance, and tissue levels of protein, DNA and RNA in gilts. Ph.D. thesis, Oklahoma State University, Stillwater, OK.

Jones, R. D. and C. V. Maxwell. 1974. Effect of protein level on growth, nitrogen balance and reproductive performance in gilts. J. Anim. Sci. 39:1067.

Jones, R. D. 1973. The effects of dietary protein levels on growth, nitrogen balance and reproductive performance in gilts. M.S. thesis, Oklahoma State University, Stillwater, OK.

Kerty, A. F., L. R. Prewitt, A. G. Lane, and J. R. Campbell. 1970. Effect of dietary protein intake on creatinine excretion and the creatinine-nitrogen ratio in bovine urine. J. Anim. Sci. 30:278.

Kline, R. D., L. L. Anderson, and R. M. Melampy. 1972. Nitrogen retention in pigs during the estrous cycle on pregnancy and after hysterectomy or ovarectomy. J. Anim. Sci. 35:585.

Kumar, I., D. G. Land, and A. W. Boyne. 1959. The determination of body composition of living animals. Brit. J. Nutr. 13:320.

Lee, M. and S. P. Lucia. 1963. Some relationships between caloric restriction and body weight in the rat. IV fecal nitrogen, fat, energy, and ash and urinary creatinine. J. Nutr. 81:117.

Lewis, J. S., M. L. Bunker, S. S. Getts, and R. Essine. 1975. Variability of creatinine excretion of normal, phenylketonuric and galatosemic children, and children treated with anit-convulsant drugs. Am. J. Clin. Nutr. 28:310.

Lofgreen, G. P. and W. N. Garrett. 1954. Creatinine excretion and specific gravity as related to the composition of the 9, 10, and 11th rib cut of hereford steers. J. Anim. Sci. 13:496.

Manchester, K. L. 1972. Effect of insulin on protein synthesis. Diabetes 21:447.

Manchester, K. L. 1970. Sites of hormonal regulation of protein metabolism. IN H. M. Munro (Ed.) Mammalian Protein Metabolism. Vol. IV. Academic Press, New York.

Miller, A. T. and C. S. Blyth. 1952. Estimation of lean body mass and body fat from basal oxygen consumption and creatinine excretion. J. Appl. Physiol. 5:73.

Miller, D. S. and P. R. Payne. 1964. Dietary factors influencing nitrogen balance. Proc. Nutr. Soc. 23:11.

Miller, D. S. and P. R. Payne. 1963. The theory of protein metabolism. J. Theoret. Biol. 5:398.

Miller, D. S. and P. R. Payne. 1961. Problems in the prediction of protein values of diets; caloric restriction. J. Nutr. 75:225.

Miller, G. M., D. E. Becker, A. H. Jensen, B. G. Harmon, and H. W. Norton. 1969. Effect of protein intake on nitrogen retention by swine during late pregnancy. J. Anim. Sci. 28:204.

Munro, H. N. 1964. General aspects of the regulation of protein metabolism by diet and by hormones. In H. N. Munro (Ed.). Mammalian Protein Metabolism. Vol. I. Academic Press, New York.

Munro, H. N. and D. J. Naismith. 1953. The influence of energy intake on protein metabolism. Biochem. J. 54:191.

O'Bannon, R. H., H. D. Wallace, A. C. Warnick and G. E. Combs. 1966. Influence of energy intake on reproductive performance of gilts. J. Anim. Sci. 25:706.

Pike, I. H. 1970. The effect on nitrogen utilization in the sow of variation in dietary protein concentration and pattern of feeding in pregnancy. J. Agr. Sci. 74:209.

Plotka, E. D. and R. E. Erb. 1969. Identification and excretion of estrogen in urine during the estrous cycle of the ewe. J. Anim. Sci. 29:934.

Pond, W. G. 1973. Influence of maternal protein and energy nutrition during gestation on progeny performance in swine. J. Anim. Sci. 36:175.

Pond, W. G., W. C. Wagner, J. A. Dunn, and E. F. Walker, Jr. 1968. Reproduction and early postnatal growth of progeny in swine fed a protein-free diet during gestation. J. Nutr. 94:309.

Rippel, R. H. 1967. Protein and amino acid nutrition of gravid swine. J. Anim. Sci. 26:526.

Rippel, R. H., O. G. Rasmussen, A. H. Jensen, H. W. Norton, and D. E. Becker. 1965a. Effect of level and source of protein on reproductive performance of swine. J. Anim. Sci. 24:203.

Rippel, R. H., B. G. Harmon, A. H. Jensen, H. W. Norton and D. E. Backer. 1965b. Response of the gravid gilt to levels of protein as determined by nitrogen balance. J. Anim. Sci. 24:209.

Smith, O. W. 1942. Creatinine excretion in women: data collected in the course of urinalysis for female sex hormones. J. Clin. Endocrinol. 2:1.

Van Niekerk, B. D. H., J. T. Reid, A. Bensadoun, and O. L. Paladines. 1963a. Urinary creatinine as an index of body composition. J. Nutr. 79:463.

Van Niekerk, B. D. H., A. Bensadoun, O. L. Paladines, and J. T. Reid. 1963b. A study on some of the conditions affecting the rate of excretion and stability of creatinine in sheep urine. J. Nutr. 79:373.

Vestergaard, P. and R. Leverett. 1958. Constancy of urinary creatinine excretion. J. Lab. Clin. Med. 51:211.

West, E. S. and W. R. Todd. 1961. In Textbook of Biochemistry. (3rd Ed.). Macmillan, New York.

VITA

Gawain Merlin Willis

Candidate for the Degree of

Master of Science

Thesis: INFLUENCE OF PROTEIN INTAKE, ENERGY INTAKE AND STAGE OF GESTATION ON GROWTH, REPRODUCTIVE PERFORMANCE NITROGEN BALANCE, CREATININE EXCRETION AND CARCASS COMPOSITION OF THE GRAVID GILT

Major Field: Animal Science

Biographical:

Personal Data: Born in Altus, Oklahoma, Septmeber 9, 1951, the son of G. D. and Blanche Willis; married Karmen Marie Karr October 23, 1970.

Education: Graduated from Altus High School 1969. Received the Bachelor of Science Degree from Oklahoma State University in December, 1974, with a major in Animal Science. Completed requirements for Master of Science Degree at Oklahoma State University in July, 1978.

Professional Experience: Reared on a farm in southwestern Oklahoma; Veterinary Tech. 1970-1971; Graduate Assistant, Oklahoma State University 1975-1977.