EFFECTS OF VARYING WATER AND PROTEIN INTAKES ON SOME ASPECTS OF BODY METABOLISM IN SWINE REPRODUCTION

A Thesis

Presented to

The Faculty of Graduate Studies and Research

University of Manitoba

In Partial Fulfillment

of the Requirements for the Degree

Master of Science

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by

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June 1967

ACKNOWLEDGEMENTS

Thanks are extended for the support and guidance provided by Dr. S. C. Stothers of the Animal Science Department during the course of this investigation.

The assistance given by Dr. R. J. Parker in the statistical analyses and Mr. J. A. McKirdy in the chemical analyses of the data are also gratefully acknowledged.

The assistance and advice provided by Dr. N. E. Stanger in the veterinary care of the animals is much appreciated.

This project was made possible by financial grants from the National Research Council and the Manitoba Department of Agriculture and Conservation.

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ABSTRACT

EFFECTS OF VARYING WATER AND PROTEIN INTAKES ON SOME ASPECTS OF BODY METABOLISM IN SWINE REPRODUCTION

Two experiments were conducted during this investigation. In Experiment I, three trials were conducted to study the effect of water intake on nitrogen utilization. Water intakes were either 6, 8 or 12 litres per sow per day with gestation rations tested containing 10 - 19 percent protein. The majority of these studies were conducted in the last third of pregnancy and involved the use of the nitrogen balance technique. In addition to the nitrogen analyses, urine was analyzed for a number of organic compounds related to protein metabolism.

In Experiment II, nine sows were studied throughout gestation using the nitrogen balance technique and urine analyses as for Experiment I. During the first 54 days (average) of pregnancy (Period I), the sows were fed a barley based gestation ration with a nitrogen balance trial conducted at the end of this period. Subsequently, a nitrogen-free purified ration was fed up to the 90th day of pregnancy (Period II). After 18 days on this feed, each sow was put on a nitrogen balance trial. From the 90th to the 115th day of gestation (Period III), three semi-purified

rations were used. For one treatment, three sows were fed an equivalent of 80 g. of protein per day (maintenance ration), another three sows were fed the equivalent of 170 g. of protein per day (production ration), and a third group was fed 120 g. of protein per day attained by adding lysine, methionine and diammonium citrate to the maintenance ration.

The low (6 l./day) or high (12 l./day) water intake had no significant effect on the apparent protein digestibility coefficient, urinary nitrogen excretion, or nitrogen retention. However, as might be expected a high level of water intake significantly increased urinary volume and lowered specific gravity compared to the low water intake.

A high protein intake reduced urinary volume apparently because of a simultaneously higher nitrogen retention in the body. In other words, the sows increased apparent water retention when a higher protein level was fed. As might be expected, urinary nitrogen and urea fluctuated most consistantly with protein intake.

A test of urinary creatinine levels as affected by temperature and storage time showed that temperatures of 27°C and 38°C brought about a rapid loss of creatinine within 8 days time. The daily creatinine and creatine excretion tended to be constant regardless of treatment. The daily urinary ammonia values, except when diammonium citrate was fed, varied considerably and with little apparent relation

to treatment. The amount of daily allantoin excreted in urine tended to be correlated with protein intake, sows fed the high protein intake having the higher allantoin values in the urine.

No significant treatment differences in the total number of pigs born, the numbers of still born, pig birth weights or three week weights were detected in experiment II. Milk protein concentration in the first week fluctuated with the gestation protein intake of the sows but no significant differences were detected.

The addition of lysine, methionine and diammonium citrate to the maintenance ration for sows during the last 25 days of pregnancy had no significant effect on sow's weight gain, reproductive performance, apparent dry matter digestibility, apparent or true crude protein digestibility and nitrogen retention as compared with sows fed the maintenance ration.

In experiment II, the total serum protein and albumin levels tended to decrease gradually during gestation.

INTRODUCTION

Theoretically, once a body's metabolic nitrogen requirement is met, the amount of urinary nitrogen excreted should fluctuate consistently with protein or non-protein nitrogen intake. However, there is some evidence that water intake and consequently volume of urinary excretion can influence urinary nitrogen output in humans, dogs, and ruminants. As far as swine are concerned such research has not been done.

Recent research at the University of Manitoba using the nitrogen balance technique, has shown that satisfactory swine reproduction was obtained on lower daily intakes of crude protein than those generally recommended by the National Research Council. Since water was supplied ad libitum in this work and no precise consumption record kept, it was considered of interest in using the nitrogen balance technique to simultaneously study water and protein intakes and nitrogen excretion in gestation. Similarly, since no comprehensive gestation data on organic urinary components related to protein metabolism are available, it was opportune to investigate this area simultaneously.

With the concept that amino acids are basic constituents of protein, by providing the animal with an ideal balance of amino acids it is possible to obtain optimum growth and reproduction at the lowest level of dietary

protein. This kind of approach stimulated much research in monogastric animals and extensive work has been done in the field of swine nutrition. However, much of these data are concerned with the use of amino acid (s) as a supplement in the diet for young growing pigs. In reproductive studies some data on amino acid requirements of gravid gilts have been published recently. However, there has been no work published to date on the use of amino acids in the diet of pregnant sows. Theoretically, pregnant gilts require protein for maintenance, reproductive purposes and growth whereas pregnant sows should not need protein for growth.

Since lysine and methionine are usually the first two limiting amino acids and are commercially available, it was decided to initiate studies in this area by supplementing a daily maintenance level of protein with these two amino acids. With methionine considered to be involved in the formation of creatine and creatinine a special interest in urinary levels of these compounts was obvious. These amino acid additions were made in the last 25 days of pregnancy having been preceded by two periods of what might be considered first a storing then a depletion of body protein. Since fetal growth occurs primarily in the last third of pregnancy, the final treatment period could be considered a good stress on the body's protein requirements. In this work, further data were collected on the organic urinary components related to protein metabolism.

LITERATURE REVIEW

The Effect of Protein Intake on Reproductive Performance of Swine

Jesperson and Olsen (1939), Meyer (1940), and Olafsson (1950) concluded that there was a strong correlation between heavy birth weight and survival rate to ten weeks. However, evidence concerning the effects of nutrition, especially protein level, upon birth weight is very contradictory.

Many investigators (Davidson 1930; Terrill et al. 1953; Lenkeit, 1957; Duncan and Lodge, 1960; Boaz, 1962) have contended that the number of pigs born alive and their birth weight are not influenced by inadequate protein intakes. In order to be more exact on this matter recent workers have tried to define more precisely the protein requirements for pregnant gilts and sows. Devilliers et al. (1958) assumed a biological value of 60 for dietary protein and suggested that 200 g. of "true digestible" protein are needed for maintenance and growth of the gilt with an additional 50 and 95 g. daily of digestible protein required to meet the intrauterine and mammary tissue needs at the 80th and 100th day postcoitum respectively. Self et al. (1960) suggested that protein requirements for fetal production, growth and maintenance can be met by a daily

supply of about 300 g. of digestible protein. Clawson et al. (1963) demonstrated that as little as 0.14 kg. of soybean protein per gilt daily in gestation did not significantly affect reproductive performance of swine. In 1965, Milne showed that pregnant sows reproduced satisfactorily by feeding 170 g. crude protein per day. Recently, in the report of Rippel et al. ($1965_{a.\ b.}$) 0.09 kg. of protein daily during the last 45 days of pregnancy was sufficient in terms of the usual reproductive criteria. They suggested that a daily intake of 0.23 kg. crude protein from a high energy, corn-soybean meal diet was sufficient to meet the gilts essential amino acid and amino nitrogen needs during the last one-third of pregnancy.

Thus the essential amino acid and non-protein nitrogen needs of the gravid gilt in terms of crude protein are considerably less than the daily requirement of 0.41 kg. suggested by National Research Council (1964).

Only limited data support a high protein intake during gestation to obtain optimum pig survival and weights in the subsequent nursing period. Hanson et al. (1955) reported that sows fed 11 or 14 percent protein had similar parturition performance, but that the pigs resistance to a scours-type disease was greater if the sows had received the higher level of protein during gestation. Stevenson and Ellis (1957) observed a significantly higher survival rate

of pigs from gilts that had received 0.30 or 0.52 kg. protein daily during gestation, than from sows fed 0.26 kg. protein daily.

Protein Retention of Swine During Gestation

Since so many workers favor more critical evaluation of the dam's performance as an indicator of the success of protein intake for reproduction, nitrogen retention of the dam during pregnancy as well as birth weight, survival rate and litter size of the young can be considered as added criteria of the adequacy of protein intake for reproduction.

Basic to a consideration of nitrogen on protein requirements for gestation is a knowledge of maintenance requirements for protein, i.e. endogenous urinary nitrogen and metabolic fecal nitrogen. After thirty-one balance trials with six sows, Henning (1959) found that the mean urinary excretion of endogenous nitrogen was 17.5 mg. per kilogram of body weight daily and the relationship between endogenous urinary nitrogen and live weight in kilograms could be expressed as;

$$\text{Endogenous Urinary Nitrogen} = 0.07619 \quad W_{kg.}^{0.7253}$$

It was noted that endogenous fecal nitrogen was related to dry matter intake as well as body weight and could be expressed as;

$$\text{Endogenous Fecal Nitrogen} = 0.1355 \quad W_{kg.}^{0.5952}$$

Armstrong and Mitchell (1955) have tabulated metabolic fecal nitrogen values ranging from 0.63 to 2.00 g. per kg. dry matter consumed while 106 mg. urinary nitrogen per $W_{kg.}^{0.74}$ was the minimum endogenous urinary nitrogen output per day for the growing pigs.

Studies by Rippel et al. ($1965_{b.}$) showed that mean daily metabolic fecal nitrogen and endogenous urinary nitrogen excretion were, respectively, 1.37 and 6.5 g. for 1.82 kg. fed daily to pregnant gilts.

Reproduction in the pig as reviewed by Duncan and Lodge (1960) gave some attention to extrauterine storage of nitrogen. Many reports (Evans 1929; Mitchell 1931; Lenkeit et al. $1955_{a, b.}$) have shown that considerable nitrogen was retained for extrauterine storage in pregnant pigs. In addition to these experiments. Penzes (1961) analyzed the organs of pregnant and nonpregnant sows, and found increases in weight during pregnancy for many tissues. These tissues partially increased nitrogen storage, but, the nitrogen concentration still decreased because of the great hydration.

Lenkeit and Gutte (1955) and Lenkeit (1956) detected a considerable extrauterine nitrogen retention during one to two weeks prior to parturition. Retention of nitrogen was especially pronounced after the 95th to 100th day of gestation, with the amount retained closely related to the

quantity fed.

Their work also indicated that the nitrogen stored by the gravid gilt was extremely labile, as evidenced by a sizeable negative balance occurring after parturition.

Milne (1965) studied nitrogen retention for pregnant sows fed 420, 240 or 170 g. protein daily. He found that nitrogen retentions during gestation were significantly lower for animals receiving 240 g. and 170 g. of protein daily. In trial II, a marked increase in protein-retention occurred immediately post-breeding with a further significant rise later in gestation for animals receiving 170 g. protein daily.

Rippel et al. (1965_b) presented a more extensive evaluation of the total nitrogen requirement of the gravid gilt as determined by nitrogen balance. They illustrated that efficiency of nitrogen retention plateaued at approximately 12.5 percent protein on both the 85th and 105th days postcoitum. Gilts fed the 9, 12 and 15 percent protein diets showed superior ability to retain nitrogen at the later stage.

Amino Acid Studies and Amino Acid Imbalance

With the knowledge that amino acids are basic constituents of protein and that animals require a proper dietary balance of amino acids in addition to crude

protein requirements, extensive research has been done on dietary amino acid requirements, balance and imbalance.

In 1906, Willcock and Hopkins, the first workers in the area of amino acid balance, reported that a supplement of tryptophan prolonged the lives of mice fed a tryptophan-free diet consisting largely of zein. Within a short time, Osborne and Mendel (1914) demonstrated specific requirements for lysine and sulfur-containing amino acids.

Subsequently, there were numerous works which studied amino acid supplements in the diet for poultry as well as young growing pigs. As a general finding, the kind and amount of amino acid added as a supplement to the diet is related to the quality and quantity of the intact protein in the diet.

Soldevila and Meade (1964) worked with growing pigs using L - lysine and DL - methionine additions to barley-soybean meal diets. These data showed a highly significant increase in nitrogen retention due to lysine supplementation, but no effect due to methionine nor a lysine x methionine interaction. Therefore, they suggested that lysine was the first limiting amino acid in a 14 percent protein barley-soybean meal diet based on a 13.3 percent protein barley. The addition of DL-methionine alone or in combination with lysine did not bring about rates and efficiencies of gain superior to those obtained with the

addition of lysine alone. Apparently barley protein supplied enough methionine and cystine to meet the needs in this experiment.

For pregnant animals, only limited research has been done in this area; Rippel et al. (1965_b) used amino acid supplementation of intact protein fed to gravid gilts to study essential amino acids requirements. They demonstrated that 0.42 percent lysine, 0.37 percent isoleucine, and 0.28 percent methionine-cystine were required for maximum nitrogen retention, with the gilt's requirements for these amino acids strikingly similar to those of the finishing pig (Becker et al., 1963).

The effect of a dietary imbalance of amino acids has been observed by many investigators. Rose (1938) and Frazier et al. (1947) emphasized that the initial response to an imbalance of amino acids is a depression in food intake. It is also known that animals will consume more of a protein-free diet than of a diet completely deficient in one amino acid (Frazier et al., 1947; Greenstein and Winitz, 1961). Recently it was shown that the plasma amino acid pattern of animals fed on an amino-acid-deficient diet is characterized by a very low concentration of the amino acid which limits growth (Longenecker and Hause, 1959; Gray et al., 1960).

Water Intake Studies

While several workers (Lepkousky and Furuta, 1960; Crizek, 1959) have studied the effect of water addition to diets, a review of the literature indicated a dearth of information.

Studies on water intake effects on nitrogen balance by Kinishi and McCay (1960) working with dogs showed that a decrease in water intake resulted in lower nitrogen excretion and that an increase in water intake had the reverse effect.

Further studies by Bressani and Braham (1964) observed that the increased water intake of dogs resulted in lower retention of nitrogen at low or high levels of nitrogen intake, and in most cases daily excretion of urinary urea decreased when water intake increased. They concluded that in nitrogen balance work, water intake should remain as constant as possible to reduce the variability in nitrogen retention.

Blood Serum Protein Studies for Swine in Gestation

Friedell *et al.* (1951) and Miller *et al.* (1961) published data on serum protein levels and composition in swine noting that total serum protein showed little change as gestation progressed but tended to fall slightly. Simultaneously, small increases in serum albumin levels

occurred and corresponding proportional decreases in globulin fraction were observed. Conversely, Smith et al. (1959) and Reboud et al. (1963) working with humans observed a decrease in serum albumin simultaneously with an increase in the proportion of gamma and beta globulin fractions while total globulin levels fell.

Brooks et al. (1964) working with growing pigs found that lysine supplementation resulted in increased serum protein and serum albumin levels. Analyses of blood protein fractions indicated that serum albumin patterns might become useful as a measure of protein adequacy. However, according to Rippel et al. (1965_b), alterations in serum protein were apparently more dependent upon the stage of pregnancy than upon adequacy of amino acid intake. The fall in serum gamma globulin prior to parturition paralleled the fall in total serum protein. The relative concentrations of alpha and beta globulin increased as pregnancy progressed, but albumin levels tended to decrease.

Urinary Studies

Specific Gravity and pH

The specific gravity of urine normally varies between 1.015 and 1.025 but is subject to wide fluctuation under various conditions. It may fall to 1.003 or lower

as the result of copious water drinking and rise to 1.040 or higher because of hemoconcentration due to excessive perspiration. Certain diseases (diabetes, nephrosis and antidiuretic hormone pitressin deficiency) can also influence the specific gravity of the urine.

The kidneys may secrete urine with a pH value as low as 4.5 and as high as 8.2 under extreme conditions. The mean pH of the normal mixed 24-hour sample is about 6.0.

pH is dependent almost entirely upon the nature of the diet, being low on a high protein diet, which yields much sulfuric and phosphoric acids upon metabolism, and high on a high vegetable and fruit diet, which when metabolized yields a basic residue from oxidation of the potassium and sodium salts of organic acids present. (West and Todd, 1964).

Organic Urinary Components

Urea and Ammonia

Urea is the principal end product of protein (amino acid) metabolism in mammals in general, and in certain lower forms of life. In the human it usually represents 80 to 90 percent of the total urinary nitrogen. Usually, the proportion of nitrogen as urea increases as the total urinary nitrogen increases, and vice versa.

On very low protein diets urea may represent 60 percent or or less of the total nitrogen. The quantity of urea excreted, in general, is proportional to the total protein metabolism, whether this protein represents food protein or the protein of tissue undergoing catabolism.

Ammonia, normally, is the second most important nitrogenous substance of urine quantitatively. Ordinarily, 2.5 to 4.5 percent of the total urinary nitrogen is composed of ammonium salts. On the average this represents about 0.7 g. per day.

Since both urinary ammonia and urea are derived from the amino groups of the amino acids, for a given quantity of nitrogen excreted, an increase in the amount of the one leads to a decrease in the amount of the other. The quantity of ammonia in the urine may be enormously increased through hydrolysis of urea by bacteria in the bladder (cystitis) or other parts of the urinary tract. This baterial production of ammonia from urea in normal urine may take place if the samples are stored without preservative alkaline fermentation. (Munro and Allison, 1964; West and Todd, 1964).

Creatine and Creatinine

Folin (1905); Shaffer (1908); Myers and Fine (1913), and Hahn and Meyer (1923) from circumstantial evidence adduced that creatine was a waste product of

protein metabolism from muscle and was elimated as creatinine in the urine.

Lofgreen and Garret (1954), Miller and Blyth (1952), and Saffle et al. (1958) showed that the "creatinine coefficient" in steers, humans, and hogs respectively is related to the amount of muscle in the live animal.

After Folin's (1905) classic studies on protein metabolism, the concept that the creatinine excretion in adult animals and man was essentially constant and not subject to change as a result of dietary protein manipulation has risen continuously.

Paton (1905); Beard (1943); Chow et al. (1963) and Nakagawa et al. (1964), reported that creatinine excretion was just as variable as any other urinary component and dependent upon the type of protein and kinds of amino acids in the diet. The recent report by Hans (1965) showed that creatinine excretion was not constant and varied with protein intake and amino acid content. On the basis of these experiments, no pattern which would permit prediction of creatinine excretion as it related to the level of dietary nitrogen or of amino acids could be distinguished. The conclusion of Beard (1943) suggested that free amino-acids stimulate creatine synthesis, and subsequently creatinine excretion, more readily than protein. In a study of sheep Van Nickerk et al. (1963)

illustrated that the increased creatinine output in response to increased intake of protein was significant. The creatinine was found to be unstable and to decay rapidly at the normal pH (8.4 to 8.7) of sheep urine at temperatures from 15 to 39°C; however, no loss of creatinine occurred during a five-month period when urine was stored at its normal pH but at 4°C.

Allantoin

Allantoin is formed in the liver by the action of the enzyme uricase upon uric acid, and the quantity of the substance in urine varies with the amount or activity of this enzyme.

Allantoin occurs in varying amounts, in the urine of practically all mammals. In the case of swine conversion of purines to uric acid is minor so that pigs excrete allantoin in relatively large amounts. (West and Todd, 1964).

PROCEDURE

These experiments were conducted with sixteen and nine sows used in experiments I and II, respectively. All sows were crossbreds (44% Landrace, 20% Wessex, 13% Welsh with small quantities of the Minnesota No. 1, Berkshire, Tamworth and Yorkshire breeds) and bred to Yorkshire boars.

There were three trials conducted in experiment I. Trial I was conducted in the summer of 1965, and used four sows weighing between 163 to 170 kg. at the start of the trial. The stage of gestation at the start of the trial was approximately the 35th day postcoitum.

Nitrogen balance trials were conducted with two protein levels and two levels of water intake in a 2 x 2 factorial arrangement, with the exception that each sow was twice tested on the high and low levels of water intake. In the first period, a sow's water intake was changed from a low to a high level with the reverse of this in the second period and vice versa for the second sow in each treatment.

The low and high protein ration contained 12.7 percent and 14.6 percent protein with 2.27 kg. of either ration fed daily per sow. For the low and high water intakes, each sow was given 6 l. or 12 l. per day during

the nitrogen balance trials. A week prior to the nitrogen balance trial and between the periods, sows were provided with a water intake of 8 l. per sow daily to standardize body water relationships.

Trial II was conducted in the winter of 1965 and used eight sows weighing between 233 to 250 kg. with the stage of gestation at the start of the trial averaging the 85th day postcoitum. Nitrogen balance trials were conducted on both low and high protein levels; with each sow tested on low and high levels of protein intake.

The low and high protein rations contained approximately 11 percent and 19 percent protein with 2.27 kg. of either ration fed daily per sow. Each sow was provided with 8 l. of water daily. In this trial, one sow from each protein level farrowed at the beginning of the last nitrogen balance trial, therefore, their data were excluded.

Trial III was conducted in the early summer of 1966 with four sows weighing between 240 to 250 kg. and the start of the trial was at an average of 92 days postcoitum. However, one sow farrowed during the first nitrogen balance trial, and the data were excluded.

This trial was conducted almost the same as trial II with the only difference being that barley instead of corn was used as the primary energy and protein source

in the rations. The low and high protein rations contained approximately 10 percent and 18 percent protein.

In all trials, feed and water were mixed and fed twice daily, with half the total allotment fed each morning and late afternoon. Feed and water wastage was seldom a problem with the sows generally consuming the entire amount within 30 minutes.

All of the nitrogen balance trials were conducted in metabolism crates with room temperature between 27 and 29°C. Ration compositions for experiment I are presented in tables I and II.

Experiment II was conducted from May to October of 1966, with three separate periods of gestation considered. Nine sows with initial weights between 181 and 227 kg. were used as experimental animals.

In period I, nine sows were fed a basal ration containing approximately 13 percent protein. All of the sows were group-hand-fed 2.27 kg. per sow daily (once per day) with water ad libutum from the date of breeding to an average 43rd day of gestation. From 43 to 54 days, sows were individually fed 2.27 kg. of the same ration with 8 l. of water per sow daily. A nitrogen balance trial was conducted with each sow in the last 6 days.

In period II, the sows were fed a nitrogen-free purified diet from approximately the 54th day to 90th

TABLE I: COMPOSITION OF RATION FOR EXPERIMENT I, TRIAL I

Crude Protein Ingredient	12.7% Low Protein %	14.6% High Protein %
Barley	88.5	48.0
Oats	-	35.0
Soybean meal	-	3.5
Alfalfa meal	10.0	10.0
Meat meal	-	2.0
Limestone	1.0	1.0
Salt	0.5	0.5

Note: 1. Dry vitamin D_2 and vitamin B_{12} were added to the ration to supply 165 I.U. and 11 ug. per kg. respectively.

2. Approximately 10 kg. of either ration was premixed with vegetable oil and chromium oxide to give a final concentration of 0.5% vegetable oil and 0.5% chromium oxide.

day of pregnancy. In order to maintain a calorie intake similar to period II, each sow was fed 1.82 kg. of the purified ration with 8 l. water daily. A nitrogen balance trial was initiated at an average of 72 days of pregnancy.

In period III, at the 90th day of pregnancy until farrowing, the sows were allotted to three treatments based

TABLE II: COMPOSITION OF RATION FOR EXPERIMENT I, TRIALS II AND III

Crude Protein	Trial II		Trial III	
	11% Low Protein	19% High Protein	10% Low Protein	18% High Protein
Ingredient	%	%	%	%
Barley	86.75	67.00		
Corn	-	-	86.75	67.00
Soybean meal	-	20.00	-	20.00
Alfalfa meal	10.00	10.00	10.00	10.00
Bone meal	1.50	1.00	1.50	1.00
Limestone	0.25	0.50	0.25	0.50
Salt	0.50	0.50	0.50	0.50
Vegetable oil	0.50	0.50	0.50	0.50
Chromium oxide	0.50	0.50	0.50	0.50

Note: 1. Dry vitamin D_2 and vitamin B_{12} were added to the ration to supply 198 I.U. and 11 ug. per kg. respectively.

2. A commercial vitamin supplement containing riboflavin, calcium pantothenate, niacin, and choline chloride was added to meet the minimum N.R.C. standards for riboflavin, pantothenic acid, and niacin. Vitamin content of ration was calculated using values in N.R.C. nutrient requirements for swine (1964) and used as basis for addition.

on body weight and previous nitrogen balance trial data. In the first treatment (Treatment I) three sows were fed daily 2.27 kg. of a semi-purified maintenance ration supplying 80 g. of crude protein daily. The name maintenance ration refers to the fact that it supplied a maintenance level of crude protein based on Hennings (1959) formula. For the second treatment (Treatment II) another three sows were fed the maintenance ration but with one percent of the amino acids lysine and methionine (in the ratio 1.56:1) and one percent diammonium citrate (D.M.C.) as a source of non-protein nitrogen included. The treatment II ration contained a crude protein equivalent of 5.3 percent, and was fed at a level of 2.27 kg. per day supplying 120 gm. of crude protein daily.

Initially, in order to bring the total nitrogen intake of Treatment II similar to Treatment III, 2 percent amino acids (same ratio) and 2 percent D.M.C. were added to the maintenance ration. However, the sows refused to ingest this after one feeding. Subsequently, a series of rations (4% amino acids with maintenance, 3% amino acids and 1% D.M.C. with maintenance, and 1% amino acids and 3% D.M.C. with maintenance) were tested with each of the three sows. After one or two feedings all of them refused to ingest the highest amounts of amino acids and non-protein-nitrogen as included in the maintenance ration. However, the sows fairly readily

accepted treatment II as outlined so they remained on this ration for the duration of the trial.

The treatment III ration was fed at a level of 2.5 kg. daily. This ration was also referred to as a maintenance and production ration since the level of protein, 170 g. per sow per day, was based on Milne's calculation (1965) of the gestation protein requirement. The nitrogen balance trials were conducted from the 103rd to 107th day of pregnancy in all these treatments. In experiment II, the feeding procedures were the same as for experiment I with all individually fed sows provided with 8 l. of water daily. After farrowing, sows were fed a standard lactation ration approximating the N.R.C. requirements. The ration and water were provided ad libutum.

The ration compositions for experiment II are presented in Table III, IV, and V.

Digestibility and Nitrogen Balance Studies

Digestibility and nitrogen balance studies were conducted in both experiments I and II. For four days prior to and during the nitrogen balance trial sows were fed the ration containing 0.5% chromium oxide. After two to three days sows were placed in the metabolism crates and urinary catheters were inserted (Cunningham 1955; Milne 1965). A one or two day adjustment period was allowed, then feces and urine were

TABLE III: COMPOSITION OF RATIONS FOR EXPERIMENT II, PERIODS I AND II

	13% Period I Basal Ration[a]		0 Period II, Nitrogen-Free-Purified Ration
Ingredient	%	Ingredient	%
Barley	81.0	Cellulose	2.0
Alfalfa	10.0	Corn Starch	93.4
Soybean Meal	6.0	Defluorinated Rock Phosphate	2.5
Defluorinated Rock Phosphate	0.5	Salt	0.5
Limestone	0.5	Vitamins[b]	0.1
Salt	0.5	Corn Oil	1.0
Corn Oil	1.0	Chromium Oxide	0.5
Chromium Oxide	0.5		

a. Dry vitamin D_2 and Vitamin B_{12} were added to basal ration to supply 198 I.U. and 11 ug. per kg. respectively

b. Vitamin mix according to Rippel et al. (1965_b).

collected separately for 3 and 4 consecutive days, respectively.

Excreta Studies

The daily volume of urine was recorded. Approximately 10 ml. of fresh urine were collected daily for total nitrogen determination by macro-kjeldahl technique. Three percent of

TABLE IV: COMPOSITION OF RATION FOR EXPERIMENT II, PERIOD III

Ingredient	Treatment I %	Treatment II %	Treatment III %
Barley	22.0	22.0	48.7
Dehydrated Alfalfa Meal	5.0	5.0	5.0
Corn Starch	68.7	66.7	42.2
Lysine & Methionine[a]	-	1.0	-
Diammonium Citrate	-	1.0	-
Defluorinated Rock Phosphate	2.2	2.2	2.0
Salt	0.5	0.5	0.5
Vitamin[b]	0.1	0.1	0.1
Corn Oil	1.0	1.0	1.0
Chromium Oxide	0.5	0.5	0.5
Daily Protein Intake, g.	80	120	170
Daily Energy Intake, cal.	8,210.6	8,208.3	8,320.0
Lysine[c]	.17%	.77%	.30%
Methionine & Cysteine[c]	.11%	.50%	.23%

a. Lysine and Methionine Ratio 1.56:1.

b. Vitamin mix according to Rippel et al. (1965)[b].

c. Calculated value.

the daily urine output of each sow was pooled and kept in the deep-freeze (approximately -22°C) until determination of various urinary components, namely, creatine, creatinine, urea, ammonia and allantoin were conducted. Creatine and creatinine determination were made on all samples of both experiment I and II, but the stability test was only determined in experiment I, trial I, using the technique described by Richardson (1959). Peters and VanSlyke's (1961) method was followed for the determination of urea and ammonia and was used on samples of trial II and III of experiment I and all the samples of experiment II. Allantoin was only determined in experiment II according to the method of Young and Conway (1942).

TABLE V: COMPOSITION OF LACTATION RATION

Ingredient	Lactation Ration %
Barley	82.0
Soybean Meal	16.0
Defluorinated Rock Phosphate	0.5
Limestone	1.0
Salt	0.5

1. Dry vitamin A, D_2 and B_{12} were added to lactation ration to supply 1650 I.U., 264 I.U. and 11 ug. per Kg. respectively.

Approximately 100 g. of green (Cr_2O_3) feces were collected every 24 hrs. and stored in the deep-freeze. At the end of three day's collection, the 3 samples from each individual sow were pooled and homogenized. About 50 ml. of 10% hydrochloric acid were mixed with each homogenized sample, in order to prevent fecal ammonia evaporation while the samples were oven dried at 60°C for 72 hrs. Dry fecal samples were ground in a Wiley mill. However, in period II of experiment II (nitrogen-free-period) the sows consumed considerable hair which appeared in the feces. It was necessary to remove this hair by a pulverizing-screening-process (Size 9½ inch, 3 times for each sample). A chromic oxide determination was made on fecal samples of experiment I and II, with the method used as described by Dunsky and Hill (1952).

Blood Serum Studies

Blood serum was only studied in experiment II. At the end of each period, approximately 20 ml. were collected from each sow's tail-vein and allowed to coagulate for two to three hours at room temperature. The samples were then centrifuged for thirty minutes at 1,500 r.p.m. and the serum was pipetted off and stored in the refrigerator. For chemical analyses, serum protein was determined by the macro-kjldhal technique and electrophoretic patterns were obtained on each

sample using the Sphinco Model R. paper electropharesis system. The patterns obtained were assessed photometrically on a Sphinco analytrol.

Milk Studies

Milk studies were only conducted in experiment II. Of the nine sows in the experiment, one sow died at farrowing in treatment I and post-mortem examination showed a gastric ulcer, two hair balls in the stomach and bladder infection. Undoubtedly the two hair balls accumulated in the period when the nitrogen-free ration was fed. Another sow on treatment II was removed from this study at term because of bladder infection. Three days after farrowing, milk samples were collected from seven sows. Pure oxytocin pituitary (P.O.P.) was injected intravenously in the ear-vein to initiate milk let-down. The milk from the first three pairs of functional udder sections referred to as the front section were pooled. The milk samples from the remaining udder section referred to as the rear udder section were pooled. Two ml. of milk sample were used for the total nitrogen determination by the macro-kjeldhal method.

Statistical Analyses

Analysis of variance was done according to Steel and Torrie (1960) with Duncan's multiple-range test (1955) used to compare treatment differences.

RESULTS AND DISCUSSION

Body Weight Changes

Average body weights and gestation weight changes of experiments I and II are presented in Tables VI and VII respectively.

In experiment I, sows were weighed at the beginning and at the end of each trial. It is difficult to compare the body weight and weight changes among the three independent trials, because of so many variables such as the stage of gestation, the levels of protein fed, length of trials and the few sows used, with consequently more biological variability.

In experiment II, when the basal ration containing 12.7 percent crude protein was fed in period I, the sows gained weight. However, the sows lost weight in period II, when the nitrogen-free ration was provided. This body weight loss could be expected, because the sows had to catabolize their body protein to supply their physiological needs since no dietary protein was available.

In period III, the sows in treatment III had the highest weight gain of the three treatments and, although not significantly different, gained more than twice as much weight as the sows on the other two treatments. When the ration of treatment II was fed, the average weight gains were

TABLE VI: SOW BODY WEIGHTS AND BODY WEIGHT CHANGES IN EXPERIMENT I

	Trial I	Trial II	Trial III
No. of sow	4	8	3[b]
Initial Wt. kg.	166.8	242.8	247.3
Wt. Gain kg.	29.20	8.64	6.37
Average Daily Gain kg.	0.40 (73)[a]	0.48 (18)	0.34 (19)

a. The number of days from the start of the first nitrogen balance trial until the end of the last nitrogen-balance trial is used in calculation of average daily gain.

b. One sow farrowed during nitrogen balance trial, and was excluded from data.

TABLE VII: GESTATION BODY WEIGHT CHANGES IN EXPERIMENT II

Treatment	I	II	III
No. of Sows	3	3	3
Prior Weaning Wt. kg.	169.5 ± 13.8	162.1 ± 12.6	177.7 ± 28.8
Period I Gain kg.	41.1 ± 17.6	24.1 ± 9.5	27.5 ± 9.8
Period II Loss kg.	-15.1 ± 13.8	-3.6 ± 1.4	-5.0 ± 2.2
Period III Gain kg.	8.2 ± 3.3	8.5 ± 5.8	16.8 ± 2.0
Post Farrowing			
No. of Sow	2[a]	2[b]	3
Post Farrowing Body Wt. kg.	178.5 ± 8.1	169.0 ± 13.9	202.0 ± 5.5
Wt. Gain of Sows in Gestation kg.	9.0	6.9	24.3

a. One sow died of gastric ulcers at term and was excluded from data.

b. One sow had a urinary tract infection at term and was excluded from data.

only slightly higher than for ration of treatment I. Based on this small difference in body weight, the added amino acids and non-protein nitrogen in the ration of treatment II apparently did not improve weight gains as might be expected.

Excluding the weight gain due to the developing litter, the average gestation weight gain of the sows fed the ration of treatment III was 24.3 kg. in comparison to 9.0 and 6.9 kg. for the sows fed the rations of treatments I and II, respectively.

Lactation Performance of Sows

The lactation perfrrmance of the sows used in experiment II is given in Tables VIII, IX and X. It should be restated that all sows received a lactation ration which met N.R.C. requirements including a minimum of 15% protein.

For the young pigs, no significant difference in the average three weeks weight due to the sow's gestation treatment could be found. A portion of the difference in average three week weights could be related to the numbers of pigs nursed in the three week-period. It is interesting to note that the heaviest average 3 week body weight was obtained with pigs whose dam had received the lowest protein intake in the last 25 days of gestation.

Milk protein concentrations of the 6th or 7th day of lactation appeared to vary with the gestation protein intake

TABLE VIII: SOW BODY WEIGHT CHANGE AND FEED CONSUMPTION IN LACTATION

Treatment		I	II	III
No. of Sows		2	2	3
Ave. Daily Feed Consumption per sow	(kg.)	4.9±0	4.3±1.0	5.1±0.3
Post Farrowing Body Wt.	(kg.)	178.5±7.8	169.0±13.5	202.0±5.5
Weaning (3 weeks) Body Wt.	(kg.)	171.6±6.9	175.3±18.7	197.0±6.6
Weight change in 3 weeks	(kg.)	+6.9	-6.3	-5.0

TABLE IX: MEAN REPRODUCTIVE PERFORMANCE OF SOWS (EXPERIMENT II)

Treatment		I	II	III
No. of Sows		2	2	3
Ave. No. Born (Alive and dead)		9.0 (8.5-0.5)[a]	11.5 (10.5-1.0)[b]	8.7 (8.3-0.3)
Ave. Birth Wt. (Alive)	(kg.)	1.75±0.11	1.51±0.03	1.39±0.13
Pig No. & Ave. Birth Wt. (3 weeks)	(kg.)	17-4.95±0.19	20-3.95±0.19	23-4.49±0.86
Total Litter Wt. (3 weeks)	(kg.)	84.2	79.0	103.3

a. One sow died of gastric ulcers at term; 15 young were born dead and excluded from the data.

b. One sow had a urinary tract infection; only 3 of a litter of 13 were born alive and were excluded from the data.

of the sows but no significant differences were detected. The rear udder section secreted a milk with a higher protein

TABLE X: MILK PROTEIN CONCENTRATION[a] IN 6TH OR 7TH DAY OF LACTATION

Treatment	I	II	III
No. of Sows	2	2	3
Front Udder Section	5.13-0.97	6.19-0.56	6.46-0.17
Rear Udder Section	6.13-0.42	7.06-0.28	7.13-0.40

a. Percent Crude Protein per 2 ml. of milk sample.

concentration than the front udder section. Although not measured, there appeared to be a larger volume of milk secretion from the front udder section as compared to the rear portion.

Milne (1965) concluded that no significant differences in number of live pigs born, the number of still born or pigs birth weights could be detected when the dams were fed as low as 170 g. of protein daily in gestation. However, the lower level of protein in the gestation ration did have a significant lowering effect on the average weights of young and the total litter weight at the three week stage of lactation. The young of the sows which received the low protein ration in gestation received almost identical quantities of milk as the high protein treatment although due to a lower protein

content, significantly less milk protein at the one week stage of lactation was received.

Rippel (1965_a) reported that neither the level nor the source of protein fed in gestation influenced litter size, number of live pigs farrowed, birth weight of live pigs or their livability.

Digestibility Coefficients

Apparent dry matter and crude protein digestibility coefficients are presented in Tables XI and XII.

Dry Matter

The dry matter digestibility coefficients of trial I, in experiment I, for low and high water intakes are very similar and no significant difference among them was noted. In comparing the dry matter digestibility of the low and high protein treatments in trial I, experiment I, the dry matter digestibility of the low protein ration was significantly higher ($P < .01$) than the digestibility of the high protein ration.

In trials II and III of experiment I, in order to minimize individual digestibility differences, sows were tested on both low and high protein rations. The high protein rations of trials II and III had higher dry matter digestibilities than the low protein rations of trials II and

TABLE XI: APPARENT DRY MATTER AND CRUDE PROTEIN DIGESTIBILITY COEFFICIENTS (EXPERIMENT I, TRIALS I, II and III)

	No. Sows	Low Protein (12.7%)		High Protein (14.6%)	
		Low Water (6 l./day) %	High Water (12 l./day) %	Low Water (6 l./day) %	High Water (12 l./day) %
TRIAL I					
Dry Matter Digestibility	4	78.3** ±0.86	78.1** ±0.67	71.8±2.50	73.9±0.79
Crude Protein Digestibility	4	75.1±1.98	75.2±2.75	78.3±2.87	80.1±1.74

	No. Sows	Low Protein (11%)	High Protein (19%)
TRIAL II			
Dry Matter Digestibility	7	74.3±0.69	76.5±1.80
Crude Protein Digestibility	7	65.7±2.06	77.7** ±1.33

	No. Sows	Low Protein (10%)	No. Sows	High Protein (18%)
TRIAL III				
Dry Matter Digestibility	2	76.6±0.55	4	77.9±2.42
Crude Protein Digestibility	2	71.4±1.06	4	79.9±1.80

** ($P \leq 0.01$)

TABLE XII: APPARENT DRY MATTER AND CRUDE PROTEIN DIGESTIBILITY COEFFICIENTS (EXPERIMENT II)

Items	Period I Basal Ration	Period II N-Free	Period III Treatment I	Period III Treatment II	Period III Treatment III
No. of Sows	9	7[a]	3	3	3
Dry Matter Digestibility %	73.3±0.83	89.1±0.83	88.7*±0.63	84.8±0.69	82.3±1.38
Crude Protein Digestibility %	79.9±0.57	- - -	49.4 ±1.09	52.8±1.95	60.0±2.7

a. In period II (n-free treatment), two sows delayed excreting feces which contained chromium oxide. Their data are excluded in this table.

Note. The statistical differences are compared between Periods I and II, and between Treatments I, II and III.

* ($P<.05$)

III, but no significant differences were detected. These data of trials II and III are in contrast with those of trial I, but are in general agreement with data by Schneider (1947).

In experiment II, the dry matter digestibility of period I would be expected to be similar to that in trial I, experiment I, because the rations used were the same. The dry matter digestibilities of periods II and III were higher than for period I and the trials in experiment I, because of the highly digestible corn starch used in the rations of these two periods.

In period II, when the sows were fed a purified nitrogen-free ration containing 93.4 percent corn starch, as a general observation, the feces from these sows were markedly reduced in quantity as well as in moisture content. Furthermore, since two of the sows had an unusually low indicator (Cr_2O_3) recovery in the feces, giving dry matter digestibilities of only 34.8 percent and 62.2 percent, their data were excluded from the table. The reason for this may be partially explained by a very slow gastro-intestinal passage rate so that fecal material was not actually excreted but had to be manually removed from the rectum. In the subsequent period, the same problem did not arise with these sows or with any of the others.

Crude Protein

In trial I, the apparent crude protein digestibility

values on the high water intake are slightly higher than the values determined for the low water intake, but there is no significant difference.

In both experiments I and II, it will be noted that mean digestibility values for high protein rations are higher than the values for low protein rations, but only in trial II, experiment I, was there a significant difference ($P < .01$).

Similar results to these were shown by Milne (1965). Since these digestibilities are only apparent digestibility values, no allowances have been made for metabolic fecal nitrogen levels. If the latter are not considered, there will be a greater lowering of the apparent digestibility coefficients of the low protein rations. Milne (1965) noted the true crude protein digestibility values determined by Armstrong and Mitchell (1955) and Henning (1959) and consequently suggested that the apparent digestibility difference obtained in the two rations he tested was not real.

In these data a daily mean value of 3.82 g. of endogenous fecal nitrogen was obtained with the N-Free ration in period II, and was used as a correction factor to calculate the true digestibility for periods I and III, For comparative purposes, the endogenous fecal nitrogen estimations of Mitchell and of Henning were also used and these values are given in table XIII. As expected, true digestibility is always higher than apparent digestibility. The most marked

change is for the true digestibility value in treatment I, which could be expected since these sows received the lowest daily intake of protein. Between treatment differences in digestibility are considerably reduced when expressed as true digestibility rather than as apparent digestibility.

TABLE XIII: CORRECTED OR TRUE CRUDE PROTEIN DIGESTIBILITIES (Experiment II)

Periods	App. Digest. %	True Digest. %	Mitchell corr.[a] True Digest. %	Henning corr.[b] True Digest. %
Period I				
Digestibility	79.9	88.0	84.7	86.5
Period III Digestibility				
Treatment I	49.4	77.5	66.1	72.9
Treatment II	52.8	71.9	64.2	68.3
Treatment III	60.0	74.0	69.2	72.1

a. Mean of Mitchell's Endogenous Fecal Nitrogen = 0.1 g./100 g. food intake.

b. Henning estimated EFN = $0.1355\ W_{kg.}^{0.5952}$

Urine Studies

The results of urinary analyses are based on daily nitrogen excretion per sow and are presented in Tables XIV, XV, XVI and XVII.

Urinary Volume

It is obvious that urinary volume can be greatly affected by water intake. In turn, voluntary water intake is influenced by environmental temperature, exercise, mineral intake as well as other factors. In these experiments, except for the daily amounts of water or protein supplied, other environmental factors were relatively constant. In experiment I, trial I, due to the large difference in daily water supplied, there was a significant difference ($P \leq .01$) in urinary volume between the sows on the low and high water intake.

In the present study, an interesting result has been found where the protein intake has affected the urinary volume. Generally, a high protein intake reduces urinary volume, or, in other words, the sows increased apparent water retention when a higher protein level was fed. The data are presented in Table XVIII. Significant differences were only noted in experiment I, trial II ($P \leq .01$) and in experiment II when comparing periods I and II ($P \leq .01$). The latter comparison ignores the stage of gestation, but if it were considered it is reasonable to assume that more water would be retained later in gestation such as in period II.

As an explanation, the sows fed high levels of protein probably increased water retention in order to keep a constant plasma protein concentration in the blood or other tissue

TABLE XIV: MEAN OF URINARY COMPONENTS PER SOW PER DAY (EXPERIMENT I, TRIAL I)

	No. Sows	Low Protein (12.7%) Low Water (6 l. per day)	Low Protein (12.7%) High Water (12 l. per day)	High Protein (14.6%) Low Water (6 l. per day)	High Protein (14.6%) High Water (12 l. per day)
Total volume (l.) excretion	4	4.0 ±0.17	10.0** ±0.24	3.9 ±0.33	9.7** ±0.12
Specific Gravity	4	1.014** ±0.0005	1.005±0.0005	1.015** ±0.0002	1.007±0.0005
pH	4	8.84 ±0.25	8.85 ±0.10	8.70 ±0.26	8.85 ±0.21
Total Nitrogen excretion (g.)	4	25.9 ±2.13	26.1 ±1.83	28.1 ±2.23	28.8 ±0.74
Creatine N. (g.)	4	0.68 ±0.21	0.28±0.06	0.45±0.06	0.40 ±0.12
Creatinine N. (g.)	4	1.75 ±0.09	1.54±1.16	1.47±0.11	1.64 ±0.16

** ($P < .01$)

TABLE XV: MEAN OF URINARY COMPONENTS PER SOW PER DAY (EXPERIMENT I, TRIAL II)

	No. Sows	Low Protein (11%)	High Protein (19%)
Total Volume excretion (l.)	7	7.7** ±0.25	5.9 ±0.23
Specific Gravity	7	1.002±0.00003	1.006**±0.00008
pH	7	7.8 ±0.16	7.51 ±0.04
Total Nitrogen excretion (g.)	7	22.9 ±0.97	30.4** ±1.22
Urea N. (g.)	7	13.45 ±0.85	20.75** ±0.89
Ammonia N. (g.)	7	2.02*±0.54	0.60 ±0.11
Creatine N. (g.)	7	0.39 ±0.07	0.74* ±0.11
Creatinine N. (g.)	7	2.10 ±0.13	2.04 ±0.19

* ($P<.05$) ** ($P<.01$)

TABLE XVI: MEAN OF URINARY COMPONENTS PER SOW PER DAY (EXPERIMENT I, TRIAL III)

	No. Sows	Low Protein (10%)	No. Sows	High Protein (18%)
Total Volume excretion (l.)	2	5.5 ±0.64	4	5.4 ±0.22
Specific Gravity	2	1.007±0.11	4	1.011±0.001
pH	2	7.68 ±0.85	4	7.9 ±0.43
Total Nitrogen excretion (g.)	2	23.7 ±5.29	4	35.21 ±3.18
Urea N. (g.)	2	10.54 ±8.08	4	27.15 ±2.18
Ammonia N. (g.)	2	4.58 ±3.30	4	4.51 ±1.69
Creatine N.. (g.)	2	0.39 ±0.01	4	0.44 ±0.15
Creatinine N..(g.)	2	2.37 ±0.38	4	2.29 ±0.49

TABLE XVII: MEAN OF URINARY COMPONENTS PER SOW PER DAY
(EXPERIMENT II, PERIODS I, II and III)

Items	Period I Basal Ration	Period II N-Free Ration	Period III Treatment I	Period III Treatment II	Period III Treatment III
No. of Sows	9	9	3	3	3
Total Volume excretion (l.)	5.4 ±0.2	7.7** ±0.1	6.2 ±0.3	5.9 ±0.2	5.71 ±0.3
Specific Gravity	1.015** ±0.0007	1.006±0	1.006±0.001	1.007±0	1.007±0.001
pH	8.3** ±0.17	6.8 ±0.07	6.55 ±0.17	6.35 ±0.4	6.92 ±0.29
Total N. (g.)	27.94** ±1.4	5.66 ±0.2	8.52 ±1.61	13.44* ±0.97	15.62* ±0.75
Creatinine N. (g.)	1.96 ±0.2	2.04 ±0.1	1.62 ±0.17	1.52 ±0.05	2.05 ±0.01
Creatine N. (g.)	0.64** ±0.1	0.016±0.002	0.10 ±0.17	0.09 ±0.0	0.24 ±0.05
Urea N. (g.)	15.36** ±1.3	1.07 ±0.2	3.67 ±0.98	5.67 ±0.23	7.42 ±1.90
Ammonia N. (g.)	3.60** ±0.9	1.92 ±0.2	2.37 ±0.57	4.36 ±0.17	3.42 ±1.09
Allantoin N. (g.)	1.65** ±0.1	0.70 ±0.03	1.39 ±0.05	1.42 ±0.06	1.75* ±0.06
Undetermined N. Material (g.)	4.73	-0.086	-0.63	0.38	0.74
Undetermined N. / Total N. x 100	16.9%	1.5%	7.4%	2.8%	4.7%

Note: The statistical difference are compared between Periods I and II, and between Treatments I, II and III.
** ($P \leq .01$), * ($P \leq .05$).

TABLE XVIII: APPARENT DAILY WATER RETENTION PER SOW ON DIFFERENT PROTEIN INTAKES

	No. Sow	Low Protein	High Protein
Experiment I			
Trial I			
12 l. water intake/day	2	2.0 l.	2.3 l.
6 l. water intake/day	2	2.0 l.	2.9 l.
Trial II			
8 l. water intake/day	8	0.3 l.	2.1** l.
Trial III			
8 l. water intake/day	4	2.5 l.	2.6 l.
Experiment II (8 l. of water intake per day during all periods)			
Period I	9	-	2.6** l.
Period II	9	0.3 l.	-
Period III			
Treatment I	3	1.8 l.	-
Treatment III	3	-	2.3 l.

** ($P<0.01$)

after the ingestion and retention of extra protein from the high protein rations. In the first two trials of experiment I, significant differences in nitrogen retention were noted when the high protein ration was fed. In trial III, a similar tendency was noted but a lack of numbers probably precluded any significant difference.

Specific Gravity

In experiment I, trial I, the urinary specific gravity values 1.014 and 1.015 from the low water intake are significantly higher ($P < .01$) than the 1.005 and 1.007 values from the high water intake treatments. This can readily be explained since sows given a greater amount of water excrete a larger volume of urine, so that, the relative urine concentration falls, giving a lower specific gravity, and vice versa.

In comparing the two protein treatments of experiment I, trials I, II and III, the urinary specific gravities are generally higher on the high protein treatment than on the low protein treatment. However a significant difference was only detected in trial II ($P < .01$). Again, in experiment II, the urinary specific gravity values investigated showed that when sows were fed more protein, the urinary specific gravities were higher and vice versa. With sows on a high protein intake, there was a larger amount of urinary nitrogen but a smaller volume of urine excreted resulting in the elevated

specific gravities of urine. In part, the higher specific gravities of urine resulting when rations containing primarily natural ingredients are used as compared to the purified or semi-purified ration in experiment II, would be due to a higher mineral content in such rations. Subsequently, a greater amount of inorganic material could be excreted in the urine.

Urinary pH

Urinary pHs are very constant comparing the treatments within each trial. However, the values are higher in trial I than in both trials II and III for no apparent reason.

In experiment II, the pH value of period I is highly significantly different ($P < .01$) from the others. The reason is that sows in period I were fed a basal ration having a larger amount of mineral than sows fed the purified-ration in period II and the semi-purified-ration in period III, and the extra minerals would tend to give a more alkaline pH.

Nitrogen Excretion and Retention

The results of the nitrogen balances are expressed as daily nitrogen retention per sow and presented in Tables XIX, XX and in Figure I.

In experiment I, trial I, the low and high water intakes had no significant effect on nitrogen excretion or

nitrogen retention. These results disagree with the studies by Konishi and McCay (1960) and Ricardo and Braham (1964), where both groups worked with dogs and reported that a decrease in water intake resulted in lower nitrogen excretion with the reverse effect with an increasing water intake. They suggested that two mechanisms could explain the effects of high and low water intake on nitrogen balance. First, high intakes of water may have a flushing effect on the kidney and other tissues washing out nitrogen metabolites. Secondly, a high water intake may have some specific influence on the formation of urea consequently increasing urea excretion. With this disagreement, the differences in species, body function, composition of the ration and nitrogen intakes have to be considered. For example, in the work with dogs, two to five times as much nitrogen per kg. of body weight was fed daily to growing animals as was fed in these studies to gestating sows.

As might be expected, in all these trials of experiment I, urinary nitrogen excretion was greater with the high protein intake. However, a significant difference was only detected in trial II ($P<.01$). Opposite to this observation was the fact that nitrogen retention was greater when the higher protein-intakes were given. Significant differences in nitrogen retention were detected in trial I ($P<.05$) and trial II ($P<.01$). The small number of animals in trial III

TABLE XIX: MEAN OF NITROGEN BALANCE (EXPERIMENT I, TRIALS I, II AND III)

Dietary Protein	No. Sow	Apparent Digest. N.[a] g.	Urine g.	Retention g.	Efficiency of Retention %
Trial I					
Low Water Intake (6 l./day)					
High Protein Intake	4	41.58±3.06	28.09±2.23	13.49*±0.7	32.44
Low Protein Intake	4	34.79±2.62	25.94±2.13	8.85±1.44	25.44
High Water Intake (12 l./day)					
High Protein Intake	4	42.44±2.23	28.78±0.74	13.66±2.22	32.19
Low Protein Intake	4	34.83±2.97	26.06±1.83	8.77±1.44	25.18
Trial II					
High Protein Intake	7	52.67±0.89	30.42±1.22	22.25**±1.33	42.24
Low Protein Intake	7	25.96±0.81	22.92±0.97	2.98 ±1.45	11.48
Trial III					
High Protein Intake	2	50.81±1.73	35.21±1.59	15.60±2.17	30.70
Low Protein Intake	4	26.58±0.03	23.66±3.67	2.92±2.53	10.99

a. Total N. intake x Apparent Crude Protein Digestibility.

** ($P<.01$), * ($P<.05$).

ABLE XX: MEAN OF NITROGEN BALANCE (EXPERIMENT II, PERIODS I, II AND III)

eriod	No. Sow	Apparent Digestible N. g.	Urinary N. g.	Retention g.	Efficiency of Retention %
eriod I, Basal ration	9	34.99±0.27	27.88±1.47	7.11**±1.37	20.32
eriod II, N-Free ration	7[c]	3.82[b]±0.34	5.51[a]±0.19	-9.33±0.45	-
eriod III, Treatment I	3	6.42±0.03	8.52±0.64	-2.10±0.64	-32.71
Treatment II	3	10.07±0.30	13.44±0.75	-3.37±1.26	-33.47
Treatment III	3	16.75±0.52	15.62±1.61	1.13±1.9	6.75

. Endogenous urinary nitrogen.

. Metabolic fecal nitrogen.

. Two sows delayed deliver of feces containing chromium oxide; their data were excluded.

* ($P<0.01$)

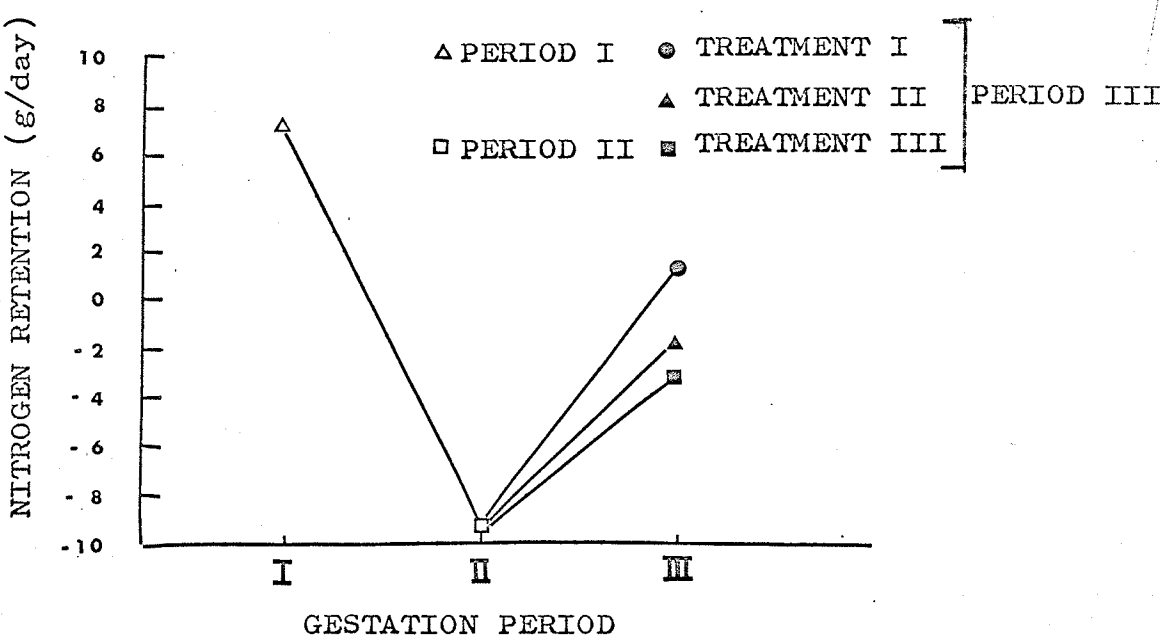

Figure 1. Nitrogen retention (experiment II).

decreased the possibility of any significant difference.

As expected in experiment II the urinary nitrogen excretion of period I was significantly higher ($P<.01$) than in the subsequent period II. Within the three treatments of period III, sows on both treatments II and III had significantly higher ($P<.05$) urinary nitrogen excretion than sows on treatment I. For nitrogen retention, sows were fed the basal ration during period I had the highest retention of all periods. When the nitrogen-free ration was fed in period II, sows had a negative nitrogen retention value. The latter value is the sum of endogenous urinary nitrogen and metabolic fecal nitrogen and its negative value is to be expected since no dietary nitrogen was supplied.

In period III, there are no significant differences in nitrogen retention between the three treatments. As might be expected, treatment I had a negative nitrogen retention since the ration contained only enough nitrogen for maintenance of the sow but not for gestation. However, treatment II, in which an extra 6.5 g. of nitrogen daily were supplied in comparison to treatment I, had a larger negative nitrogen retention than treatment I and this would not be expected. Apparently the lysine, methionine and diammonium citrate added to the ration of treatment I had no benefit for nitrogen retention in the present study. In explaining this, first of all the ration of treatment II contained 0.77 percent lysine and

0.50 percent sulphur amino acid. These levels are approximately two-fold higher than the requirements of 0.42 percent lysine and 0.28 percent methionine for gravid gilts suggested by Rippel (1965_c) so the extra lysine and methionine may have caused an amino acid imbalance. Soldevida and Meade (1964) working with growing pigs suggested that methionine was neither the first nor the second limiting amino acid in barley. Therefore, when methionine was added to the barley, it would proportionally increase the limiting amino acid imbalance. Theoretically, either taking methionine out or adding limiting amino acids, the imbalance should be decreased or eliminated. The sows tested may have been in amino acid imbalance when the amino acids and diammonium citrate were given at the 90th day of pregnancy to test their acceptability. This amino acid imbalance might have existed during the nitrogen balance studies, although this suggestion is less probable than the previous possibilities given.

The efficiency of nitrogen retention figure given in the table is a means by which nitrogen excretion and retention may be considered simultaneously. The values given demonstrate that for the higher protein intakes tested, a better efficiency of nitrogen retention was obtained. The lower levels of protein intake studied in experiment II, period III were not sufficient to maintain nitrogen balance in all the sows tested, although the subsequent lactation performance of these sows

was not adversely affected. It is reasonable to assume that at some greater protein intake, not tested here, the efficiency of nitrogen retention would be impaired, i.e. the extra nitrogen would simply be excreted.

Due to the range in protein intakes studied and the great number of nitrogen balance trials conducted, it was of interest to evaluate these collectively as well as in the individual experiments. Therefore, in the present study a total of 58 nitrogen retention values from experiments I and II, adjusted for metabolic size ($W_{kg.}^{0.75}$), are presented in table XXI and in figure II. Thirty-four of the nitrogen balance trials were conducted at 72 days postcoitum or later.

Regression analysis showed that nitrogen retention has a highly significantly linear effect with nitrogen intake. Regarding nitrogen retention efficiency, a daily nitrogen intake of 56.3 g. is the optimum level or when expressed as crude protein, 0.35 kg. (N x 6.25). Based on the data this level gave optimum efficiency of retention for the developing young and some tissue storage of protein in the sow.

Rippel et al. (1965_b) had the same response on nitrogen intake and retention, i.e. a linear relationship; however, a daily intake of 0.23 kg. crude protein was suggested as adequate for pregnant gilts. DeVilliers et al. (1958) assumed a biological value of 60 for dietary protein and suggested that 250 and 295 g. of digestible protein were required at the

TABLE XXI: MEAN OF NITROGEN RETENTION AND RETENTION EFFICIENCY ON DIFFERENT NITROGEN INTAKE DURING SOW'S GESTATION

	Daily Nitrogen Intake per Sow (g.)					
	0	13.39	28.22	37.23	39.50	46.80
Nitrogen[a] Retention (g.)	-0.181±0.02	-0.038±0.03	0.020±0.06	0.041±0.13	0.072±0.07	0.111±0.07
Efficiency[b] of Retention (%)	- - -	-.28379	.07087	0.11013	0.18228	0.24089
	47.44	49.71	51.53	56.30	63.56	67.87
Nitrogen[a] Retention (g.)	0.213±0.03	0.233±0.03	0.272±0.04	0.356±0.05	0.253±0.12	0.359±0.04
Efficiency[b] Retention (%)	0.44899	0.46872	0.52785	0.63233	0.39804	0.52895

a. Nitrogen Retention = $\frac{\text{Daily Nitrogen Retention per Sow (g.)}}{\text{B. } W_{\text{kg.}}^{0.75}}$

b. Efficiency of Retention = $\frac{\text{Daily Nitrogen Retention per sow/B.} W_{\text{kg.}}^{0.75}}{\text{Daily Nitrogen Intake per Sow}} \times 100$

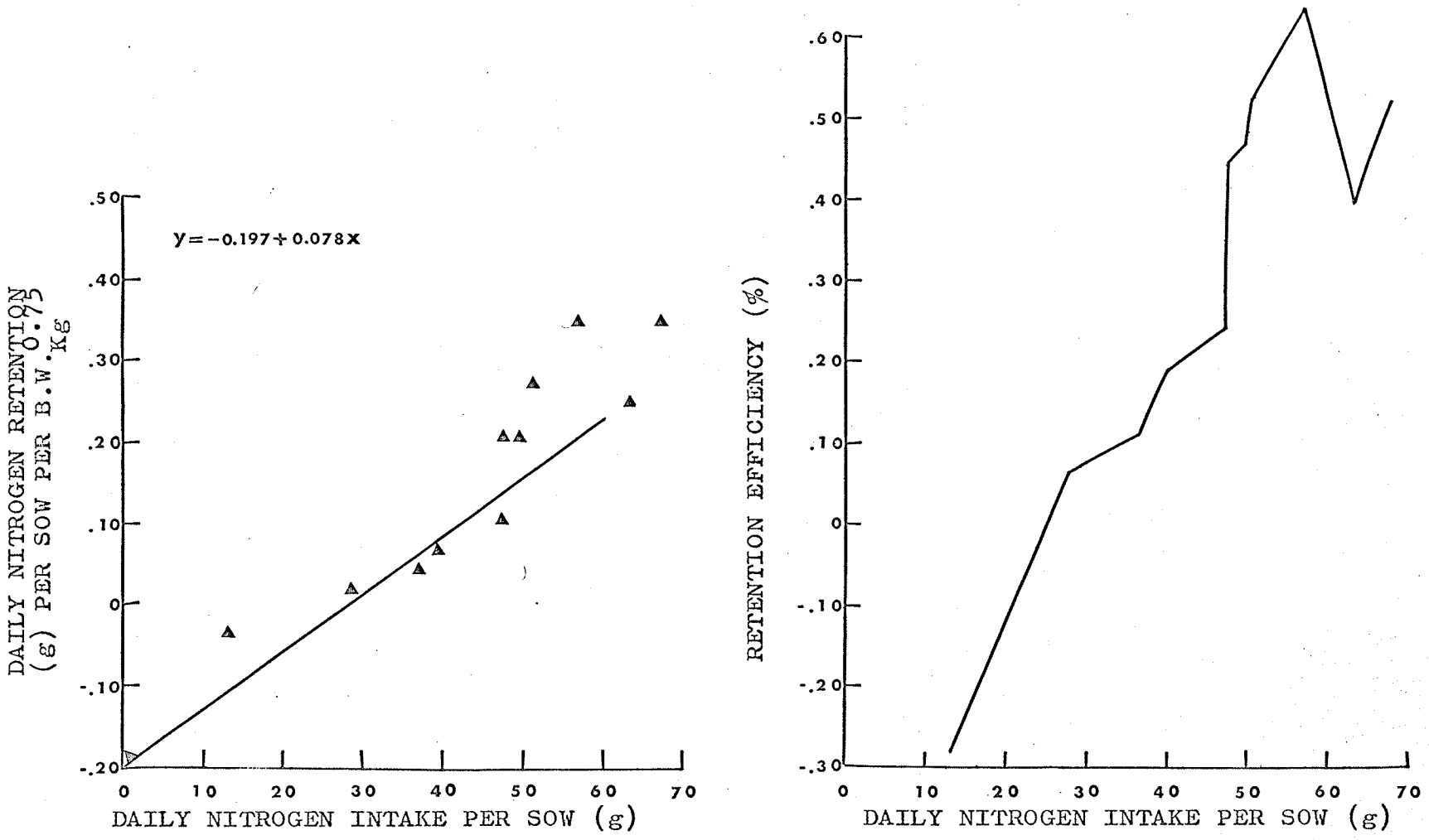

Figure 2. The response of nitrogen retention and retention efficiency to different nitrogen intakes in gestating swine.

80th and 100th days postcoitum, respectively. Self et al. (1960) suggested that protein requirements can be met by a daily supply of about 300 g. of digestible protein.

Urea and Ammonia

Urea and ammonia were determined for all the samples except for trial I of experiment I.

Urea is a primary end product of protein catabolism in mammals, and its excretion is closely related to the daily protein intake. This fact is clearly shown in both experiments I and II. In experiment II, period II when the sows were fed a nitrogen-free ration the smallest urea value of 1.9 g. is noted as compared with the largest urea values of 20.8 g. and 27.2 g. when the sows were provided ration containing approximately 19 percent protein.

It is interesting to note that next to total nitrogen excretion, urea excretion varied most directly with protein intake. In experiment II, period II, on the nitrogen-free ration urea made up approximately 19 percent of the total nitrogen excreted while on the high protein rations it made up 66 to 75 percent of the total nitrogen excreted. Daily urea excretion levels might be considered a good measure of exogenous catabolism.

Ammonia is one of the most variable urine components. Several factors such as the composition of the ration, temper-

ature of chemical determination, and pH of urine could have affected these results. In experiment II, treatment II during period III, the highest daily ammonia value of 4.36 g. is noted. This result was probably caused by adding diammonium citrate to the ration. This would infer that at the level fed, it was not used completely as a factor for protein synthesis.

Creatine and Creatinine Stability

As previously mentioned, because of the work with sheep urine of Van Nickerk et al. (1963) which showed that urinary creatinine was unstable under certain conditions, a similar study was carried out with pregnant sow urine. However, this study considered urinary creatine as well as creatinine. The results are presented in Tables XXII, XXIII and in Figure III.

The changes in urinary creatine at three different temperatures (4, 27 and 38°C) show no definite trend and in some cases vary greatly from the original value.

Regarding the creatinine changes, Figure III illustrates the percentage of creatinine remaining when stored at one of three different temperatures for a period of 13 days. The amount of urinary creatinine was markedly reduced where the urine samples were stored at 27 and 38°C; after 8 days of storage only 10 - 20 percent of the original creatinine

TABLE XXII: URINARY CREATINE CHANGE WITH STORAGE AT DIFFERENT TEMPERATURES

Temp.	Days of Storage				
	1st Day	2nd Day	4th Day	6th Day	13th Day
	ug./500 ul	ug./500 ul	ug./500 ul	ug./500 ul	ug./500 ul
4°C	2.54±2.4 (100%)	2.25±1.9 (88.58%)	2.66±2.2 (104.72%)	2.89±2.5 (113.79%)	1.96±1.5 (77.17%)
27°C	2.36±1.9 (92.91%)	2.18±1.3 (85.83%)	2.90±1.8 (114.17%)	3.48±2.1 (137.01%)	2.79±2.1 (109.84%)
38°C	2.58±1.9 (101.57%)	2.65±2.1 (104.33%)	5.14±4.1 (202.36%)	3.36±2.2 (132.29%)	2.61±2.1 (102.76%)

Experiment I, Trial I.

TABLE XXIII: URINARY CREATININE CHANGE WITH STORAGE AT DIFFERENT TEMPERATURES

Temp.	Days of Storage				
	1st Day	2nd Day	4th Day	6th Day	13th Day
	ug./500 ul	ug./500 ul	ug./500 ul	ug./500 ul	ug./500 ul
4°C	14.13±6.1 (100%)	11.43±6.1 (80.89%)	13.48±5.7 (95.40%)	11.84±7.0 (83.79%)	12.27±5.6 (86.84%)
27°C	12.63±5.3 (89.38%)	10.05±4.6 (71.13%)	6.50±5.2 (46.00%)	1.55±2.3 (10.97%)	0.77±0.9 (5.45%)
38°C	11.45±5.0 (81.03%)	7.45±4.8 (52.72%)	3.64±3.5 (25.76%)	0.93±0.4 (6.58%)	0.50±0.3 (3.54%)

Experiment I, Trial I.

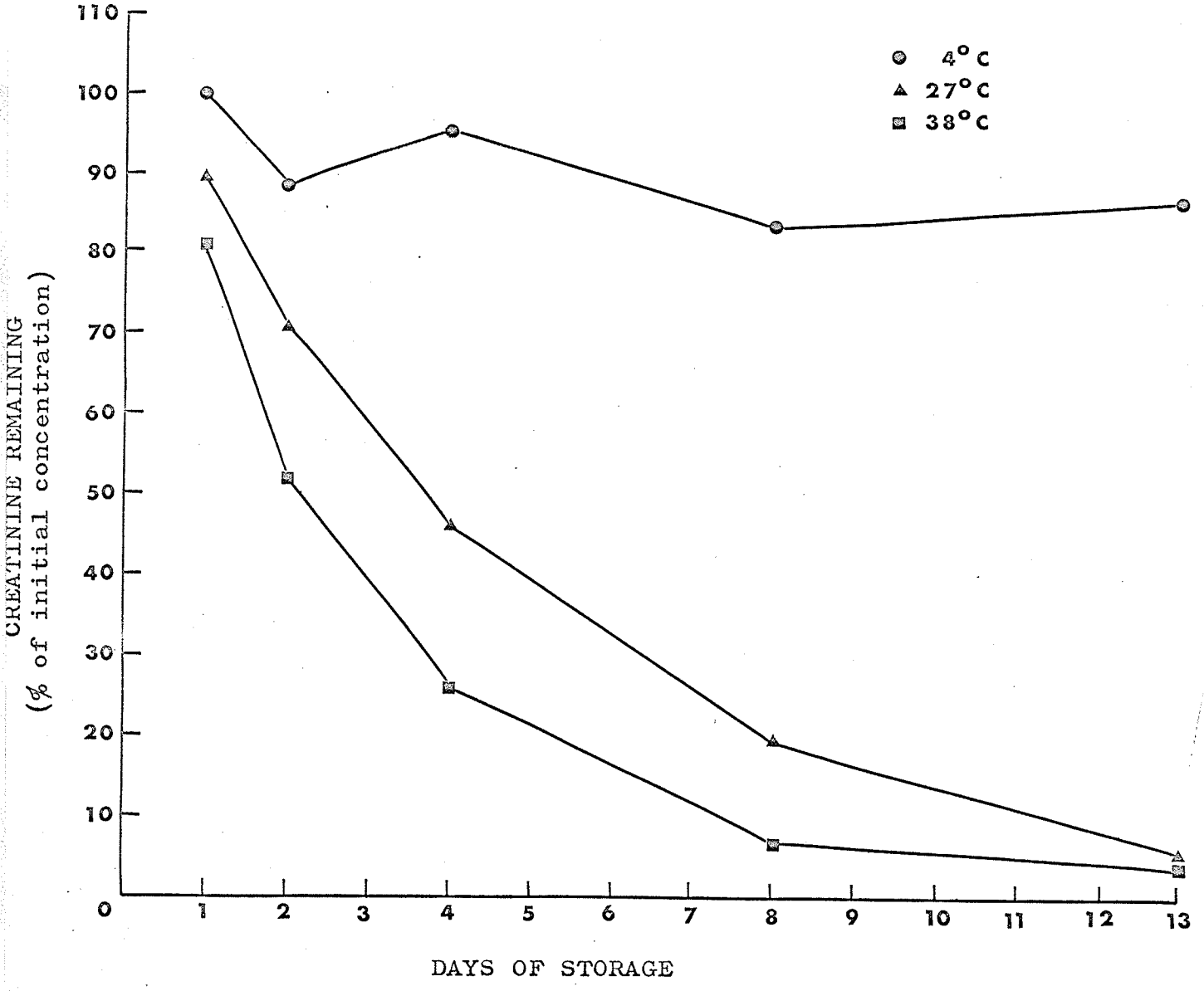

Figure 3. Percent of urinary creatinine retained with varying storage times and temperatures.

remained. The creatinine content was reduced approximately 15 percent when urine samples were stored at 4°C for 8 days.

In general, the creatinine data in this study support the results of Van Nickerk et al. (1963) although even at 4°C there appeared to be a loss of 10 - 15 percent in comparison to the original value. In subsequent nitrogen balances, all sows' urine was stored at 4°C.

Creatine and Creatinine

In experiment I, trial I, the data show that a low water intake gives a higher creatine excretion than a high water intake, but no significant difference was detected. The creatine value of 0.68 g. from the low water and low protein treatment is higher and more variable than the others and is not readily explained.

Urinary creatine is noted to vary consistently with protein intake in both experiments I and II. Generally a higher creatine value is observed on the high protein intake than with a low protein intake except for the value 0.68 g. previously mentioned. In experiment I, trial II, and in experiment II, comparing periods I and II, a significant difference is noted in favor of the high protein ration.

One reason could be that a high protein intake induced a higher creatine turn-over rate in the body tissue, consequently elevating urinary creatine excretion. Secondly, perhaps the high protein ration contains a larger amount of creatine, with this extra creatine in the diet elevating the

creatine intake and subsequently causing higher creatine excretion from urine.

Creatinine is one of the most constant components in urine with neither protein intake nor water intake affecting creatinine excretion in experiments I and II.

In creatinine excretion studies there are two points of view based on completely different theories. After Folin (1905), several research reports have demonstrated that creatinine is excreted constantly and is closely related to muscle tissue in the live animal (Lofgreen and Garret 1954; Miller and Blyth 1952; and Saffle et al. 1958). However, other workers have claimed that creatinine excretion was just as variable as any other urinary component depending upon the type and level of protein intake and the kinds and amounts of amino acids in the diet, (Paton 1905; Beard 1943; Chow et al. 1963; Nakagawa et al. 1964; and Hans 1965).

Based on biochemical reactions in the animal's body, the synthesis of creatine involves the amino acids glycine, arginine, and methionine. Most of the creatine in normal tissue of mammals (red cells excepted) occurs as the high-energy compound creatine phosphate and is subsequently excreted in the form of creatinine in the urine. Therefore, in experiment II, in treatment II of period III, with methionine added to the ration, higher values for both creatine and creatinine might be expected. However, the results in this

study show that the excretion of both creatine and creatinine was not affected with methionine added to the ration.

The results of creatinine coefficients are given in Table XXIV. Comparing the creatinine coefficients with those of Milne (1965), all of the values are at least twice as high in this study. However, these values are approximately one-half the value of 19.5 mg. per kg. based on a body weight of 67.6 kg. as reported by Smuts (1935).

In experiment II, period II the creatinine coefficient value might be expected to be higher than the value of period I and a slight increase was noted which could be interpreted to mean that some catabolism of lean tissue occurred. The body weight losses noted in this period substantiate this observation, but to be more specific body composition data would have to be obtained.

In period III, all of the values were lower than for period I and II similar to the observation of Milne (1965). He suggested that two factors which could combine to emphasize this fall in creatinine coefficient were the alternative utilization of amino acids and a possible edematous condition which occurs as parturition approaches. In the latter case an increasing water percentage in mammary tissue and in the uterus would bring about a decrease in the creatinine coefficient.

TABLE XXIV: CREATININE COEFFICIENTS IN EXPERIMENTS I AND II

Experiment I	Low Protein Intake	High Protein Intake
Trial I: Low Water Intake	10.50	8.82
High Water Intake	9.24	9.84
Trial II	8.65	8.40
Trial III	9.58	9.26

Experiment II				
Period I	9.77	Period III:	Treatment I	7.95
			Treatment II	7.95
Period II	10.58		Treatment III	9.45

Note: Creatinine Coefficients = $\frac{\text{Mean of the daily excretion (mg.)}}{\text{Mean of the body weight (kg.)}}$

Allantoin

Allantoin was only determined in experiment II. The amount of allantoin excreted in urine tends to be correlated with protein intake; sows fed the high protein intake have the higher allantoin values in the urine. A highly significant difference ($P<.01$) was detected comparing periods I and II. Within the treatments of period III, treatment III was significantly higher ($P<.05$) than the treatments I and II.

Morris and Ray (1939) working with ruminants suggested a positive correlation between the level of protein intake and the allantoin output in urine. This may possibly be due to the presence in the protein molecule of amino acids capable of being synthesized to purine compounds. The results in

this study generally support these data, except the value of 1.75 g. for treatment III in period III which was higher than the value of 1.65 g. for the highest protein intake of period I. However, as a partial explanation, it might be stated that these sows had a faster purine turn-over rate when sufficient protein intake was provided after the protein-depletion period.

Undetermined Nitrogen

In experiment II, undetermined nitrogen was estimated from the total nitrogen values minus the nitrogen values from the analysis of urinary components. In the present study, undetermined nitrogen would contain primarily uric acid, some purine compounds and small amounts of free amino acids.

For undetermined nitrogen, the two lowest protein intakes, namely, the nitrogen-free ration of period II and treatment I in period III, showed that some "extra" nitrogen was found when the individual analyses were summed. All of the other undetermined nitrogen values indicated that some nitrogen was not determined in the individual analyses. This undetermined nitrogen was greatest in the case of period I and can be partially explained by the fact that it was a natural diet rather than a purified or semi-purified diet. This assumes that the diet of period I, with two protein and non-protein nitrogen sources, could cause a greater variety of nitrogen compounds to be excreted and remain undetermined.

Endogenous Urinary Nitrogen and Metabolic Fecal Nitrogen

Since the composition of the nitrogen-free ration of experiment II, period II was identical with that used by Rippel et al. (1965_b) certain data from period II need further comparison with the Illinois' data.

Rippel et al. (1965_b) reported that the mean daily endogenous urinary nitrogen and metabolic fecal nitrogen excretions were 6.5 g. and 1.4 g. respectively. He noted that the endogenous urinary nitrogen value of 6.5 g. was higher than the expected value of 5.0 g. and 5.3 g. based on the formula by Brody (1945). However, the endogenous urinary nitrogen 5.5 g. of period II is very similar to the estimated value of Brody (1945) but lower than for Rippel's gilts (1965_b).

Regarding metabolic fecal nitrogen, Rippel (1965_b) considered his daily value of 1.4 g. lower than the value of 0.63 to 2.00 g. per kg. dry matter consumption based on Armstrong and Mitchell (1955). Although the metabolic fecal nitrogen value of 3.82 g. in this study is about three-times higher than Rippel's value 1.37 g. it is close to the estimated value of 3.0 g. based on Henning's formula (1959). In period II, the sows ingested some of their own hair during the nitrogen-free period and it was found to be very difficult to separate it from the feces, even though a pulverizing-screening method was used. This hair could have elevated the

metabolic fecal nitrogen value.

Blood Serum Studies

The total protein and the electrophoretic profile of blood serum were only determined in experiment II and are presented in Table XXV.

Regardless of the different protein intakes fed in the three periods, total serum protein decreased gradually during pregnancy. Rippel et al. ($1965_{b,c}$) and Milne (1965) reported that total serum protein decreased as gestation progressed. The reason may be that total blood volume increases during pregnancy, causing a decreased total serum protein concentration. Excluding the value of period I, the total serum protein is generally lower than the values ranging from 6.5 g. to 9.1 g. percent reported by Milne (1965), and a partial reason may be that these sows were depleted of body protein during period II. In period III comparing the total serum protein values within treatments, the treatments II and III were slightly higher than treatment I. Though no significant differences were detected, the greater the protein and non-protein nitrogen intake, the higher the serum protein values.

Again, the albumin levels are lower than the values ranging from 2.4 g. to 3.9 g. percent reported by Milne (1965) and may be explained for the same reason as for the total

serum protein. In all cases, albumin levels showed a tendency to fall as parturition approached. A significant difference was detected between period I and II ($P<.01$). In period III, the higher protein and non-protein nitrogen intakes gave the higher values for albumin. However, only treatment III was significantly greater than treatment I and II.

The total serum globulin levels obtained are very similar to values presented by Milne (1965) where it ranged from 3.3 g. to 5.0 g. percent. Beta and gamma globulins were slightly decreased during pregnancy, but no significant differences were detected. In period III, the alpha globulin value for treatment II was significantly lower ($P<.05$) than treatments I and II, though it is not readily explained.

Rippel et al. (1965_b) reported that the relative concentrations of gamma and alpha globulins increased linearly as the level of dietary protein was decreased, as a significant quadratic effect was noted for percent change of beta globulin when different protein intakes were fed. However, Foster et al. (1950) and Friedell et al. (1951) reported that variations in globulin concentration were more a reflection of gestation phase than of dietary variables in gravid swine.

TABLE XXV: SERUM PROTEIN AND ELECTROPHORETIC PROFILE OF SOWS DURING GESTATION (EXPERIMENT II, PERIODS I, II AND III)

Items	Period I	Period II	Period III Treatment I	Period III Treatment II	Period III Treatment III
No. of Sows	9	9	3	3	3
Total Serum Protein g./100 ml.	7.72±0.2	6.87±0.17	5.63±0.17	5.81±0.11	6.17±0.11
Albumin g./100 ml.	3.48** ±0.13	2.57±0.07	1.74±0.06	1.94±0.06	2.15* ±0.06
Globulin g./100 ml.			Alpha		
	1.37±0.03	1.31±0.07	1.44* ±0.06	1.18±0.06	1.44* ±0.006
			Beta		
	1.29±0.07	1.11±0.03	0.93±0.06	1.06±0.11	0.97±0.06
			Gamma		
	1.58±0.1	1.88±0.1	1.52±0.04	1.63±0.06	1.61±0.05

Note: The statistical differences are compared between Period I and II, and between treatment I, II and III.

** ($P < .01$), * ($P < .05$)

SUMMARY

In experiment I, three trials were conducted to study the effects of water intake on nitrogen metabolism in the gestating sow. The conclusions are as follows:

1) The low (6 l./day) and high water (12 l./day) intakes had no significant effect on nitrogen excretion, nitrogen retention, apparent dry matter or apparent protein digestibilities.

2) The high water intakes resulted in significantly greater urinary volumes ($P<.01$) and lower specific gravity ($P<.01$) than the low water intakes.

3) When urine samples were stored at 4°, 27° and 38°C for a period of 13 days the amount of urinary creatinine was very markedly reduced with storage time. After 8 day's storage the urine samples at 27° and 38°C had 10 - 20 percent of the original creatinine. Creatinine was reduced 15 percent for urine samples stored at 4°C for 8 days. However, creatine values at the same three temperatures and storage time showed no predictable trend and in some cases varied appreciably from the original value.

In experiment II, three periods were used to study the effect of varying levels of protein and non-protein nitrogen intakes on nitrogen metabolism, blood serum and reproductive performance. The results of this experiment were as follows:

4) There were no significant treatment differences ($P<.05$) in the total number of pigs born, the number of still born, pig birth weights or three week weights. Milk protein concentrations of the first week fluctuated with the gestation protein intake of the sows but no significant differences were detected.

5) The addition of one percent lysine and methionine (in the ratio 1.56:1) and one percent diammonium citrate to a barley-maintenance ration for sows from the 90th day of pregnancy until farrowing had no significant effects on sow weight gains, reproductive performance, apparent dry matter and apparent and true protein digestibilities or nitrogen retention.

6) In period II, sows fed a nitrogen-free ration had a mean daily endogenous urinary nitrogen value of 5.51 g. and a mean daily metabolic fecal nitrogen value of 3.82 g.

7) The amount of allantoin excreted in urine tends to be correlated with protein intake with sows fed the high protein intake having the higher allantoin values in the urine.

8) Although there was no significant difference detected, the total serum protein decreased gradually during gestation. Albumin showed the same trend as the total serum protein, with a significant difference between periods I and II ($P<.01$), and within the treatments of period III. ($P<.05$).

Considering experiments I and II jointly, some common results are as follows:

9) Regression analysis showed that nitrogen intake had a highly significant linear effect on nitrogen retention. For nitrogen retention efficiency, a daily nitrogen intake of 56.3 g. was the optimum level [expressed as crude protein - 0.35 kg. daily (N x 6.25)] for developing young and some tissue storage of protein in the pregnant sow.

10) A high protein intake reduced urinary volume apparently because of a simultaneously higher nitrogen retention in the body. In other words, the sows increased apparent water retention when a higher protein level was fed, but a significant difference was only detectd in experiment I, trial II and in experiment II when comparing periods I and II ($P \leq .01$).

11) As might be expected, urinary nitrogen and urea fluctuated most consistantly with protein intake. In these studies daily creatinine and creatine excretion tended to be constant regardless of treatment. The daily urinary ammonia values, except when diammonium citrate was fed, varied considerably and with little apparent relation to treatment.

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